FINAL OUTPUT JOURNAL

DairyUP

Unlocking potential

Unlocking the potential of milk, cow and water
to increase productivity and profitability,
de-risk the dairy industry and develop new markets.

A COLLABORATIVE DAIRY RD&E PROGRAM FOR NSW

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Who is Dairy UP

DairyUP
Unlocking potential

Dairy UP has developed a new and innovative model of collaborative dairy R&D, based on four pillars: **Co-investment, Collaboration, Co-delivery, and Rejuvenation.**

Who is Dairy UP?

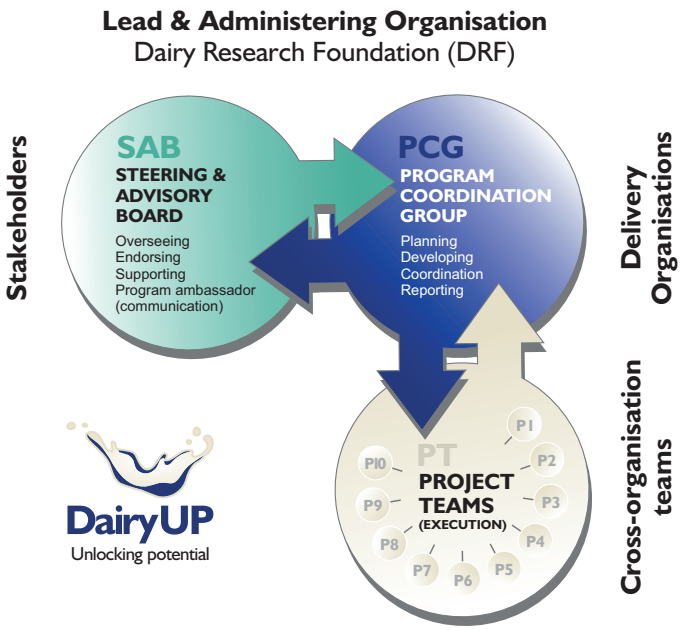
Dairy UP phase one was a collaborative program led by the University of Sydney's Dairy Research Foundation, and delivered through the New South Wales Department of Primary Industry, Scibus, Dairy Australia, and the University of Sydney.

Dairy UP phase one was funded through the NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash and in-kind contributions from the delivery partners and additional supporter organisations below.

The Program Coordination Group (PCG) had and executive role and was responsible for delivering the program and manging the budget. The key delivery organisations were the University of Sydney's Dairy Research Foundation, Scibus, NSW DPIRD, and Dairy Australia.

The projects were delivered by individual Project Teams (PT's), with the Steering & Advisory Board (SAB) overseeing the whole program and providing support to PCG and PT's.

Governance Model



Delivery organisations

The University of Sydney Dairy Research Foundation | NSW Department of Primary Industries and Regional Development | Scibus | Dairy Australia

Partner organisations

NSW Farmers Growing the best | eastAUSmilk | Dairy NSW / Dairy Australia | AFMH | Leppington Pastoral Company Pty Ltd | NSW Government Local Land Services | Norco

Additional program supporters, collaborations or partnerships

Charles Sturt University | DairyBio | DataGene | Eagle Direct | Entegra
Macquarie University | NSW EPA | smaXtec | UC Davis | University of Technology Sydney

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What is Dairy UP

What is Dairy UP?

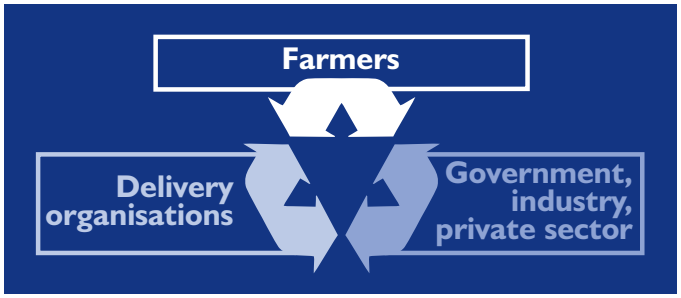
Dairy UP phase one was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aimed to realise three primary objectives:

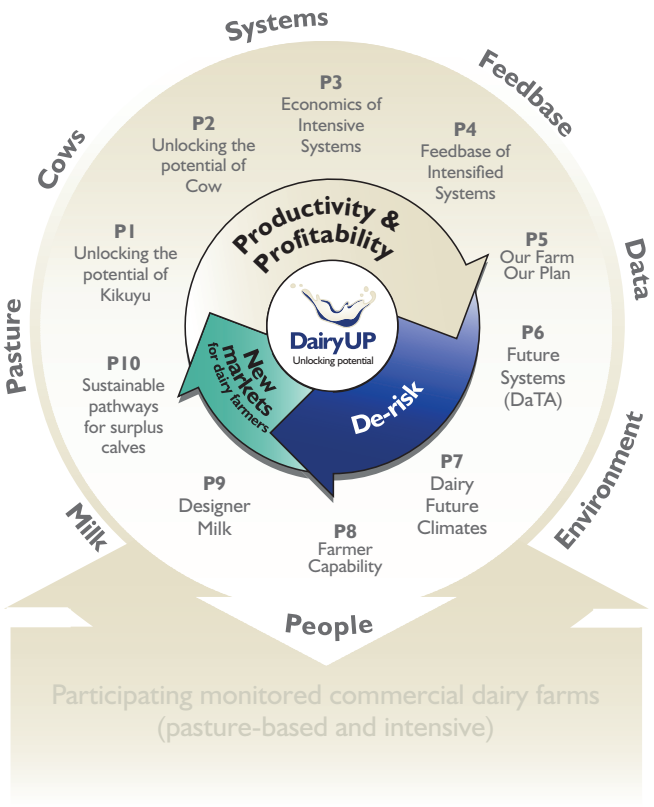
- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP phase one was a coordinated network of partner farms across New South Wales (and beyond) that provided valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP phase one made a significant contribution to the rejuvenation of dairy research and development, attracting new researchers, PhD students and Industry investment.

The program also attracted over $2.5M of additional income, including $1,098,280 in cash funding.





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Achievements report

DairyUP
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Building resilience in the NSW dairy industry

Dairy UP generated practical, farm-relevant evidence giving NSW dairy farmers greater confidence in decisions about their feedbase, cows, farm systems and future markets.

ACHIEVEMENTS REPORT

Largest dairy research project
- 10 world first projects
- 23 R&D sub-projects
- 12 PhD graduates
- 30+ farms monitored
- 70+ people involved
- 11 partner organisations

Innovation in pasture yields
Up to ~4t/ha/year potential additional kikuyu utilisation

Cutting carbon emissions
Comparable emissions intensity between pasture and intensive systems

Longer lasting, more productive cows
~80% lower culling risk - associated with omega-3 blood profiles in older cows

Advances in breeding
New early indicators for heat tolerance and milk composition

New insights from farm data
Earlier identification of underperforming calves using automated feeding systems

New market opportunities
New pathways for surplus calves and milk lactose

Dairy UP demonstrated the value of coordinated, industry-led research and development delivered by:

THE UNIVERSITY OF SYDNEY
Dairy Research Foundation

NSW GOVERNMENT
Department of Primary Industries and Regional Development

Scibus

Dairy Australia

Building resilience through evidence-based farm decisions

Dairy UP generated practical, farm-relevant evidence to help NSW dairy farmers improve pasture utilisation, cow performance and long-term productivity.

FEEDBASE

- Tested satellite-based advanced monitoring methods for kikuyu
- Evaluated new kikuyu varieties
- Refined management strategies to improve kikuyu pasture performance
- Generated independent NSW data on BMR maize yield and feed quality
- Discovered new insights about the links between plant stress, microbes and kikuyu toxicity.

Benefits to farmers

Greater confidence in:

- **Grazing and pasture management**
- **Choice of crop and pasture varieties**
- **Feed planning decisions**

COW PERFORMANCE

- Identified new markers for heat tolerance and milk composition
- Demonstrated that well-grown heifers can perform well when joined from 12 months
- Identified the main drivers of cows leaving the herd
- Discovered that blood omega-3 lipid profiles are associated with lower culling risk in older cows
- Identified early-life calf feeding and growth patterns linked to higher milk production and longer survival in the herd
- Developed new data-based reports for farmers.

DAIRY SYSTEMS

- Found similar emissions intensity across pasture-based and confinement systems, but different priorities for reducing emissions
- Identified different economic strengths and trade-offs across pasture-based and intensive systems
- Confirmed digital soil mapping could support farm-scale decision-making
- Investigated practical pathways for adapting dairy systems to future climates
- Demonstrated the value of structured whole-farm planning for business decision-making
- Identified viable market pathways for surplus calves and beef-on-dairy systems.

Benefits to farmers

Greater confidence in:

- **Breeding decisions**
- **Age of joining**
- **Disease prevention and management**
- **Feeding strategies**
- **Animal welfare**

Benefits to farmers

Greater confidence in:

- **System choices**
- **Climate adaptation**
- **Farm planning**
- **Resource management**
- **Surplus calf pathways**

DairyUP

Unlocking potential

Project 1:
Unlocking the potential of Kikuyu based pastures.

Project 2:
Unlocking the potential of cows.

Project 3:
The economics of intensive systems.

Project 4:
Feedbase of intensive systems (maize).

Project 5:
Our Farm, Our Plan

Project 6:
Future systems DaT.A..

Project 7:
Dairy future climates

Project 8:
Farmer capability.

Project 9:
Designer milk.

Project 10:
Sustainable pathways for surplus calves.

WHAT MADE DAIRY UP DIFFERENT?

- Whole-of-system research
- Commercial farm validation
- Industry-led collaboration
- Practical on-farm focus
- Integrated data and technology
- Multiple investors, delivery partners and collaborating organisations
- Capability building for future dairy R&D

Delivery organisations

The University of Sydney Dairy Research Foundation

Dairy Australia

Scibus

NSW Government Department of Primary Industries and Regional Development

Funding organisations

Australian Government

NSW Government

The University of Sydney Dairy Research Foundation

NSW Government Department of Primary Industries and Regional Development

Scibus

Dairy Australia

NSW Farmers Growing the best

eastAUSmilk Advocacy · Unity · Service

Dairy NSW

Dairy Australia

AFMH

Leppington Pastoral Company Pty Ltd

NSW Government Local Land Services

Norco Quality since 1895

Additional program supporters, collaborations or partnerships

CharlesSturtUniversity | DairyBio | DataGene | EagleDirect | Entegra
MacquarieUniversity | NSW EPA | smaXtec
UC Davis | University of Technology Sydney

Dairy UP is located at the Centre for Carbon, Water and Food (CCWF) on Camden Campus, School of Life and Environmental Sciences, Faculty of Veterinary Science, The University of Sydney.

www.dairyup.com.au

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FINAL OUTPUT JOURNAL

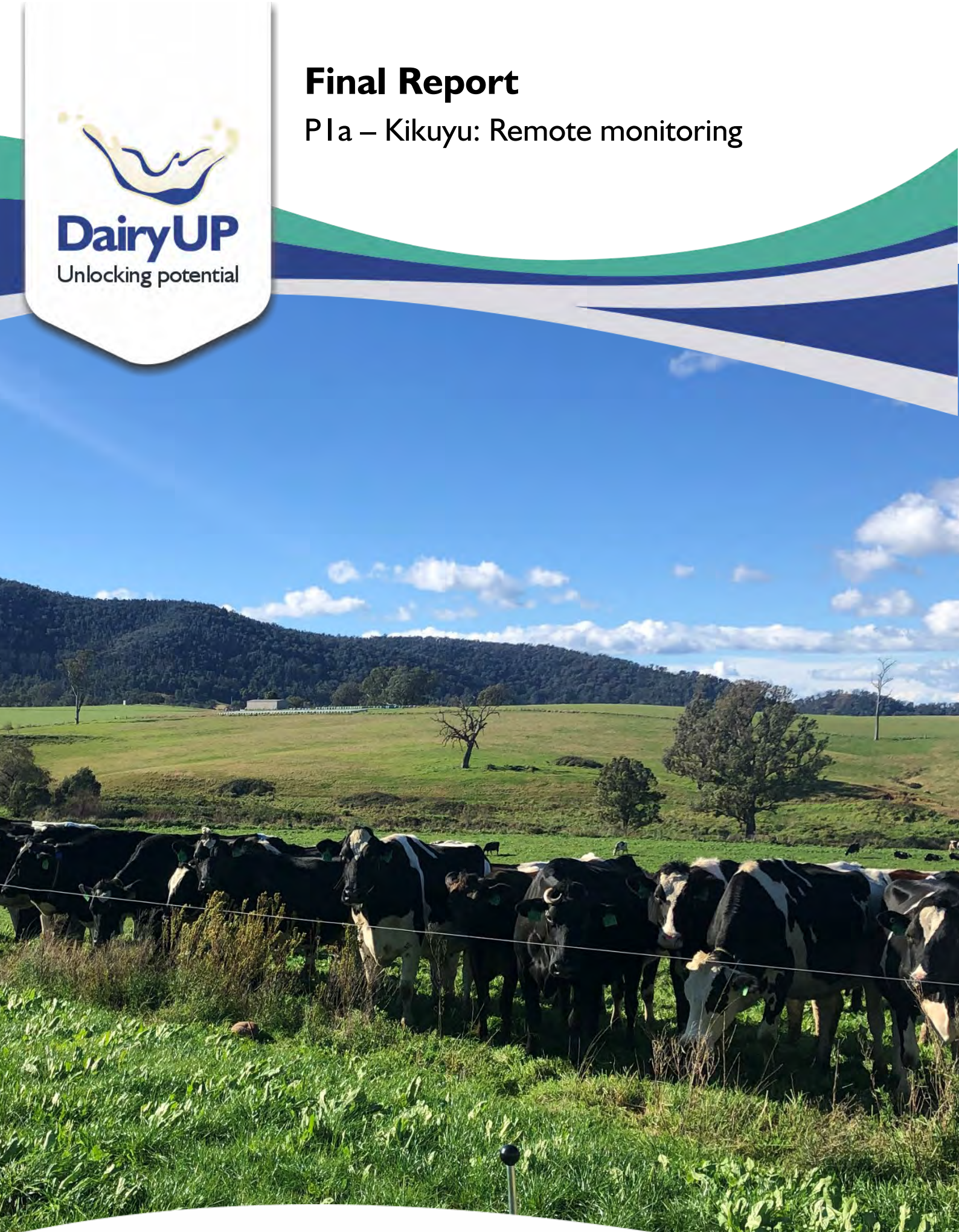
P1
Unlocking the potential of Kikuyu

P1a – Kikuyu: Remote monitoring
P1b – Kikuyu: Investigating toxicity
P1c – Kikuyu: Developing new varieties
P1d – Kikuyu: Carbon on NSW dairy farms
P1e – Kikuyu: Nutritional Value

DairyUP
Unlocking potential

Final Report

P1a – Kikuyu: Remote monitoring



THE UNIVERSITY OF SYDNEY
Dairy Research Foundation

This project was led by Dr. Martin Correa-Luna (calibration phase) and Dr. Tori Alexander (advanced monitoring phase to project completion) from the University of Sydney Dairy Research Foundation

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

Proudly funded by

Australian Government · NSW Government · Dairy Australia · Department of Primary Industries and Regional Development · Scibus · The University of Sydney – Dairy Research Foundation · nsw Growing the best · eastAUSmilk · Dairy Australia · AFMH · Leppington Pastoral Company · NSW Local Land Services · Norco

Delivery organisations

The University of Sydney – Dairy Research Foundation · NSW Department of Primary Industries and Regional Development · Scibus · Dairy Australia

Additional program supporters, collaborations or partnerships

Charles Sturt University | DairyBio | DataGene | Eagle Direct | Entegra | Macquarie University | NSW EPA | smaXtec | UC Davis | University of Technology Sydney

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1. Executive Summary

This report includes a synopsis of the P1a Dairy UP project "Kikuyu: Remote monitoring", offering a description of the sampling procedures and methodologies, key findings, implications and future considerations of the research, informed by results and farmer feedback.

The project's goal was to utilise a remote monitoring technology on NSW dairy farms to support pasture management through providing accurate and timely observations of pasture biomass, and to test and validate the reliability of the technology. Secondary to this, the project also conducted an exploration into further information that could be produced from the data generated by the remote monitoring technology which could aid decision making and provide a snapshot of pasture production and utilisation.

Farms participating in the project (n=16) were located across three distinct dairying regions of NSW, being the north coast, mid coast and south coast. Data were recorded over a two-year period, with farms being monitored by a commercially available remote monitoring satellite technology (RMST; Pasture.io) as well as on-ground monitoring with a calibrated rising plate meter (RPM). Other on-ground observations (e.g. farmer perception, farm management activities, pasture samples) were also made, with regular contact and dialogue between participating farmers and project officers.

The project confirmed the RMST could measure pasture biomass with comparable predictive precision to a calibrated RPM, although this level of accuracy was only achievable with fortnightly training of the RMST (involving measuring five representative paddocks with a RPM every fortnight; Correa-Luna, M., *et al*, 2024). The time required to measure the paddocks (approx. 1.5 h fortnightly) with a RPM was relatively low (particularly compared to the potential benefits), however is still perceived to be a barrier to adoption. A potential solution would be to develop an automated strategy to take on-ground pasture measurements for calibrating the technology, or alternately to improve the technology to a point where calibration is no longer required.

Through objectively monitoring pasture biomass using a RMST ("advanced monitoring"), the project identified that, on average, farms grew over 4 tonnes DM/ha more pasture than what they utilised through grazing-based activities (could also be perceived as "wastage"). Reducing this gap between pasture grown and pasture used through increasing pasture use would not only improve pasture utilisation but also have a potential large-scale positive impact in the business by more effectively utilising home-grown feed (pasture).

Further exploration of the data was able to quantify the relationship between pasture grown and pasture utilised. As expected, higher volumes of pasture grown were tied to higher pasture utilised. However, there was variation in pasture used between farms with the same or similar pasture grown. Some management factors were found to impact pasture utilisation (pasture use efficiency), being pre- and post-grazing biomass and grazing frequency (number of grazing events per year). Farm characteristics such as size and number of paddocks were not shown to be significant factors in pasture use.

Future research should focus on working with farmers, while monitoring their pasture through advanced monitoring techniques, to capture the steps/strategies and management decisions which lift pasture productivity and utilisation, the changes in management due to incorporating advanced monitoring techniques and the impacts on business performance (e.g. milk yield and, in turn, profitability).

Outside of the outcomes of the project, participating farmers received individualised reports outlining their farm's performance (including benchmarking with their region and all farms in the project) and were given the opportunity to provide feedback on their experience and value of the information generated. During data collection and following receipt of their individual farm report, farmers perceptions and confidence in the technology were observed. At the conclusion of the project, and after receiving their report, farmer confidence in the technology increased from 2.6 to 3.6, a rise of 38% (scored out of 5, with 1 being not confident and 5 being very confident). Commonly mentioned reasons why farmers felt information presented in the report was important included that it supported management decisions, offered reassurance and identified issues/areas of improvement.

Results illustrated how data from the trained RMST can aid in highlighting biomass and paddock variability, along with room for improvement in pasture production and utilisation. This work showed that satellite technology is a potential tool to assist farmers when making decisions related to pasture management, as it can not only monitor pasture in every paddock across the farm, but also provide information which can be used to quantify potential pasture losses, highlight inefficiencies and support decisions around productivity and profitability.

2. Project Overview

Item	Description
Project Title	Kikuyu: Remote monitoring
Funding Body	Dairy UP
Dairy UP Project	P1a
Project Duration	2022-2026
Lead Organisation	The University of Sydney
Project Lead	Dr. Martin Correa-Luna (calibration phase) Dr. Tori Alexander (advanced monitoring phase to project completion)
Research team	Prof. Sergio Garcia, Dr. Fernando Masia, Dr. Juan Gargiulo, Blessing Azubuike, Prof Cameron Clark, Peter Beale, Josh Hack, Chloe Wilson, Zac Geldof, Mikayla Woods, Kate Ireland
Key Collaborators	NSW Department of Primary Industries and Regional Development, NSW Local Land Services

3. Abbreviations

DM — Dry matter
GPS — Global positioning system
Ha — Hectare
Kg — kilogram
MAE — Mean absolute error
NSW — New South Wales
RPM — Rising plate meter
RMSE — Root-mean-square error
RMSPE — Root-mean-square prediction error
RMST — Remote Monitoring Satellite Technology
t — Tonne
TO — Technical Officer

4. Project Background and Rationale

Pasture productivity and utilisation across most pasture-based dairy farms in Australia has stagnated for some time and has long been seen as an area where improvements are possible but are currently unrealised. It is well understood that pasture management can be challenging and multifaceted, and losses contributing to differences between potential and realised pasture production and utilisation occur for a variety of reasons (Figure 1). These losses are not only in quantity (e.g. reduced production), but also in quality, and are often cumulative and difficult to quantify (hidden) (Garcia, S., *et al*, 2014).

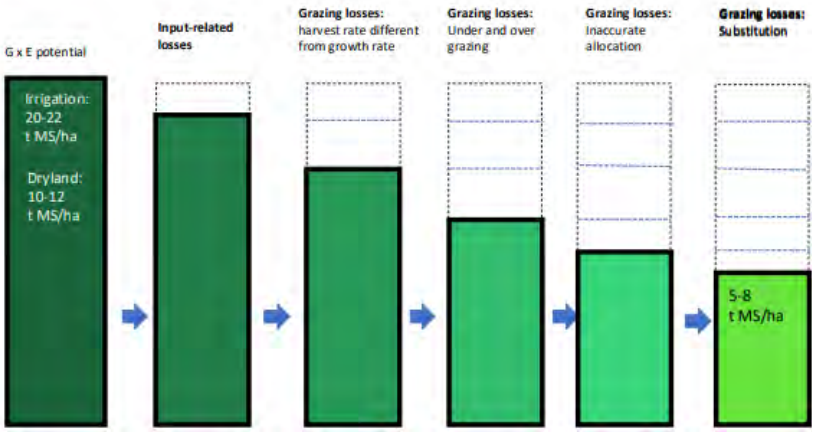

Figure 1: Illustration of some of the sources contributing to lost pasture productivity and utilisation, from research completed by the FutureDairy program (Farina et al., 2011).

Traditional monitoring (e.g. weekly pasture walks with a RPM) can help identify some of these losses but is not commonly performed on commercial dairy farms, with anecdotal evidence suggesting farmers may perceive objectively monitoring pastures as time consuming, may not be fully aware of the benefits or value of objectively monitoring, or may even lack trust in tools (Heins, B. J., *et al, 2023)*. Traditional methods of monitoring pasture are also limited somewhat in the level of detail they can provide when it comes to detecting and quantifying the full extent of hidden losses. However, technological advancements mean that farmers have access to tools that offer the option to monitor pasture remotely (via satellite, referred to here as an RMST).

This project set out to assess an RMST (the commercially available tool "Pasture.io") for suitability in monitoring pasture production in dairy systems. This tool (in its premium version) specifically uses satellites (Sentinel and Planet) for remote monitoring of pasture biomass and growth rates and integrates the use of cow-worn global positioning system (GPS) collars to automatically track grazing events and cow location on the farm.

The assumption was that utilising objective and reliable information captured by the RMST; farmers and operators could make informed decisions about their pasture management which are not influenced by subjective factors (e.g. estimates from eye assessments). It was also anticipated that using a RMST would encourage and support better communication between farmers and their farm advisors, staff, and researchers, and provide whole-of-farm pasture measurements at any given time. With progressive updates in system performance over time, problems can be identified and addressed, and outcomes improved.

5. Project Objectives

The primary goal of this project was to support NSW dairy farmers with their pasture management through testing an automated RMST which measures and estimates pasture growth and biomass. Current management practices and pasture biomass data were passively observed on the milking platform for all participating farms to assist in identifying excelling from underperforming paddocks and management settings and provide an independent assessment of the accuracy of the RMST being used. Once this had been achieved, the project then intended to actively work with the participating farmers to use the data and information produced by the RMST to make informed decisions to enhance pasture management. Unfortunately, the findings and outputs of the primary goal prohibited the project from embarking on its intention to actively work with farmers, as will be explained in this report. However, it is recommended that this work occur in the future.

A second objective of this project was to explore current pasture productivity and utilisation for the participating farms, as well as delving into the data obtained to better understand additional pasture management factors such as potential losses (grown vs grazed/harvested) and differences in pasture utilisation between paddocks under the same management.

To achieve the primary goal of the project, it was important to consider:

- Calibration of satellite data with field measurements – confirming how accurate the RMST was at measuring on-ground biomass, and then to determine how many field measurements are needed to calibrate the satellite to achieve accurate pasture cover and growth estimations.
- On-farm factors related to calculation of growth rates and pasture cover – requiring accuracy of the data flow – e.g. correct recording of when and where grazing events occurred (signalling pasture regrowth) or when farm operation occurred (e.g., mulching, spraying, sowing, fertiliser or missed fertiliser).
- Off-farm factors related to calculation of growth rates and pasture cover – requiring the software program to be adaptable and deal with missing data (e.g., missed readings from cloud cover, or irregular satellite flyovers).

6. Materials and Methods

Initial farm set up

The project monitored **pasture produced** (total amount of pasture grown, measured through objectively estimated weekly growth rate throughout the year) and **pasture utilised** (or harvested, being total pasture depletion from grazing events including direct grazing and cutting where it occurred) on 16 commercial dairy farms over a two-year period between July 2022 and June 2024. Data were collected remotely (from the RMST) and on ground with a Technical Officer (TO) taking measurements (RPM, pasture samples). Farmers were not directly engaged in the project activities, and data collection was largely passive in nature.

To capture representation across key dairying regions in NSW (including different climates), participating farms were located across 3 distinct geographical regions, being North Coast (n = 4), Mid Coast/Hunter (n = 7) and South Coast/Bega (n = 5) (Figure 2).

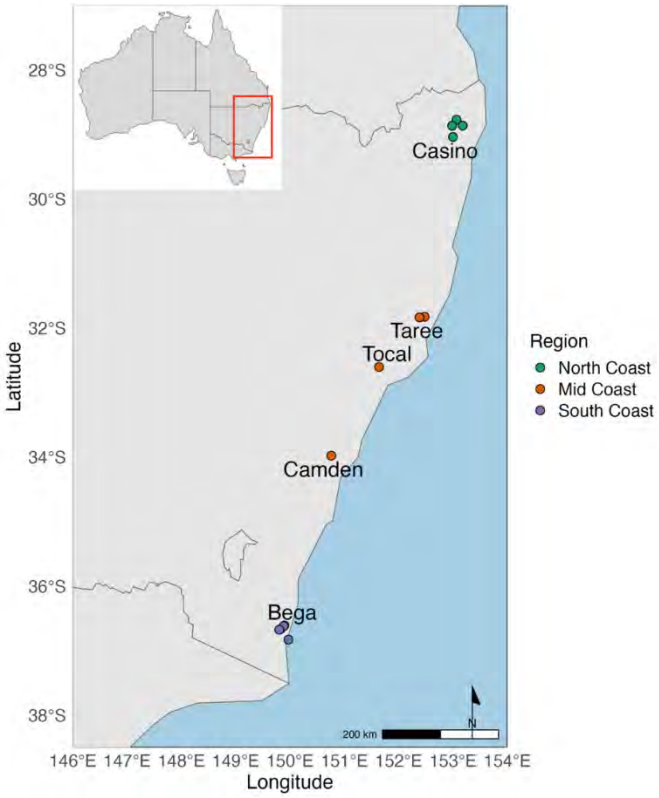

Figure 2. Location of the participating farms in New South Wales, Australia.

Farms were selected to participate in the project based on specific criteria, which included that they operate on an annual ryegrass–kikuyu grazing rotation. These systems typically involve grazing temperate C3-grass species such as annual ryegrass (*Lolium multiflorum L.*), which is generally sown in early autumn and grazed during winter and spring months, followed by grazing of the tropical perennial C4-grasses such as kikuyu (*Cenchrus clandestinum*) over the warmer months of the year (Garcia, S., *et al*, 2014).

Other criteria for participation in the project included:

- Willingness of the farmer/family to work with the project team; as well as the openness to try new things and/or work with the project constructively.
- Having sound cow performance data, including health, reproduction, and milk production data at the individual cow level.
- Having a continuous calving system with variable supplementary feed included throughout the year.
- Being located within reasonable distance from each other to facilitate travel and monitoring.
- Being reflective of typical dairy farm businesses (e.g. farm size).

Once a farm was formally recruited, the TO initiated the farm set up in the RMST. Set up included using a farm map to objectively determine paddock locations and sizes, which was then confirmed by the farmer. Table 1 includes a brief description of the initial farms included in the project. Out of all the farms, one utilized a robotic milking system and half of them had irrigation capability in at least some of their paddocks.

Table 1. Description of the participating farms in the project.

Farm	Area (ha)	Paddocks	Paddocks with irrigation capability	Milking system	Milking herd
North coast					
Farm 01	90.3	60	38	Conventional	175
Farm 02	130	35	-	Conventional	250
Farm 03	131.8	76	6	Automatic	210
Farm 04	105.6	43	-	Conventional	190
Mid coast					
Farm 05~	218.8	64	-	Conventional	388
Farm 16*	229.1	60	-	Conventional	440
Farm 06	174.8	46	46	Conventional	280
Farm 07	64.5	22	-	Conventional	200
Farm 08	134.1	27	-	Conventional	300
Farm 09	88.9	39	-	Conventional	105
Farm 15	83.4	39	-	Conventional	258
South coast					
Farm 10	260.4	53	8	Conventional	291
Farm 11	199.2	77	16	Conventional	160
Farm 12	152	48	35	Conventional	300
Farm 13	313	83	48	Conventional	580
Farm 14	149.4	45	14	Conventional	200

~This farm was sold and left the project after Year 1. *This farm replaced Farm 05 in March 2023.

Manual farm monitoring activities (on ground)

The procedural protocols for the project were meticulously formulated and deliberated upon with the project team at the project's inception. The following section includes the main considerations of the methodology.

Year 1 data collection procedure

Five paddocks per farm were monitored weekly by a TO during year 1, which ran from July 2022 until June 2023. The paddocks were selected based on the below criteria, enabling comparison across farms and regions and at the same time representing the baseline of a typical grazing platform for the dairy systems in the project. Selection criteria included:

- Pasture composition: ryegrass presence from autumn to late spring and thick and evenly distributed kikuyu presence during summer months.
- Proximity to the dairy: avoid paddocks too close to the dairy (greater risk of being night-paddocks or sacrificed paddocks).
- Being relatively flat.
- Had permanent fences.
- Sites convenient to road access to enable quick entry in and out of paddocks.

For each paddock, the TO measured and collected:

- Weekly pasture cover, using a calibrated RPM (Jenquip EC20 Electronic Pasture Meter, Fielding, New Zealand).
- Monthly quadrant cuts at 5cm height, with 9 samples taken from two of the five paddocks (18 samples total each month) to calibrate the RPM. The two paddocks were pre-selected and always the same.
- Visual assessment of pasture botanical composition.
- Weekly farm management data (e.g. fertilisation, irrigation, sowing and mulching).
- Regular checking of cow GPS collars for technical issues.

Cutting pasture to 5 cm height enabled samples to be dirt-free, desirable for water content determination and minimised the risk of collecting dried and dead biomass. Pasture cut samples were lab tested to measure metabolisable energy, crude protein, acid- and neutral-detergent fibre, and water-soluble carbohydrates content among other nutritional indicators. Pasture cuts were also used to calibrate the RPM.

Visual assessments of pasture botanical composition involved assessing photos along with the field notes taken on the date the paddock was monitored. A scoring system representing a percentage applied to each pasture type was implemented with the following classes: “annual ryegrass”, “kikuyu”, “white clover”, “other grasses”, “weeds” (predominantly broadleaf species), and “dead material” (of any species). This allowed a general characterisation of the pasture type (based on the main functional group of grasses) to be made throughout the duration of the project, with ryegrass-based pastures being any pasture with annual ryegrass and white clover presence higher than 70%. Similarly, when kikuyu and other grasses different from annual ryegrass were the main grasses present, pastures were categorized as being kikuyu-based.

Grazing records were automatically made (usually daily) using cow collars with integrating GPS trackers. These trackers generated a GPS point every three minutes, and data was automatically fed

into the RMST. Each farm used 3 trackers across the milking herd, although the AMS farm used 5 trackers.

Year 2 data collection procedure

Results from Year 1 indicated that fortnightly data were the minimum required for satellite calibration (see below and Correa-Luna *et al*., 2024). Thus, commencing in July 2023, TO's began fortnightly visits to participating farms. During each visit, the following were measured with a calibrated RPM:

- Pasture cover from the same 5 paddocks as Year 1 (data used to calibrate the RMST) – same methodology as outlined for Year 1.
- Pasture cover from the paddock about to be grazed on the visit day (pre-grazing paddock). Pasture samples from this paddock (hand plucking at grazing height observed when visiting the farm) were also collected for quality analysis.
- Pasture cover of nearby grazed paddocks (post-grazing paddocks) were collected for quantification of grazing intensity, pasture allocation assessment, pasture use efficiency.

These visits provided an opportunity for the TO to engage with interested farmers, and to gather informal feedback from farmers on the functionality of the RMST. Insights into recent farm management practices, including cow feeding strategy and pasture allocation management, were also obtained.

Similarly to year 1, grazing events were monitored using GPS cow collars. In cases where GPS trackers failed to generate grazing records, TO's collected records from the dairy. These records were obtained by contacting farmers or accessing existing records and were manually entered into the RMST.

Remote farm monitoring activities

Weather data

Climatic data (daily rainfall, maximum and minimum air temperature, and global solar radiation) for each farm were obtained from the closest Australian Bureau of Meteorology Weather Station via the SILO service (www.longpaddock.qld.gov.au/silo/). This was important information to collect, as pasture growth is strongly linked with weather and climate factors.

RMST raw data

The RMST used works on algorithms originating from a series of machine learning models utilising neural networks developed from: (i) satellite optics and sensors; (ii) weather and atmospheric conditions; (iii) farm management activities (i.e., grazing records, fertilisation, pasture silage making); and (iv) pasture biomass measurements manually added by the user (which were strictly controlled for the purpose of this project). The remote sensing data originated from the PlanetScope CubeSats constellation (Planet Team, 2021) and the Copernicus Sentinel-2 mission (European Space Agency, 2022). While the PlanetScope CubeSats constellation provides multispectral data at 3–4m spatial resolution with a 30h global median average revisit interval, Copernicus Sentinel-2 images provide a lower spatial (10–60m) and temporal (five days) resolution imagery. By utilising a combination of the remote sensing data mentioned, the machine learning process generates algorithms for estimating pasture biomass and growth rate employing multiple bands (i.e., NDVI, EVI, ENDVI, SAVI) which are embedded within the satellite images.

Bulk raw data for all participating farms was accessed every two months. This data was mainly comprised of pasture biomass cover estimated by the RMST, grazing records generated with GPS trackers, and any farm management practices entered manually into the RMST by the farmer or the TO.

To facilitate smooth collaboration and data sharing between the research team and the operator of the

RMST, a contractual partnership was established. This also ensured that reporting remained unbiased and transparent, without any conflicts of interest. Additionally, the research project had full access to the raw data produced by the RMST, enabling thorough analysis and evaluation. This collaboration reflects a dedication to scientific integrity and advancement in enhanced pasture management using remote sensing.

Evaluation metrics

The results of the manual monitoring were compared with predictions made by the RMST, which used high resolution satellite images to estimate pasture biomass. It could also incorporate local information (e.g. weather, soil and farm management data) which can be used to assist farmers in making informed pasture management decisions. The RMST had been developed and validated for perennial ryegrass pastures however NSW pastures are a mix of C3 (e.g. ryegrass) and C4 (e.g. kikuyu) grasses, and it was therefore important to compare the RMST pasture predictions with the on-ground manual measurements to independently evaluate the RMST for use in these types of pasture-based dairy systems (predominant in NSW, but common in many other dairying regions across Australia).

All evaluation undertaken included the root-mean-square error (RMSE) expressed in biomass (kg DM/ha) and in percentage of the observed pasture biomass. The RMSE was also decomposed by the three types of error: error in central tendency which is a measure of precision, error due the slope regression which refers to the accuracy, and error due to the disturbance which cannot be reduced. Given the contrasting range of conditions across farms in this study, the mean absolute error (MAE) was also reported (kg DM/ha and as a %) since it is less sensitive to potential outliers. For those comparisons with different numbers of observations, the RMSE to standard deviation of observed values (RSR) was also included.

7. Key Findings

Weather observations

Key observation: Weather patterns fluctuated throughout the project, and many farms experienced significant wet weather, or even flooding, throughout the project.

All three regions experienced an excessively wet autumn and spring in 2022 (start of Year 1), which continued into Year 2 (Table 2). These wet conditions (including flooding in some areas) were a significant challenge for farmers to manage and made quadrant cuts (to calibrate the RPM) difficult for TO's, resulting in some monitoring activities being impacted temporarily.

Table 2. Mean monthly rainfall (mm) during the project, separated into the three farm regions of New South Wales, Australia. Historical mean monthly rainfall produced from records starting on 01/01/1900.

Region	North coast		Mid coast		South coast	
	Monthly rainfall	Historic monthly rainfall	Monthly rainfall	Historic monthly rainfall	Monthly rainfall	Historic monthly rainfall
			Year 1 (2022-2023)			
July	64	51	327	65	15	49
August	14	38	27	52	31	43
September	94	38	125	54	108	45
October	106	70	175	76	178	63
November	16	86	47	86	46	71
December	50	119	37	92	21	75
January	55	131	108	113	52	74
February	52	151	84	131	82	85
March	110	146	146	144	36	89
April	28	88	120	108	16	60
May	49	79	24	88	27	62
June	14	68	8	93	2	66
Total	652	1065	1228	1102	614	782
			Year 2 (2023-2024)			
July	13	51	28	65	1	49
August	13	38	22	52	19	43
September	17	38	11	54	4	45
October	39	70	87	76	43	63
November	139	86	86	86	263	71
December	68	119	103	92	255	75
January	155	131	82	113	61	74
February	110	158	182	135	44	88
March	80	115	67	116	53	75
April	188	87	189	108	100	61
May	100	80	109	87	144	62
June	6	66	103	93	49	67
Total	928	1039	1069	1077	1036	773

Pasture quality and composition

Key results: There was noticeable variability in pre-grazing pasture biomass, particularly in the spring/summer months (in line with knowledge of the prolific growth of summer grasses). The months where biomass was most variable was also linked to the pasture quality falling (higher fibre, lower metabolisable energy and crude protein).

Pre-grazing pasture biomass and chemical composition averages per farm and per season are presented in Table 3. The missing data in autumn for the north coast is due to a delayed start for the TO in that region. The relatively lower biomass in autumn (compared to other seasons) is reflective of the general reduction in pasture availability at this time (summer pastures dying back while winter pastures are establishing), meaning farmers often need to open a paddock for grazing "early" (i.e. before it reaches a standard biomass target of around 3,000 kg DM/ha). Conversely, in spring and summer the winter pastures die back, and summer pastures emerge, with summer pastures commonly growing more prolifically and are often linked to higher pre-grazing biomass as farmers may not be able to graze paddocks quickly enough (can lead to paddocks being dropped from the grazing rotation). The higher biomass is almost always linked with a higher NDF content, as is seen in these results, due to a higher portion of stem and more mature/older leaf.

Table 3. Average pre-grazing pasture cover and chemical composition results reported during year 1.

		Autumn					Winter					Spring					Summer				
Region	Farm	Pasture cover	ME[1]	CP[2]	NDF[3]	ADF[4]	Pasture cover	ME[1]	CP[2]	NDF[3]	ADF[4]	Pasture cover	ME[1]	CP[2]	NDF[3]	ADF[4]	Pasture cover	ME[1]	CP[2]	NDF[3]	ADF[4]
North coast	Farm 01						2,655	12.6	20.2	41.07	20.10	3,436	11.3	20.9	46.47	23.38	3,384	9.6	20.9	55.41	26.30
	Farm 02						3,223	12.5	27.4	44.59	20.79	2,995	11.2	25.9	47.78	22.31	3,279	10.1	21.6	53.71	23.63
	Farm 03						2,359	12.1	29.4	46.80	20.16	2,863	11.5	27.1	47.44	19.98	2,665	10.2	22.8	51.70	22.56
	Farm 04						2,488	11.4	25.3	38.54	17.05	3,014	11.2	22.6	48.55	21.98	3,213	9.5	21.1	55.52	25.30
Mid coast	Farm 05	2,794	8.7	21.3	58.89	25.24	3,111	11.4	22.9	54.81	25.55	3,087	10.1	23.2	47.69	25.23	2,692	9.8	21.8	53.23	23.31
	Farm 06	3,108	9.6	26.3	48.60	19.61	3,143	12.9	29.3	39.03	20.29	3,073	10.6	25.0	46.58	26.99	3,477	8.5	18.4	63.21	29.56
	Farm 07	3,155	8.9	22.4	59.32	23.45	4,003	11.5	27.7	40.51	20.47	3,586	10.4	22.3	49.28	23.50	3,673	9.6	23.1	52.56	23.09
	Farm 08	2,666	8.5	22.8	58.46	23.81	3,239	12.1	29.2	44.01	22.51	3,207	10.5	25.4	50.46	23.85	3,140	9.8	25.8	51.55	22.11
	Farm 09	3,296	8.2	23.3	59.20	23.35	3,155	11.4	27.7	47.99	22.27	3,477	10.1	24.1	51.61	23.01	3,363	10.1	24.2	49.11	21.27
South coast	Farm 10	2,929	11.0	22.6	49.53	24.80	2,914	13.1	24.6	42.53	20.56	3,037	11.9	23.0	41.84	21.21	3,321	9.9	19.2	52.76	26.81
	Farm 11	2,517	11.4	23.0	46.53	22.83	2,711	12.5	27.6	52.04	26.31	2,838	11.5	23.3	46.58	23.09	3,967	9.5	22.1	53.40	25.49
	Farm 12	2,569	10.7	24.3	49.35	21.90	3,342	12.9	21.2	42.16	21.48	3,207	11.6	19.6	46.65	24.02	3,923	10.1	19.8	51.25	24.72
	Farm 13	2,602	10.5	24.2	46.67	22.87	2,812	12.6	25.5	45.16	23.24	3,238	11.6	22.6	46.30	23.67	3,088	10.2	20.6	51.03	25.25
	Farm 14	2,527	10.5	22.5	52.66	23.74	2,749	12.4	24.3	48.29	23.80	2,837	11.5	20.9	44.89	22.73	3,117	9.7	19.1	52.86	25.52

1Metabolizable energy content, MJ ME kg DM-1. 2Crude protein concentration, %. 3Neutral detergent fibre concentration, %. 4Acid detergent fibre concentration, %. Note, farms 15 and 16 are not included as they were not part of the project in year 1.

Figure 3 summarises average values for pre-grazing pasture quality collected during the first year of the project. The "whiskers" (vertical lines at each graph point) indicate the variability between samples (standard error) collected for that specific measurement, where the larger the whisker the higher the variability. There is variability in pre-grazing pasture cover (Figure 3a), which was also shown in the pre-grazing biomass values presented in Table 3. This variability indicates that the pre-grazing biomass target (3,000 kg DM/ha) wasn't being consistently met at the time of grazing and is an area for further investigation and potential improvement. Energy (Figure 3b) and crude protein (Figure 3c) both dropped over summer, occurring during the same period when pasture cover variability tended to be highest (Figure 3a). Fibre content (Figure 3d) also increased at this time. This aligns with our understanding that the higher the pasture biomass (particularly above 3,000 kg DM/ha), the lower the nutritive value and digestibility (i.e. the lower the quality).

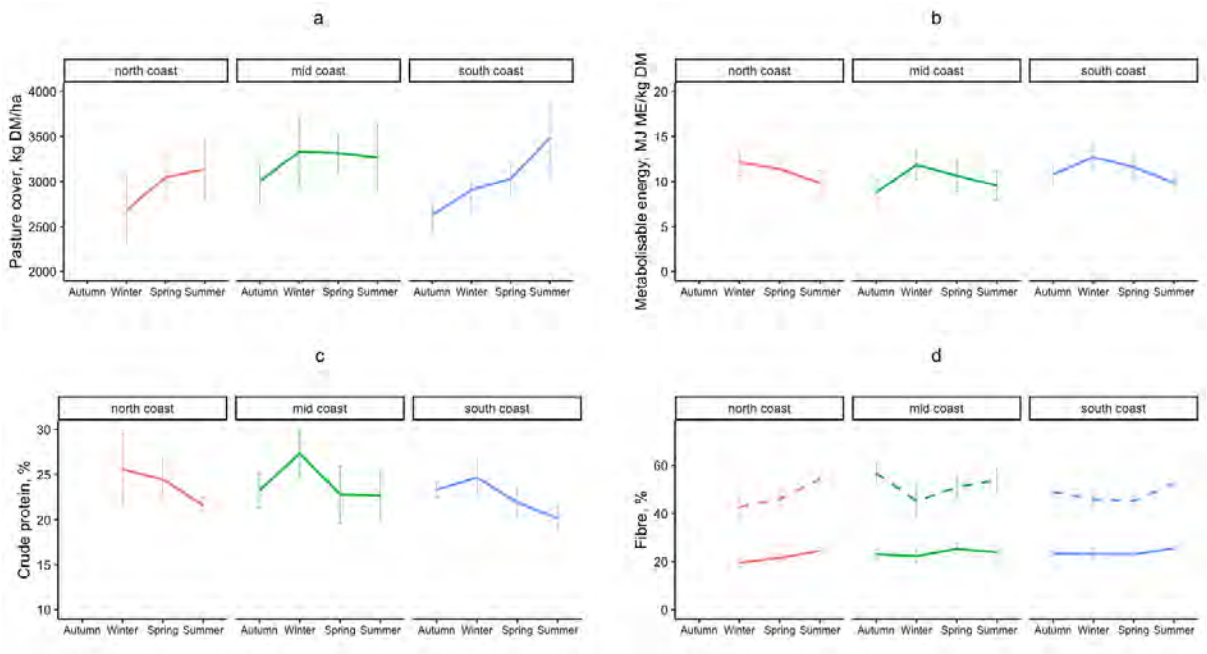

Figure 3. Mean pasture quality per season and region; a: pre-grazing pasture cover (kg DM ha-1), b: pasture energy content (MJ ME kg DM-1), c: crude protein concentration, d: neutral detergent fibre (solid line) and acid detergent fibre (dashed line).

As expected, pasture composition substantially changed during the year in all regions (Table 4). Annual ryegrass was the most predominant grass species in winter and spring, and kikuyu in summer. Autumn exhibited variability in the proportion of ryegrass and kikuyu present, with this being a period of transition where kikuyu starts to ease off and ryegrass becomes more prevalent. The further north (and typically the warmer the climate), the longer kikuyu persists, which is shown in the results with the north coast having the highest prevalence of kikuyu in Autumn out of the three farming regions. Other grasses noted included *Setaria sphacelata* var. *sericea*, *Paspalum dilatatum* Poir., and *Chloris gayana* in the North coast, and temperate grasses such as *Festuca arundinacea Schreb*., *Dactylis glomerata*, and perennial brome grasses in the South coast.

Table 4. Mean ± SD pasture composition content (%) throughout the year on farms located in three coastal dairy regions of New South Wales, Australia.

Region	Season	Annual ryegrass	Kikuyu	White clover	Other grasses	Weeds	Dead material*
North coast	Autumn	14.7 ± 0.01	70.0 ± 0.01	-	15.3 ± 0.01	-	-
	Winter	53.0 ± 28.1	15.3 ± 13.7	8.2 ± 6.2	17.3 ± 24.9	4.7 ± 10.1	1.6 ± 3.6
	Spring	50.9 ± 37.2	14.7 ± 23.2	7.1 ± 7.2	24.2 ± 26.1	2.9 ± 4.7	0.1 ± 0.7
	Summer	1.6 ± 6.0	61.5 ± 25.9	0.0 ± 0.0	33.6 ± 20.3	2.0 ± 4.1	1.3 ± 3.5
Mid coast	Autumn	38.5 ± 19.4	27.0 ± 18.9	7.8 ± 3.8	5.3 ± 5.7	8.0 ± 4.2	13.4 ± 7.4
	Winter	64.2 ± 22.7	20.0 ± 20.2	4.4 ± 5.3	3.3 ± 4.9	4.0 ± 4.9	4.1 ± 6.3
	Spring	55.3 ± 25.6	39.3 ± 22.3	1.3 ± 2.3	1.7 ± 4.3	0.0 ± 0.0	2.4 ± 6.1
	Summer	60.3 ± 21.5	34.5 ± 24.9	1.4 ± 2.2	1.9 ± 3.0	0.2 ± 0.7	1.7 ± 5.0
South coast	Autumn	13.7 ± 24.6	53.1 ± 32.0	1.2 ± 1.3	7.1 ± 11.1	6.1 ± 7.6	18.9 ± 19.9
	Winter	37.7 ± 24.5	26.6 ± 27.0	6.5 ± 6.5	4.7 ± 6.5	1.8 ± 2.2	22.7 ± 11.8
	Spring	72.8 ± 18.0	3.1 ± 3.7	10.0 ± 10.7	6.3 ± 6.1	3.4 ± 4.6	4.4 ± 4.9
	Summer	31.6 ± 31.9	43.6 ± 31.2	11.6 ± 12.8	3.8 ± 3.1	5.0 ± 2.1	4.4 ± 5.2

*Of any species.

Evaluation of the RMST

Calibrating the RPM

Key results: The RPM was calibrated with an acceptable error. The accuracy of the RPM remains well aligned when biomass is less than approx. 2,500-3,000 kg DM/ha but begins to fall once biomass exceeds this point (by 4,000 kg DM/ha, reliability is much lower).

A total of 2,261 quadrant cuts were taken during the first year of the project, and once outliers and unlikely observations were removed, 1,891 observations remained. These were used to create monthly linear regressions, along with compressed height measurements from the RPM, to calibrate the RPM in preparation for evaluation of the RMST. Details on the performance of the analysis are presented in Table 5. It is important to note that the residuals exhibited a normal distribution, and the MAE (a simple way to measure accuracy in the model, representing the average amount the model's predictions are off from the actual, true value) was 356kg DM/ha. The lower the MAE is, the better the predictions, and when looking at MAE%, <10% is considered excellent while 10-20% considered acceptable (in this case, the MAE% was 11.99%, making the results acceptable and close to excellent for the model).

Table 5. Overall, regional, and seasonal level evaluations of the performance of the monthly linear regression equations to calculate pasture biomass from compressed height using a rising plate meter in three coastal dairy regions of New South Wales, Australia.

Item	N	MAE, kg DM/ha	MAE, %	RMSE, kg DM/ha	ECT, %	ER, %	ED, %	RMSE, %	RSR
Overall	1,891	356	11.992	468.8	0.214	0.027	99.759	15.773	0.56
Region									
North coast	507	232	9.538	298	0.003	0.000	99.997	12.261	0.60
Mid coast	672	395	12.742	501	0.120	0.004	99.875	16.180	0.66
South coast	712	409	12.629	532	0.568	0.597	98.835	16.441	0.59
Season									
Autumn	297	407	12.691	497	0.628	0.000	99.372	15.499	0.60
Winter	518	332	12.520	432	6.539	3.014	90.447	16.296	0.72
Spring	586	347	11.830	473	1.072	1.707	97.221	16.117	0.58
Summer	490	363	11.288	484	0.001	0.001	99.998	15.037	0.52

n = number of observations. MAE = Mean-absolute error expressed in kg dry-matter (DM) per ha and as a proportion of mean observed pasture biomass (%). RMSE = Root-mean square error expressed in kg dry-matter (DM) per ha and as a proportion of mean observed pasture biomass (%). Equation error was decomposed into: ECT = error in central tendency (%), ER = error due the slope regression (%), and ED = error due to random disturbance (%). RSR = RMSE to standard deviation of observed values ratio.

In Figure 4, all calibration cuts and RPM pasture covers are presented along with the fitted calibration equation. Box plots (orange box and whiskers) illustrating the distribution of observations are also included. Confidence in the accuracy of the RMP measurements at lower biomass is high, as variability between measurements and the fitted calibration equation is small (i.e. dots are close to the equation line). However, as the biomass increases (e.g. >2,500-3,000 kg DM/ha), that variability increases and indicates that the accuracy falls, particularly once biomass exceeds around 4,000 kg DM/ha. Importantly, both the quadrat cuts and the corresponding biomass estimates obtained via the RMP display outliers at these higher biomasses and highlights the challenges and potential inaccuracies that are likely to occur at high biomass.

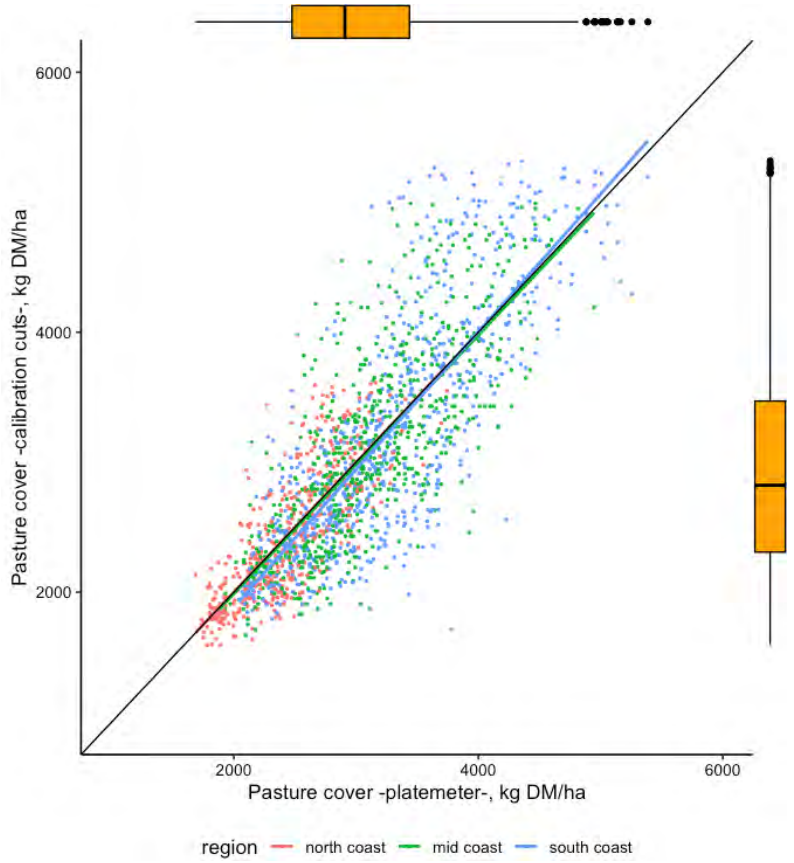

Figure 4. Fitted calibration equations (coloured lines) along with the distribution of fitted pasture cover estimates (RPM) and observed pasture cover (calibration cuts) (dots). Histograms (orange bars) show the distribution of observations for calibration cuts and RPM covers.

Evaluating the estimated pasture cover from the RMST in Year 1

Key result: The RMST was found to have an average MAE of 460 kg DM/ ha and appeared to lose predictive accuracy at both low and high pasture biomass levels. Therefore, its ability to accurately predict pasture biomass was initially perceived to be lower than that of the calibrated RPM (gold standard). Additional training of the RMST was necessary (Year 2).

Estimated pasture cover from the RMST (approximately 60,500 individual records) were evaluated against the mean pasture cover measured with the calibrated RPM (approximately 1,600 measurements, although only 612 measurements matched exactly with the date the RMST record was created – i.e. the TO farm visit was not always on the same day as a satellite flyover). This was done

using the correlation coefficient (R), which describes the proportion of variance in the measured data (RPM) explained by the estimated data (RMST), with the root mean square prediction error (RMSPE) expressed as both kg DM ha/ha and as a percentage of the observed mean pasture cover. In addition, the mean absolute error (MAE) was also reported (expressed as kg DM ha/ha and in %) since it is less sensitive to potential outliers.

Initially (before training with the on-ground measurements from the RPM), the overall evaluation revealed a prediction error of approximately 20% (Table 6), which represents an MAE of approximately 460 kg DM/ ha on average (590 kg DM/ha of RMSPE). When running the evaluation by region, the mid coast region had a higher prediction error relative to other regions. For all results, the MAE was lower than the RMSPE and is likely due to the MAE being less sensitive to outliers.

Table 6. Goodness of fit of the pasture cover estimations at the paddock level for the RMST during year 1.

Item	n	MAE kg DM/ha	MAE %	RMSPE kg DM/ha	RMSPE %	mean predicted	mean observed
Overall	1350	458.3	17.1	590.2	22	2,471	2,680
By region							
North coast	309	403.8	17.5	510.6	22.2	2,413	2,302
Mid coast	381	527.8	18.5	650.8	22.9	2,454	2,846
South coast	553	417.7	15.2	557.2	20.3	2,551	2,744
By farm							
North coast							
Farm 01	79	468.4	19.2	556.9	22.9	2,257	2,434
Farm 02	84	263.8	10.7	326.8	13.3	2,396	2,459
Farm 03	92	439.5	21.7	555.3	27.4	2,418	2,029
Farm 04	54	466.3	20	588.9	25.3	2,658	2,332
Mid coast							
Farm 05	107	578.4	20.3	727.5	25.5	2,288	2,854
Farm 06	119	462.6	17.3	596.4	22.2	2,292	2,681
Farm 08	71	491.9	18.2	598.8	22.1	2,582	2,708
Farm 09	85	536.9	18.2	656.3	22.2	2,597	2,955
Farm 07	106	617.5	20.3	733.3	24.2	2,435	3,036
South coast							
Farm 10	113	447	16.2	671.4	24.3	2,416	2,767
Farm 11	128	403.2	14.5	501.7	18	2,782	2,787
Farm 12	101	431.6	14.5	609.3	20.5	2,596	2,968
Farm 13	105	394.3	15.3	471.1	18.3	2,473	2,573
Farm 14	106	413.7	15.8	511.7	19.5	2,451	2,622

Note: farms 15 and 16 are not presented here as they were not part of the project in Year 1.

The error obtained could be interpreted as a large error but does not necessarily mean that this remote monitoring system is unsuitable for pasture management. The RMST used in the project collects information from two separate satellite missions (Sentinel and Planet), local weather, and with farm management activities (i.e., grazing records, fertilisation, silage making), and uses the information to generate pasture cover and growth estimates. Regular input of on-farm activities will likely assist in refining the estimates from the RMST.

Figure 5 provides a visual analysis of the residuals obtained by subtracting estimated pasture biomass (RMST) from measured pasture biomass (RPM). The residuals displayed a normal distribution (Figure 5a), with a slight rise in values evident as pasture biomass surpassed the 3,000 kg DM/ha mark. Variation in residuals was fairly even when within "normal" pasture biomass range (approx. 2,000-3,000 kg DM/ha, Fariña, S., *et al, 2011*) but increased both below and above this biomass (Figure 5b and c). Lastly, the homoscedasticity test indicated notable variations between farms (Figure 5d), indicating imbalances in the data (i.e. each farm was different, and employed different management practices). This analysis offers an indication that the RMST likely overpredicts biomass at lower true ground biomass, while simultaneously underpredicting biomass at high biomass levels.

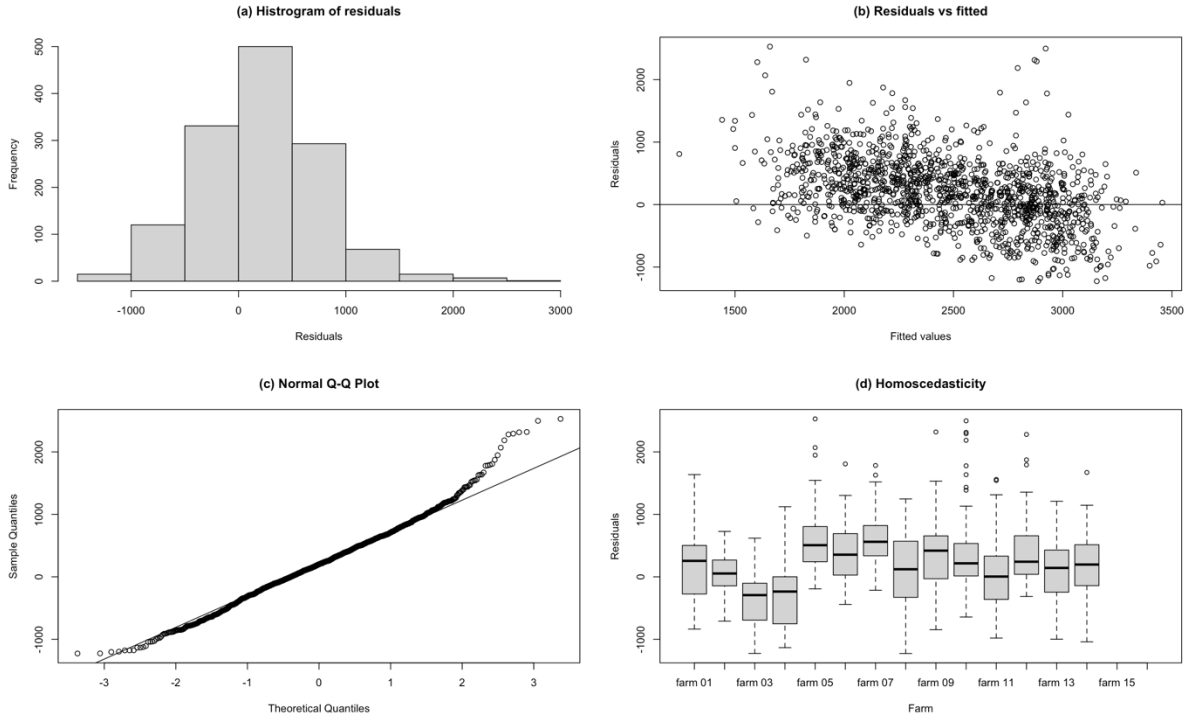

Figure 5. Analysis of residuals for the initial RMST evaluation with calibrated RMP data.

Training and re-evaluating the RMST in Year 2

Key result: training improved the accuracy of the RMST's pasture biomass estimates, where using 2 weeks per month training (from 5 paddocks per farm, or 10% of the grazing platform) significantly reduced the error (i.e. improved accuracy). The error continued to reduce when training increased further, but this change was not significant. Therefore, from a practical perspective, using 2 weeks a month of RPM data to train the RMST is likely to be the most effective approach (lowest level of input to achieve acceptable gain in accuracy).

The data collected in both years of the project were used to train and then re-evaluate the RMST after training to determine the impact and effectiveness of training on pasture biomass estimates and what level of training is needed to obtain the results.

Pasture biomass measurements recorded with a calibrated RPM were filtered to ensure the date of each RPM record matched the date of the RMST data for accurate comparisons. The RPM data was split into groups, called "training" (80% of the measurements) and "evaluation" (20% of the measurements). Training data was then sorted into four groups, being 1) 1 Week (1W; 25% of the data); 2) 2 Weeks (2W; 50% of the data); 3) 3 Weeks (3W; 75% of the data); and 4) 4 Weeks (4W; 100% of the data). These data groups were fed into the RMST sequentially, starting at W1, which enabled a clear assessment of the impact of increasing training frequency on RMST biomass estimates accuracy. The evaluation data was used to validate the RMST estimates across each of the 4 training groups.

For Year 1 data, there was minimal difference in MAE between the control error (evaluation data) and 1W (training data; Table 7), while 2W resulted in a significant drop in error – i.e. the accuracy of the RMST improved significantly at 2 weeks per month of training. Additional reductions in error were seen with further training (3W and 4W) but were not significantly different to 2W.

Table 7. Evaluation of pasture biomass performance of a machine learning algorithm decision support tool based on satellite imagery, weather, and farm management data after progressive model training intensities using calibrated rising plate meter data of 5 fixed paddocks per farm.*

Data	MAE kg DM/ha	MAE %	RMSE kg DM/ha	ECT %	ER %	ED %	RMSE %
Control	498a	19.1	602a	4.8	15.9	79.3	23.1
1W	518a	19.9	642a	0.8	28.0	71.2	24.6
2W	342b	13.1	431b	3.1	0.4	96.5	16.5
3W	298b	11.4	371b	3.6	0.9	95.5	14.2
4W	293b	11.2	362b	3.3	0.6	96.1	13.9

n = number of observations. MAE = Mean-absolute error expressed in kg dry-matter (DM) per ha and as a proportion of mean observed pasture biomass (%). RMSE = Root-mean square error expressed in kg dry-matter (DM) per ha and as a proportion of mean observed pasture biomass (%). Equation error was decomposed into: ECT = error in central tendency (%), ER = error due the slope regression (%), and ED = error due to random disturbance (%). Different superscripts in MAE and RMSE indicates statistical differences with respect to the mean differences at a p-value of < 0.05 according to the Tukey's HSD test. *Training intensities: Control (no training), 1W: training based on field data from week 1 of a given month, 2W: training based on field data from weeks 1 and 3 of a given month, 3W: training based on field data from weeks 1, 3, and 2 of a given month, 4W: training based on field data from 4 weeks of a given month.

When shown visually (Figure 6), the evaluation data (control – no training; Figure 6a) and 1W training data (Figure 6b) were poorly aligned with the RPM data. Like Table 8, they show high levels of error, but they also show that the RMST estimates were underpredicting pasture biomass compared to the mechanical measurements of the RPM. Figures 6c-6e show a strong alignment between the estimates of the RMST and the measurements of the RPM, again indicating improvement in accuracy of the estimates from the RMST.

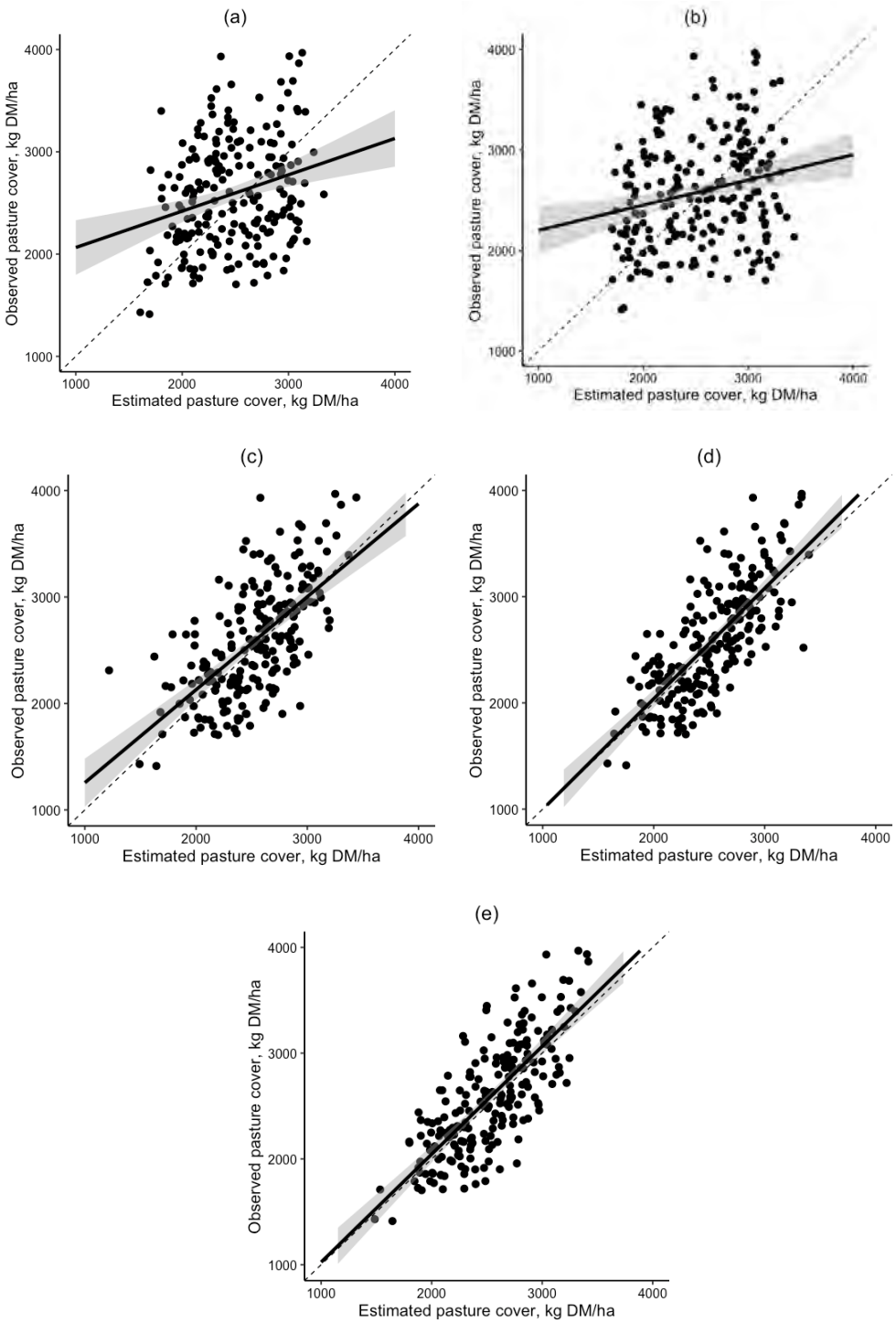

Figure 6. Scatter plots of observed (RPM) versus predicted (RMST) pasture biomass (in kg DM/ha) on four training intensities with calibrated RPM data using 5 fixed paddocks per farm: (a) control (no training), (b) 1W: RMST trained with RPM data of one week per month, (c) 2W: RMST trained with RPM data of two weeks per month, (d) 3W: RMST trained with RPM data of three weeks per month, and (e) 4W: RMST trained with RPM data of four weeks per month. Grey-shaded areas show the uncertainty in the relationship of observed vs estimated pasture biomass. Dashed line is the one-on-one line.

Even with training, the RMST had difficulty accurately estimating pasture biomass at higher volumes (Table 8), where the error at the highest biomass group (3,580 kg DM/ha) was much higher than at the lower two biomass groups. Like Table 7, training the RMST reduced the error (i.e. improved the accuracy).

Table 8. Evaluation of the accuracy of pasture biomass estimates from the RMST with RPM data at different pasture biomass volumes (tercile groups).

Item	Pasture biomass observed kg DM/ha				MAE	MAE	RMSE		ER	ED	RMSE
	N	Mean	Min	Max	kg DM/ha	%	kg DM/ha	%	%	%	%
First tercile	75	2,031	1,413	2,314							
Control					411a	20.2	509a	42.3	43.2	14.5	25.0
1W					521a	25.6	655a	36.0	54.3	9.7	32.3
2W					322ab	15.9	395ab	30.5	44.2	25.3	19.4
3W					261ab	12.9	307ab	30.9	30.6	38.5	15.1
4W					241b	11.9	303b	28.8	33.4	37.9	14.9
Second tercile	110	2,748	2,330	3,222							
Control					453a	16.5	531a	21.5	55.9	22.6	19.3
1W					430a	15.6	534a	8.3	69.4	22.3	19.4
2W					280ab	10.2	353ab	13.2	42.6	44.2	12.9
3W					250ab	9.1	311ab	10.5	36.3	53.2	11.3
4W					262ab	9.5	315ab	9.4	38.2	52.4	11.5
Third tercile	29	3,580	3,248	4,183							
Control					895a	25.0	967a	85.8	7.1	7.1	27.0
1W					848a	23.7	920a	84.8	8.1	7.1	25.7
2W					632b	17.6	702b	81.1	6.0	12.9	19.6
3W					577b	16.1	637b	82.1	4.4	13.5	17.8
4W					541b	15.1	597b	82.0	3.7	14.3	16.7

Different superscripts in MAE and RMSE indicate statistical differences with respect to the mean differences at a p-value of < 0.05 according to the Tukey's HSD test for each pasture biomass tercile separately. *Training intensities: Control (no training), 1W: training based on field data from week 1 of a given month, 2W: training based on field data from weeks 1 and 3 of a given month, 3W: training based on field data from weeks 1, 3, and 2 of a given month, 4W: training based on field data from 4 weeks of a given month.

Figure 7 also demonstrates the change in accuracy as biomass changes. Between 1,500 and 3,000 kg DM/ha, the MAE obtained was approx. 250 kg DM/ha with variation being quite low. Below 1,500 kg DM/ha, MAE doubled (500 kg DM/ha), while between 3,000 and 3,500 kg DM/ha the prediction error experienced a linear increase (MAE starting at 260 and reaching nearly 600 kg DM/ha). Beyond 3,500 kg DM/ha, MAE experienced a four-fold increase in the prediction error relative to the MAE obtained between 1,500 and 3,000 kg DM/ha. It is important to acknowledge that most of the training data used was within the 1,500 and 3,000 kg DM/ha range (potential to provide higher accuracy within this range, compared to the lower or upper ends). However, even if the RMST loses accuracy outside this range (as was the case in this project), pastures are intended to be maintained at a biomass between 1,500 and 3,000 kg DM/ha to avoid over- and under-grazing and maximise nutritional value in the pasture, and the results indicate the RMST is capable of predicting biomass in this range with an acceptable accuracy.

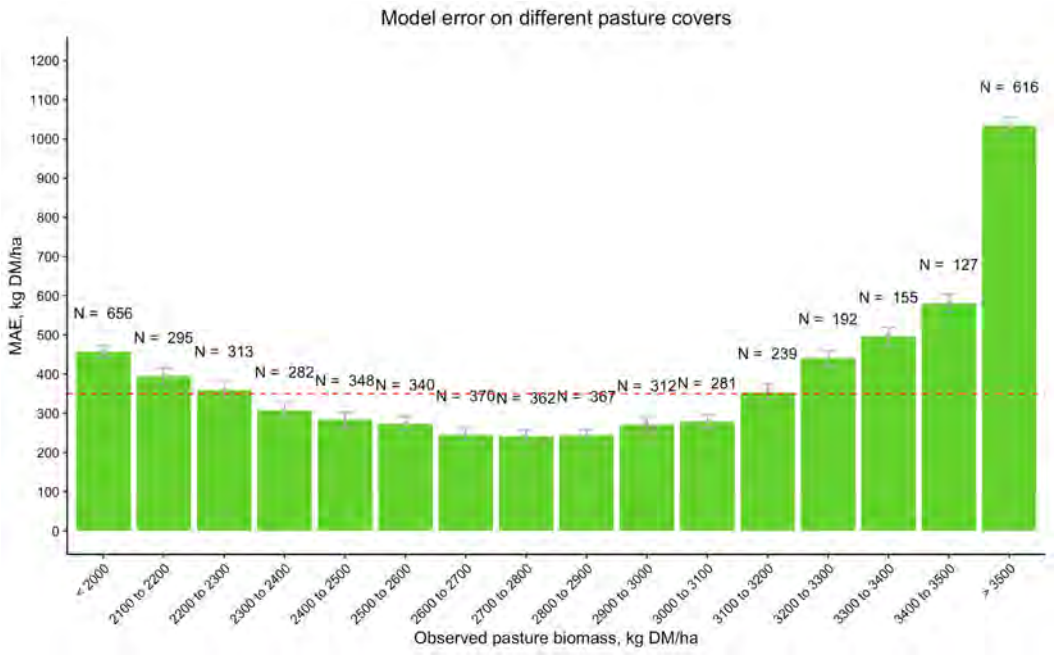

Figure 7. Mean-absolute error (in kg DM/ha) of the RMST at observed pasture biomass levels (RPM) after training is conducted. The dashed line is the average MAE.

RMST performance after training ends

The following result was not intended to be formally part of the project but was conducted and reported on out of interest. At the conclusion of Year 2, the RMST training (using the on-ground calibrated RPM data) ended. However, there was still data available from one participating farm which enabled a very rough assessment of the performance of the RMST without the fortnightly training. The results (Table 9) provide a very rough indication of what happens to the accuracy once training ends, but care must be taken when considering these results are they are from a single farm with an extremely small sized data set. The informal analysis indicated that MAE increases once training ends (i.e. the error increases, or conversely the accuracy reduces). The reduction of MAE in August 2025 is likely due to ryegrass being the predominant pasture species at that time, and (as mentioned at the start of this report) these types of technologies were created on ryegrass-based pastures, so it isn't surprising that performance is higher at this time.

Table 9. Informal evaluation of the accuracy of the RMST once training concluded.

Data	N	MAE kg DM/ha	MAE %	MAEse kg DM/ha	CCC	R	R2
Total	13	542.4	21	139.1	0.111	0.21	0.04
Dec 2024	4	481.3	20	217.5	0.269	0.77	0.59
Feb 2025	5	800.1	26	233.9	0.068	0.33	0.11
Aug 2025	4	281.5	13	162.9	0.297	0.47	0.22

n = number of observations. MAE = Mean-absolute error expressed in kg dry-matter (DM) per ha and as a proportion of mean observed pasture biomass (%). MAEse = standard error. CCC = concordance correlation coefficient. R = correlation coefficient. R2 = coefficient of determination. Note: data is from a single farm only.

Farm performance (pasture productivity and utilisation)

Key result: Average pasture utilisation as a percentage of pasture grown was 58.9%. The average difference between pasture grown and harvested was just over 4,500 kg DM/ha meaning that farms grew approx. 4.5 t DM/ha more than they utilised.

Growth rates were used to calculate pasture production throughout the year, while grazing records and biomass estimates enabled the calculation of pasture harvested. This in turn determined pasture utilisation (what proportion of grown pasture was harvested) and presented in Table 10. There were a range of results between farms, reflecting the intricate interplay of factors such as climate (particularly considering the unusually wet conditions experienced during the project period), soil composition, management strategies, and stocking rates. The far north coast exhibited the highest production and utilisation, while the mid and south coast regions were more similar. Production averaged 11,181kg DM/ha across all farms. The results indicated high consistency in pre-grazing biomass between farms (i.e. average pre-grazing biomass was close to target pre-grazing biomass, set at 3,000 kg DM/ha), which may indicate that farmers were opening paddocks for grazing when pasture was close to target. However, it is important to consider that the tools used (both the RPM and RMST) rapidly lose accuracy once biomass approaches and exceeds 3,000kg DM/ha, meaning pre-grazing biomass variation may be masked and should be interpreted with caution.

Table 10. Average yearly pasture production, pasture harvested and pasture management results for all farms and regions in the project (across both years of the project).

Unit	Pasture Production	Pasture disappearance	Pasture use efficiency (%)	Pre-grazing biomass	Post grazing biomass
Average overall	11,330	6,773	58.9	2,901	2,152
By region					
North coast	13,452	8,868	65.5	2,816	2,049
Mid coast	10,780	6,307	58.3	2,941	2,208
South coast	10,403	5,751	54.4	2,913	2,155
By farm					
North coast					
Farm 01	11,119	6,744	61.0	2,830	2,003
Farm 02	16,330	11,376	70.0	2,891	2,129
Farm 03	14,120	9,357	66.0	2,788	2,081
Farm 04	12,238	7,996	65.0	2,753	1,982
Mid coast					
Farm 05	10,772	6,175	57.0	2,896	2,244
Farm 16	10,861	6,842	63.0	2,966	2,165
Farm 06	9,612	3,838	40.0	2,883	2,285
Farm 07	11,599	7,093	61.0	3,010	2,299
Farm 08	11,735	7,699	66.0	3,002	2,215
Farm 09	12,082	6,687	55.0	2,835	2,163
Farm 15	8,798	5,816	66.0	2,992	2,083
South coast					
Farm 10	10,539	5,569	53.0	2,902	2,160
Farm 11	8,103	3,231	40.0	2,944	2,314
Farm 12	12,138	8,569	71.0	2,975	2,014
Farm 13	10,453	6,143	59.0	2,910	2,125
Farm 14	10,783	5,241	49.0	2,835	2,163

Presented as kg DM/ha for all measurements (unless specified)

Grazing events were tracked using manual records and GPS collars. Including grazing events in the yearly pasture growth patterns enabled a visual depiction of pasture management and highlights pre- and post-grazing biomass levels (Figure 8). This visual depiction assists in identifying where pasture management is excelling (e.g. paddocks or seasons where biomass targets were met) and where there is room for improvement (e.g. paddocks or seasons where biomass targets were not met).

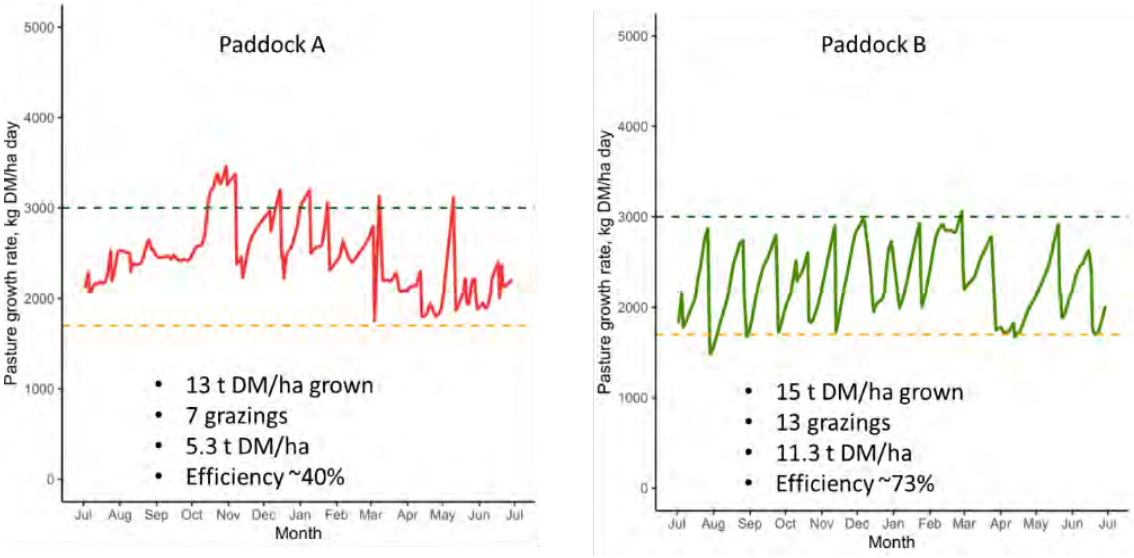

Figure 8. Example pasture management from two paddocks on the same farm (i.e. same management), showing the pasture biomass throughout the year, along with the pre- (green dotted line) and post-grazing (orange dotted line) biomass targets.

Relationship between pasture production and grazing performance

Key result: Pasture production strongly influenced total grazed, but differences between farms were mainly explained by pasture utilisation efficiency. Farms with higher pre-grazing pasture mass and lower post-grazing pasture level performed better, while farm size and paddock number had little effect.

To further understand differences between farms, the relationship between pasture production and total grazed was analysed (Figure 9). This analysis shows a strong and consistent pattern, where farms that produce more pasture generally harvested more pasture. The line on the figure represents the expected grazing outcome based on pasture production, indicating the average relationship observed across all farms.

While most farms follow this trend, there are clear differences in how closely individual farms align with the expected outcome. Some farms are positioned above the line, meaning they graze more pasture than expected for their level of production, while others fall below the line, indicating lower than expected utilisation. These differences highlight variation in grazing performance that cannot be explained by pasture production alone.

To better interpret these differences, farms were grouped based on both total grazed and their position relative to the expected line, which reflects how well pasture is being utilised. This allows separation of farms that achieve high grazing outcomes mainly through high production from those that achieve similar or better results through more effective use of pasture. The results suggest that grazing management plays a key role. In particular, farms that enter grazing with higher pasture mass and achieve lower post grazing residuals tend to perform better. In contrast, structural factors such as farm size or number of paddocks showed little influence on grazing performance.

Overall, the analysis reinforces that while pasture production sets the potential for grazing, differences between farms are largely driven by how effectively that pasture is utilised.

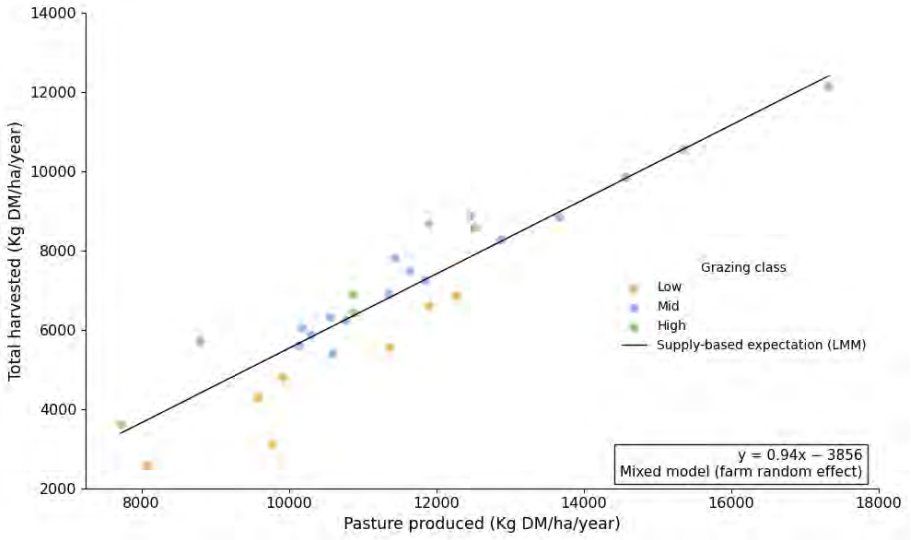

Figure 9. Relationship between pasture produced and total pasture harvested at the farm-year level. The line shows the expected level of harvesting based on pasture production.

Farmer perceptions and feedback

Key result: farmer confidence in the RMST increased by an average of 38% at the end of the project, and many indicated this type of technology has more than one purpose (i.e. more than just pasture monitoring).

Throughout the project, farmers were asked to indicate their confidence in the RMST (noting that confidence was contingent on both the RMST itself and the remote sensing approach, which can't be differentiated in the responses). Except for one farmer, farmers in the North Coast region generally improved their perception of the RMST. Similarly, nearly all farmers in the Mid Coast region reported an enhanced perception of the RMST. In contrast, farmers in the South Coast region reported a reduced perception, while Mid Coast farmers had mixed responses. Overall, confidence sat at 2.6/5 at this point.

At the conclusion of the project, farmers were sent individual farm reports which summarised their pasture production and utilisation. It also benchmarked their farm with those of their region, and the entire cohort of participating farms. After receiving their reports, farmers were given the opportunity to provide feedback on both the report and the project experience. Confidence in the RMST increased by 38% to 3.6/5 after farmers saw their individual report and the additional information the data could provide (outside what was available through the real-time RMST interface). Farmers also commonly indicated they would use the information from the RMST and presented in their report to assist with grazing and support reducing wastage, with more specific responses including:

- Assist with grazing (e.g. setting allocations, grazing more rigidly).
- Reduce wastage.
- Checking/confirming growth rates.
- Make decisions based on objective data (particularly useful in multi-manager set ups, and where managers/family members have different views on how to proceed).
- Can see peaks in growth/production and therefore support decision making and considerations into how to better use the excess pasture during those times.

Many farmers also found the RMST useful for other purposes, appreciating its capability as a comprehensive platform to document all farm activities. Notably, since pasture biomass is predominantly calculated from widely used remote sensing indexes such as NDVI and EVI, the RMST provided guidance to farmers in relative terms, allowing them to organise paddocks from highest to lowest biomass.

8. Outputs

The project produced several high-quality outputs, including:

- A Ph.D thesis (The Unrealised Potential of AI Solutions for Pasture-based Dairy Systems, Blessing Nnenna Azubuike).
- A high-quality pasture dataset spanning two years, collected using remote monitoring technology providing objectively measured pasture production, growth rates and pasture depletion data. Farm characteristics and grazing management information is also included. This is the first dataset of this nature compiled for NSW pasture-based farms that we are aware of.
- An informative, industry focused video outlining the key results of the project and application potential of those results on farm.
- Publication of research in multiple international journals, including Nature's Scientific Report (see list in Section 11).
- Conference presentations, industry reports and individualised participating farmer reports, disseminating findings to researchers, producers and industry audiences.

9. Applications and Impacts

The findings have direct implications for pasture-based dairy farms, not only in NSW but across Australia, and are particularly relevant for farms in regions that experience the C3-C4 winter-summer pastures, as previous work into remote pasture monitoring has focused largely on C3 pastures.

- **At the farm level**, the project demonstrated that the pasture monitoring technology tested could estimate pasture biomass with comparable accuracy as the RPM. To monitor the entire farm, the RMST required approx. 1.5h a fortnight of manual labour (to measure 5 paddocks with a RPM) for continuous training, a reduction in labour required for pasture monitoring using traditional methods. The technology was able to provide a clear and accurate estimate of pasture biomass (availability and depletion) in near-real time, and enabled visualisation of pasture performance and identification of well performing paddocks and paddocks that require additional attention to reduce inefficiency. This information could be used by farmers and advisors to support pasture management decisions and increase pasture use efficiency through increasing pasture production or increasing the amount of grown pasture which is consumed (e.g. more closely hitting pre- and post-grazing biomass targets).
- Being objective, this type of technology can help farm businesses maintain consistency in pasture management across different staff, aiding in stable pasture productivity and utilisation.
- **At the industry level**, the outcomes from this project offer a pathway forward for supporting increasing pasture utilisation, which has been identified as an important element of feedbase management. Farms in the project were found to grow, on average, 4.5 tonne DM/ha per year more pasture than they consumed through grazing, and "advanced monitoring" has huge potential to enable farmers to identify areas for improvement in their pasture management and work towards increasing pasture use efficiency (i.e. use more of what is grown).
- **At the technology level**, accuracy of tools needs to be factored into decision-making. Both tools (the RPM and RMST) were limited in their accuracy at very low (<1,500 kg DM/ha) and high (>3,000 kg DM/ha) biomass. However, pastures should be managed within that range to achieve best outcomes in terms of pasture nutrition and regrowth (reducing risk of under or over-grazing). The pasture species will still determine target biomass, along with the method of monitoring the pasture (e.g. type of tool).

10. Future Research Opportunities and Actions

- Work with farmers to implement objective monitoring practices on-farm and capture measurable gains in productivity, pasture use efficiency and profitability achieved through monitoring pasture (this project demonstrated that the average "gap" between pasture grown and pasture utilised was more than 4t DM/ha, meaning there are substantial gains to be made in pasture utilisation). Work should consider also aligning this information with milk production to ensure comprehensive monitoring of the entire farm system, aiding in discerning between suitable and poor strategic and tactical management decisions.
- One type of remote monitoring tool was evaluated in this project, although there are many more on the market. More work should be directed to better understanding the performance and reliability of other tools on the market so farmers can make informed decisions on how they may incorporate objective monitoring into their pasture management routines.
- Work with private consultants/agronomists to develop practical strategies for implementation of objective monitoring on dairy farms.

11. Project-wide Dissemination

The project has produced one PhD thesis, five peer-reviewed publications in high-impact international journals (Table 11).

Findings were also presented at multiple conferences and scientific events, including the American Dairy Science Association (ADSA) Annual Meeting in the USA, the Australasian Dairy Science Symposium (ADSS), and the Dairy Research Foundation Symposium (see Section 13 for details). These presentations collectively reached an estimated audience of thousands of people, spanning industry professionals, researchers, farmers and policymakers.

Beyond peer-reviewed and conference outputs, the project produced several other outputs. Of note was a video, highlighting the main findings and implications of the research as well as several project updates on the Dairy UP website and via the Dairy UP newsletter. Participating farmers were sent reports throughout the project, as well as a final report at the conclusion of the project detailing their farm's performance and providing benchmarking against other farms in the project. Farmers were also contacted and met with via phone call to discuss results and their perspective on the project, technology and farm data.

Table 11. Peer-reviewed publications.

Author	Title	Journal	Year Published
Correa-Luna, M. *et al*	Accounting for minimum data required to train a machine learning model to accurately monitor Australian dairy pastures using remote sensing	Nature, Scientific reports	2024
Azubuike, B.N., *et al*	Data Augmentation and Interpolation Improves Machine Learning-Based Pasture Biomass Estimation from Sentinel-2 Imagery	Remote sensing	2025
Azubuike, B.N., *et al*	Machine Learning for Grazing Event Detection and Pasture Utilisation Quantification from Sentinel-2 Data	Smart Agricultural Technology	2026
Azubuike, B.N., *et al*	Transfer learning and stacking ensembles for biomass estimation from smartphone imagery in pasture-based dairy systems	Journal of Agriculture and Food Research	2026
Azubuike, B.N., *et al*	AI in Pasture-Based Dairy Systems: Advances in Precision Feeding, Remote Sensing, and Grazing Management	Computer and Electronics in Agriculture	In review

12. Conclusions and Recommendations

The results of the evaluation and validation across a range of data collected on commercial dairy farms in NSW provided a rich dataset from which the following key messages, conclusions and recommendations can be taken.

- Clear and accurate estimates of pasture availability and depletion can be captured using remote monitoring technology (comparable to a calibrated RPM). This in turn can be used by farmers and farm advisors to make informed pasture management decisions, both predictively and retrospectively (i.e. predict future allocations/pasture availability, and review and address pasture management based on captured data such as pre- and post-grazing biomass).
- The RMST assessed had reduced accuracy initially. Following fortnightly training with calibrated RPM data (using 10% of the farms' grazing area), it produced pasture biomass estimates that were of comparable accuracy to the RPM (gold standard tool). This is encouraging as it indicates the RMST can be relied on, however still requires the approximately 1.5h a fortnight labour to generate the RPM data for training. This also does not guarantee that every technology on the market will perform the same way. It may be necessary to conduct additional research into understanding the performance of other commercially available products. Additionally, research into the performance of the RMST once training ends (i.e. had a period of training, which concludes) is recommended to better understand how training impacts performance if training is not planned to be continuous.
- Beyond approx. 2,800 kg DM/ha, pasture quality was seen to drop. This is in line with previous knowledge but sits slightly less than the standard 3,000 kg DM/ha target for pre-grazing biomass. For full results on pasture quality related to biomass, please see results from Project 1e "Understanding and predicting changes in nutritional value of Kikuyu" (https://dairyup.com.au/resource-library/). Importantly, the take home message here is that it is critical to graze (or harvest for silage) pasture before quality starts to drop and reinforced the key message of hitting pre-grazing biomass targets.
- Tools used in this project (both the RPM and the RMST) were limited in their accuracy at very low (<1,500 kg DM/ha) and high (>3,000 kg DM/ha) biomass. However, pastures should be managed within that range to achieve best outcomes in terms of pasture nutrition and regrowth (reducing risk of under or over-grazing), linking directly to the point above. The pasture species will still determine target biomass, along with the method of monitoring the pasture (e.g. type of tool).
- There is a need to work with farmers to implement objective monitoring practices on-farm and capture measurable gains in productivity, pasture use efficiency and profitability achieved through monitoring pasture (this project demonstrated that the average "gap" between pasture grown and pasture utilised was more than 4t DM/ha, meaning there are substantial gains to be made in pasture utilisation). Work should consider also aligning this information with milk production to ensure comprehensive monitoring of the entire farm system, aiding in discerning between suitable and poor strategic and tactical management decisions.
- Similarly, it is important to work with private consultants/agronomists to develop practical strategies for implementation of objective monitoring on dairy farms, recognising that farmers are likely to select technology that meets multiple farm and business needs, and so may not rely solely on accuracy when choosing a tool to support their pasture management.
- The concept of 'advanced monitoring' developed by this project can also help to improve management of C4 pasture for silage as well as help farmers to be more in control over the critical transition periods between C3 and C4 and vice versa. These will be assessed in Dairy UP2.

13. Annexes

Table 12. Conference presentations, abstracts, published material and other media engagements.

Authors	Title	Presentation Type	Conference/ Event	Location	Year	Audience
Dairy UP	Remote pasture monitoring P1a	Video		Online	2024	Industry/ farmer
Dairy UP	Improving pasture utilisation using satellite technologies	Case study		Online – Dairy UP	2024	Industry/ farmer
Sergio Garcia	The hidden losses of pasture utilisation	Written article	Magazine article	The Australian Farmer	2024	Industry/ farmer
Dairy UP	Mid-project insights: Remote pasture monitoring P1a	Project update		Online	2024	Industry/ farmer
Azubuike, B.N., *et al*	Machine learning increases the accuracy of satellite derived pasture cover estimation	Conference abstract	American Dairy Science Association	Louisville, KY, USA	2025	Scientific/industry
P1a team	Key results from Remote Pasture Monitoring Project (P1a)	Video		Online	**2025**	Industry/ farmer

14. References

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Final Project Summary

P1a – Kikuyu: Remote monitoring

May 2026

The power of pasture utilisation

Improving pasture utilisation is one of the most cost-effective ways Australian dairy farmers can improve business profitability. Dairy Australia (2026) estimates that increasing homegrown feed utilisation by one tonne per miking hectare would deliver $230m in total annual gross benefits for dairy farmers. Despite this, pasture utilisation on Australian dairy farms has stagnated in recent years.

Regular monitoring of available pasture and precise allocation of the herd's daily allowance is a key to improving pasture utilisation. Until recently, pasture monitoring involved the laborious and regular task of estimating coverage in paddocks using a rising plate meter or visual assessment.

Remote monitoring satellite technology

The emergence of remote monitoring satellite technology is a potential gamechanger by automating the task of regular pasture assessment.

The technology uses high resolution satellite images and local information (e.g. weather soil and farm management data) to help farmers monitor their pastures and make grazing management decisions. It can be used to predict pasture cover biomass and forecast production.

The technologies available were originally developed and validated for perennial ryegrass pastures.

Dairy UP's work aimed to expand the application of this technology to Kikuyu pastures for NSW dairy farms, working with a company supplying the technology commercially in Australia and overseas.

This project had three aims:

- To test and validate the reliability of a remote monitoring technology (Pasture.io) for Kikuyu pastures.
- To explore the potential to use remote sensing data to provide a snapshot of pasture production and utilisation to aid decision making.
- To support NSW dairy farmers to use the tool for improved pasture management.

Unlocking the potential of Kikuyu

Dairy UP's P1 project aimed to unlock the potential of Kikuyu pastures used by NSW dairy farmers. P1 was a suite of five projects that collectively explored new management options to grow and utilise more Kikuyu over summer and increase the productivity of Kikuyu-based pastures.

P1a – Kikuyu: Remote monitoring

P1b – Kikuyu Investigating toxicity

P1c – Kikuyu: Developing new varieties

P1d – Kikuyu: Carbon on NSW dairy farms

P1e – Kikuyu: Nutritional value

This document is the final update on P1a – Kikuyu: remote monitoring.

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DairyUP
Unlocking potential

Key findings

The project confirmed that this remote monitoring satellite technology can measure Kikuyu-based pasture biomass with comparable predictive precision to a calibrated rising plate meter.

In addition to monitoring pasture in every paddock across the farm, it also provided information that can be used to quantify potential pasture losses, highlight inefficiencies and support decisions around productivity and utilisation.

The more the pasture grown, the more the pasture utilised by grazing. But pre- and post-grazing biomass and grazing frequency also influenced pasture utilisation, while farm size did not.

The current need for fortnightly training/calibration of the technology is a significant barrier to adoption.

More info

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Research and results

Monitor farms

The Dairy UP team worked with a network of 16 dairy farmers in the north, mid and south coast NSW regions to adapt the Pasture.io monitoring system for Kikuyu-based pastures.

Over a two-year period, farms were monitored with the remote monitoring technology as well as physical monitoring of pasture cover, composition, quality and management. The team visited regularly to measure pasture (with a rising plate meter), collect samples and record observations about farm management activities and farmer perceptions about the technology.

The rising plate meter pasture cover results were compared with the predictions made by the remote monitoring technology.

Participating farmers received reports outlining their farm's performance (including benchmarking with their region and all farms in the project). They provided feedback on their experience and value of the information generated.

Pasture utilisation

The project determined that, on average, pasture utilised was 4 tonnes DM/ha *less* than pasture grown. This represents a significant opportunity for NSW dairy farmers to boost the efficiency, and therefore potential profitability, of their systems (Figure 1).

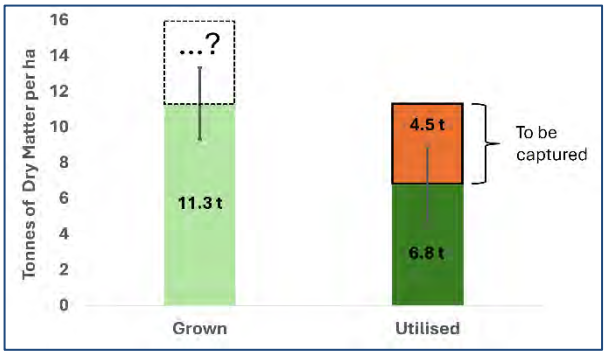

Figure 1: Up to 4 tonne DM/ha extra feed is potentially capturable; and that is without considering the potential to grow more [left bar].

Analysis showed that pasture utilisation varied between farms. Pasture grown was closely related to pasture consumed (Figure 2, over page). However, the amount of pasture consumed varied between farms with similar pasture production. Pre- and post-grazing biomass, and grazing frequency, explained some of the variation while characteristics such as farm size did not.

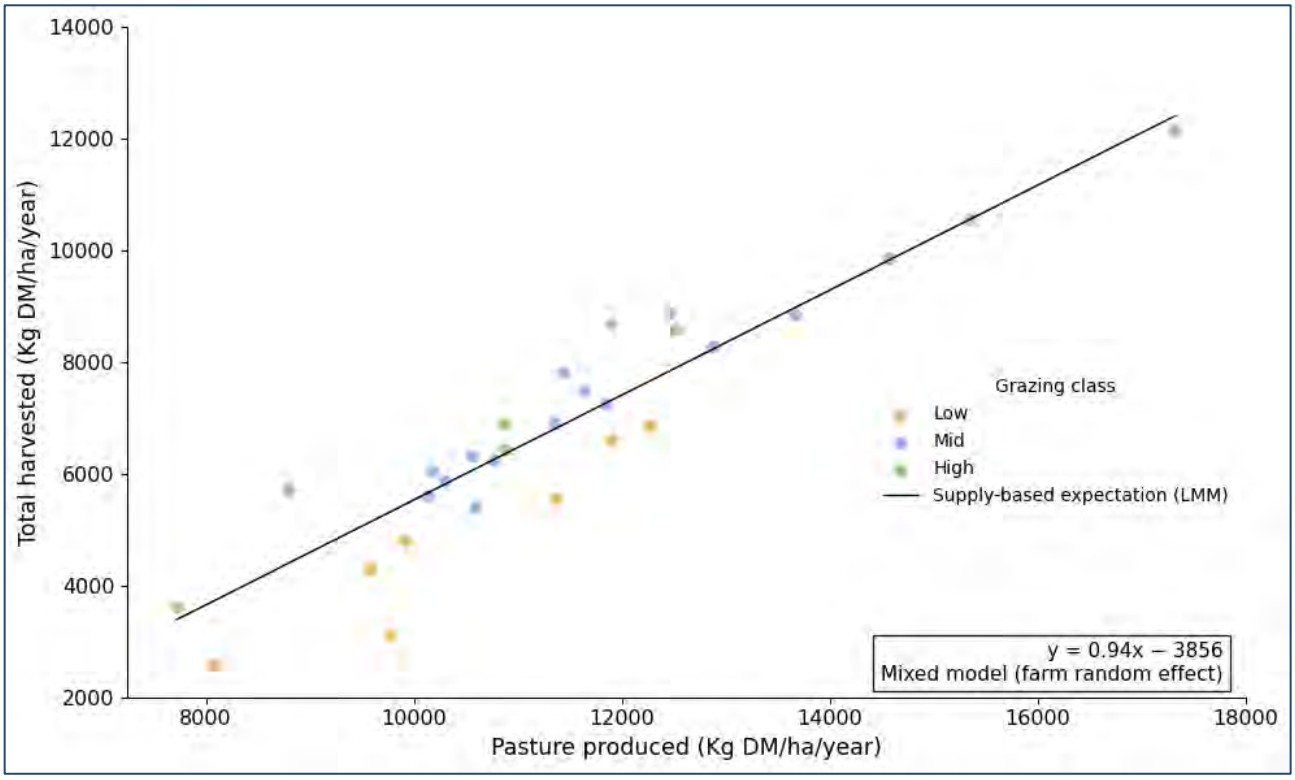

Figure 2: Although pasture grown is a key driver of pasture harvested, some farms achieved higher utilisation than others with similar pasture production highlighting the importance of management.

Remote monitoring satellite technology
The project confirmed the satellite technology tested can measure pasture biomass with comparable predictive precision to a calibrated rising plate meter. However, this level of accuracy was only achievable with fortnightly training of the satellite technology (involving measuring five paddocks with a rising plate meter). The time involved in measuring the paddocks with a rising plate meter was about 1.5 hours fortnightly. While this is relatively low compared to the potential benefits, it is a significant barrier to adoption.

The information from the trained remote monitoring satellite technology can aid in highlighting biomass and paddock variability and opportunities to improve pasture production and utilisation.

Confidence in technology
On average, farmers' confidence in the technology increased by 38% over the two years of the project.

Participating farmers acknowledged that the information presented in their individualised report supported management decisions, offered reassurance and helped identify opportunities for improvement.

Read more

P1a Final Report

Dairy Australia Strategic Plan 2030

Martin Correa-Luna M. et. al., Accounting for minimum data required to train a machine learning model to accurately monitor Australian dairy pastures using remote sensing. Nature Scientific Reports (2024) 14:16927

Azubuike, B.N. et.al. A data-driven approach for optimising supplement allocation to individual lactating dairy cows in pasture-based systems, Smart Agricultural Technology, Volume 12, 2025, 101669

Azubuike, B.N. et.al. Data Augmentation and Interpolation Improves Machine Learning-Based Pasture Biomass Estimation from Sentinel-2 Imagery. *Remote Sens.* **2025**, *17*, 3787

Azubuike, B.N. et.al. Machine learning for grazing event detection and pasture utilisation quantification from sentinel-2 data, Smart Agricultural Technology, Volume 14, 2026, 101954

DairyUP
Unlocking potential

Delivery organisations

The University of Sydney – Dairy Research Foundation

NSW Government – Department of Primary Industries and Regional Development

Scibus

Dairy Australia

Partner organisations

NSW Farmers – Growing the best

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Hunter Local Land Services
Dairy UP P1a Case Study

Improving pasture utilisation using satellite technologies

April 2024

Introduction

Dairy Up's P1- Unlocking the potential of kikuyu-based systems project has been investigating the benefits of using remote sensing and data-based management (P1a) technologies to optimise pasture growth, quality and utilisation. It aims to increase farmer use of real-time monitoring systems to better inform grazing and harvesting decisions that maximise year-round feed production and quality.

The team has partnered with 15 farms along the NSW coastal dairy regions, from Bega to Lismore to ground truth satellite data, input farm data to better inform the tool, and upskill local farmers in the technology.

Pasture.io is one of several decision-support tools that use satellite-derived pasture measurement. The company is working with the Dairy Up team to modify the technology for dairy farms using kikuyu/ryegrass mixed pasture systems. Each of the fifteen farms is supported by a local project technical officer.

Pasture.io maps and predicts pasture growth rate (Kg DM/ha.day) and current available feed (Kg DM/ha or TDM) through the use of satellite imagery, soil characteristic, weather and farm management data. Accessible via smartphone, this decision-support tool allows both farmer and advisor to enter information on daily pasture management.

To further enhance accuracy of Pasture.io for NSW mixed pasture system, ground-based rising plate measurements are also taken fortnightly using electronic plate meters. The data is helping in two ways:

- To train the models that underpin the technology by entering the paddock-level data that is collected manually.
- To validate the accuracy of Pasture.io by comparing the satellite measurements against manual measurements.

The farmer- Tim Bale

Tim Bale operates a year-round calving herd of 280 cows on a 150-hectare mixed pasture system at Stewarts River on NSW's mid-coast. He is working with local Project Officer, Josh Hack (Ag Farming Systems) to trial Pasture.io over 18 months.

Tim said that grazing rotation can often be guesswork. "Pasture.io has given me something to compare my decisions against at this stage. It has certainly focused my rotations- what I'm grazing and how much pasture I am allocating to the herd," Tim said.

Tim Bale and Josh Hack inspect the pasture quality after taking the fortnightly pasture plate meter readings.

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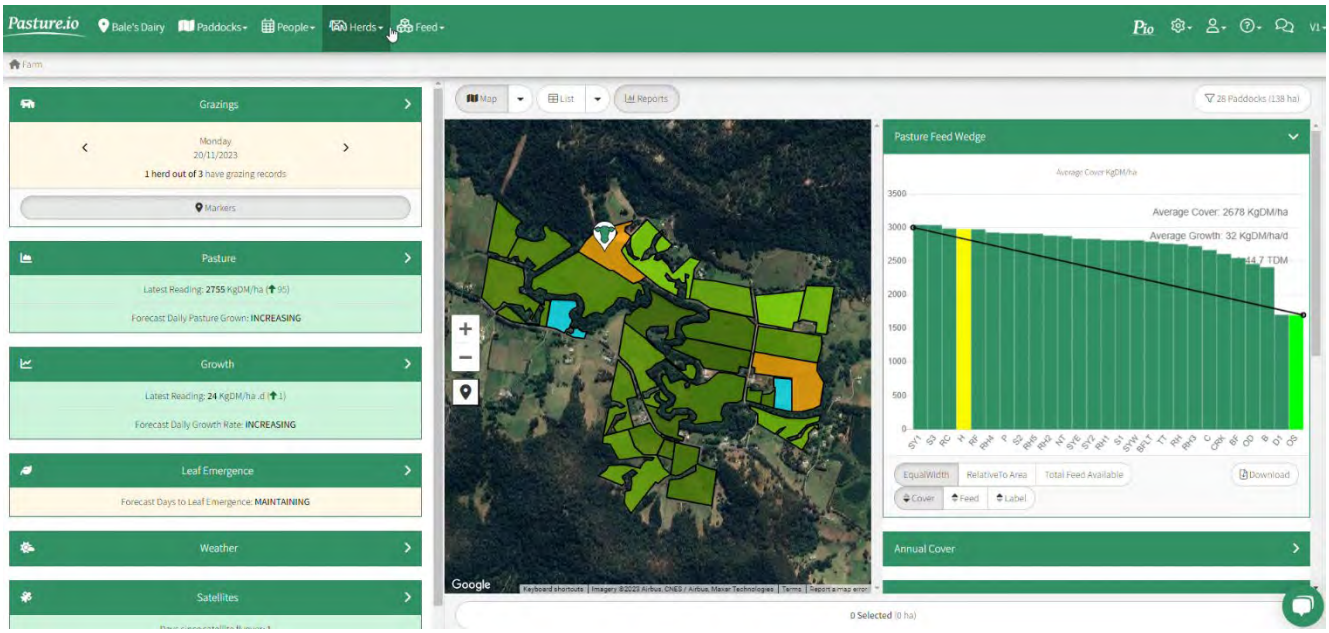

The Pasture.io dashboard shows current pasture availability and forecast information per paddock as a feed wedge.

Tim said he had more confidence in the decisions he made on where to graze next and where to cut silage as he can easily determine what feed he has ahead if he allocates paddocks to be grazed or taken out of the rotation.

"The feed wedge is a confidence booster. I can select 'milkers only' and it will show the milking platform and I can click on paddocks to take-out for silage at this time of year, then it automatically recalculates the new milking area."

Pasture.io tracks cow movements spatially and daily by GPS. With a myriad of daily decisions to be made, this removes pressure upon Tim's memory bank''.

"I like the GPS on the cows as I have an accurate record of where they have been and how long they have been there. I can easily find out when I last grazed certain paddocks."

> *"The feed wedge is a confidence booster."*
>
> Tim Bale, Stewarts River, NSW

The advisor- Josh Hack

From the advisor perspective, Josh Hack, of Ag Farming Systems, said having his clients use Pasture.io allowed him to provide a more effective and efficient service.

"I can come onto farm and together we can easily bring all the information up on my phone. I can see where the cows have been and what we have on-hand. Together, Tim and I can chat things over whilst in the paddock or in his office," he said.

While Tim has yet to explore the full capabilities of the system, he and Josh see real potential in moving paper-based nutrient application records across to Pasture.io.

"The benefit is that Pasture.io quickly tallies fertiliser applications per nutrient. For example, we can accurately see how much nitrogen has gone onto individual paddocks or across the farm to monitor against both the farm nutrient and financial budgets," Josh said.

Tim has off-farm business interests so Pasture.io is potentially a good tool for him to communicatewith his employees.

"I want to set-up Pasture.io access on their smartphones so we are all on the same page

Figure 3. Josh Hack and Tim Bale review the paddock data of Pasture.io in the farm office.

when it comes to allocating paddocks and tracking nutrient inputs. I can also start to empower my employees to make informed decisions if I can't be contacted."

Accuracy of the technology

While Pasture.io has been developed to predict pasture growth rate (KgDM/ha.day) and current available feed (KgDM/ha or TDM), both Josh and Tim believe that it's not entirely accurate….yet.

"The absolute figures are not necessarily reflective of what we are seeing in the manual readings, though what is really accurate is the trends," Tim said.

"What Pasture.io shows as having the most feed is always the paddock we know we should be feeding next. So, the feed wedge trend is definitely accurate."

Josh said there is no doubt the project was improving the accuracy of Pasture.io on mixed pasture systems.

"The plate meter data is being entered into Pasture.io and this is definitely improving the accuracy of the model. Effectively what we are doing is helping to calibrate the machine learning of the platform so Pasture.io can become a useful tool for review and improvement across these systems in NSW."

Results to date

The Dairy UP team has used 12 months of calibrated ground-based data (e.g., rising plate meter readings) to improve the machine learning capabilities of Pasture.io. The results demonstrate enhanced accuracy of the tool. Well-managed pastures haven been shown to be measured by Pasture.io with an accuracy of +/- 250350 kg DM/ha. This comparison has been made using entire paddock imagery data from Pasture.io versus specific transect data from paddocks. Therefore, it may be likely that even greater accuracy is being achieved.

Farmers and advisors involved in the project are excited about the potential to have accurate current and forecasted available feed information at their finger-tips in mixed pasture systems.

"There is no doubt production and profitability w350ill benefit from me and the staff being able to make more informed and confident decisions, " Tim said.

More information

Dairy Up Website

Watch a video

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Machine learning for grazing event detection and pasture utilisation quantification from sentinel-2 data

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ARTICLE INFO

Keywords:
Anomaly detection
Biomass
Cross-year validation
Semi-supervised learning
Temporal transferability

ABSTRACT

Effective pasture management in pasture-based dairy systems (P-BDS) requires accurate knowledge of when and where grazing occurs and how much biomass is consumed, yet manual recording methods are time consuming and prone to error. This study compared machine learning approaches for automated grazing event detection and pasture utilisation quantification from both rising plate meter and interpolated Sentinel-2 satellite data. Performance was evaluated against GPS-tracked cow grazing records across 12 commercial dairy farms in New South Wales, Australia (July 2022–June 2024). Eleven approaches were evaluated across within-year and cross-year validation scenarios to assess temporal transferability. Random Forest achieved optimal within-year detection performance (F1 score = 0.878, precision = 0.938, recall = 0.825), whilst One-Class Support Vector Machine (OCSVM) demonstrated superior cross-year transferability (F1 score = 0.692), outperforming supervised models by 7.6% on independent Year 2 data despite supervised models experiencing average performance degradation of 24.2%. Farm-level detection variability ranged from F1 = 0.500 to 0.815, with site-specific factors exerting stronger influence than regional characteristics. Independent pasture utilisation agreement validation on 218 GPS-confirmed events demonstrated strong biomass quantification concordance between satellite and ground measurements (pre-grazing biomass $R^2 = 0.966$; post-grazing biomass $R^2 = 0.998$; biomass removal $R^2 = 0.922$), establishing proof-of-concept that satellite systems can both identify when grazing occurs and accurately quantify biomass changes. Temporal alignment constraints between satellite revisit schedules and ground measurements limited validation to 19.4% of total events, highlighting the need for daily or near-daily observations through multi-sensor fusion approaches to achieve deployment in commercial P-BDS.

1. Introduction

Effective pasture management in dairy systems requires accurate knowledge of when and where grazing occurs, as this information directly influences feed budgeting, rotational grazing schedules and overall farm productivity [1,2]. In pasture-based dairy systems (P-BDS), the ability to detect grazing events (GE) rapidly and accurately, combined with precise quantification of pasture utilisation (PU), enables farmers to optimise forage management and improve economic returns. Pasture utilisation refers to the amount of herbage mass consumed during a GE, typically quantified as biomass removal (kg DM ha^{-1}) and harvesting rate (%), calculated as (pre-grazing biomass − post-grazing biomass) / pre-grazing biomass × 100. Accurate PU quantification enables farmers to optimise stocking rates, ensure adequate residuals for pasture recovery, balance feed demand with supply, and improve feed budgeting accuracy. Regular monitoring of herbage mass combined with accurate records of grazing timing and intensity can improve farm profitability by up to 15 % through better alignment of pasture availability with animal requirements and reduced wastage from overgrazing or under utilisation [1]. Traditional approaches to recording GE rely on manual documentation by farmers, a process that is time consuming, prone to error and often incomplete, particularly on larger operations managing multiple paddocks simultaneously [3]. As precision agriculture technologies advance, automated methods for detecting GE could

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A data-driven approach for optimising supplement allocation to individual lactating dairy cows in pasture-based systems

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ARTICLE INFO

Keywords:
Differential evolution
Feed optimisation
Individual concentrate allocation
Machine learning
Pasture-based systems

ABSTRACT

With feed costs accounting for about 40-60 % of milk production expenses in Australia, efficient supplementary concentrate allocation is crucial for profitability. Despite an increase in concentrate use per cow over the past decade, the average milk yield response remains about 1 L per kg of dry matter concentrate. While machine learning and data-driven optimisation are widely utilised in sectors such as engineering, healthcare, and finance, their application in feed optimisation within dairy farming has not been extensively researched. This study aims to develop a machine learning-based method to optimise individual cow supplement allocation, using similar total daily concentrate with a tolerance range allowing for a 2-10 % decrease, to maximise milk yield. Data from a controlled field study involving 130 lactating Holstein-Friesians (32,504 records) were analysed.

Sixteen machine learning algorithms were evaluated to predict milk yield based on concentrate allocation and available cattle data (days in milk, daily milk yield and liveweight, and parity number). The Random Forest (RF) model was the best performer, achieving an R^2 of 0.60 and RMSE of 4.20 L/cow/day. Then 7371 records from 81 cows over 91 days were employed to run the concentrate levels optimisation using the Dirichlet-Rescale (DRS) algorithm and Monte Carlo simulation. The RF model and SciPy optimisation determined optimal individual cow allocations (5-9 kg/cow.day^{-1}). Implementing this resulted in a herd-level 8 % increase in daily milk yield. This study highlights the potential benefits of adopting data-driven algorithms for individualised dairy feed optimisation based on observed correlations within existing management practices. While results suggest improvement over conventional flat-rate methods, the study is limited by the nature of the dataset, and findings reflect associations under standard practice rather than experimental manipulation of concentrate levels, requiring validation through controlled field trials to confirm practical efficacy and economic impact in actual dairy farming contexts.

1. Introduction

Pasture-based dairy farms are the predominant dairy production systems in Australia. In these systems, the cost of milk production is inversely related to profitability [1]. Feed accounts for about 40-60 % of the cost of producing milk and grain-based concentrates are typically 3 to 5 times more expensive per kg DM than pasture in Australia [2,3]. Cow nutrition is affected by seasonal variations in both pasture quality and quantity, weather-related fluctuations. To counter this, supplementary feed, including fodder and concentrates are used by dairy farmers to uphold cow health and production during periods of low pasture availability. Over the last 10-15 years, there has been a notable increase in the allocation of supplementary grain-based concentrates (including grain/s, mixes of grains, commercial pellets) per cow [4–6]. Despite the potential for higher milk production (1 kg DM of concentrate contains 11-13 MJ of Metabolisable Energy (ME), which is equivalent to the ME required for 2 to 2.2 litres of milk), the average response of individual cows remains at approximately ~1 L of milk per kg DM of concentrate [7].

There are two major concentrate feeding strategies adopted by dairy farmers. The 'flat rate' feeding strategy, where every cow in the herd is allocated the same average amount of supplement whilst being milked, is used by about 78 % of dairy farms in Australia and New Zealand [8]. The alternative strategy is the individualised feeding system, where each

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remote sensing

MDPI

Article

Data Augmentation and Interpolation Improves Machine Learning-Based Pasture Biomass Estimation from Sentinel-2 Imagery

Blessing N. Azubuike [1,2,*], Anna Chlingaryan [2,3], Martin Correa-Luna [1,2], Cameron E. F. Clark [2,4] and Sergio C. Garcia [1,2]

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Highlights

What are the main findings?

- Full-band Sentinel-2 reflectance combined with weather variables substantially improved pasture biomass prediction accuracy, outperforming vegetation indices and achieving up to 70% variance explained after interpolation.
- Multiquadric interpolation and progressive temporal training strengthened temporal consistency in the data, reducing prediction errors by approximately 30% relative to sparsely sampled baselines.

What are the implications of the main findings?

- These findings demonstrate that integrating physically meaningful spectral information with biologically constrained data augmentation enhances the reliability and scalability of satellite-based biomass estimation across farms and seasons.
- The resulting open-source modelling framework provides a robust foundation for real-time pasture monitoring and can be readily incorporated into automated decision-support tools for precision grazing management.

check for updates

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Abstract

Accurate pasture biomass (PB) estimation is critical for tactical grazing management, yet traditional satellite-derived vegetation indices such as Normalised Difference Vegetation Index (NDVI) saturate when canopy density exceeds about 3 t DM ha^{-1}. This limits predictive accuracy because the spectral signal plateaus under dense vegetation, masking further biomass increases. To address this limitation, this study integrated multiple data sources to improve PB estimation in dairy systems. The dataset combined Sentinel-2 spectral bands, rising plate-meter (RPM) PB measurements, daily weather data, and paddock management features. A total of 3161 paired RPM–satellite observations were collected from 80 paddocks across 16 New South Wales dairy farms between November 2021 and July 2024. Eight regression algorithms and four predictor configurations were evaluated using robust cross-validation, including an 80:20 farm/paddock-stratified train–test-set split. The XGBoost model using full-band reflectance and concurrent weather data achieved strong baseline performance (R^2 = 0.63; MAE = 243 kg DM ha^{-1}) on non-interpolated data, outperforming NDVI-based models. To address temporal gaps between field readings and

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Accounting for minimum data required to train a machine learning model to accurately monitor Australian dairy pastures using remote sensing

Martin Correa-Luna[1]✉, Juan Gargiulo[2], Peter Beale[3], David Deane[3], Jacob Leonard[3], Josh Hack[4], Zac Geldof[5], Chloe Wilson[1] & Sergio Garcia[1]

Precision in grazing management is highly dependent on accurate pasture monitoring. Typically, this is often overlooked because existing approaches are labour-intensive, need calibration, and are commonly perceived as inaccurate. Machine-learning processes harnessing big data, including remote sensing, can offer a new era of decision-support tools (DST) for pasture monitoring. Its application on-farm remains poor because of a lack of evidence about its accuracy. This study aimed at evaluating and quantifying the minimum data required to train a machine-learning satellite-based DST focusing on accurate pasture biomass prediction using this approach. Management data from 14 farms in New South Wales, Australia and measured pasture biomass throughout 12 consecutive months using a calibrated rising plate meter (RPM) as well as pasture biomass estimated using a DST based on high temporal/spatial resolution satellite images were available. Data were balanced according to farm and week of each month and randomly allocated for model evaluation (20%) and for progressive training (80%) as follows: 25% training subset (1W: week 1 in each month); 50% (2W: week 1 and 3); 75% (3W: week 1, 3, and 4); and 100% (4W: week 1 to 4). Pasture biomass estimates using the DST across all training datasets were evaluated against a calibrated rising plate meter (RPM) using mean-absolute error (MAE, kg DM/ha) among other statistics. Tukey's HSD test was used to determine the differences between MAE across all training datasets. Relative to the control (no training, MAE: 498 kg DM ha^{-1}) 1W did not improve the prediction accuracy of the DST ($P > 0.05$). With the 2W training dataset, the MAE decreased to 342 kg DM ha^{-1} ($P < 0.001$), while for the other training datasets, MAE decreased marginally ($P > 0.05$). This study accounts for minimal training data for a machine-learning DST to monitor pastures from satellites with comparable accuracy to a calibrated RPM which is considered the 'gold standard' for pasture biomass monitoring.

Keywords Machine learning, Model training, Remote sensing, Pasture grazing management

Pasture-based dairy systems remain appealing for a number of reasons, including the initial low investment needed on infrastructure[1] and their relatively low reliance on market commodity prices[2]. They are also socially well-perceived because of their positive environmental and animal welfare reputation[3], superior food nutritional quality produced from human-inedible feed[4–6], and the opportunity to be implemented where human edible crops cannot be economically cultivated[6]. For these systems to remain profitable, pasture conversion efficiency into milk would need to be maximized while maintaining a low production cost[7]. For this, it is especially important to have a balance between feed demand and supply while maintaining high proportion of grass eaten by lactating cows[2,8,9]. Since pasture is a 'living feed source', thus perishable, its quantity and quality changes throughout

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DairyUP
Unlocking potential

Final Report

P1b – Kikuyu: Investigating toxicity

Department of Primary Industries and Regional Development

This project was led by Krista Plett and Barbara Brito Rodriguez from DPIRD - EMAI

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

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1. Executive Summary

Kikuyu grass (Cenchrus clandestinus) is a vital forage species for Australian dairy and beef industries, particularly in coastal New South Wales and Queensland. However, its widespread adoption has been constrained by kikuyu poisoning, a sporadically occurring, potentially fatal syndrome in cattle characterised by forestomach necrosis and severe dehydration, with mortality rates up to 32%. Despite decades of reports, the causative agent has remained unknown.

This project, comprising three complementary subprojects, applied cutting-edge fungal microbiome sequencing and untargeted metabolomics to investigate the biological basis of kikuyu toxicity. Together, the studies examined five kikuyu genotypes under controlled drought and rehydration conditions, tracked the behaviour of the suspected fungal agent *Fusarium torulosum,* and compared the microbiome and metabolite profiles of pastures actively causing cattle poisoning with healthy farms across NSW.

Key outcomes across the three subprojects are:

- Sudden rehydration after drought, rather than drought itself, is the dominant trigger of biological change in kikuyu grass, consistent with field observations of poisoning events.
- *F. torulosum*, long implicated as the causative agent, was found to decline significantly after rehydration and was not consistently enriched in toxic pastures. The mycotoxin wortmannin, attributed to this fungus, was not detected in any sample across the pot studies.
- Specific metabolite signatures, including seven compounds uniquely elevated in both leaf and stem of actively toxic pastures, were identified in the field study. These represent the first candidate toxic compounds identified in kikuyu using untargeted metabolomics from real poisoning events.
- A distinct fungal species, *Acremonium polychromum*, was significantly elevated in leaf, stem, and soil of toxic pastures and is capable of producing toxic alkaloids, making it a strong candidate for further investigation.
- Kikuyu genotype significantly influenced metabolite profiles, suggesting that some cultivars may carry inherently lower toxicity risk, an important finding for future breeding programs.

These results collectively redirect the field away from *F. torulosum* as the primary causal agent and toward a more nuanced picture involving plant stress responses to overwatering and specific fungal interactions. The findings provide a scientific foundation for developing practical tools for farmers, including early-warning metabolite markers and evidence-based pasture management guidance.

2. Project Overview

Item	Description
Project Title	Kikuyu: Investigating toxicity
Funding Body	Dairy Australia, University of Sydney, NSW DPIRD
Dairy UP Project	P1c
Project Duration	
Subprojects	1. Effect of Water Stress on Microbiome and Metabolite Profiles in Three Kikuyu Genotypes 2. Response of Fusarium torulosum to Water Stress in Five Kikuyu Genotypes 3. Fungal Profiles Associated with Kikuyu Exhibiting Toxicity Field Study
Lead Organisation	DPIRD - EMAI, USyd
Project Lead	Krista Plett, Barbara Brito Rodriguez
Key Collaborators	Vivien Tan (PhD), Percy Wong, Richard Trethowan, Pedro Pincowski, Aki Kawasaki

3. Abbreviations

CV — Coefficient of Variation
ASV — Amplicon Sequence Variant
DADA2 — Divisive Amplicon Denoising Algorithm 2
EMAI — Elizabeth Macarthur Agricultural Institute
ESI — Electrospray Ionisation
ITS — Internal Transcribed Spacer (fungal barcode region)
LC-MS/MS — Liquid Chromatography tandem Mass Spectrometry
OUT — Operational Taxonomic Unit
PCA — Principal Component Analysis
PCoA — Principal Coordinates Analysis
PERMANOVA — Permutational Multivariate Analysis of Variance
PLS-DA — Partial Least Squares Discriminant Analysis
WHC — Water Holding Capacity

4. Project Background and Rationale

Kikuyu grass is one of the most important pasture species for Australian dairy and beef producers, particularly along the coastal regions of New South Wales and Queensland. Its drought tolerance, rapid growth, and adaptability to acidic soils make it highly valued in grazing systems. However, the threat of kikuyu poisoning has long restricted farmer confidence in the species.

Kikuyu poisoning presents as a sudden-onset syndrome in cattle, characterised by forestomach necrosis, severe dehydration, salivation, incoordination, and ruminal fluid accumulation. Mortality rates can reach 32%, with significant economic consequences for affected farms. The syndrome typically occurs following rapid grass regrowth after heavy rain or irrigation that breaks a prolonged dry period, a pattern that is becoming more frequent with the increasing intensity of drought-and-flood cycles driven by climate change.

Despite reports of kikuyu poisoning dating back to the 1960s across Australia, New Zealand, South Africa, and Kenya, the causative agent has never been definitively identified. Earlier research using culture-based methods proposed *Fusarium torulosum*, a fungus capable of producing the mycotoxins wortmannin and butanolide, as a likely cause. However, this fungus has not been consistently found in toxic pastures across studies, and no definitive mechanistic link has been established.

This project was motivated by the need to apply modern, culture-independent genomic and metabolomic tools to comprehensively characterise the microbiome and chemistry of kikuyu grass under conditions that mirror real poisoning events. By combining controlled pot experiments with field sampling from active poisoning events, the project aimed to identify the biological signatures of kikuyu toxicity and test the *F. torulosum* hypothesis rigorously.

5. Project Objectives

The overarching objective of this project was to determine the biological basis of kikuyu poisoning in cattle by characterising the fungal microbiome and metabolite profiles of kikuyu grass under toxic and non-toxic conditions. Specific objectives were:

- To determine how drought stress followed by sudden rehydration affects the fungal community and metabolite profiles of different kikuyu genotypes under controlled conditions.
- To test whether *F. torulosum* abundance and wortmannin production are influenced by water stress and rehydration, and whether this is consistent with its proposed role as the causative agent.
- To compare the microbiome and metabolome of kikuyu grass collected from active kikuyu poisoning events with historically affected and unaffected pastures across NSW.
- To identify specific fungal taxa or metabolites consistently associated with confirmed toxic kikuyu, providing candidate targets for further mechanistic investigation.
- To assess whether kikuyu genotype influences toxicity-relevant biological responses.

6. Subproject P1b.1 – Effect of Water Stress on Microbiome and Metabolite Profiles in Three Kikuyu Genotypes

Research team: Vivien Tan, Percy Wong, Richard Trethowan, Barbara Brito & Krista L. Plett

6.1 Background

Kikuyu poisoning events are consistently reported following rapid grass regrowth after a prolonged dry period, suggesting the environmental trigger, drought followed by rehydration, plays a central role. However, it was unknown whether this pattern reflects a change in the fungal microbiome, a shift in plant chemistry, or both. Earlier research had also speculated that kikuyu poisoning may vary between different cultivars, but no systematic comparison had been made.

This subproject aimed to establish a baseline understanding of how drought stress and rehydration alter the fungal community and metabolite profiles in three kikuyu genotypes under controlled conditions. It was designed to test whether the environmental conditions associated with poisoning events produce measurable biological changes in kikuyu, and whether those changes differ by genotype.

6.2 Objectives

- To characterise shifts in fungal microbiome composition in soil, root, stem, and leaf tissues of three kikuyu genotypes following drought stress and rehydration.
- To identify metabolites significantly altered by water treatment and rehydration.
- To determine whether genotype influences microbial or metabolite responses to water stress.
- To test whether the conditions associated with kikuyu poisoning produce biological changes consistent with a toxicity mechanism.

6.3 Materials & Methods

A pot experiment was conducted at the Elizabeth Macarthur Agricultural Institute (EMAI), NSW, from November 2022 to June 2023. Three kikuyu genotypes: Whittet (commercial cultivar) and two breeding lines (lines 11 and 13), were established in 1.1 L pots using commercial potting mix with 1% soil from a kikuyu paddock with a history of poisoning to introduce a field soil microbiome. Six replicate pots per genotype were assigned to each of two treatments after four months of establishment.

Treatments consisted of a reduced-watering group maintained at 30% field capacity and a control group maintained at 80% field capacity. A gradual conditioning period was applied prior to treatment commencement to avoid physiological shock. After six weeks of treatment, pots were rehydrated to full saturation for one week to simulate drought-breaking rainfall. Grass was trimmed fortnightly throughout to maintain approximately 15 cm height.

Soil, root, stem, and leaf samples were collected before and after the rehydration week and stored at −80°C. Fungal community composition was analysed using ITS amplicon sequencing on the Illumina MiSeq platform, with taxonomy assigned against the UNITE fungal database. Metabolite profiling was conducted by untargeted LC-MS/MS using a Sciex ExionLC AD system coupled to a TripleTOF 6600+ mass spectrometer in both positive and negative electrospray ionisation modes. Data were processed using QIIME2/DADA2 for microbiome and MSDial/MetaboAnalystR for metabolomics, with statistical analysis including PERMANOVA, ANOVA with Tukey post-hoc, and PLS-DA.

6.4 Key Findings

- Based on the PERMANOVA results, rehydration, not drought, was the dominant driver of change in the fungal microbiome across leaf, stem, and root tissues. Timepoint (before vs. after rehydration) was the most significant factor in leaf and stem while genotype was the most significant factor in root and soil.
- Genotype was the strongest factor influencing metabolite profiles. Genotypes 11 and 13 were characterised by elevated amino acids, organic acids, and lipids, while genotype 15 showed higher cholines, dicarboxylic acids, and alpha-diketones.
- Water stress produced limited treatment-specific metabolite changes. In leaf tissue, alkaloids and nitrogen-containing compounds were slightly elevated under the reduced-watering treatment. In stem tissue, six metabolites (some only putatively identified), including amino acids, hydroxy fatty acids, and aldoximes were higher under drought conditions.
- A total of 174 metabolites were significantly elevated in genotypes 11 and 13 (leaf tissue), and 108 in genotype 15, suggesting a strong effect of the genetic background on the metabolome.

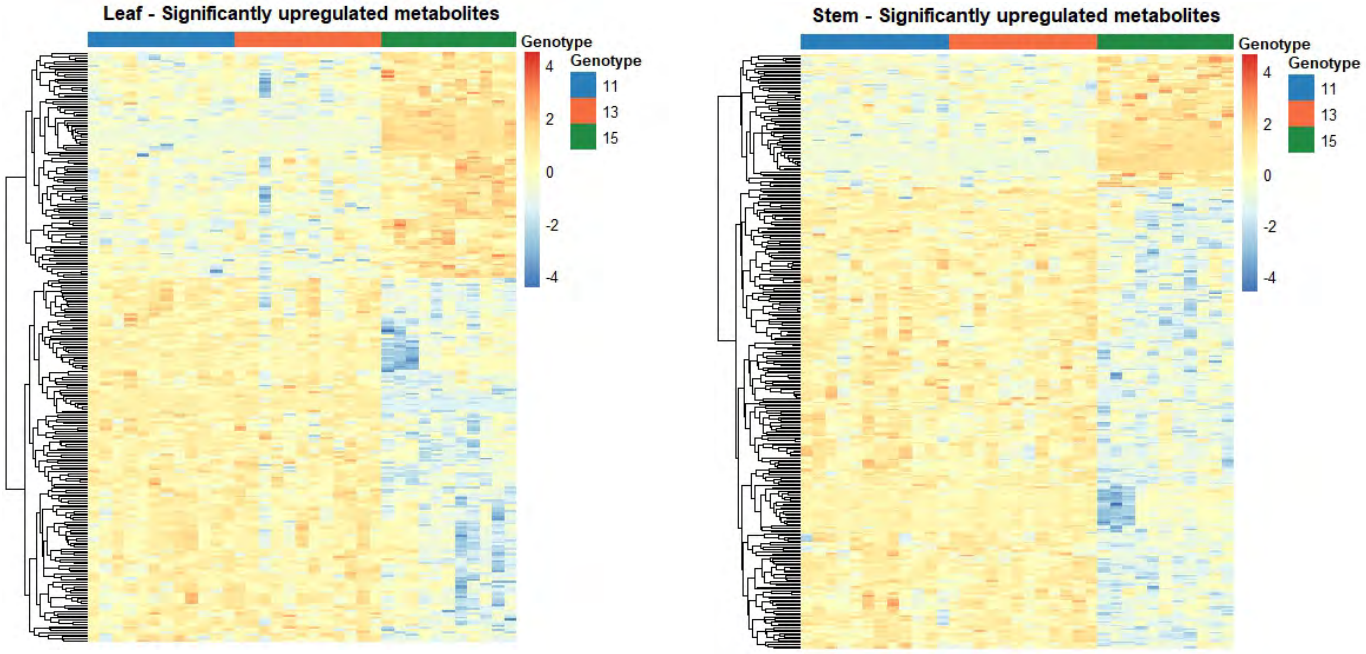

Figure 6.5. Abundance of significantly upregulated metabolites in leaf and stem across genotypes.

6.5 Outputs

- Dataset: ITS amplicon sequencing data and LC-MS/MS metabolomics dataset for three kikuyu genotypes under water stress and rehydration conditions.
- Manuscript in preparation for peer-reviewed publication.
- Contribution to project-wide database of kikuyu microbiome and metabolite profiles.

6.6 Applications & Impacts

The finding that rehydration, not the dry period, is the primary biological trigger is directly relevant to farming practice. It confirms that the critical risk window is in the days immediately following irrigation or rain after drought, rather than during the dry period itself.

The genotypic differences in metabolite profiles are significant for breeding programs. If specific chemical signatures are associated with toxicity risk, selecting against these profiles in breeding could yield safer cultivars. The absence of wortmannin across all conditions further calls into question management strategies or monitoring approaches that have been built around *F. torulosum* as the causal agent.

6.7 Future Research Opportunities and Actions

- Investigate the functional significance of the genotype-specific metabolite differences identified, particularly whether they are linked to susceptibility to toxin accumulation.
- Explore the mechanistic basis of the rehydration-triggered metabolite response, including gene expression analysis.
- Develop farmer-facing resources on safe reintroduction timeframes following drought-breaking rain, based on these and subsequent study findings.

7. Subproject P1b.2- Response of *Fusarium torulosum* to Water Stress in Five Kikuyu Genotypes

Research team: Vivien Tan, Percy Wong, Richard Trethowan, Barbara Brito & Krista L. Plett

7.1 Background

Fusarium torulosum has been the most frequently cited candidate cause of kikuyu poisoning, primarily because of its known capacity to produce wortmannin and butanolide, mycotoxins that produce symptoms in cattle resembling kikuyu poisoning when administered experimentally. However, *F. torulosum* has not been consistently found in toxic pastures, and prior studies were constrained by culture-based methods that may have missed or misrepresented fungal communities.

This subproject expanded on the findings of Subproject P1b.1 by directly inoculating all pots with a verified *F. torulosum* isolate and tracking its abundance alongside the broader microbiome and metabolome across five kikuyu genotypes. This allowed a direct test of whether drought and rehydration conditions influence *F. torulosum* abundance and wortmannin production in a way consistent with its proposed role as the causative agent.

7.2 Objectives

- To test the hypothesis that *F. torulosum* abundance is favoured by conditions associated with toxicity, namely a prolonged dry period followed by rehydration.
- To detect wortmannin or butenolide production under drought and rehydration conditions using metabolomics.
- To assess whether genotype influences *F. torulosum* colonisation or plant metabolite responses.
- To characterise fungal community and metabolite shifts across a broader set of five kikuyu genotypes.

7.3 Materials & Methods

A pot experiment was conducted at the University of Sydney Camden Campus, NSW, from July to November 2024. Five kikuyu genotypes, Whittet, Fulkerson, and three breeding lines (2, 8, and 10) were established in 1.1 L pots. At the start of the experiment, all 60 pots were inoculated with a spore suspension of *F. torulosum* (isolate accession number 11062, obtained from the Botanic Gardens of Sydney), confirmed by PCR amplification and Sanger sequencing of the ITS and Tef-1 gene regions. Spore concentration was 9.04×10^4 spores/mL.

After a two-week conditioning period at 80% field capacity, pots were assigned to either a reduced-watering treatment (30% field capacity) or a control (80% field capacity) in a randomised block design. After eight weeks, at which point reduced-watering plants had gone dormant, all pots were rehydrated to full saturation for one week to simulate drought-breaking rainfall. Soil, root, stem, and leaf samples were collected at two timepoints: immediately before rehydration and one week after.

F. torulosum abundance was quantified from ITS sequencing data. Fungal community composition was analysed using ITS sequencing on the Illumina MiSeq i100 Plus (QIIME2/DADA2 pipeline, UNITE database). Metabolomics was performed using LC-MS/MS in positive and negative ionisation modes, with data processed via MSDial and MetaboAnalystR. A correlation network analysis between microbial taxa and significantly upregulated metabolites was performed using Spearman rank correlation.

7.4 Key Findings

- Drought did not significantly reduce *F. torulosum* abundance over eight weeks. The organism persisted under prolonged dry conditions across all genotypes and tissue types.
- Rehydration caused a significant and consistent decline in *F. torulosum* abundance in both the control and reduced-watering treatments. This is the opposite of the pattern expected if *F. torulosum* were driving post-rehydration toxicity.
- Genotype had no significant effect on *F. torulosum* colonisation, but a significant interaction between genotype and treatment ($p = 0.04995$) was observed, suggesting that *F. torulosum* responds differently across genotypes under water stress.
- Neither wortmannin nor butanolide were detected in any sample. No metabolite fragment consistent with wortmannin or butenolide was identified under either dry or rehydrated conditions.
- As in Subproject 1, rehydration was the dominant driver of change in the fungal community across leaf, stem, and root tissues. Treatment (drought) had the greatest effect in soil only.
- Rehydration triggered upregulation of fatty acids, flavonoids, alkaloids, and nitrogen-containing compounds in leaf and stem tissue, consistent with a plant stress response to overwatering rather than a fungal toxin response.
- Correlation analysis between microbial taxa and upregulated metabolites was weak (maximum Spearman $r = 0.69$), indicating the metabolite changes are more likely driven by the plant itself than by the fungal community.
- A weak correlation ($r = 0.63$) was observed between the *F. torulosum* inoculum and hexadecanoic acid, a fatty acid with antimicrobial properties, in leaf control samples, possibly reflecting a plant defensive response.

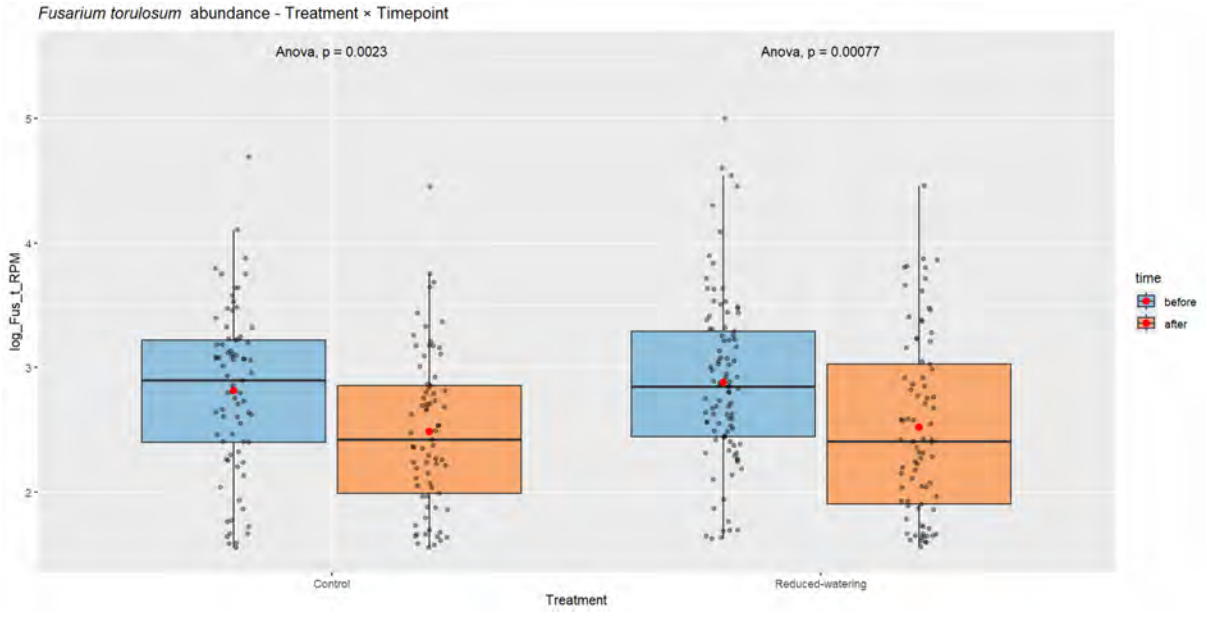

Figure 7.5. F. torulosum abundance across treatments and timepoints.

7.5 Outputs

- Dataset: ITS sequencing and LC-MS/MS metabolomics data for five kikuyu genotypes with *F. torulosum* inoculation under water stress and rehydration conditions.
- Quantified *F. torulosum* abundance data across genotypes, treatments, and tissue types.
- Manuscript in preparation for peer-reviewed publication.
- Contribution to the project-wide kikuyu microbiome and metabolite database.

7.6 Applications & Impacts

The results of this subproject substantially weaken the case for *F. torulosum* as the primary causative agent of kikuyu poisoning. Its decline, rather than proliferation, following rehydration, combined with the absence of wortmannin under any tested condition, suggests that monitoring or management strategies built around this fungus are unlikely to be effective.

These findings redirect attention toward alternate fungal species or the plant's own stress response as the source of toxic compounds, and open the possibility that overwatering, not merely drought, stresses kikuyu sufficiently to trigger the production of toxic metabolites. For farmers, this reinforces the importance of managing reintroduction of cattle to pastures after heavy rain and highlights the need for practical risk indicators that do not rely on *F. torulosum* detection.

7.7 Future Research Opportunities and Actions

- Examine the identity and biological activity of the alkaloids and nitrogen-containing compounds upregulated post-rehydration, to determine if any could cause the clinical signs of kikuyu poisoning.

8. Subproject P1b.3. Microbial and Metabolite Profiles Associated with Kikuyu Grass Exhibiting Kikuyu Toxicity

Research team: Krista L. Plett, Vivien Tan, Deirdre Hanrahan-Tan, Aki Kawasaki, Pedro Pinsczowski, Barbara Brito Rodriguez

8.1 Background

While the pot experiments (Subprojects P1b.1 and P1b.2) provided controlled evidence that rehydration drives the most significant biological changes in kikuyu, they could not directly identify which microbes or metabolites are present during actual poisoning events. Previous field studies had relied on culture-based methods, which detect only a subset of the fungal community and may miss the true causal organism or toxic compounds.

Subproject P1b.3 addressed this gap by sampling kikuyu from commercial farm pastures with confirmed, historical, suspected, and no history of kikuyu poisoning across multiple geographic regions and seasons in NSW. Using unbiased ITS and 16S sequencing and untargeted metabolomics, this study produced the first comprehensive molecular profiles of toxic kikuyu grass from confirmed field poisoning events.

8.2 Objectives

- To characterise the full fungal and bacterial microbiome of kikuyu pastures across active, historical, suspected, and unaffected toxicity classifications.
- To identify metabolites consistently and significantly elevated in actively toxic kikuyu compared to non-toxic pastures.
- To identify candidate fungal or bacterial taxa whose abundance is significantly associated with confirmed kikuyu toxicity events.
- To narrow down potential causative agents, microbial or chemical, for further targeted investigation.

8.3 Materials & Methods

Kikuyu pasture samples were collected from cattle farms across NSW between November 2021 and April 2024, spanning the Hunter Valley, Mid-Coast, North Coast, Shoalhaven, and Bega regions across multiple seasons. A total of 47 sampling sites were included, classified as: Toxic (active poisoning event confirmed by cattle necropsy, n = 9 sites from 1 farm), Historical (prior confirmed poisoning events, n = 12, from five farms), Suspected (symptoms consistent with poisoning but unconfirmed, n = 2; 1 farm), and Unaffected (no history of poisoning, n = 24, 17 farms).

At each site, intact swards (approximately 20 × 20 × 20 cm) were collected and sub-sampled within 24 hours into leaf, stem/stolon, fine root, and adhering soil fractions, then frozen at −80°C. Suspected samples were retained for comparative visualisation only and excluded from statistical analyses due to unconfirmed status.

Metabolomics was performed on stem and leaf samples using LC-MS/MS (Sciex ExionLC AD / TripleTOF 6600+) in positive and negative ionisation modes. Metabolite identification and peak alignment were conducted with MS-DIAL, and statistical analysis (PCA, ANOVA, heatmaps) with MetaboAnalyst 6.0. Microbiome profiling used ITS or 16S amplicon sequencing with taxonomy assigned against the UNITE database. Community diversity and composition were assessed using the vegan and ecodist R packages, and differential abundance of ASVs in toxic vs. non-toxic samples was determined using ALDEx2 with Benjamini-Hochberg correction.

8.4 Key Findings

Metabolomics:

- Toxic kikuyu leaf samples had a significantly different metabolite profile from both historical and unaffected samples, primarily in negative ionisation mode. Toxic samples separated on PC2 (11.7% of total variation) in PCA (Figure 1).

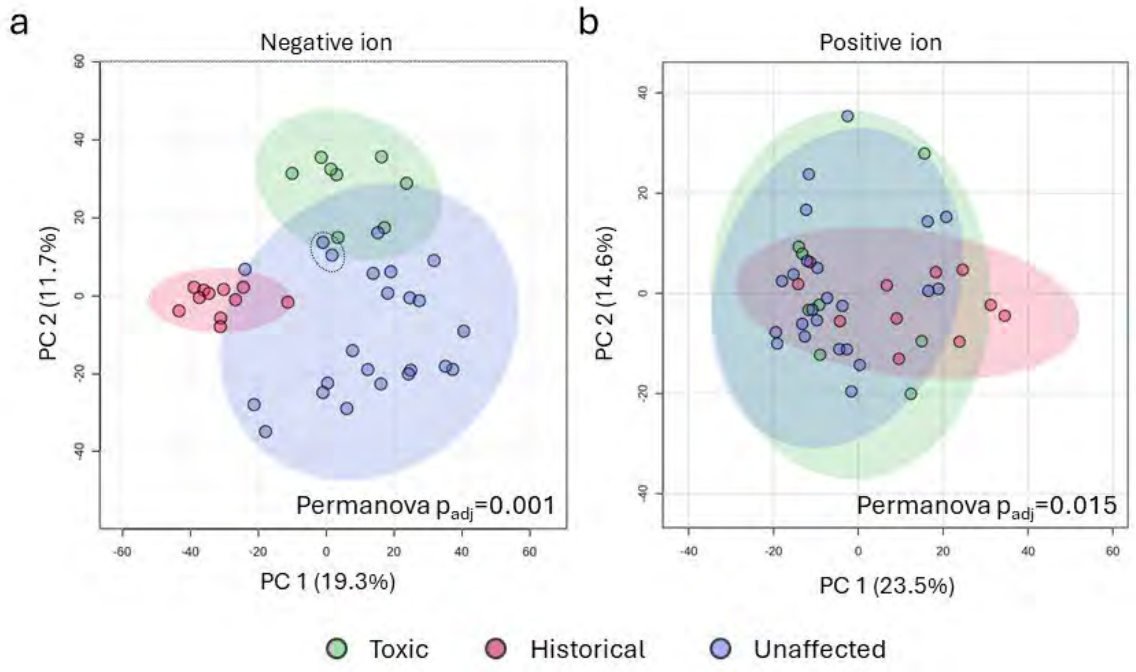

Figure 8.4 Actively toxic, historically toxic and unaffected Kikuyu leaf samples have significantly different metabolomic profiles. PCA of total metabolites present in Kikuyu leaf samples identified with negative (a) or positive (b) ionisation mode. Ellipses delineate the 95% confidence limit and results of PERMANOVA indicate significance of separation ($p<0.05$).

- ANOVA identified 259 negative-mode and 53 positive-mode metabolites significantly elevated in toxic kikuyu compared to both other groups.
- A refined subset of 17 (negative mode) and 6 (positive mode) metabolites showed consistently high abundance in toxic samples and low abundance in historical and unaffected samples. These represent the strongest candidate biomarkers of toxicity.
- Seven of these 23 metabolites were also significantly elevated in stem tissue, making them particularly robust candidates, a compound present in both leaf and stem during active toxicity is more likely to be causally relevant than one confined to a single tissue.
- Putative identities of the most elevated compounds include enterodiol, tridesacetoxykhivorin, a compound related to macrocyclic structures (resembling trichothecene-type mycotoxins), and several unidentified metabolites with high fold changes (up to 59-fold in leaf tissue; Table 1).

Table 1. Metabolites showing specific elevation in Toxic Kikuyu leaf and stem samples relative to non-toxic (unaffected) grass samples.

Metabolite ID	Ionisation Mode	Fold Change (Leaf/Stem)	Retention time	m/z	Putative ID (ontology/compound)
1927	Negative	40.5/11.1	10.00	303.15	Unknown
2482	Negative	27.3/3.5	14.78	347.15	Lignols/Enterodiol
4187	Negative	59.0/17.1	23.40	505.23	Hydroxybenzaldehydes/[(E,2R)-5-(3-chloro-5-formyl-2,6-dihydroxy-4-methylphenyl)-3-methyl-1-[(1S,2R,6R)-1,2,6-trimethyl-3-oxocyclohexyl]pent-3-en-2-yl] 3-methylbutanoate
4191	Negative	11.4/1.1	14.84	505.23	Hydroxybenzaldehydes/ [(E,2R)-5-(3-chloro-5-formyl-2,6-dihydroxy-4-methylphenyl)-3-methyl-1-[(1S,2R,6R)-1,2,6-trimethyl-3-oxocyclohexyl]pent-3-en-2-yl] 3-methylbutanoate
4199	Negative	20.6/4.5	12.41	506.23	Macrolactams/3,15-dibenzyl-1,4,12-trimethyl-1,4,7,10,13-pentazacyclopentadecane-2,5,8,11,14-pentone
4314	Negative	19.6/4.2	12.20	519.25	Liminoids/Tridesacetoxykhivorin
4143	Positive	14.4/5.0	3.11	423.81	Dicarboxylic acid derivative/2-[5-[2-[2-[5-(2-hydroxybutyl)oxolan-2-yl]propanoyloxy]propyl]oxolan-2-yl]propanoic acid

Microbiome:

- A total of 5,693 independent fungal ASVs were identified across soil, root, stem, and leaf tissue. Species richness was highest in soil and lowest in stem samples.
- Toxic pastures showed a trend toward lower fungal species richness compared to historical ($p = 0.096$) and unaffected ($p = 0.068$) sites, though not statistically significant.
- Six fungal taxa were significantly more abundant in leaves of toxic pastures (padj < 0.05), including Acremonium polychromum, Dothideomycetes sp., Sordariomycetes sp., Derxomyces longicylindricus, Dioszegia sp., and Neocucurbitaria ribicola.
- *Acremonium polychromum* was significantly elevated across leaf, stem, and soil of toxic pastures, a consistent multi-tissue signal that distinguishes it from organisms enriched in only one tissue. More detailed analysis of the ITS sequence associated with this ASV suggests that it may be more appropriately assigned to *Clonostachys rosea*, a common plant endophyte.

- An additional 12 fungi were significantly enriched in toxic stem and soil samples, including *Microascus chinensis, Amauroascus niger* (known producer of zaragozic acids), and *Hypomyces semicircularis*.
- *F. torulosum* was not identified as a significantly enriched fungus in toxic pasture samples.
- The bacterial community showed less variation in the toxic samples than the fungi with no species showing significant enrichment in toxic leaves.

8.5 Applications & Impacts

This subproject delivers the most directly actionable findings of the project. By identifying specific metabolite candidates and fungal taxa associated with confirmed poisoning events, it provides a starting point for developing a diagnostic toolkit for kikuyu toxicity risk. If the candidate metabolites or fungi can be validated as reliable markers, they could form the basis of a rapid field test or sampling protocol that allows farmers to assess pasture toxicity risk before introducing cattle.

The identification of *Acremonium polychromum* (or *Clonostachys rosea*) as a consistently enriched organism across multiple tissue types in toxic pastures is a significant finding. This organism belongs to a fungal group capable of producing toxic alkaloids. *C. rosea* is considered a beneficial endophyte promoting plant growth and disease suppression but also produces potent metabolites with known antifungal and insecticidal activity. This ASV's consistent presence, across leaf, stem, and soil, at toxic sites warrants targeted investigation into whether it produces compounds capable of causing kikuyu poisoning symptoms.

8.6 Outputs

- Dataset: ITS amplicon sequencing and LC-MS/MS metabolomics data from 47 kikuyu pasture sites across NSW, spanning confirmed toxic, historical, suspected, and unaffected classifications.
- Candidate list of 23 metabolite markers consistently elevated in toxic kikuyu (7 validated in both leaf and stem tissue).
- Candidate list of 18 fungal taxa significantly enriched in toxic pasture samples, with *Acremonium polychromum* (or *C. rosea*) identified as the highest-priority candidate for follow-up.
- Peer-reviewed manuscript in preparation (Plett et al.).
- Data contributed to cross-project kikuyu toxicity reference database.

8.7 Future Research Opportunities and Actions

- Expand field sampling to include more confirmed active poisoning events across a greater range of geographic locations and seasons, to strengthen the statistical power of metabolite and microbiome associations.
- Trial *A. polychromum* and *C. rosea* species on kikuyu in conditions associated with toxicity, and capture metabolites for *in vitro* trials to assess impact on cattle stomach cell lines.

9. Project-wide Dissemination

Table 9. Peer-reviewed publications.

Author	Title	Journal	Year Published	Citations (to April 26)
Jia Ling Vivien Tan, Barbara Brito, Percy T.W. Wong, Amit Singh, Richard Trethowan, and Krista L. Plett.	Effect of water-stress on the microbiome and metabolite profiles in three kikuyu (*Cenchrus clandestinus*) genotypes.	In preparation		
Jia Ling Vivien Tan, Barbara Brito, Percy T.W. Wong, Amit Singh, Richard Trethowan, and Krista L. Plett.	Response of *Fusarium torulosum* to water-stress in five kikuyu (*Cenchrus clandestinus*) genotypes. Microbial and metabolite profiles associated with *Cenchrus clandestinus* (Kikuyu) exhibiting Kikuyu toxicity	In preparation		

10. Conclusions and Recommendations

This project represents a comprehensive investigation of kikuyu toxicity, integrating controlled pot experiments with field sampling and applying modern, culture-independent sequencing and metabolomics tools that were unavailable to earlier researchers. Taken together, the three subprojects have substantially advanced understanding of the biological basis of kikuyu poisoning and meaningfully redirected the field toward productive new hypotheses.

The three subprojects converge on several clear conclusions:

The environmental trigger matters, and the risk window is rehydration, not drought. Across both pot experiments, rehydration, not the dry period itself, was the dominant driver of biological change in the kikuyu fungal microbiome and plant metabolome. This is consistent with the epidemiology of kikuyu poisoning, which is characteristically associated with rapid grass regrowth following drought-breaking rain or irrigation, and has direct implications for when cattle are most at risk.

Fusarium torulosum is not the primary causative agent. The controlled inoculation experiment (P1b.2) provided the most direct test of this hypothesis to date. *F. torulosum* did not proliferate under conditions associated with toxicity; instead, its abundance declined significantly following rehydration. Neither wortmannin nor butenolide was detected under any experimental condition. F. torulosum was also absent from the list of fungi significantly enriched in field-confirmed toxic pastures. Together, these findings make a compelling case that management or monitoring approaches built around this organism are unlikely to reduce poisoning risk, and that the field should move on from this hypothesis.

Candidate toxic compounds have been identified from real poisoning events for the first time. The field study (P1b.3) identified seven metabolites significantly elevated in both leaf and stem tissue of actively toxic pastures relative to historical and unaffected sites. These include putative lignols, limonoids, macrolactams, and hydroxybenzaldehyde-related compounds, some with fold changes as high as 59-fold in leaf tissue. Their consistent elevation across two tissue types in confirmed poisoning events makes them the strongest candidate biomarkers or causative compounds yet identified.

A specific fungal candidate has emerged. *Acremonium polychromum*, which may represent *Clonostachys rosea* based on refined phylogenetic analysis, was significantly enriched across leaf, stem, and soil of toxic pastures, making it the most consistently associated fungal organism identified to date. While *C. rosea* is broadly regarded as a beneficial plant endophyte, it is capable of producing potent secondary metabolites with antifungal and insecticidal activity, and its role in kikuyu poisoning warrants direct investigation.

Genotype shapes the chemical environment of kikuyu independently of water stress. Genotype was the strongest driver of metabolite profiles across both pot experiments, with substantial differences between commercial cultivars and breeding lines in the classes and quantities of metabolites produced. This raises the possibility that cultivar selection could be a practical long-term strategy for reducing toxicity risk.

Recommendations

Prioritise validation of the seven candidate toxic metabolites. Targeted chemical analysis and bioassays, including testing in bovine rumen or stomach cell lines, should be conducted to determine whether any of the identified compounds, particularly the macrolactam and limonoid candidates, are capable of causing the clinical signs of kikuyu poisoning.

Directly investigate *Acremonium polychromum* / *Clonostachys rosea*. Controlled inoculation

experiments on kikuyu under conditions associated with toxicity, combined with metabolite profiling of the fungus in culture, are the necessary next step to determine whether this organism produces compounds consistent with the toxicity syndrome.

Expand field sampling. The toxic sample set is currently limited to confirmed events from the mid coast (additionally, data is being analysed from two recent events). Expanding to additional farms, regions, and seasons will improve the statistical robustness of the metabolite and microbiome associations and determine whether the identified signatures generalise across NSW and beyond.

Move toward a practical on-farm diagnostic. The candidate metabolite markers identified in P1b.3 provide a foundation for developing a rapid field test, for example, a lateral flow assay or portable spectrometry approach, that would allow farmers and veterinarians to assess toxicity risk before cattle are introduced to recently rehydrated kikuyu pastures.

Update industry guidance on *F. torulosum* and rehydration risk. Current Dairy Australia and state veterinary guidance on kikuyu poisoning should be reviewed in light of these findings. In particular, the rehydration trigger and the lack of evidence for *F. torulosum* should be communicated to farmers, agronomists, and veterinary practitioners through extension channels.

Integrate genotype screening into breeding programs. The strong genotypic effect on metabolite profiles observed across both pot experiments should be incorporated into kikuyu cultivar development programs, with screening for toxicity-associated chemical signatures as a selection criterion alongside existing disease resistance traits.

11. Annexes

Table 2. Conference presentations, abstracts and other meetings

Authors	Title	Presentation Type	Conference/ Event	Location	Year	Audience
Jia Ling Vivien Tan, Barbara Brito, Percy T.W. Wong, Amit Singh, Richard Trethowan, and Krista L. Plett.	Effect of drought on microbiome community and metabolite profiles in kikuyu (Cenchrus clandestinus)	Oral	ADSA	Louisville,KY	2025	
Richard Trethowan, Krista Plett, Barbara Brito, Percy Wong, Amit Singh	Good grass gone bad – kikuyu poisoning	Oral	Annual District Veterinarians Conference	Wollongong	2024	
Krista Plett	Kikuyu Toxicity: A pasture out of balance?	Oral	DRF Symposium	Camden	2023	
Barbara Brito, Krista Plett, Vivien Tan.	DiaryUP Team meeting update	Oral	Virtual meeting		2022, 2025,	

Table 3. Technical reports, pamphlets, published material and other media engagements

Authors	Title	Place Published	Year
Lee-Ann Monks, Monks Communication	P1b Project Update: Investigating Kikuyu Toxicity	https://dairyup.com.au/wp-content/uploads/2026/03/Proj-Update-P1b-kikuyu-toxicity-2026.pdf	Feb 2026
Lee-Ann Monks, Monks Communication	Research aims to unlock Kikuyu grass benefits	https://www.dpi.nsw.gov.au/about-us/media-centre/releases/2026/general/research-aims-to-unlock-kikuyu-grass-benefits	May, 2026
McPherson Media Group	Testing kikuyu to solve an industry mystery	https://www.dairynewsaustralia.com.au/news/testing-kikuyu-to-solve-an-industry-mystery/	May, 2024

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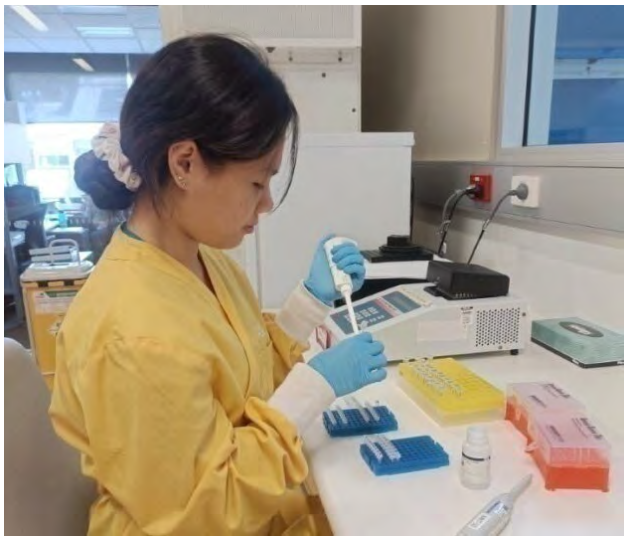
Final Project Summary

P1b – Kikuyu - Investigating toxicity

May 2026



Kikuyu is a perennial pasture species commonly grown in NSW for livestock production. It is classified as a C4 grass meaning it is adapted to warm or hot conditions. Kikuyu is fast growing and produces more dry matter – of higher quality – than other C4 grass varieties.

However, there's still a lot to learn about kikuyu toxicity – a risk for livestock producers.

Unlocking the potential of Kikuyu

Dairy UP's P1 project aimed to unlock the potential of Kikuyu pastures used by NSW dairy farmers. P1 was a suite of five projects that collectively explored new management options to grow and utilise more Kikuyu over summer and increase the productivity of Kikuyu-based pastures.

P1a – Kikuyu: Remote monitoring

P1b – Kikuyu Investigating toxicity

P1c – Kikuyu: Developing new varieties

P1d – Kikuyu: Carbon on NSW dairy farms

P1e – Kikuyu: Nutritional value

This document is the final update on P1b – Kikuyu: Investigating toxicity.

Kikuyu toxicity

Cattle deaths and illness officially linked to kikuyu poisoning are rare but have been recorded in NSW over the past 20 years.

Kikuyu poisoning, or toxicity, is frequently associated with specific environmental conditions and new growth following heavy rainfall, often after a summer drought or prolonged dry conditions.

Cause

The compound or mechanism that makes kikuyu toxic to cattle under these conditions is unknown. Kikuyu toxicity is difficult to research because of the unpredictability and rarity of kikuyu poisoning. The only definitive way to determine if cattle have kikuyu poisoning is a combination of the history, clinical signs, and specifically looking for damage to the forestomach (necropsy findings).

Specific signs linked to kikuyu toxicity include dehydration, sham drinking (trying to drink but unable), abdominal pain, unsteady gait, drooling and death. But not all cattle suffering from kikuyu poisoning present with these signs. In some instances, animals may recover, although they will not be as productive as before, given the damage to the digestive system.

Some affected cattle are reported as kikuyu toxicity cases; however, many remain unreported, with their illness blamed on other causes or not investigated.

Project aim

DairyUP researchers wanted to find out why kikuyu can become toxic, to help find ways to prevent or manage it.

Providing clarity around kikuyu toxicity would

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inform future grazing management practices and research.

Ultimately, project findings could be used to develop new kikuyu varieties that are less susceptible to toxicity events, diagnostic tests to evaluate pasture safety, in-field preventative treatments and treatments for affected cattle.

Benefits

Although kikuyu poisoning is infrequent, it remains a risk and can prevent dairy farmers from unlocking all the benefits of kikuyu.

Understanding the underlying reasons for kikuyu poisoning would give dairy farmers more confidence using kikuyu as a feed option – potentially providing more homegrown feed alternatives year-round.

Any findings could benefit Australia's entire livestock sector. As well as affecting dairy and beef cattle, kikuyu poisoning can also affect sheep and goats, although they are less susceptible.

Key findings

Findings from this research weaken the evidence for *Fusarium torulosum* as the primary cause of kikuyu toxicity. Although this fungus was previously found in affected pasture, our results suggest other plant or microbial compounds may be involved.

This work provides a foundation for future diagnostic tools and evidence-based pasture management strategies to help reduce toxicity risk.

Supporting findings:

- Kikuyu variety strongly influenced plant metabolite profiles, suggesting some cultivars may carry lower toxicity risk.
- Seven compounds were consistently elevated in leaf and stem samples from toxic pastures, making them strong candidate biomarkers of toxicity.
- Re-watering after drought triggered greater biological change in kikuyu grass than drought alone.
- *Fusarium torulosum* declined after re-watering and was not consistently associated with toxic pastures.
- One fungal species was consistently elevated in toxic pastures and warrants further investigation.

More info

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Research and results

On-farm monitoring

The team collected and analysed grass and soil samples from properties across NSW. An average sample of grass from a paddock contains hundreds of microbial species and thousands of secondary metabolites.

Samples from unaffected and historically affected farms provided a reference "baseline" for comparison with samples collected during active or suspected kikuyu toxicity events.

Dairy UP researchers visited the farms affected by the following suspected kikuyu poisoning events:

- 2018-19: Hunter Valley.
- March 2024: Bega (unconfirmed).
- April 2024: Hunter Valley.

Farmers on affected farms gave their observations of kikuyu poisoning.

Samples collected from the reference and affected farms were analysed for fungal and bacterial communities (microbiome analysis).

A technology called metabolomics was used to examine samples from the reference and affected farms. Metabolomics gives a "fingerprint" of the many compounds present in a grass sample. Researchers identified potentially toxic compounds by comparing compounds in the database from unaffected and affected farms.

Findings
Several fungal species were significantly more abundant in toxic samples. One fungal species was consistently elevated across leaf, stem and soil samples from toxic pastures. It is now being investigated further as a potential contributor to kikuyu toxicity.

Importantly, *Fusarium torulosum* – a fungus frequently found in affected kikuyu pasture – was not significantly enriched in toxic pasture samples. This fungal species is known to produce metabolites that can cause stomach lesions in laboratory animals.

Metabolites: The team identified seven metabolites that were consistently elevated in both leaf and stem tissue from toxic samples but low or absent in the unaffected and historical samples. These compounds are considered the strongest candidate biomarkers or potentially toxic compounds identified in the project to date.

Fungi: Samples from toxic pastures appeared to have less diversity and richness of fungi in the leaf and stem tissue.

Greenhouse trial

In addition, PhD student, Vivien Tan set up a greenhouse trial to create the environmental conditions that are understood to precede kikuyu toxicity. The trial pots contained soil collected from farms with a history of toxicity cases. It included three varieties:

- Whittet, the most common commercial kikuyu cultivar.
- a drought-resistant variety in development.
- a drought-tolerant variety in development.

She examined how the different varieties and their associated microorganisms, including fungi, respond to drought and re-watering conditions. The aim was to determine whether genetic background influences the fungal microbiome or plant chemistry associated with kikuyu toxicity risk.

An additional pot trial involved inoculation with *Fusarium torulosum* to investigate how its abundance is affected by a drought/re-water cycle, which typically precedes a kikuyu toxicity event.

Findings
Fungi: There was also little difference in overall fungal community composition between the control and drought treatment pots. However, the fungal community composition changed significantly following re-watering. This suggests rehydration has a much greater impact on the microbiome than drought conditions alone.

Genotype also influenced some fungal communities, particularly in root and soil samples, while plant genotype was the strongest factor influencing metabolite profiles.

When *Fusarium torulosum* was added, there was no significant difference in *Fus. torulosum* abundance between the control and reduced-watering treatments. Its abundance declined significantly after re-watering following drought treatment. This suggests the conditions associated with kikuyu toxicity do not favour proliferation of this fungus.

Metabolites: Analysis also showed that kikuyu genotype strongly influenced plant chemistry, with clear differences in the types and abundance of compounds produced by different varieties. Re-watering after drought triggered larger biological changes than drought alone, supporting field observations that toxicity events are most commonly associated with rapid regrowth following drought-breaking rain or irrigation.

Importantly, two metabolites previously linked to *Fusarium torulosum* (wortmannin and butanolide) were not detected in the pot trials.

Together, the findings weaken the evidence for *Fus. torulosum* as the primary cause of kikuyu toxicity and suggest other plant or microbial compounds may be involved.

Collaborators

The P1b project was a collaboration between Dairy UP, University of Sydney and NSW Department of Primary Industries and Regional Development's Elizabeth Macarthur Agricultural Institute (DPIRD- EMAI).

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Read more

P1b final report

Tan et Al. 2025 Effect of drought on microbiome community and metabolite profiles in kikuyu. ADSA abstract.

Delivery organisations

The University of Sydney Dairy Research Foundation
NSW Government Department of Primary Industries and Regional Development
Scibus
Dairy Australia

Partner organisations

NSW Farmers Growing the best
eastAUSmilk
Dairy NSW
Dairy Australia
AFMH
Leppington Pastoral Company Pty Ltd
NSW Government Local Land Services
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Additional program supporters, collaborations or partnerships

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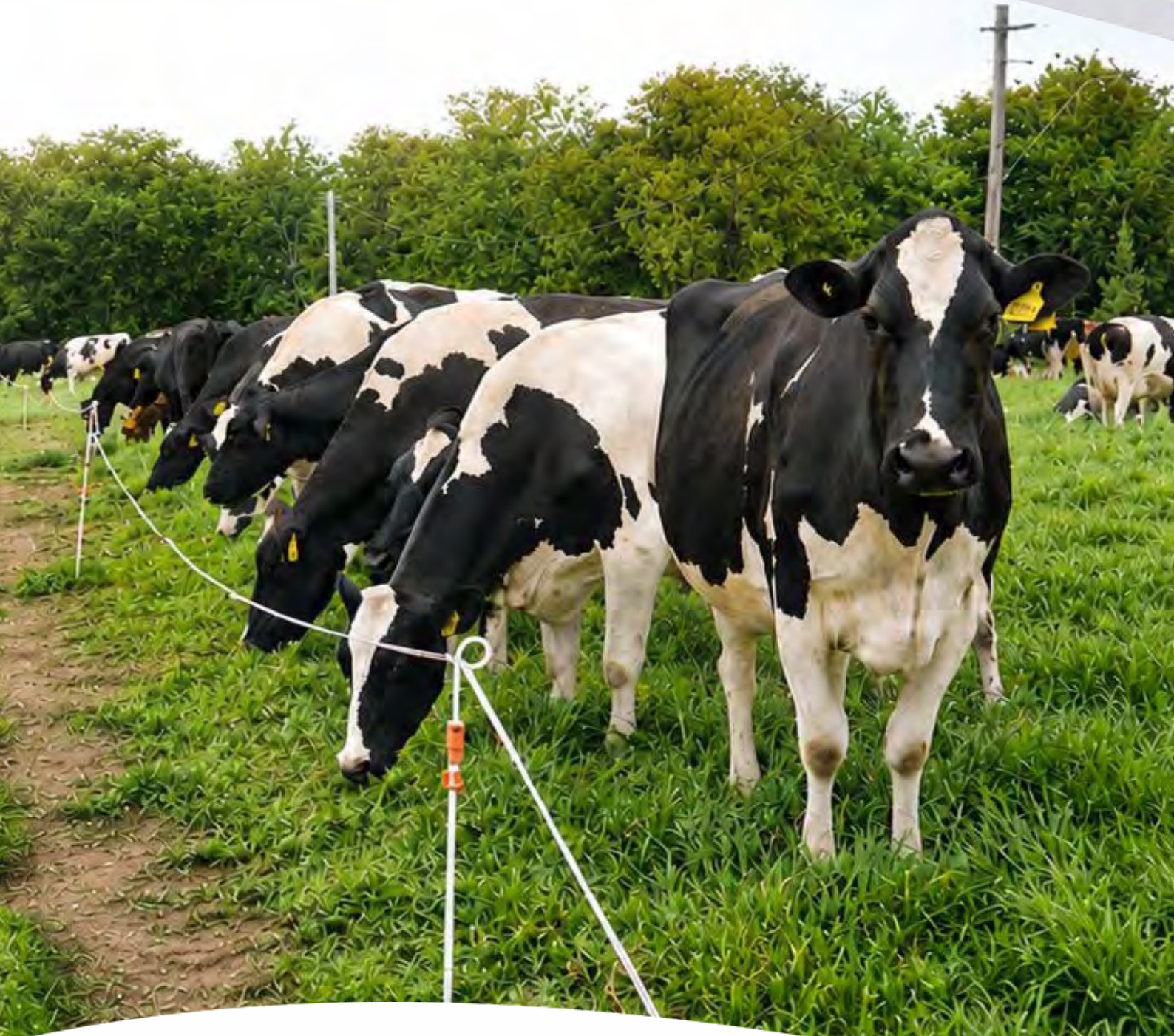
Macquarie University | NSW EPA | smaXtec | UC Davis | University of Technology Sydney

DairyUP
Unlocking potential

Final Report

P1c – Kikuyu: Developing new varieties



THE UNIVERSITY OF SYDNEY
Dairy Research Foundation

This project was led by Richard Trethowan from the University of Sydney Dairy Research Foundation

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

Proudly funded by

Australian Government | NSW Government | Dairy Australia | Department of Primary Industries and Regional Development | Scibus | The University of Sydney Dairy Research Foundation | NSW Growing the best | eastAUSmilk | Dairy Australia | AFMH | Leppington Pastoral Company | NSW Local Land Services | Norco

Delivery organisations

The University of Sydney Dairy Research Foundation | Department of Primary Industries and Regional Development | Scibus | Dairy Australia

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1. Executive Summary

New kikuyu genotypes developed by the University of Sydney in collaboration with Hatton's Turf Research have been screened for drought and salinity tolerance. However, the pasture potential of these lines was unknown including their dry matter production, disease resistance and nutritional value. To put these materials into context, a DNA study was conducted that included the new candidate lines and kikuyu leaf samples collected from farms around NSW. The DNA fingerprints showed that the new materials were genetically distinct from the cultivars Whittet and Fulkerson, and that 35% of all pastures were related to these two cultivars. An experiment was established to screen 13 candidate lines at Lansdowne in a 2-year small plot experiment. The results identified 3 candidate lines with competitive dry matter production, suitable nutritional profiles and potentially better disease resistance. These candidates were then sown on farm in strip plots across 2 seasons and grazing was allowed on one half of each strip. Dry matter production was assessed at regular intervals across the two growing seasons and a chemical analysis conducted to determine the nutritional value of the new materials. At the same time, the disease reactions of the new candidates to black spot and kikuyu yellows disease were assessed. Results showed that the new candidate lines produced equivalent dry matter to Whittet across the season at each location. However, physical analysis of dry matter samples for nutritional traits identified two lines with better profiles. These two lines were also more resistant to black spot and kikuyu yellows disease than Whittet. Both lines offer dairy farmers valuable new genetic diversity with enhanced nutritional and disease profiles at an equivalent dry matter production per ha.

2. Project Overview

Item	Description
Project Title	Kikuyu: Developing new varieties
Funding Body	Dairy UP
Dairy UP Project	P1c
Project Duration	2022-2026
Lead Organisation	The University of Sydney
Project Lead	Richard Trethowan
Key Collaborators	NSW Department of Primary Industries: Elizabeth Macathur Agricultural Institute

3. Abbreviations

USYD — The University of Sydney
NDF - neutral detergent fibre
ADF - acid detergent fibre
OM - organic matter
DM – dry matter
CDM – cumulative dry matter
DOMD - dry organic matter digestibility
WSC - water soluble carbohydrates
ME - metabolizable energy
CP – crude protein
NDVI – normalized difference vegetation index

4. Project Background and Rationale

The University of Sydney in collaboration with Hatton's Turf Research has been selecting kikuyu grasses for biomass production, disease resistance and tolerance to salinity and drought for many years. These new materials represent potential valuable genetic diversity for the dairy industry. P1c aims to evaluate these promising kikuyu lines for commercially important traits in comparison with two commonly grown cultivars of kikuyu. Testing is two-tiered, with larger numbers of lines initially tested at one location across two summers, followed by a selected subset evaluated across multiple environments in strip plots under farmer practice. The results of a DNA analysis of kikuyu pastures in NSW and the evaluation of kikuyu lines, including a selected subset tested across seasons and environments, are presented in this report.

5. Project Objectives

Dairy UP's P1c project aims to unlock the potential of Kikuyu pastures used by NSW dairy farmers by exploiting genetic diversity. This project explored and evaluated promising lines of kikuyu that have been selected for tolerance to salinity and drought.

6. The genetic diversity of kikuyu pastures and the new materials

Research team: Richard Trethowan, Vivien Tan, Amit Singh, Chong Mei Dong and Laura Espinosa

6.1 Background

While the University of Sydney in collaboration with Hatton's Turf developed new kikuyu materials with potential for the dairy industry, little was known of genetic diversity among existing pastures in NSW. We hypothesised that most pastures would be related to Whittet (L15 in this report), a successful variety released more than 50 years ago and widely adopted, presenting a potential issue with genetic vulnerability in our changing climate. This study also examined the genetic diversity of existing pastures compared to the new genetic materials to better understand their potential.

6.2 Objectives

To compare the DNA fingerprints of kikuyu materials collected on farm with the new genetic materials developed at the University of Sydney

6.3 Materials & Methods

Tissue samples were collected from dairy farms on the mid north coast (MC), north coast (NC) and south coast (SC) of NSW. These samples included the varieties Whittet and the Bill Fulkerson line and the 13 candidate kikuyu lines. Kikuyu DNA was extracted from dried leaves using the CTAB method and the samples subsequently genotypes using DArTseq. A total of 91 kikuyu samples were genotyped, and the locations of these samples are given in Table 1. Genetic distance and the dendrogram were generated by computing pairwise genetic distance (Hamming-style distance) between samples using their SNP genotypes. Distances between two samples were calculated per SNP as follows:

- 0 if both samples have the same homozygote or the same heterozygote
- 1 if the samples have different homozygotes (e.g., 0/0 vs 1/1)
- 0.5 if one sample is homozygous and the other is heterozygous
- SNPs with missing values in either sample are excluded

The final distance between two samples was the average per-SNP distance across all markers with valid genotypes. All analysis were conducted using R.

Table 1. Location of samples collected for DNA analysis on the mid-north coast (MC), north coast (NC) and south coast (SC) of NSW. New genotypes are marked TC. Toxicity is indicated if reported in the pasture at time of sampling.

Sample code	Sample information	Toxicity reported
MC-1	Neals Dairy, Patrick and Louise Neal, 31.53138 S, 152.33593 E, 40 Neals Ln, Oxley Island NSW 2430 (close to Taree).	
MC-2	Tocal LLS, Tocal, Paddock 9, 32.37492 S, 151.35288 E, Tocal College 815 Tocal Road PATERSON NSW 2421 (The Hunter)	
MC-3	Nicholson Dairy and Silage Contracting, Sam Nicholson, House, 31.51433 S, 152.35534 E,NSW. (20km North of Taree)	

Sample code	Sample information	Toxicity reported
MC-4	Bale's dairy, Tim Bale, House, 31.43344 S, 152.38344 (between Taree and Port Macquarie ,Stewarts River NSW 2443	
MC-5	Neilson dairy, Emily and Matt Neilson, 31.43527 S, 152.41440 E, farm at Bendolba, north of Dungog.	
MC-6	Glen Martin farm, Hunter Valley (Doug Cowan) mid level Kikuyu paddock, site of original toxicity outbreak.	Yes
MC-7	Glen Martin farm, Hunter Valley (Doug Cowan) mid level Kikuyu paddock, site of original toxicity outbreak.	Yes
MC-8	Glen Martin farm, Hunter Valley (Doug Cowan) low level Kikuyu paddock, near water hole where animals died.	Yes
MC-10	421 Marshdale Rd, Hunter Valley, main site of Kikuyu toxicity (2018) 16/40 cows dead.	Yes
MC-11	421 Marshdale Rd, Hunter Valley, secondary site of Kikuyu toxicity (2018) 2/3 cows dead. Lower elevation, wetter field than main site, other grasses present	Yes
MC-12	Stuart and Lorraine, Hunter Valley, Main site of cattle death (2018), low site, dense Kikuyu.	Yes
MC-13	Stuart and Lorraine, Hunter Valley, Main site of cattle death (2018), low site, dense Kikuyu.	Yes
MC-14	Geoff and Narelle, Hunter Valley, 6/15 cattle lost (2018), affected paddock, short Kikuyu mixed with rye and clover	Yes
MC-15	Geoff and Narelle, Hunter Valley, 6/15 cattle lost (2018), upper paddock unaffected mid length Kikuyu mixed with rye	Yes
MC-16	Glen Martin, Hunter Valley 2019 outbreak, 9/250 cattle (all young), paddock on hill down to water, mixed grasses and clover with Kikuyu	Yes
MC-17	Glen Martin, Hunter Valley 2019 outbreak, 9/250 cattle (all young), paddock on hill down to water, mixed grasses and clover with Kikuyu, mid hill	Yes
MC-18	James Neal, 75 Templetons Ln Oaxley Island, -31.886972079332907, 152.58513364202872 sample 1	
MC-19	James Neal, 75 Templetons Ln Oaxley Island, -31.886972079332907, 152.58513364202872 sample 2	
MC-20	James Neal, 75 Templetons Ln Oaxley Island, -31.886972079332907, 152.58513364202872 sample 3	
MC-21	Simon Scowen, Manning Valley Dairy, Warriwillah, 700 Bungay Rd, Whingham, Mid Coast DAGs, -31.906336037683786, 152.34403923104526, sample 1	

Sample code	Sample information	Toxicity reported
MC-22	Simon Scowen, Manning Valley Dairy, Warriwillah, 700 Bungay Rd, Whingham, Mid Coast DAGs, -31.906336037683786, 152.34403923104526, sample 2	
MC-23	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, 4 Paddock 1 (affected) yellow (looks like the grass from before the rain), confirmed kikuyu poisoning	Yes
MC-24	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, 3.1 paddock 1 (affected) (shoot), confirmed kikuyu poisoning	Yes
MC-25	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, 3.2 paddock 1 (affected) (long grass), confirmed kikuyu poisoning	Yes
MC-26	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, #2 Paddock 1 (affected), confirmed kikuyu poisoning	Yes
MC-27	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, #1 1404 76C Bucketts Way Paddock 1 (affected), confirmed kikuyu poisoning	Yes
MC-28	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, 5.1 Paddock 2 (less affected) Grazed (shoot) patch, confirmed kikuyu poisoning	Yes
MC-29	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, #6 Paddock 2 (less affected) Yellow, long, ungrazed, confirmed kikuyu poisoning	Yes
MC-30	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, 5.2 Paddock #2 Long not-grazed, confirmed kikuyu poisoning	Yes
MC-31	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, 8 Paddock 3, confirmed kikuyu poisoning	Yes
MC-32	CW & JI Edwards, 1404 The Bucketts Way, Stroud Road, Hunter Valley, 3 cows dead, 4 sick, 7 Paddock 3 Unaffected Grazed okay, confirmed kikuyu poisoning	Yes
MC-33	Midgy Gharret Rd, -31.9227848, 152.5283997	
NC-1	Merryvale Jerseys, Jo and Lee Behrens, 28.40285 S, 152.54458 E, 431 Knights Road 2474 Ettrick NSW	
NC-2	Kupidabin farm, Fleur and Sam Tonge, 28.48341 S, 153.04110 E, Kupidabin, NSW	
NC-3	Loongana Park, Paul and Wayne Clarke, 28.48492 S, 152.57323 E,	

Sample code	Sample information	Toxicity reported
	Loongana Park,Dobies Bight NSW	
NC-4	Riverside, Krystie and Adam Gould, Paddock 15, 28.54299 S, 153.06529 E, Greenridge NSW 2471	
NC-5	220 Elfords Rd, Dobies Bight NSW 2470	
NC-6	Near Brombin, -31.4721884, 152.6406919	
NC-7	Comboyne, -31.6174141, 152.4422854	
NC-8	Near Brombin, -31.4538014, 152.6426120	
NC-9	634-600 Bowman Farm Rd, -31.9327622, 151.9312421	
NC-10	Comboyne, -31.5919996, 152.4978439	
NC-11	Near Wingham, -31.9024723, 152.3411391	
NC-12	Bowman Farm, 31.55592 S, 151.55540 E,Bowman Farm NSW 2422	
NC-13	Dobies Bight, Fleur and Sam Tonge, 28.48197 S, 152.59120 E,220 Elfords Rd Dobies bight 2470, Suspected case of toxicity in Feb 2019	Yes
NC-14	Lamont terra B, 829 Barrington west rd Darringlton	
SC-1	Old Bemboka, Kev and Brodie Game, L Paddock, 36.65758 S, 149.60966 E,Bemboka NSW 2550	
SC-2	Hillgrove dairy, Phil Ryan, Paddock C2, 36.76422 S, 149.75675 E,Toothdale NSW 2550	
SC-3	Jelgowry, Will Russell, Paddock 'Center Pivot 3', 36.71750 S, 149.86821 E,Jellat Jellat NSW 2550	
SC-4	Yarranung, Ryan Apps, Paddock Rd 12, 36.64427 S, 149.83831 E,Angledale NSW 2550	
SC-5	Ferncliff dairy, Sam Holmes, Paddock 32, 36.81076 S, 149.71243 E,Candelo NSW 2550	
SC-6	Hillgrove Dairy, Phil Ryan, Little Irrigation Paddock, 36.76241 S, 149.75510 E,Whittet variety sowed 3 years ago, Toothdale NSW 2550	
SC-7	Ferncliff Dairy, Sam Holmes, 24a Paddock, 36.81358 S, 149.70649 E Candelo NSW 2550	
SC-8	36.37093628600237 S, 149.95213265366874 E, 5 Narira Park Ln, Cobargo NSW 2550	
SC-9	Phil Ryan, 36.76241 S, 149.75510 E, Trial at Bega	
SC-10	Phil Ryan, Hillgrove Dairy - Front Irrigation Paddock, Toothdale NSW 2550, 3 mature aged cows (7-10 years), kikuyu based with Italian ryegrass and legume mix	Yes

Sample code	Sample information	Toxicity reported
SC-11	Ferncliff/Sam Holmes, Bega, 5 cows, pure kikuyu pasture, suspected kikuyu bloating	Yes
SC-12	Ken Kimber, Ottonville Rd, Angledate, -36.64167, 149.879134	
SC-13	Ken Kimber, Ottonville Rd, Angledate, -36.64167, 149.890167	
SC-14	Brodie Game, "Blackjack Holsteins", 2367 Snowy Mountains Hwy, Bemboka, -36.6523647, 149.6064037	
SC-15	Andrew Irvin, "Aljo", Snowy Mountains Hwy, Bemboka, -36.6583577, 149.6350883	
SC-16	Brad Smith, "Oakleigh", 69 Jews Creek Rd, Brogo, -36.5949042, 149.8483728	
SC-17	Sam Holmes, "Ferncliff", Towridgee Lane, Candelo, -36.8072846, 149.7073084	
SC-18	Sam Holmes, "Ferncliff", Towridgee Lane, Candelo, -36.8126446, 149.712964	
SC-19	Hayden Parbery, "Willowdene", Tantawangalo Mtn Rd, Candelo, -36.811284, 149.712964	
SC-20	Hayden Parbery, "Willowdene", Tantawangalo Mtn Rd, Candelo, -36.782057, 149.644978	
SC-21	Phil Ryan, "Hillgrove", Wyndham Lane, Kanooma, -36.7627400, 149.7561343	
SC-22	Ryan Apps, "Yarranung", 180 Angledale Rd, Bega, -36.6571331, 149.8338638	
SC-23	Darren Wallis, Kameruka Lane, Candelo, -36.7513585, 149.7240622	
SC-24	Will Russell, "Jelgowry", 1159 Tathra Rd, Bega, -36.718967, 149.873522	
SC-25	Wal Jasper	
TC-1	Line 43, small plot experiment in Cobbitty	
TC-2	Line 51`, small plot experiment in Cobbitty	
TC-3	Line 133, small plot experiment in Cobbitty	
TC-4	Line 134, small plot experiment in Cobbitty	
TC-5	Line 6, small plot experiment in Cobbitty	
TC-6	Line 119, small plot experiment in Cobbitty	
TC-7	Line 96, small plot experiment in Cobbitty	
TC-8	Line 90, small plot experiment in Cobbitty	

Sample code	Sample information	Toxicity reported
TC-9	Line 7, small plot experiment in Cobbitty	
TC-10	Line 143, small plot experiment in Cobbitty	
TC-11	Line 44, small plot experiment in Cobbitty	
TC-12	Line 73, small plot experiment in Cobbitty	
TC-13	Line 59, small plot experiment in Cobbitty	
TC14	Bill Fulk from runners	
TC15	Whittet	
Bill S	Seedling source	
CRF	Corstorphine Dairy Farm, University of Sydney, Mid of Paddock 1 & Paddock 2, 34.024116 S, 150.654738 E, 335 C07BP, Brownlow Hill NSW 2570, Australia	
GD	Gallpen Dairy, 35.31417 S, 144.57237 E,Deniliquin NSW 2710	
RT	Galston farm, 33.38135 S, 151.04012 E, Galston NSW 2159	
UN	Brown pot, Unknown	

6.4 Key Findings

A dendrogram was constructed based on DNA diversity and this is presented in Figure 1. Four main groups of materials were identified. The cultivars Whittet and Fulkerson fell into one group (Group 1), whereas the new lines were distributed in Groups 2, 3 and 4 with a higher number in Group 4. None (apart for the two check cultivars, TC-14 (Fulkerson) and TC-15 (Whittet)) appeared in Group 1. Results indicate that there is significant diversity in kikuyu pastures across NSW with just 35% of locations sampled similar to Whittet and Fulkerson. No distinct grouping based on region (MC, NC, SC) was evident with sites spread across the groups. Interestingly, these appears to be a genetic relationship with reported toxicity with most instances appearing in Group 1 along with Whittet and Fulkerson.

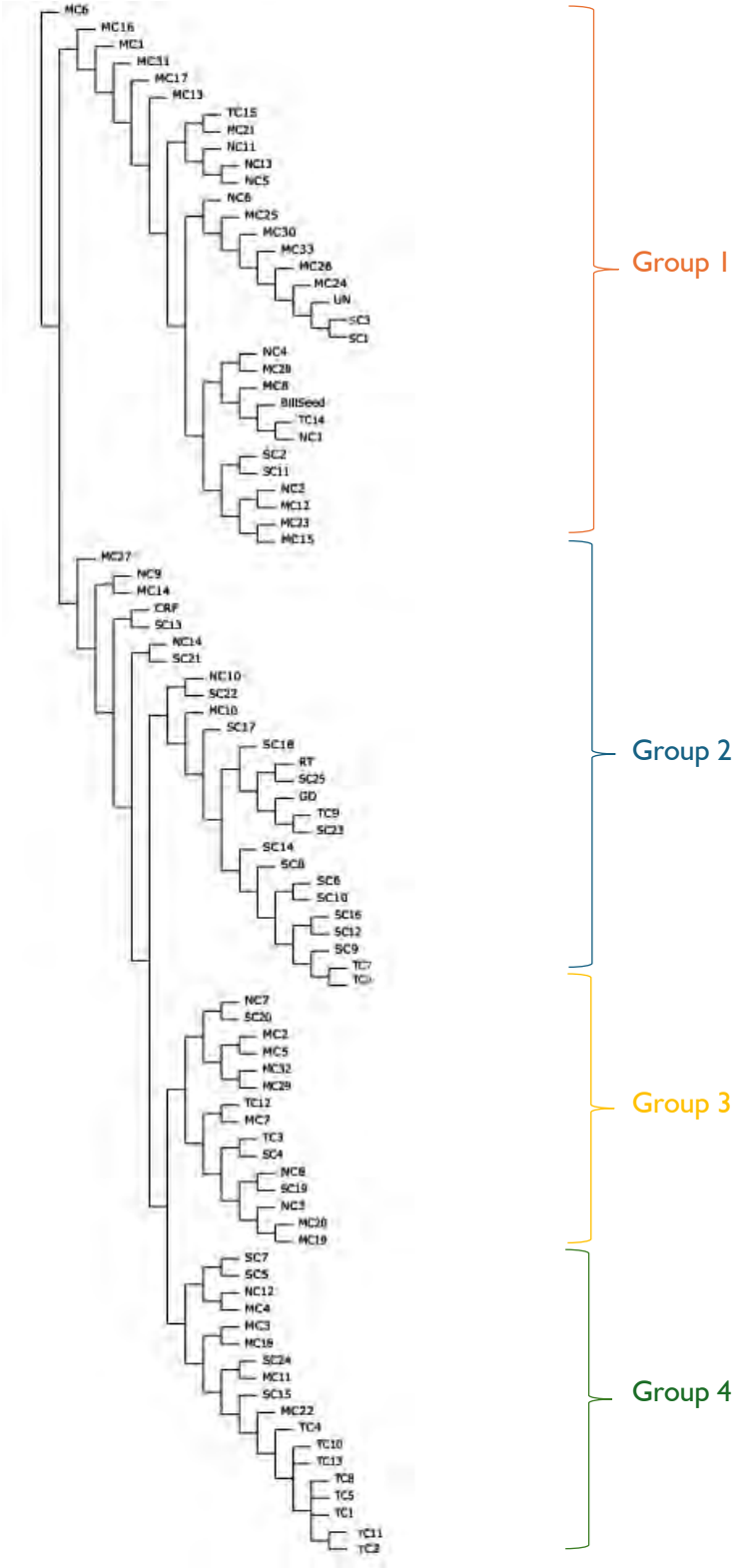

Figure 1. Dendrogram of kikuyu diversity at sampled sites in NSW. Clusters are indicated in four groups. A location/genotype key is given in Table 1.

7. Dry matter production of the new materials

Research team: Richard Trethowan, Laura Espinosa, Amit Singh, Vivien Tan

7.1 Background

The new kikuyu genotypes were identified from a previous study conducted by Duncan Fraser, a PhD student. He screened materials for drought and salinity tolerance (Fraser, 2020). During this process, 13 genotypes with pasture potential were identified based on biomass production and habit. These 13 genotypes were then compared to the cultivars Whittet and Fulkerson at one location and selected candidate lines subsequently evaluated across locations.

7.2 Objectives

- To evaluate the dry matter production of the new genetic materials throughout the growing season compared to Whittet and the Fulkerson varieties

7.3 Materials & Methods

(i) Initial evaluation of kikuyu genotypes

A small plot experiment was established at Landsdowne near Camden. Plots were generated from single runners of 15 genotypes (13 candidate lines plus the two checks) in 1 m^2 plots in a randomised complete block design with 4 replicates on the 17th of January 2021. The runners were grown in pots in the glasshouse and later transferred to the field and 1 m^2 plots established (Figure 2). Plots were harvested for biomass using a mower throughout the growing period and the fresh and dry weights determined. NDVI was assessed on the same date prior to mowing.

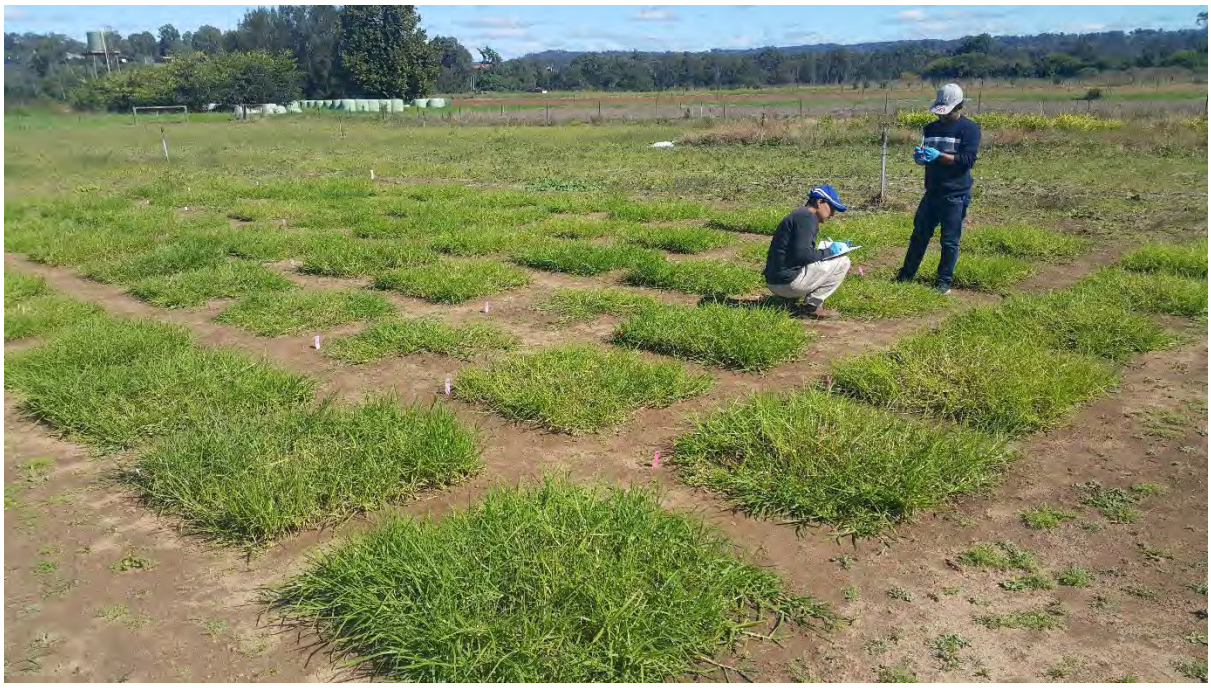

Figure 2. Small plot trial at Lansdowne in April 2022

(ii) Multi-environment strip trials of selected kikuyu genotypes

Based on small plot testing at Lansdowne in 2022-2023, three genotypes with potential were identified (L2, L8 and L10). On-farm strip trials were established of these three lines plus Whittet (L15) at three locations (Berry, Bega and Corstorphine near Camden) in the Eastern of NSW.

The strip trials were replicated and divided laterally using electric fencing to provide grazed and mowed treatments. Trials were established successfully (see Figure 3). Kikuyu biomass was harvested from the mowed treatment in each strip on a 1.5 m^2 area using a hand mower and dry matter estimated (see Figure 5). The sub-samples were subsequently analysed for nutritional traits using NIR and wet chemistry. The mowing and grazing treatments were conducted as close as possible to the same date. The trials were sampled at up to 30 different dates between November 2024 and May 2026. NDVI and a Plate Meter were used to capture estimates of greenness (chlorophyl) and biomass prior to cutting.

Figure 3. Cows grazing at Berry on the strip plot trial (photo: Laura Espinosa)

7.4 Key Findings

(i) Small plot experiment at Lansdowne

NDVI and dry matter was assessed across a 12-month period to identify lines with consistent dry matter production (Figure 4). The cumulative dry matter across the same period was also calculated (Figure 5). Three candidate lines, L2, L8 and L10 were selected for further testing in different environments with and without grazing pressure based on dry matter production at different dates of sampling and cumulative biomass production in a 12-month period.

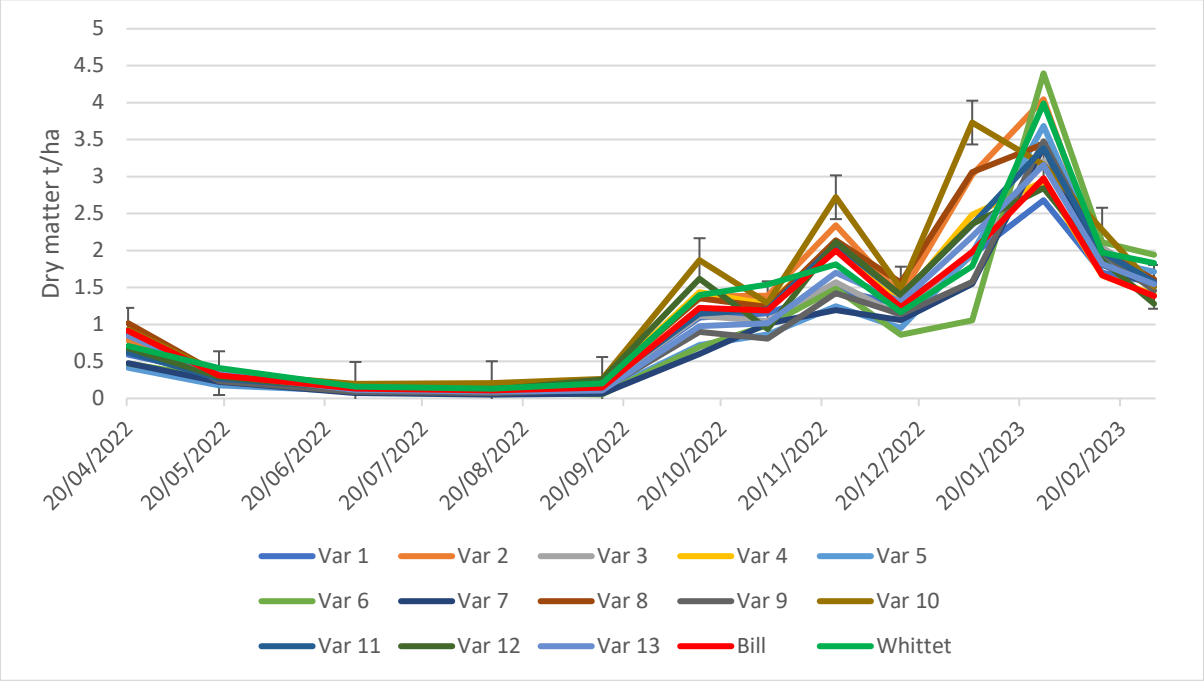

Figure 4. Dry matter production over time for 15 genotypes including Whittet and the Bill Fulkerson varieties

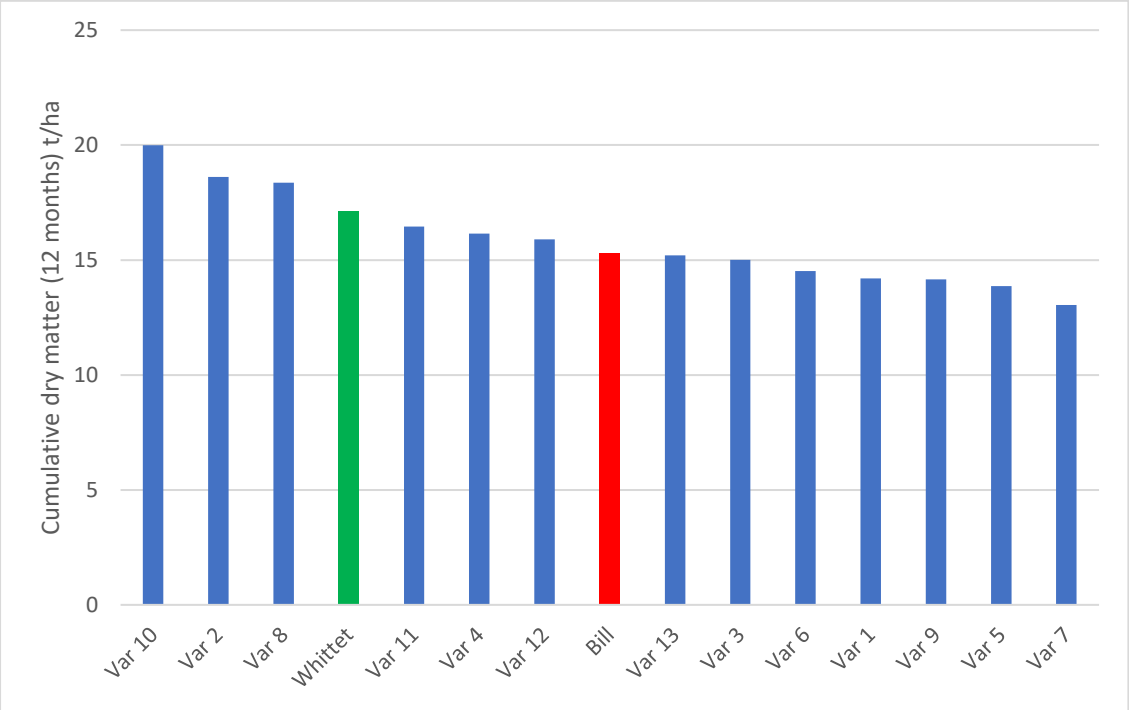

Figure 5. Cumulative dry matter production over time for 15 genotypes

(ii) Multi-environment strip trials of selected kikuyu genotypes

Based on small plot testing at Lansdowne in 2022-2023, three genotypes with potential were identified (L2, L8 and L10). These candidate lines and Whittet also referred as L15 were evaluated at

three sites in strip trials across two years. The dry matter production over time at Corstorphine, Berry, and Bega are shown on Figure 6. (Kikuyu grew under irrigated condition in Corstorphine while conditions were rainfed in Berry and Bega). As expected, the dry matter production peaked in summer and declined into winter. The 4 genotypes followed a similar pattern at all three sites.

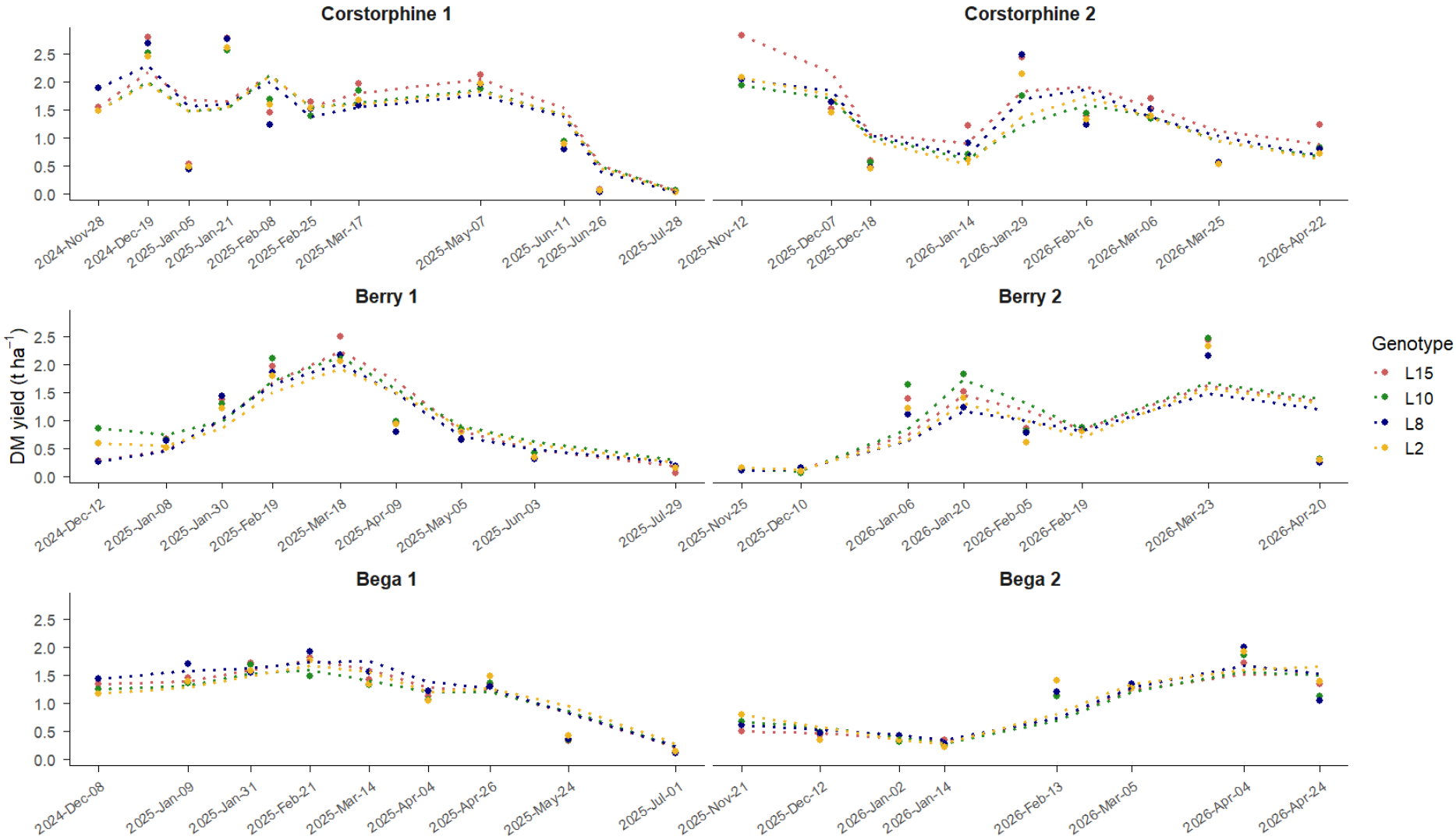

Figure 6. Dry matter production over two years (summer) at Corstorphine, Berry, and Bega. Points represent observed mean dry matter (DM) yields. Dotted lines represent the moving-average trend for each genotype within each summer season.

NDVI and the plate meter were assessed on each strip throughout the growing period in each year. No difference between treatments (moved and grazed) were observed. The plate meter was much more discriminating than NDVI for most effects (Table 2). For the plate meter, year, location and genotype main effects were highly significant, as were all two-way interactions with genotype (Table 2), indicating differences in genotype performance among environments. Whittet and L8 had the highest plate meter values across sampling dates, sites and years (Figure 7). Figure 8 shows the genotype responses at each site in each year with changes in ranking at each site over time.

Table 2. Probabilities from tests of fixed effects for NDVI and Plate Meter over years using date as a random term

	NDVI	**Plate Meter**
Fixed term	F pr	F pr
Year	<0.001	<0.001
Site	0.044	<0.001
Treatment	0.199	0.866
Genotype	0.296	<0.001
Year.FarmName	0.927	<0.001
Year.Treatment	0.305	0.235
FarmName.Treatment	0.01	<0.001
Year.Genotype	0.345	<0.001
FarmName.Genotype	0.193	<0.001
Treatment.Genotype	0.414	<0.001
Year.FarmName.Treatment	0.076	<0.001
Year.FarmName.Genotype	0.38	<0.001
Year.Treatment.Genotype	0.404	0.277
FarmName.Treatment.Genotype	0.345	0.015
Year.FarmName.Treatment.Genotype	0.393	<0.001

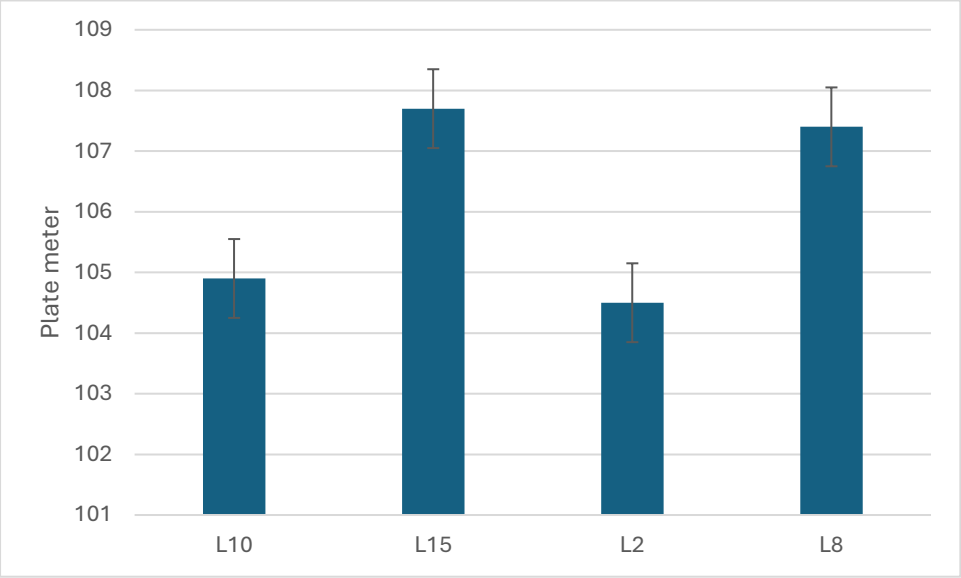

Figure 7. Plate meter results for each genotype across years, dates and locations

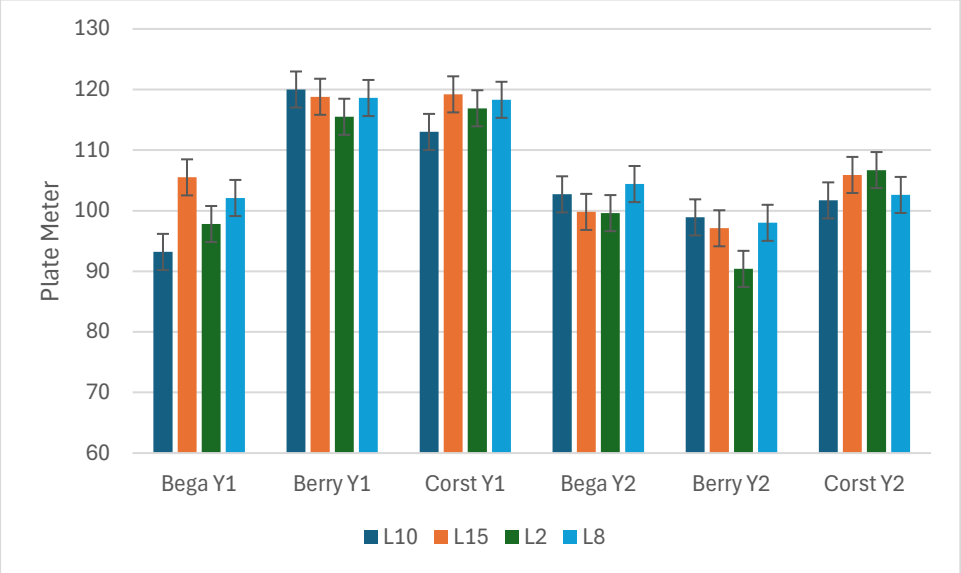

Figure 8. Plate meter results for each genotype in each year (Y1 and Y2) and location

Dry matter was assessed at regular intervals across both seasons using a lawn mower to collect fresh biomass that was later dried and weighed. No differences between seasons were observed, however, location and genotype main effects were significant as was the two-way interaction location x genotype (Table 3).

Table 3. Test of fixed effects for dry matter across years and locations using date as a random term

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Year	0.61	1	0.61	159	0.213
Location	14.58	2	7.29	159	0.013
Genotype	9.63	3	3.21	477	0.006
Year.Location	2.1	2	1.05	159	0.746
Year.Genotype	6.39	3	2.13	477	0.499
Location.Genotype	19.08	6	3.18	477	<0.001
Year.Location.Genotype	14.78	6	2.46	477	0.086

Mean dry matter across all sites, years and sampling dates showed that Whittet was in general was slightly higher than other genotypes (Figure 9).

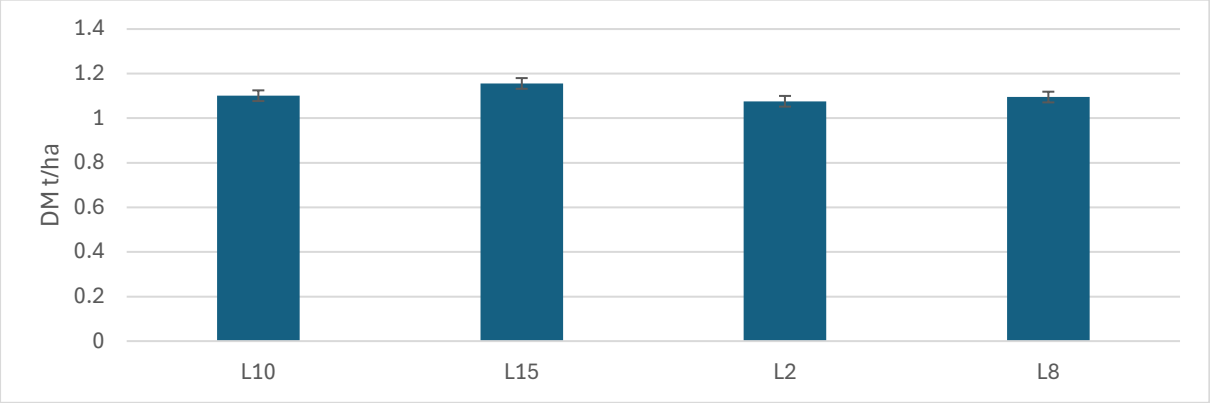

Figure 9. Mean dry matter for each genotype across years, site and dates

However, the slightly higher mean dry matter of Whittet was driven by exceptional biomass production and Corstophine in the second season (Figure 10).

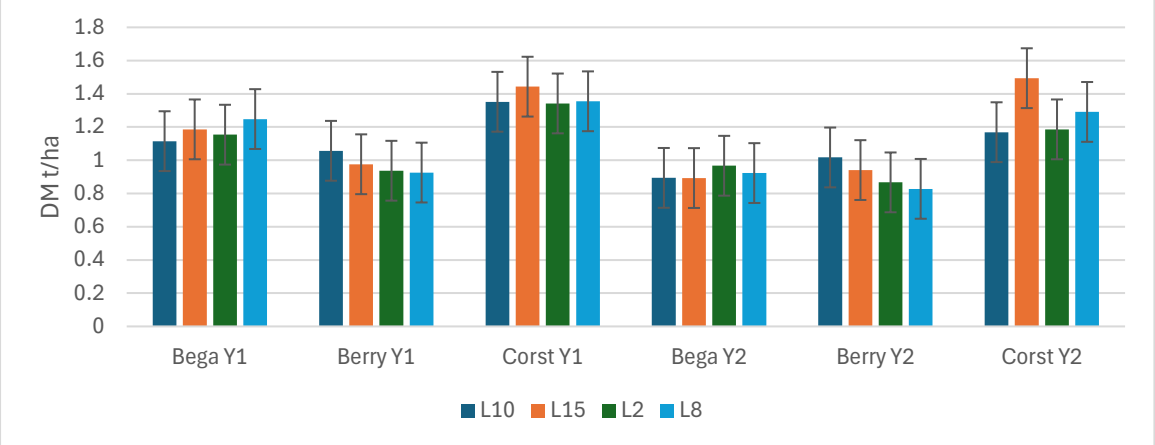

Figure 10. Mean dry matter for each genotype in each location and year

Cumulative dry matter across both seasons at each location was also calculated. There was a significant year and location main effect and genotype x location interaction (Table 4). However, the genotype main effect was not significant.

Table 4. Test of fixed effects for cumulative dry matter

Fixed term	Wald statistic	n.d.f.	F statistic	d.d.f.	F pr
Year	53.35	1	53.35	12	<0.001
Location	151.56	2	75.78	12	<0.001
Genotype	7.1	3	2.37	36	0.087
Year Location	5.94	2	2.97	12	0.089
Year.Genotype	0.98	3	0.33	36	0.806
Location.Genotype	22.26	6	3.71	36	0.006
Year.Location.Genotype	5.27	6	0.88	36	0.52

As genotype main effects were not significant, the cumulative dry matter production of each genotype in year and location was assessed (Figure 11). Once again, there was little difference among the genotypes at all sites in all years with the exception of Corstorphine in year 2, where Whittet produced significantly more dry matter across the season.

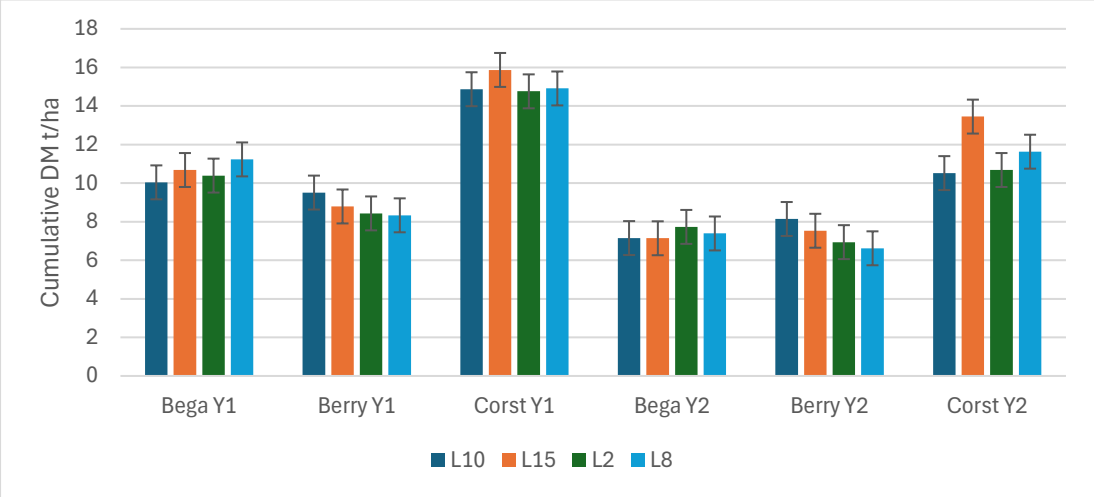

Figure 11. Cumulative dry matter for each genotype in each year and location

To examine the usefulness of NDVI and the plate meter as indirect assessments of dry matter, they were correlated across sampling dates for each genotype at each site (Table 13). The plate meter was best correlated with dry matter at Corstophine and Berry. However, the NDVI was also correlated at Corstophine. No significant associations were found in Bega, although the plate meter correlations were positive. These results suggest that the plate meter can be used to assess dry matter with a reasonable degree of accuracy.

Table 13. Correlation between dry matter and NDVI and the Plate meter at each location for each genotype across dates of sampling.

Site	**Genotype**		**NDVI**	**Plate Meter**
Corstophine	L10	Dry Matter	0.690**	0.676**
	Whittet	Dry Matter	0.467*	0.577**
	L2	Dry Matter	0.607**	0.643**
	L8	Dry Matter	0.585**	0.542*
	Mean	Dry Matter	0.65**	0.550*
Berry	L10	Dry Matter	-0.294 ns	0.804***
	Whittet	Dry Matter	0.427ns	0.777**
	L2	Dry Matter	0.451ns	0.781***
	L8	Dry Matter	0.435ns	0.819***
	Mean	Dry Matter	-0.84ns	0.801***
Bega	L10	Dry Matter	-0.004ns	0.25ns
	Whittet	Dry Matter	0.069ns	0.477ns
	L2	Dry Matter	0.056ns	0.397ns
	L8	Dry Matter	-0.028ns	0.376ns
	Mean	Dry Matter	0.0222ns	0.376ns

8. Nutritional value

Research team: R Trethowan, Laura Espinosa, Sergio Garcia, Rafiq Islam

8.1 Background

The new candidate materials have potentially competitive biomass and better disease resistance profiles compared to Whittet and the Fulkerson varieties. However, while they were screened earlier for drought and salinity tolerance by a PhD student (Duncan Fraser), nothing is known of their nutrition profile and suitability for feeding to dairy cattle.

8.2 Objectives

To evaluate the selected candidate lines for nutritional quality using biomass sampled at multiple sites

8.3 Materials & Methods

Dry matter sampled at each site over time was retained and ground for chemical analysis. The chemical analysis including NIR and physical testing using NDFD 24h was conducted by the Department of Primary Industries and Regional Development Feed Quality Service.

8.4 Key Findings

A preliminary chemical analysis of the original 15 lines was conducted using dry matter harvested per plot in 2022. The ranks of these lines for a series of nutritional traits are given in Table 5. Three candidate lines, L2, L8 and L10 were selected for further testing in different environments with and without grazing pressure using biomass, disease information and the preliminary chemical analysis. These three candidate lines and Whittet and Fulkerson were tested again in 2023 for nutritional traits (Table 6).

Table 5. Ranks of each genotype from the chemical analysis of the Lansdowne trial in 2022

RANK	DM	CDM	NDVI	ADF	CP	DMD	DOMD	ME	NDF	WSC
1	L 10	L 10	L 10	L12	L 10	L 12	L 12	L 12	L12	L 13
2	L 2	L 2	L 2	L2	L 12	Whittet	Whittet	Whittet	L11	L 12
3	L 8	L 8	L 8	L8	L 8	L 11	L 10	L 10	L8	L 9
4	Whittet	Whittet	L 11	L11	Whittet	L 8	L 11	L 11	L10	L 5
5	L 11	L 11	Whittet	L4	L 2	L 10	L 8	L 8	L13	L 1
6	L 4	L 4	L 4	Whittet	L 11	L 2	L 2	L 2	Whittet	L 2
7	L 12	L 12	L 12	L10	L 5	L 5	L 5	L 1	L5	L 3
8	Bill	Bill	L 3	L1	Bill	L 9	L 1	L 5	L4	L 11
9	L 13	L 13	L 13	L13	L 4	L 1	L 9	L 9	L2	L 10
10	L 3	L 3	L 1	L3	L 3	L 13	L 13	L 13	L1	L 7
11	L 6	L 6	Bill	L5	L 7	L 3	L 3	L 3	L3	Whittet
12	L 1	L 1	L 6	L7	L 6	Bill	Bill	Bill	L9	L 4
13	L 9	L 9	L 7	L9	L 13	L 4	L 4	L 4	L7	L 6
14	L 5	L 5	L 5	Bill	L 9	L 7	L 7	L 7	Bill	L 8
15	L 7	L 7	L 9	L6	L 1	L 6	L 6	L 6	L6	Bill

Note: DM, CDM, NDVI, adf, ash, cp, me, dmd, domd, me, ndf, wsc are dry matter, cumulative dry matter, NDVI, acid detergent fibre, ash content, crude protein, metabolizable energy, dry matter digestibility, dry organic matter digestibility, neutral detergent fibre, water soluble carbohydrates, respectively.

Of the three candidate lines, L8 and L10 both showed equivalent or slightly better chemical profiles than Whittet based on the NIR analysis (Table 6). All lines were superior to the Fulkerson variety with the exception of ash content.

Table 6. Significant traits from chemical analysis of candidate kikuyu lines and check cultivars from Lansdowne 2022 and 2023.

Variety	adf	ash	cp	me	ndf	wsc	om	Av Rank
L8	31.78 (2)	19.98 (5)	17.13 (2)	8.138 (2)	61.91 (1)	2.700 (4)	80.02 (1)	2.43
L10	31.99 (4)	21.03 (2)	17.30 (1)	8.096 (3)	62.02 (2)	3.338 (2)	78.97 (4)	2.57
Whittet	31.87 (3)	20.39 (4)	17.12 (3)	8.188 (1)	62.52 (3)	3.189 (3)	79.61 (2)	2.71
L2	31.68 (1)	20.54 (3)	16.72 (4)	7.981 (4)	62.92 (4)	3.419 (1)	79.46 (3)	2.86
Fulkerson	34.49 (5)	23.26 (1)	16.30 (5)	7.827 (5)	64.77 (5)	2.544 (5)	76.74 (5)	4.43
Std error	0.8544	1.242	0.5156	0.1297	0.9043	0.2416	1.072	

Note: the rank is given in brackets. adf, ash, cp, me, ndf, wsc, and om are acid detergent fibre, ash content, crude protein, metabolizable energy, neutral detergent fibre, water soluble carbohydrates and organic matter, respectively.

The three candidate lines plus Whittet were sown in multi-environment strip trials in 2024 and 2025 and dry matter retained from each plot and different sampling times. These materials were analysed for nutritional traits using NIR and the probabilities from the test of fixed effects are given in Table 7. Genotype main effects were all significant as were location effects with the exception of NDF. All two-way interactions were significant except for inorganic ash and water-soluble carbohydrates.

Chemical analysis of multi-environment trial samples

Table 7. Probabilities from tests of fixed effects for chemical traits assessed on the multi-environment trial in one year using NIR

	Trait (probability)								
Fixed term	**ADF**	**CP**	**DMD**	**DOMD**	**Inorganic ash**	**ME**	**NDF**	**OM**	**WSC**
Location	0.052	<0.001	<0.001	<0.001	<0.001	<0.001	0.071	<0.001	<0.001
Genotype	0.031	<0.001	0.006	0.006	0.016	0.006	<0.001	<0.001	0.002
Location.Genotype	0.032	0.004	0.017	0.012	0.382	0.012	0.008	0.004	0.24

Note: adf, ash, cp, me, ndf, wsc, and om are acid detergent fibre, ash content, crude protein, metabolizable energy, neutral detergent fibre, water soluble carbohydrates and organic matter, respectively

The means of each nutritional trait for each genotype are given on Table 8. Little difference was observed among genotypes with L8 showing highest WSC and Whittet lower NDF.

Table 8. Means of each genotype from the chemical analysis of multi-environment trial samples assessed using NIR

	Genotype				
Trait	L10	Whittet	L2	L8	Std error
ADF	23.85	23.40	23.95	23.54	0.1985
CP	20.29	21.26	20.09	20.47	0.2171
DMD	64.71	65.44	64.68	65.20	0.2482
DOMD	61.99	62.61	61.93	62.42	0.2190
Inorganic ash	10.39	10.60	10.36	10.42	0.0844
ME	9.58	9.71	9.57	9.67	0.0444
NDF	56.07	54.85	56.21	55.28	0.2944
OM	20.29	21.26	20.09	20.47	0.2170
WSC	4.64	4.54	4.67	4.92	0.1016

Note: adf, cp, dmd, domd, ash, me, ndf, wsc are acid detergent fibre, crude protein, dry matter digestibility, dry organic matter digestibility, ash content, metabolizable energy, neutral detergent fibre, organic matter, water soluble carbohydrates, respectively.

However, a subset of materials was select for wet chemistry analysis to confirm NIR observations. All genotype and location main effects were significant (Table 9). However, the two-way interactions were generally not significant indicating similar ranking at each environment.

Table 9. Probabilities from tests of fixed effects for chemical traits assessed on the multi-environment trial in one year using wet chemistry

	DMD	dNDF_DM_24	dNDF_OM_24	DOMD_24	NDF_DM	NDF_OM	NDFD_24_DM	NDFD_24_OM	OMD_24	uNDF_DM_24	uNDF_OM_24
Fixed term											
Location	0.001	0.019	0.016	0.009	0.003	0.002	0.007	0.006	0.003	0.001	<0.001
Genotype	0.02	0.02	0.023	0.008	0.059	0.08	0.025	0.022	0.022	0.018	0.023
Location xGenotype	0.052	0.077	0.096	0.011	0.104	0.135	0.103	0.1	0.03	0.111	0.09

The means from the physical analysis of samples is given in Table 10. This analysis showed that the candidate lines were superior to Whittet for most traits including DMD, DOMD, NDFD, OMD and NDF. Lines 2 and 8 in particular have a better nutritional profile than Whittet.

Table 10. Means of each genotype from the chemical analysis of multi-environment trial samples assessed using wet chemistry

	Genotype				
Trait	L10	L15	L2	L8	Std error
DMD	0.612	0.588	0.627	0.613	0.0117
dNDF_DM_24	30.03	27.66	30.08	29.19	0.8119
dNDF_OM_24	29.50	27.17	29.48	28.63	0.7996
DOMD_24	0.514	0.493	0.530	0.523	0.0101
NDF_DM	57.13	56.12	55.53	56.05	0.5642
NDF_OM	55.82	54.85	54.13	54.97	0.6197
NDFD_24_DM	0.526	0.494	0.542	0.523	0.0146
NDFD_24_OM	0.529	0.497	0.546	0.521	0.0149
OMD_24	0.577	0.554	0.596	0.581	0.0125
uNDF_DM_24	27.10	28.47	25.46	26.85	0.8748
uNDF_OM_24	26.32	27.68	24.65	26.34	0.9080

Note:

9. Disease resistance

Research team: Richard Trethowan, Percy Wong, Vivien Tan, Laura Espinosa

9.1 Background

The disease resistance of the new candidate materials was unknown beyond field observations during the PhD study of Duncan Fraser. The value of the new materials will only be understood if their disease reactions is known.

9.2 Objectives

To confirm the disease resistance of the new kikuyu materials under kikuyu yellows and black spot inoculations.

9.3 Materials & Methods

(i) Controlled environment testing

The three candidate lines plus Whittet and the Fulkerson variety were grown in pots under disease inoculation in controlled conditions to induce an epidemic. Black spot and Kikuyu Yellows diseases were assessed using standard inoculation procedures and the incidence and severity were scored.

(ii) Field evaluation

The candidate materials and the Whittet and the Fulkerson varieties were inoculated in the field on the small plot trial described in section 7. Incidence and severity were assessed as possible.

9.4 Key Findings

(i) Controlled environment testing

A. Black spot (under inoculation)

Scoring of each plot was done by manually counting the number of pustules on 10 randomly selected leaves in each pot. Lines 2, 8 and 10 showed significantly fewer symptoms compared to Whittet and Fulkerson (Figure 14).

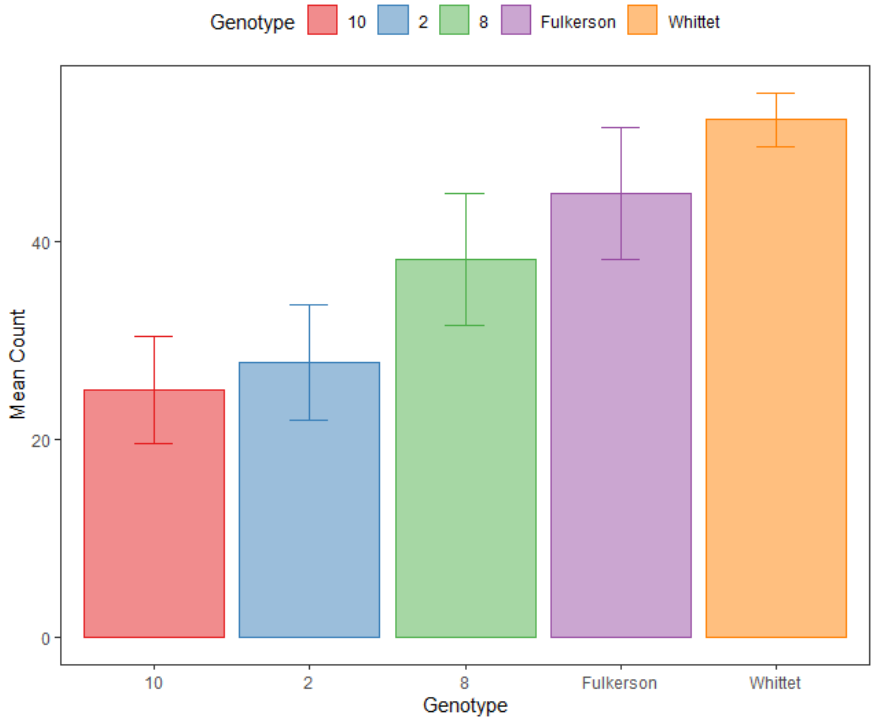

Figure 14. Blackspot disease expression on 5 genotypes

B. Kikuyu Yellows (under inoculation)

Although no significance difference between the 5 genotypes was noted for disease severity, there was a significant difference in disease progression. Line 2 had significantly slower disease progression compared to Whittet (Figure 9). The lack of differences in disease severity reflected the excessive level of infection achieved under controlled conditions. For this reason a field assessment was conducted in the 2025/26 summer.

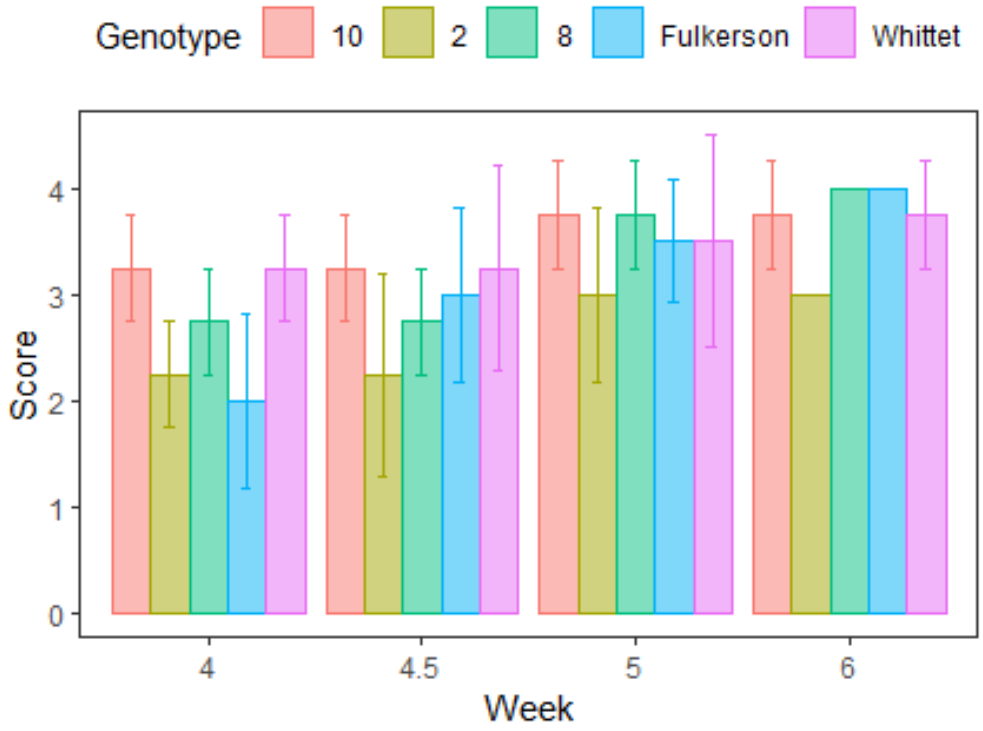

Figure 15. Kikuyu yellows expression on 5 genotypes

(ii) Field evaluation of kikuyu yellows disease at Lansdowne

Unfortunately, disease severity could not be assessed as the season cooled too quickly post the first incidence of infection. However, observations on first incidence in each replicate of the field experiment was possible. Whittet and L10 showed the first incidence of yellows disease around week 9, this was followed by L8 and L2 at week 11. The Fulkerson variety did not show incidence until week 14 when all four replicates recorded disease incidence. Based on these field results, it appears that Whittet and L10 are more susceptible than L2 and L8, which are less resistant than the Fulkerson variety. However, these are incidence only and severity still needs to be assessed.

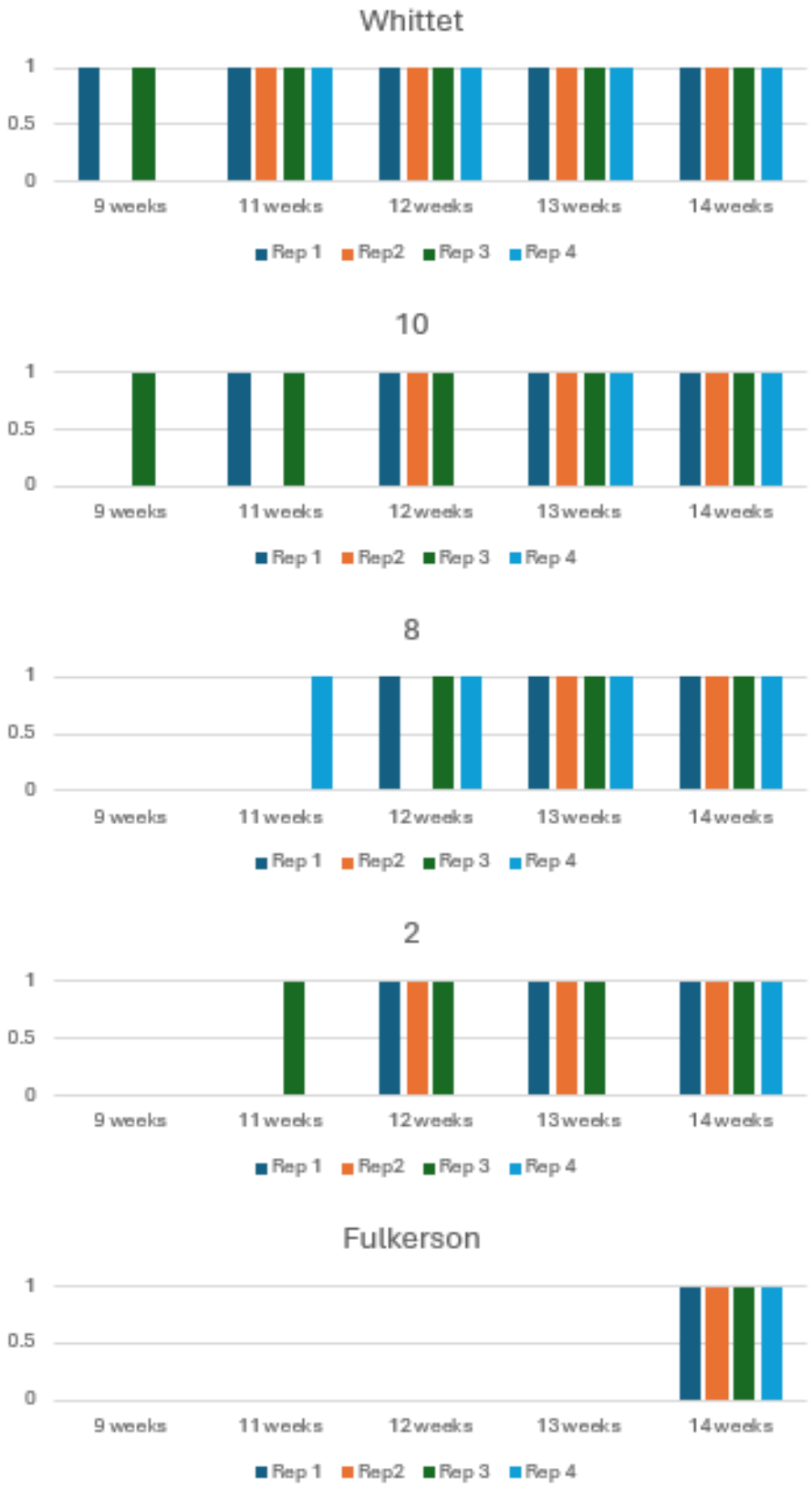

Figure 16. Boxplots of kikuyu yellows disease incidence in the field at Camden. 1 = positive and 0 = negative.

10. Key Outputs

- Results confirm the uniqueness of the new genetic materials compared to kikuyu pastures in NSW
- The diversity of kikuyu pastures across NSW was confirmed
- Two candidate lines with potential for release to dairy farmers based on dry matter production, disease resistance and nutritional profile identified

11. Conclusions and Recommendations

- Significant genetic diversity in kikuyu pastures was observed across the sampled sites in NSW with just 35% of sites clustering with Whittet and the Fulkerson line
- The new candidate kikuyu lines are genetically different and distinct from Whittet and the Fulkerson variety and generally different to most other pastures
- A relationship between kikuyu toxicity and kikuyu genotype was suggested with most instances occurring in Group 1.
- The genotypes did not differ significantly for dry matter at each date of sampling nor cumulative dry matter across two summer seasons.
- L2 and L8 have potential for release as new varieties based on genetic diversity, dry matter production (equivalent to Whittet), better black spot resistance (better than Whittet), resistance to yellows disease (better then Whittet and equivalent to Fulkerson) and superior chemical profile (better than both Whittet and Fulkerson).

Recommendations for future investment should prioritise:

- L2 and L8 should be evaluated further in the Hunter region and the NSW north coast
- The kikuyu yellow severity and incidence of the new lines need to be retested and confirmed
- The suggested link between kikuyu genotype and toxicity incidence should be investigated

Final Project Summary

P1c – Kikuyu: Developing new varieties

May 2026

Kikuyu cultivars

Kikuyu grass is a C4 grass adapted to both the tropics and temperate climates. It is fast growing and produces more dry matter of higher quality than most C4 grasses.

However, since the development of Whittet more than 50 years ago, only two new cultivars for grazing had been released (Acacia and Fulkerson).

Kikuyu genetic research

The University of Sydney in collaboration with Hatton's Turf Research have been breeding improved kikuyu grasses for more than 15 years. These new materials have greater biomass production, tolerance to salinity and drought and represent significant new diversity for nutritional factors.

Industry needs

Current kikuyu pasture cultivars are limited in adaptation, nutritional quality and scope.

However, the extent of adaptation in the new materials developed by the University of Sydney and Hatton's was largely untested in most dairy producing areas in terms of dry matter production, disease resistance and nutritional value. Such information could lead to the commercial release of improved lines and provide a basis for continued breeding and selection.

Unlocking the potential of Kikuyu

Dairy UP's P1 project aimed to unlock the potential of Kikuyu pastures used by NSW dairy farmers. P1 was a suite of five projects that collectively explored new management options to grow and utilise more Kikuyu over summer and increase the productivity of Kikuyu-based pastures.

P1a – Kikuyu: Remote monitoring

P1b – Kikuyu Investigating toxicity

P1c – Kikuyu: Developing new varieties

P1d – Kikuyu: Carbon on NSW dairy farms

P1e – Kikuyu: Nutritional value

This document is the final update on P1c – Kikuyu: Developing new varieties.

Project aims

This project explored and evaluated promising lines of kikuyu that had been selected for increased biomass production and tolerance to salinity and drought.

Key findings

The Dairy UP team identified three candidate lines of kikuyu with potential commercial value for dairy pastures.

All three candidate lines had similar dry matter production to the varieties currently used on NSW dairy farms (Whittet and Fulkerson).

Two lines had better nutritional value and were

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more resistant to black spot and kikuyu yellows than Whittet.

These two lines offer dairy farmers valuable new genetic diversity with enhanced nutritional quality and disease resistance at an equivalent yield.

Path to market

Collaboration with a commercial partner provides a clear path to market.

Once superior lines have been confirmed Hatton's Turf have the option to progress with multiplication, dissemination and marketing.

Benefits

Given that Australia's pasture lands are increasingly subject to moisture stress and warmer temperatures, this climate-ready pasture could provide greater flexibility to the animal industries including dairy.

More info

P1c Final report

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www.dairyup.com.au

Research and results

Thirteen distinct kikuyu genotypes owned by Hatton's Turf Research Pty Ltd, were selected to be evaluated in small, replicated plots at the Plant Breeding Institute at Cobbity during 2021/22. Two commercially available varieties (Whittet and Fulkerson) were included as controls.

The plots were exposed to natural conditions. Soil nutrient/moisture and daily weather were assessed throughout the experimental period. Plots were cut for biomass assessment at key periods of the year and rate and extent of re-growth assessed. Materials were screened for nutritive value and local adaptation. Concurrent pot studies were conducted in a hydroponic system for more detailed assessment of nutritive traits.

The same traits were assessed in the field at both locations for comparison and determination of stability.

Lines with potential

Based on the results, three new kikuyu lines were identified as candidates with potential commercial value. These were screened with larger plot trials in 2022/23.

Results from the initial plot trials indicated the three candidate lines were resistant to disease and grow quickly with high yields over 12 months (more than 14 tonnes/ha/yr). Their nutritional values were high in terms of crude protein, water soluble carbohydrates and fibre.

The 2022/23 plots were used to produce enough runners to establish field trials.

Field trials

The three candidate lines (and Whittet as a control) were further assessed in 2024/25 through replicated strip trials on three dairy farms in southern NSW (Bega, Berry, Corstorphine).

The trials ran from November to May with the strips managed as closely as possible to the farm's practices. Biomass was monitored using remote sensing technology and a rising plate meter. Grazing was allowed on one half of the trial and samples were collected for analysis from the mowed treatment. Biomass yield from mowing was used as a proxy for grazing and to compare with the grazing treatment.

Given the wet conditions during the 2024/25 trials, further field trials were conducted over the 2025/26 summer to better understand the performance of the candidate lines under different seasonal conditions.

Disease resistance

Lines were assessed for disease resistance in glasshouse trials where plants were inoculated with either black spot or kikuyu yellows.

To further investigate disease resistance, the 2025/26 trials included in field inoculation of black spot and kikuyu yellows.

Two of the three candidate lines were more resistant than Whittet to black spot and kikuyu yellows.

DNA testing

Throughout all trials, samples were collected for DNA testing. In addition, samples have been collected from regional dairy farms for testing.

The DNA analysis revealed more genetic diversity than expected with just 35% of sampled sites aligning with the Whittet and Fulkerson lines.

The three candidate lines are genetically distinct from the ones currently available commercially (largely Whittet, Fulkerson and Acacia).

Kikuyu toxicity

Several lines were concurrently tested in Dairy Up project P1b for differential toxicity response. PhD student, Vivien Tan, worked across both P1b and P1c to document outcomes.

There appears to be a relationship between kikuyu toxicity and kikuyu lines.

Conclusions and recommendations

Significant genetic diversity in kikuyu pastures was observed across the sampled sites in NSW with just 35% of sites clustering with Whittet and the Fulkerson line.

The new candidate kikuyu lines are genetically different and distinct from Whittet and the Fulkerson variety and generally different to most other pastures.

There appears to be a relationship between kikuyu toxicity and kikuyu lines.

Kikuyu lines do not differ significantly for dry matter at each date of sampling nor cumulative dry matter across two summer seasons.

Two lines have potential for release as new varieties based on genetic diversity, dry matter production (equivalent to Whittet), better black spot resistance (better than Whittet), resistance to yellows disease (better than Whittet and equivalent to Fulkerson) and superior nutritional profile (better than both Whittet and Fulkerson).

Priorities for future

- Further evaluate the two identified lines in the Hunter region and the NSW north coast.
- Retest and confirm the kikuyu yellow resistance.
- Further investigate the suggested link between kikuyu genotype and toxicity.

Collaborators

The P1c project was a collaboration between researchers from Dairy UP, University of Sydney and Hatton's Turf.

Delivery organisations

The University of Sydney Dairy Research Foundation | NSW Government Department of Primary Industries and Regional Development | Sebus | Dairy Australia

Partner organisations

NSW Farmers Growing the best | eastAUSmilk | Dairy NSW / Dairy Australia | AFMH | Leppington Pastoral Company Pty Ltd | NSW Local Land Services | Norco

Additional program supporters, collaborations or partnerships

Charles Sturt University | DairyBio | DataGene | Eagle Direct | Entegra
Macquarie University | NSW EPA | smaXtec | UC Davis | University of Technology Sydney

DairyUP
Unlocking potential

Final Report

P1d – Kikuyu: Carbon on NSW dairy farms

THE UNIVERSITY OF SYDNEY
Dairy Research Foundation

This project was led by Luciano Gonzalez
from the University of Sydney Dairy Research Foundation

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

Proudly funded by

Delivery organisations

Additional program supporters, collaborations or partnerships

Charles Sturt University | DairyBio | DataGene | Eagle Direct | Entegra | Macquarie University | NSW EPA | smaXtec | UC Davis | University of Technology Sydney

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1. Executive Summary

Australian dairy systems play a vital role in national food security and rural economies but contribute significantly to greenhouse gas (GHG) emissions, particularly enteric methane (CH_4) and nitrous oxide (N_2O). Achieving carbon-neutral dairy production requires system-specific, evidence-based strategies that reduce emissions while supporting productivity and long-term farm viability. This Dairy UP project, supported by the NSW Environmental Protection Authority, generated integrated data on whole-farm emissions, feeding strategies, CH_4-monitoring technologies, soil carbon (C) and others nutrient stocks, digital soil product accuracy, and pasture-level GHG fluxes across Australian dairy systems.

Whole-farm assessments showed that emissions intensity was similar between confinement and pasture-based systems, averaging 1.02 ± 0.038 and 1.07 ± 0.069 kg CO_2-eq/kg fat- and protein-corrected milk (FPCM), respectively. Enteric CH_4 was the dominant emission source (54–58%) in both systems. Secondary emissions differed by production system: manure-related emissions were higher in confinement systems, while fertiliser-related N_2O and upstream feed emissions were higher in pasture-based systems. Tree sequestration offset up to 6% of emissions in pasture-based systems but only 1% in confinement systems. These findings underscore where priorities may lie to mitigate GHG emissions in each production system such as improved manure management in confinement systems, fertiliser optimisation in pasture systems, and enteric CH_4 reduction across both systems.

Analysis of 120 commercial pasture-based farms showed that 2–3 t DM/cow/year concentrate supplementation increased milk production and reduced emissions intensity by approximately 12%. Although farm profits varied, the overall trend shows that this level of concentrate feeding can improve emissions efficiency, with actual profitability depending on each farm's management and market conditions.

Low-cost CH_4 sensors demonstrated moderate agreement with GreenFeed but lower repeatability and sensitivity to environmental conditions. While not yet suitable as a replacement for established systems, the results indicate strong potential for future low-cost, scalable CH_4 monitoring to support benchmarking and mitigation verification.

Soil assessments across nine NSW dairy farms showed that pasture-based systems stored substantially more soil organic carbon (75% higher) and nitrogen (65% higher) than confinement systems, particularly under permanent pasture and tree cover. In contrast, confinement systems accumulated significantly higher phosphorus stocks (3.7-fold higher), underscoring the need for targeted nutrient management. These findings highlight how land use, climate, and farming system influence soil C and nutrient stocks.

Validation of the Soil and Landscape Grid of Australia (SLGA) showed moderate accuracy for C and nitrogen at whole-farm scales but poor performance for phosphorus and fine-scale variation, limiting its suitability for carbon crediting or paddock-level planning. While SLGA can assist with broad baseline assessments where direct sampling is not possible, its accuracy is not sufficient for C crediting or for sub-farm planning and nutrient management decisions.

Measurements of GHG fluxes from C3 (ryegrass) and C4 (kikuyu) pastures revealed that pasture type and management strongly influence short-term C dynamics. Ryegrass pastures acted as net CO_2 sinks during regrowth, whereas kikuyu pastures became net GHG sources when surplus biomass required slashing. Shorter chamber closure times improved flux accuracy, and non-linear models were

necessary for reliable CO_2 estimation under longer closures. These findings highlight the importance of pasture species, grazing management, and measurement methodology in assessing pasture-level contributions to whole-farm C balances.

Overall, the project shows that achieving C neutrality in Australian dairy systems is possible only through integrated, system-specific pathways that combine enteric CH_4 reduction, productivity-enhancing nutrition, improved manure and fertiliser management, and long-term stewardship of soil and vegetation C. One-size-fits-all mitigation approaches are unlikely to deliver optimal outcomes.

2. Project Overview

Item	Description
Project Title	Kikuyu: Carbon on NSW dairy farms
Funding Body	Dairy UP and NSW EPA
Dairy UP Project	PhD research component
Project Duration	2022–2026
Lead Organisation	The University of Sydney
Project Lead	Luciano Gonzalez
Key Collaborators	The University of Sydney and The University of Japan

Research team: Prof. Luciano González (University of Sydney), Mulisa Dida (University of Sydney), Prof. Sergio García (University of Sydney), Madison Luke (University of Sydney), Milad Bagheri Shirvan (University of Sydney), Budiman Minasny (University of Sydney), Ho Jun Janga (University of Sydney), Feike A. Dijkstra (University of Sydney) and Toshikazu Kawaguchi (Hokkaido University, Japan).

3. Abbreviations

ADCC	**Australian Dairy Carbon Calculator**
ADSA	**American Dairy Science Association**
AARN	**Australian Association of Ruminant Nutrition**
C	**Carbon**
ASAS	**American Society of Animal Science**
CH_4	**Methane**
CO_{2eq}	**Carbon dioxide equivalent**
DM	**Dry matter**
DRF	**Dairy Research Foundation**
EBIT	**Earnings before interest and tax**
FPCM	**Fat protein corrected milk**
g	**Gram**
GHG	**Greenhouse gases**
HDR	**Higher Degree by Research**
Kg	**Kilogram**
LCA	**Life cycle assessment**
n	**Number of observations**
N_2O	**Nitrous oxide**
NSW	**New South Wales**
NZSSS	**New Zealand Society of Soil Science**
SLGA	**Soil and Landscape Grid Australia**
SOC	**Soil organic carbon**
SOLES	**School of Life and Environmental Science**
SSA	**Soil Science Australia**
t	**Tone**
USYD	**The University of Sydney**

4. Project Background and Rationale

The Australian dairy industry is a major contributor to national food security and rural economies, but it also generates substantial greenhouse gas (GHG) emissions, particularly enteric methane (CH_4), manure-derived CH_4, and fertiliser-related nitrous oxide (N_2O) (Christie, 2019; Garnett and Eckard, 2024). Nationally, dairy farming produces an estimated 9–10 Mt CO_2-eq annually, accounting for around 12–14% of agricultural emissions, with enteric CH_4 consistently representing more than half of total farm-level emissions (Australian Government DCCEEW, 2023; Christie, 2019). In response to climate change and obligations under the Paris Agreement, Australia has committed to net-zero emissions by 2050 and an interim 43% reduction below 2005 levels by 2030. Consistent with these national targets, Dairy Australia aims to reduce the emissions intensity of dairy production by 30% relative to 2015 levels by 2030 (Dairy Australia & Agriculture Victoria, 2021).

Australian dairy farms operate under diverse production systems, ranging from predominantly pasture-based operations to increasingly intensive confinement and hybrid systems (Christie, 2019; Joubran et al., 2021). These systems differ in feeding strategies, manure management, fertiliser use, land-use patterns, and reliance on external inputs, resulting in distinct emissions profiles and mitigation opportunities (Christie, 2019; Garnett and Eckard, 2024). Feeding strategies particularly concentrate supplementation play a central role in productivity and emissions intensity, yet their combined environmental and economic impacts under commercial Australian conditions remain insufficiently quantified (Arndt et al., 2022).

Progress toward emissions reduction is further constrained by limited access to affordable CH_4-monitoring technologies. Gold-standard systems such as GreenFeed provide accurate measurements but are costly and impractical for widespread adoption (Hammond et al., 2016; Hristov et al., 2018). Soils and vegetation also contribute significantly to whole-farm C balances, but reliable farm-scale information on soil C and nutrient stocks is scarce (Guo and Gifford, 2002; Eckard and Cullen, 2011). Digital soil products such as the Soil and Landscape Grid of Australia (SLGA) offer potential for scalable assessment, yet their accuracy for dairy systems has not been well validated. Pasture systems themselves influence C dynamics through CO_2 exchange (Liang et al., 2021, Bagheri Shirvan et al., 2025), but empirical data on GHG fluxes from C3 and C4 dairy pastures in Australia remain limited.

This Dairy UP project was established to address these knowledge gaps. By integrating whole-farm emissions assessment, feeding strategy analysis, CH_4-monitoring technology evaluation, soil and nutrient stock quantification, digital soil product validation, and pasture-level GHG flux measurement, the project provides practical, regionally relevant evidence to support climate-smart dairy production and inform pathways toward carbon-neutral dairy systems.

5. Project Objectives

The overall aim of this project was to generate practical, region-specific evidence to support reductions in GHG emissions while maintaining productivity and profitability in Australian dairy systems. The specific objectives were to:

- Quantify whole-farm GHG emissions from pasture-based and confinement dairy systems and identify the major emission sources.
- Assess how different levels of concentrate supplementation affect milk production, emissions intensity, and farm profitability in commercial farms.
- Evaluate the performance of low-cost CH_4 sensors against the GreenFeed system to determine their potential for scalable on-farm CH_4 monitoring.
- Quantify soil C and nutrient stocks across key land-use types in pasture-based and confinement systems.
- Test the reliability of the SLGA for farm- and sub-farm-scale assessment of soil C, nitrogen, and phosphorus.
- Quantify GHG fluxes from C3 and C4 pastures and evaluate how pasture type, grazing, and measurement methodology influence short-term carbon dynamics.

Together, these objectives were designed to inform practical, whole-farm mitigation strategies and support evidence-based decision-making for industry, policymakers, and producers working toward carbon-neutral dairy pathways.

6. Subproject 1: Whole-farm GHG emissions of confinement vs pasture-based dairy systems

Research team: Prof. Luciano González (University of Sydney), Mulisa Dida (University of Sydney), and Prof. Sergio García (University of Sydney),

6.1. Background & Rationale

Australian dairy farms operate under diverse production systems that differ in feeding, manure management, fertiliser use, and land-use patterns, leading to distinct emissions profiles. However, there is limited Australian-specific data comparing whole-farm GHG emissions across confinement and pasture-based systems using consistent boundaries. This subproject addressed this gap by quantifying system-level emissions and identifying mitigation priorities.

6.2. Objectives

- Quantify whole farm GHG emissions from pasture based and confinement dairy systems.
- Identify major emission sources and system specific mitigation priorities.
- Assess the contribution of tree-based C sequestration to whole farm emissions.
- Provide evidence to support system specific pathways toward C neutral dairy production.

6.3. Methods

Farm sample: Ten commercial NSW dairy farms were assessed (five pasture-based and five confinement).

Emissions modelling: Whole-farm GHG emissions were estimated using the Australian Dairy Carbon Calculator (ADCC), incorporating:

- Herd structure and productivity
- Feed used and purchased feed emissions
- Manure management systems
- Fertiliser inputs and N_2O emissions
- Energy use
- Tree cover and carbon sequestration

Emission sources included:

- Enteric CH_4
- Manure CH_4 and N_2O
- Fertiliser-related N_2O
- Upstream feed emissions
- Tree-based carbon offsets
- All results were expressed as kg CO_2-eq per kg fat- and protein-corrected milk (FPCM).

6.4. Key Findings

- Emission intensity was similar across systems
  - *Pasture-based: 1.07 ± 0.069 kg CO_2-eq/kg FPCM*
  - *Confinement: 1.02 ± 0.038 kg CO_2-eq/kg FPCM*
- Enteric CH_4 dominated emissions, confirming its central role in mitigation.
- Manure emissions were substantially higher in confinement systems *(31% of total vs 13% in pasture-based)* due to stored manure and anaerobic conditions.
- Fertiliser and pre-farm emissions were higher in pasture-based systems, reflecting greater reliance on N fertiliser.
- Tree-based carbon offsets were modest, contributing:
  - *~1% of total emissions in confinement systems*
  - *~6% in pasture-based systems*
- System-specific mitigation priorities emerged:
  - Confinement → manure management
  - Pasture-based → fertiliser optimisation
  - Both → enteric CH_4 reduction

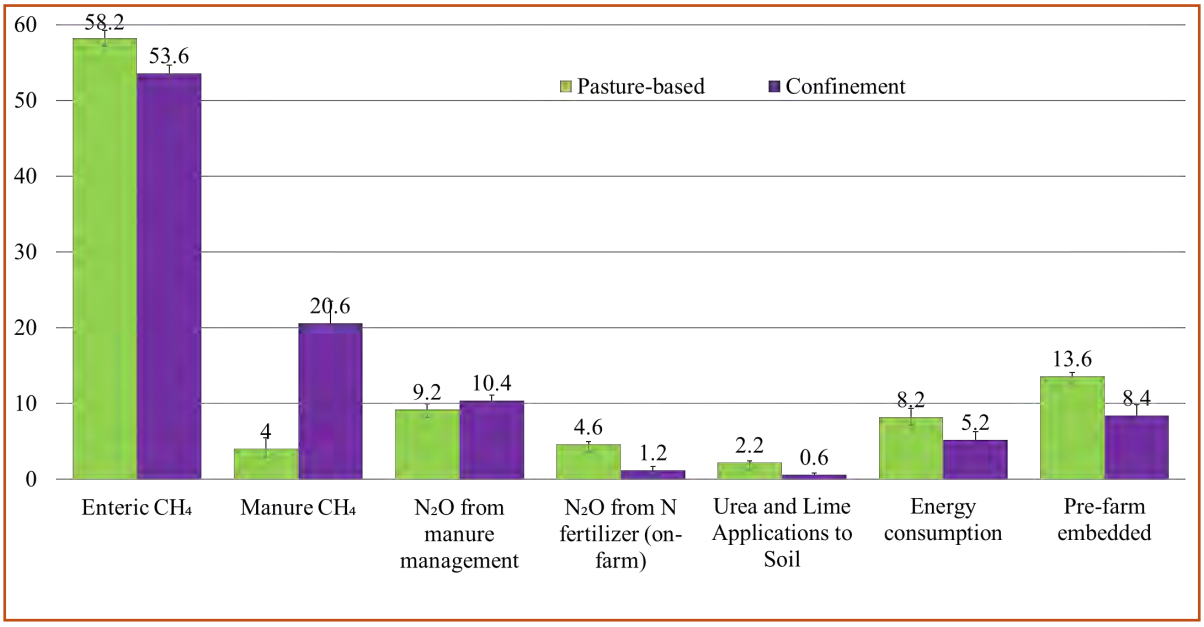

Figure 1. Breakdown by source, % of total farm CO_{2eq}

6.5 Applications & Impacts

Farm-level:

- Confinement farms can reduce emissions through improved manure storage and handling.
- Pasture-based farms can benefit from fertiliser efficiency strategies and optimised N use.
- Enteric CH_4 mitigation (e.g., feed additives, genetics, diet optimisation) is essential across all systems.

Industry-level:

- Results support improved national GHG inventories and C-accounting frameworks.
- Evidence informs Dairy Australia's sustainability reporting and net-zero planning.

Environmental:

- Tree cover provides modest offsets but cannot replace direct mitigation.
- System-specific strategies are required; one-size-fits-all approaches are ineffective.

6.6. Limitations of the study

- SOC sequestration was not included in whole-farm balances.
- Sample size (n=10 farms) limits national representativeness.
- Tree sequestration estimates rely on default factors.

6.7. Key Outputs

- Whole-farm emissions dataset for 10 NSW dairy farms.
- Peer-reviewed publication in Journal of Dairy Science (2025).
- Integrated modelling framework combining herd, feed, manure, fertiliser, and tree-cover data.
- Conference presentations at ADSA, AARN, and DRF Symposium.

6.8. Future Research Opportunities

- Expand whole-farm assessments across more regions and production systems.
- Integrate soil C sequestration into whole-farm C balances.
- Evaluate combined mitigation strategies (feeding, manure, fertiliser, genetics).
- Develop system-specific emission factors for Australian conditions.
- Support industry adoption through training and improved C-accounting tools.

7. Subproject 2: Concentrate supplementation, emissions intensity, and profitability

Research team: Prof. Luciano González (University of Sydney), Mulisa Dida (University of Sydney), and Prof. Sergio García (University of Sydney).

7.1. Background & Rationale

Concentrate supplementation is widely used to support milk production in pasture-based dairy systems, yet its combined effects on emissions intensity and profitability under commercial Australian conditions remain poorly quantified. Variation in supplementation practices across regions creates uncertainty about optimal feeding levels. This subproject examined how different supplementation rates influence productivity, emissions, and economic outcomes.

7.2. Objectives

- Quantify how different levels of concentrate supplementation affect milk yield.
- Assess the impact of supplementation on emissions intensity (CO_2-eq/kg FPCM).
- Evaluate economic outcomes (gross margin, EBIT) across supplementation levels.
- Identify supplementation ranges that optimise both productivity and emissions efficiency.

7.3. Methods

Dataset: A stratified sample of 120 pasture-based dairy farms across South Australia, New South Wales, Tasmania, and Victoria was analysed using Dairy Australia's Dairy Farm Monitoring Project dataset.

Farm classification: Farms were grouped by annual concentrate feeding level:

- Low: <1 t DM/cow/year
- Moderate: 1-2 t DM/cow/year
- High: 2–3 t DM/cow/year
- Very high: >3 t DM/cow/year

Emissions modelling: Emissions were estimated using the Australian Dairy Carbon Calculator (ADCC) following National Greenhouse Gas Inventory methodologies.

Economic analysis: Gross margin and EBIT were calculated using data extracted from the DFMP dataset and compared across supplementation groups.

7.4. Key Findings

- Supplementation of 2–3 t DM/cow/year increased milk yield and reduced emissions intensity by approximately 12% compared with supplementation of <1 t DM/cow/year
- Economic indicators (gross margin, EBIT) were generally highest in the 2–3 t DM/cow/year.
- Supplementation above 3 t DM/cow/year showed diminishing productivity returns and higher manure-related emissions.
- Productivity gains were the primary driver of improved emissions efficiency.

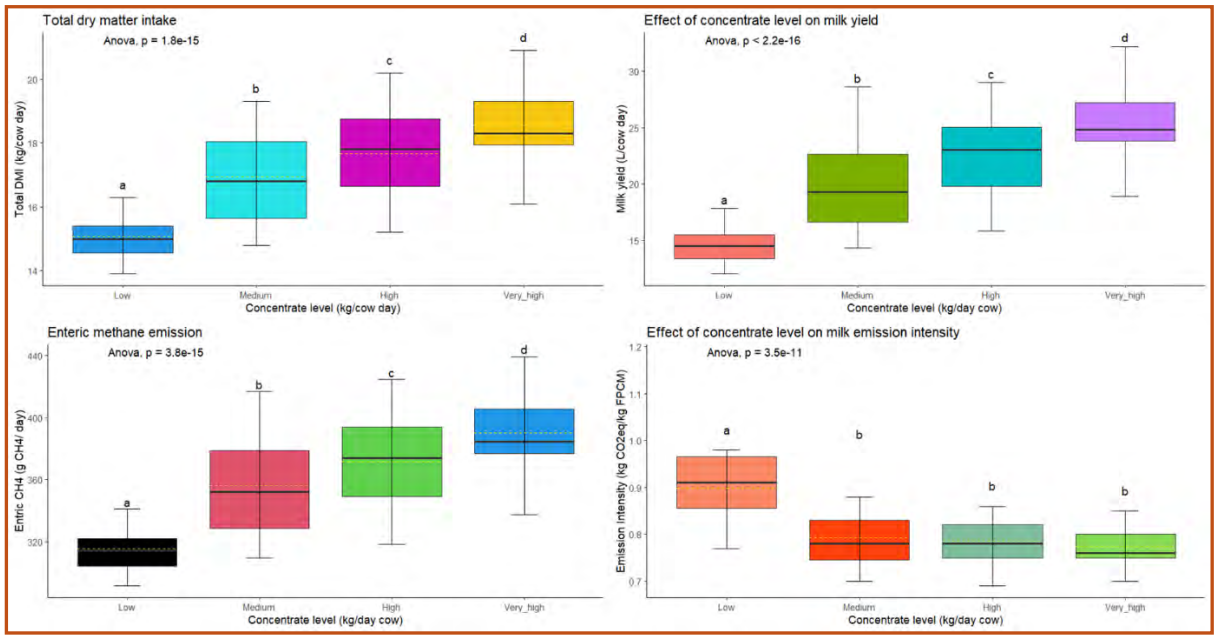

Figure 2. Effects of concentrate feeding on dry matter intake (DMI), Milk yield, enteric CH_4, FPCM emission intensity

7.5. Applications & Impacts

Farm-level:

- 2–3 t DM/cow/year supplementation is a practical, scalable strategy to reduce emissions intensity.
- Over-supplementation (>3 t) increases costs.
- Feeding strategies should be integrated with manure management to avoid shifting emissions between sources.

Industry-level:

- Results support refinement of feeding guidelines and benchmarking tools.
- Evidence informs Dairy Australia's sustainability reporting and extension programs.

Environmental:

- Improved emissions efficiency contributes to national net-zero targets.
- Strategic supplementation reduces emissions per litre of milk without compromising profitability.

7.6. Limitations of this study

- Feed quality and dry matter intake were not directly measured.
- Economic outcomes vary with milk price and feed cost fluctuations.

7.7 Key Outputs

- Integrated dataset linking supplementation, emissions, and profitability across 120 farms.
- Peer-reviewed publication in *Journal of Dairy Science* (2024).
- Conference presentations at ADSA, AARN, and DRF Symposium.
- Evidence-based recommendations for feeding strategies in pasture-based systems.

7.8 Future Research Opportunities

- Conduct controlled trials measuring dry matter intake, feed quality, and CH_4 emissions.
- Develop diet-specific emission factors for Australian pasture-based systems.
- Evaluate combined feeding and manure-management strategies.
- Model long-term economic and environmental impacts of supplementation strategies.
- Integrate supplementation data into digital decision-support tools.

8. Subproject 3: Low-cost Methane Sensor Validation

Research team: Prof. Luciano González (University of Sydney), Mulisa Dida (University of Sydney), Prof. Sergio García (University of Sydney), Milad Bagheri Shirvan (University of Sydney) and Toshikazu Kawaguchi (Hokkaido University, Japan).

8.1. Background

Accurate CH_4 measurement is essential for benchmarking emissions and verifying mitigation strategies, but existing systems such as GreenFeed are costly and impractical for widespread farm use. Low-cost sensors offer potential for scalable monitoring, yet their performance under real farm conditions is not well understood. This subproject evaluated the accuracy and suitability of a low-cost CH_4 sensor for dairy applications.

8.2. Objectives

- Test the performance of a low-cost CH_4 sensor against the GreenFeed system.
- Assess whether the sensor can detect meaningful differences in CH_4 output.
- Determine whether the technology is suitable for on-farm monitoring or requires further development.

8.3. Materials & Methods

A 45-day trial was conducted with 28 dairy heifers. Each animal was monitored using:

- A low-cost MQ-4 CH_4 sensor module
- A GreenFeed system was used as reference method

Sensor performance was evaluated using:

- Correlations between the two systems
- Repeatability (how consistent the sensor is over time)
- Daily and weekly CH_4 patterns

8.4. Key Findings

- Moderate agreement with GreenFeed: Weekly correlation was 0.62, meaning the sensor captured general CH_4 patterns but not precise values.
- Lower repeatability than GreenFeed: The sensor's repeatability was 0.13 compared with 0.31 for GreenFeed, indicating more variability.
- Environmental sensitivity: Temperature, humidity, and airflow affected sensor readings, reducing accuracy.
- Potential for future use: While not yet suitable as a replacement for GreenFeed, the sensor showed enough promise to justify further development.

8.5. Applications & Impacts

Farm-level:

- Low-cost sensors could eventually allow farmers to track CH_4 trends without expensive equipment.
- They could support decision-making around feed changes, additives, or herd management.

Industry-level:

- Affordable CH_4 monitoring is essential for verifying mitigation strategies and supporting carbon-neutrality programs.
- This work provides the first Australian evidence on the potential of low-cost sensors.

Environmental:

- Scalable CH_4 monitoring is a key enabler of national emissions-reduction goals.

8.6. Limitations

- Sensor performance was tested under a single season and production system, limiting broader applicability.
- Readings were sensitive to temperature, humidity, and airflow, which were not fully corrected for.
- Calibration relied on one reference method (GreenFeed) and one animal group.
- The study assessed short-term performance only; long-term sensor drift was not evaluated.
- The sensor measured relative CH_4 concentrations rather than absolute emissions.

8.7. Key Outputs

- First Australian field evaluation of a low-cost CH_4 sensor for dairy cattle.
- Peer-reviewed publication in *Smart Agricultural Technology* (2025).
- Dataset linking sensor readings with GreenFeed measurements.

8.8. Future Research Opportunities

- Improve sensor calibration and environmental correction algorithms.
- Test sensors across different feeding systems, climates, and herd types.
- Integrate sensors into digital farm platforms for real-time monitoring.
- Develop low-cost systems suitable for commercial adoption.

9. Subproject 4: Soil Carbon and Nutrient Stocks Across Dairy Land-Use Types

Research team: Prof. Luciano González (University of Sydney), Mulisa Dida (University of Sydney), Prof. Sergio García (University of Sydney), Madison Luke (University of Sydney), Milad Bagheri Shirvan (University of Sydney), and Budiman Minasny (University of Sydney),

9.1. Background

Soil C and nutrient stocks are critical components of whole-farm C balances and nutrient management, yet farm-scale data for Australian dairy systems are limited. Differences in land use, climate, and production system likely influence soil properties, but these patterns are not well quantified. This subproject assessed soil C, N, P and other soil physiochemical properties and stocks across major land-use types in NSW dairy farms.

9.2. Objectives

- Quantify soil C, and other soil physiochemical properties and stocks across major land-use types.
- Compare soil C and other soil physiochemical properties and stocks stocks between pasture-based and confinement dairy systems.
- Identify land-use drivers of soil C and nutrient variation.

9.3. Methods

Soil samples (n=810) were collected from nine NSW dairy farms across permanent pasture, mixed pasture-cropping, cropping, tree areas, and natural pasture. Laboratory analyses quantified SOC, total N, total P and other soil physiochemical properties. Stocks were calculated based on land-use proportions.

9.4. Key Findings

- Pasture-based farms stored more carbon and nitrogen:
    - *SOC: 75% higher than confinement farms*
    - *Total N: 65% higher*
- Tree areas and permanent pastures had the highest SOC and N stocks.
- Confinement farms had much higher phosphorus stocks (3.7-fold higher).
- Rainfall and land-use history were major drivers of SOC differences.

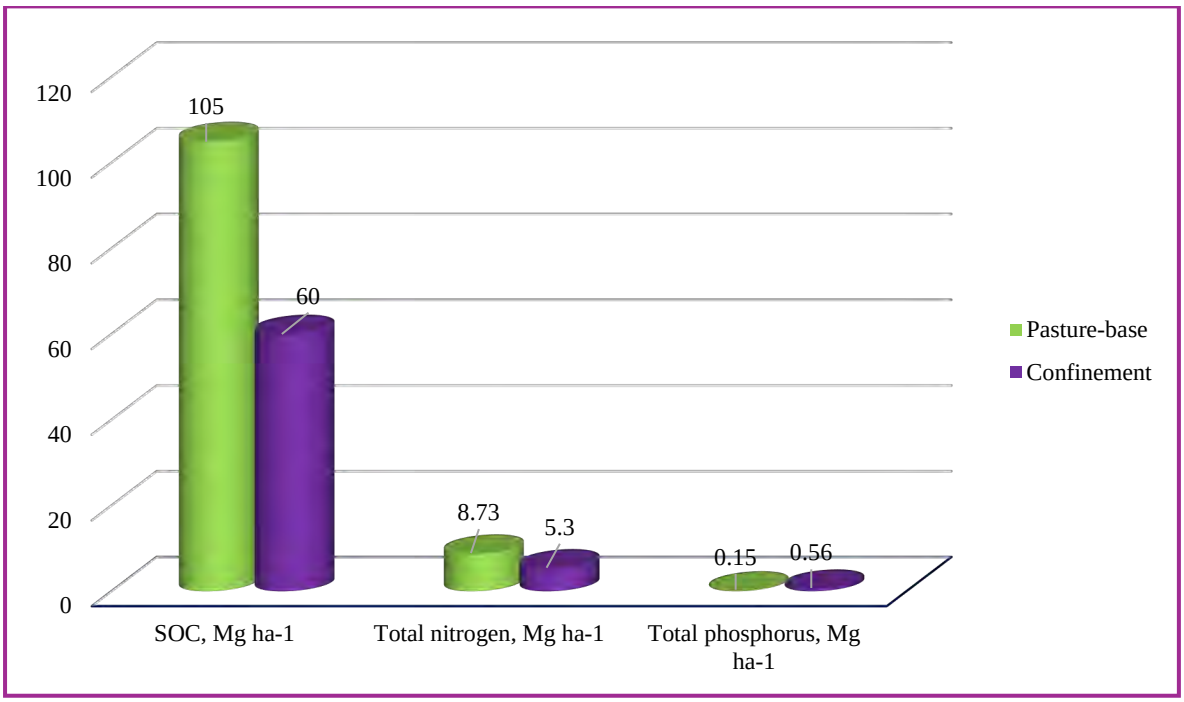

Figure 3. Effect of farming system on SOC stock and total nitrogen and phosphorus (Mg ha⁻¹) in dairy farms

9.5. Applications & Impacts

Farm-level:

- Maintaining permanent pastures and tree areas helps build soil C.
- Confinement farms need careful phosphorus management to avoid nutrient overload.
- Soil testing supports better fertiliser decisions and cost savings.

Industry-level:

- Results support improved C accounting and nutrient-management guidelines.
- Evidence helps refine national soil-C models for dairy systems.

Environmental:

- Healthy soils contribute to climate resilience and long-term sustainability.

9.6. Limitations

- The study assessed soil stocks at one point in time; changes in soil C sequestration rates were not measured.
- Sampling was limited to NSW dairy farms, reducing national representativeness.
- Sampling occurred across farms with different rainfall zones, soil types, and management histories, which may influence comparability

9.7. Key Outputs

- Comprehensive soil dataset across nine NSW dairy farms.

- Peer-reviewed publication in Environmental Impact Assessment Review (2026).
- Evidence supporting soil stewardship in dairy systems.

9.8. Future Research Opportunities

- Repeat sampling to measure soil C change over time.
- Expand assessments to other states and production systems.
- Integrate soil data into whole-farm C-neutrality models.
- Develop improved nutrient-management tools for confinement systems.

10. Subproject 5: Validation of the Soil and Landscape Grid of Australia

Research team: Prof. Luciano González (University of Sydney), Mulisa Dida (University of Sydney), Prof. Sergio García (University of Sydney), Budiman Minasny (University of Sydney), and Ho Jun Janga (University of Sydney).

10.1. Background

Digital soil products such as the Soil and Landscape Grid of Australia (SLGA) are increasingly used for C accounting and environmental reporting, but their accuracy for dairy farms has not been validated. Dairy landscapes have complex land-use patterns and high nutrient turnover, which may challenge model performance. This subproject tested SLGA predictions against measured soil data at farm and sub-farm scales.

10.2. Objectives

- Evaluate the accuracy of SLGA predictions for soil C, nitrogen, and phosphorus at both sub-farm (individual Carbon Estimation Areas, CEAs, i.e., paddocks or groups of paddocks with similar land use and management within a farm) and farm scales (aggregated across all CEAs within a farm).

10.3. Materials & Methods

The Soil and Landscape Grid of Australia (SLGA) was evaluated using the same soil dataset from the study of soil C and nutrient stocks across dairy land use types. SLGA predictions for soil C, nitrogen, and phosphorus were compared with laboratory measurements from 810 soil samples collected across nine NSW dairy farms at both point and farm scales. Standard accuracy and error metrics were used to assess the suitability of SLGA for farm-level C accounting.

10.4. Key Findings

- Moderate accuracy for soil C and nitrogen: SLGA captured broad differences between farming systems.
- Poor accuracy for phosphorus: Large errors and weak correlations make SLGA unsuitable for P-related decisions.
- Unreliable for sub-farm planning: SLGA resolution is too coarse for paddock-level management.
- Useful for broad baselines: SLGA can support preliminary assessments where direct sampling is not possible.

10.5. Applications & Impacts

Farm-level:

- SLGA can provide a starting point for understanding soil C and nitrogen.
- Ground-truthing is essential for phosphorus and for paddock-level decisions.

Industry-level:

- Results clarify when national digital tools are appropriate.
- Findings support development of improved digital soil products.

Environmental:

- Better soil information supports C accounting and nutrient stewardship.

10.6. Limitations

- •SLGA predictions were evaluated using soil samples from a limited number of NSW dairy farms, reducing national representativeness.
- The resolution of SLGA products is coarse relative to paddock-scale variability, limiting fine-scale comparison.
- SLGA models were not calibrated specifically for dairy landscapes, which have high nutrient turnover and complex land-use patterns.
- Soil samples and SLGA predictions were not always matched under identical temporal or environmental conditions.
- The study assessed SLGA accuracy at one point in time; temporal changes in soil properties were not evaluated.

10.7. Key Outputs

- First dairy-specific validation of SLGA using real farm soil data.
- Peer-reviewed publication under review in Environmental Technology & Innovation.
- Practical guidance for using digital soil tools in dairy systems.

10.8. Future Research Opportunities

- Improve digital soil mapping for nutrient-specific accuracy.
- Develop dairy-system-specific calibration datasets.
- Integrate digital tools with targeted soil sampling strategies.
- Support industry training on appropriate use of digital soil products.

11. Subproject 6: Greenhouse gas fluxes from C3 and C4 pastures

Research team: Prof. Luciano González (University of Sydney), Milad Bagheri Shirvan (University of Sydney), and Feike A. Dijkstra (University of Sydney).

11.1. Background

Pasture systems influence whole-farm carbon balances through CO_2 exchange, yet empirical data on greenhouse gas fluxes from C3 and C4 dairy pastures in Australia are limited. Differences in species, growth patterns, and management may affect short-term carbon dynamics. This subproject quantified GHG fluxes from ryegrass and kikuyu pastures and evaluated methodological factors affecting flux measurement.

11.2. Objective

This project included two studies examining methodological and ecological aspects of GHG flux measurement in pastures:

- To examine the effect of different mathematical models on GHG flux estimation and identify the optimal chamber closure period in both C_3 and C_4 pastures,
- To investigate the relationship between GHG fluxes and aboveground biomass changes in C_3 and C_4 pastures.

11.3. Methods

These studies were conducted in two pasture types in Western Sydney, NSW, a ryegrass (C_3) pasture and a kikuyu (C_4) pasture. An area of 12 × 12 m was selected as trial site in each pasture. The measurements were performed in spring in C_3 pasture, and in summer in C_4 pasture. The C_3 pasture was dominated by annual ryegrass (*Lolium multiflorum L.*), and the C_4 pasture by kikuyu (*Pennisetum clandestinum* Hoschst. ex Chiov). Eight automated chambers connected to a gas analyser (G2508, Picarro Inc., USA) were used to measure CO_2, CH_4, and N_2O concentrations.

The fluxes were estimated by fitting linear and non-linear models to gas concentrations recorded over closure time. Each measurement was divided into three closure periods to examine the effect of closure period on flux estimation. To assess the effect of grazing, each trial site was divided into two areas, one exposed to cattle grazing (grazed) and one fenced to exclude cattle grazing (ungrazed), with four chambers located in each area.

Aboveground biomass was sampled at the beginning of the measurement period, following the grazing event, and at the end of the measurement period to quantify change in aboveground biomass.

11.4. Key findings

Chamber effects on measurement accuracy

- Closed chambers can alter the micro-environment by changing temperature, humidity, and light, which can influence gas measurements.
- Shorter closure periods minimise these artefacts and improve data quality.
- At longer closure periods, CO_2 concentrations became increasingly non-linear ($p < 0.001$), meaning non-linear models were more appropriate for estimating CO_2 fluxes.
- This non-linearity occurred in both pasture types but was more pronounced in C4 (kikuyu) pasture.
- For short closure periods, a simple linear model was sufficient and reliable.

Effects on CH_4 and N_2O

- Closure period length did not significantly affect CH_4 or N_2O fluxes in either pasture type.
- Longer closure periods may still be useful because CH_4 and N_2O fluxes are highly variable, and extended measurement time can improve detection.
- Greenhouse gas dynamics in C3 vs C4 pastures
- Kikuyu (C4) pasture produces surplus biomass that often requires slashing, and during this period the pasture acted as a net source of GHG emissions.
- Ryegrass (C3) pasture acted as a net sink of CO_2, capturing carbon during regrowth.

Carbon capture per kg of aboveground biomass change:

- C3 grazed: 0.85 kg CO_2-eq/kg
- C3 ungrazed: 3.25 ± 0.31 kg CO_2-eq/kg
- C4 grazed: 0.41 ± 0.252 kg CO_2-eq/kg
- C4 ungrazed: 0.73 ± 0.218 kg CO_2-eq/kg

11.5. Applications and Impacts

Farm-level:

- Understanding when C3 and C4 pastures act as carbon sinks or sources supports better grazing and slashing decisions.

Industry-level:

- Results help refine GHG measurement protocols can be used in research and extension programs.
- Findings support more accurate carbon-accounting models for pasture-based dairy systems.

Environmental:

- Improved measurement practices strengthen confidence in reported GHG reductions.
- Identifying carbon-sink periods in pastures supports climate-smart pasture management.

11.6. Limitations

- Further research over longer periods of time and multiple pasture species are required to generate the information for estimating the full carbon balance of livestock production.

11.7. Key Outputs

- Dataset of CO_2, CH_4, and N_2O fluxes across C3 and C4 pastures.
- Evidence on optimal chamber closure periods and modelling approaches.
- Peer-reviewed publication in Agriculture, Ecosystems & Environment (Bagheri Shirvan et al., 2025).

11.8. Future Research Opportunities

- Extend flux measurements across seasons and additional pasture types.
- Quantify long-term carbon sequestration potential under different grazing strategies.
- Integrate pasture flux data into whole-farm carbon models.
- Evaluate mitigation options (e.g., biomass removal, grazing timing) to reduce GHG emissions from C4 pastures.

12. Project-wide Dissemination

The project's findings were disseminated through multiple channels, including a PhD thesis, peer-reviewed journal publications, newsletter, and presentations at national and international conferences (ADSA, AARN, NZSSS/SSA, DRF Symposium). Industry engagement occurred through Dairy UP communication channels, workshops, and direct interactions with producers and stakeholders. These activities ensured the research reached researchers, farmers, and policymakers, supporting knowledge exchange on sustainable dairy practices.

Table 1. Peer-reviewed publications.

Author	Title	Journal	Year Published	Citations (to May 13)
Dida et al., 2026	Environmental impacts of dairy farming intensification and land use on soil organic carbon stocks and physicochemical properties	Environmental Impact Assessment Review	2026	
Dida et al., 2025	Potential applications of a low-cost gas sensor to monitor enteric methane emission from ruminant animals	Smart Agricultural Technology	2025	3
Dida et al., 2025	Greenhouse gas emissions of confinement and pasture-based dairy farms: Implications for mitigation	Journal of Dairy Science	2025	5
Bagheri et al., 2025	Short-term effect of grazing on net ecosystem exchange and fluxes of greenhouse gases in C3 and C4 pastures during the growing season	Journal of Agriculture, Ecosystems & Environment	2025	1
Dida et al., 2024	Dietary concentrate supplementation increases milk production and reduces predicted greenhouse gas emission intensity in pasture-based commercial dairy farms	Journal of Dairy Science	2024	14

13. Conclusions and Recommendations

The findings of this Dairy UP project demonstrate that achieving carbon-neutral dairy production in Australia is possible but requires coordinated, system-specific strategies that address the major sources of GHG emissions while supporting productivity and economic resilience. Across confinement and pasture-based systems, emissions intensity was broadly similar, with enteric CH_4 consistently the dominant source.

This confirms that meaningful progress toward carbon neutrality will depend on effective enteric CH_4 mitigation across all production systems. System-level differences highlight the need for tailored approaches. Confinement systems require improved manure management to reduce CH_4 and N_2O emissions from stored effluent, while pasture-based systems benefit most from optimising nitrogen fertiliser use and improving nutrient efficiency. Moderate concentrate supplementation (2–3 t DM/cow/year) emerged as a practical strategy to improve emissions intensity through productivity gains, although over supplementation increases costs and manure-related emissions without proportional benefits.

The project also demonstrated the importance of enabling technologies and natural resource stewardship. Low-cost CH_4 sensors showed promising potential for future on-farm CH_4 monitoring, although further development is required. Soil assessments revealed that pasture-based systems store substantially more C and nitrogen than confinement systems, particularly under permanent pasture and tree cover, while confinement systems accumulate higher phosphorus stocks, underscoring the need for targeted nutrient management. Validation of the Soil and Landscape Grid of Australia (SLGA) showed moderate accuracy for carbon and nitrogen at whole-farm scales but insufficient precision for phosphorus or paddock-level decision-making.

Pasture-level GHG flux measurements further highlighted that species composition and management influence short-term C dynamics, with ryegrass acting as a CO_2 sink during regrowth and kikuyu becoming a source when surplus biomass is removed. Together, these findings show that no single mitigation strategy is sufficient. Instead, progress toward C-neutral dairy systems requires integrated actions across feeding, manure and fertiliser management, soil and vegetation stewardship, and improved emissions measurement.

Recommendations

1. Prioritise enteric CH_4 mitigation across all dairy systems
    - Support adoption of proven CH_4-reducing feed additives, genetics, and diet optimisation.
    - Invest in research and extension to accelerate commercial readiness of emerging technologies.
2. Implement system-specific mitigation strategies
    - Confinement systems: Improve manure storage, handling, and effluent treatment to reduce CH_4 and N_2O.
    - Pasture-based systems: Optimise nitrogen fertiliser use through soil testing, timing, and precision application.
3. Promote productivity-enhancing feeding strategies
    - Encourage 2–3 t DM/cow/year concentrate supplementation where economically viable.
    - Avoid over supplementation that increases emissions and costs without improving milk solids.
4. Strengthen soil and nutrient stewardship
    - Maintain permanent pastures and tree areas to enhance soil carbon and nitrogen stocks.
    - Improve phosphorus management in confinement systems to prevent nutrient accumulation and environmental risk.
5. Improve emissions measurement and monitoring capacity
    - Continue development and validation of low-cost CH_4 sensors for scalable on-farm use.
    - Integrate sensor data into digital decision-support tools for benchmarking and mitigation verification.
    - 6. Use digital soil products cautiously
    - Apply SLGA for broad, whole-farm baseline assessments where direct sampling is not feasible.
7. Expand integrated research and industry adoption pathways
    - Conduct multi-site trials combining feeding, manure, fertiliser, and land-use strategies.
    - Develop training, extension programs, and carbon-accounting tools tailored to diverse production systems.

14. Annexes

Table 2. Conference presentations, abstracts and other meetings

Authors	Title	Presentation Type	Conference/Event	Location	Year	Audience
Mulisa D.	Greenhouse Gas Emissions and Soil Carbon Stocks in Dairy Farming Systems	Final PhD Presentation (oral)	SOLES HDR Showcase (USYD)	Sydney	2025	40
Luciano G.	Are we understanding the "hoofprint" of pastures?	Oral presentation	DRF Symposium	Wollongong	2025	150
Luciano G.	Towards net zero dairy production in NSW: a case study approach for a deeper understanding of carbon emissions in NSW dairy farms	Oral presentation	Carbon workshop	Online (Zoom)	2025	25
Mulisa D.	Greenhouse Gas Emissions of Confinement and Pasture-Based Dairy Farms: Implications for Mitigation	Refereed abstract/poster presentation	ADSA Annual Meeting	Louisville, USA	2025	1000
Mulisa D.	Dairy Farming System and Land Use Influence Soil Organic Carbon Stocks	Refereed abstract/poster presentation	ADSA Annual Meeting	Louisville, USA	2025	1000
Mulisa D.	Dietary Concentrate Supplementation Increases Milk Production and Reduces Predicted Greenhouse Gas Emission Intensity in Pasture-Based Commercial Dairy Farms	Oral presentation	Australian Association of Ruminant Nutrition (AARN)	Melbourne	2024	100
Milad B.	Fluxes of greenhouse gases from pastures and methane emissions from beef cattle in Australia.	Oral presentation	ASAS Annual Meeting	Calgary, Canada.	2024	500
Mulisa D.	Farming System and Land Use Type Influence Soil Organic Carbon Stocks of Dairy Farms	Refereed abstract/poster presentation	Joint Conference of the NZSSS and SSA	Rotorua, New Zealand	2024	500
Luciano G.	Pasture alive! Towards net-zero in Kikuyu-based pasture systems – is it possible?	Oral presentation	DRF Symposium	Camden	2023	100
Mulisa D.	Dietary Concentrate Supplementation Benefits Both Dairy Farmers and the Climate: Evidence from the Australian Dairy Farms.	Oral presentation	DRF Symposium	Camden	2023	100

Milad B.	Effect of grazing on net ecosystem exchange and carbon balance in ryegrass pasture	Oral presentation	Soil Science Australia Conference	Darwin, Australia	2023	200
Mulisa D.	Does Dietary Concentrate Supplementation Benefit Both Dairy Farmers and Climate: Evidence from the Australian Dairy Farms	Poster presentation (Intermediate PhD)	SOLES HDR Showcase (USYD)	Sydney	2023	100
Mulisa D.	Greenhouse Gas Emissions and Carbon Soil Stocks in Dairy Farming Systems	Oral presentation (Introductory PhD)	SOLES HDR Showcase (USYD)	Sydney	2022	50
Milad B.	How are carbon dioxide and methane fluxes mediated by grazing in Kikuyu pastures?	Poster presentation	DRF Symposium	Forster, Australia	2022	100

Table 3. Technical reports, pamphlets, published material and other media engagements

Authors	Title	Place Published	Year
Luciano G.	A path to carbon-neutral dairy	Dairy UP Website	2025
Mulisa Dida	Sustainable Dairy Farming: Insights from My PhD Research	Sydney Institute of Agriculture, AgriCola	2025
Luciano G.	Carbon emissions and dairy systems	Dairy UP Website	2025
Luciano G.	Carbon on NSW dairy farms	Dairy UP Website	2023

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Final Project Summary

P1d – Kikuyu: Carbon on NSW dairy farms

May 2026

A path to carbon-neutral dairy

Reducing greenhouse gas emissions from dairy farming requires a better understanding of where emissions come from, how they vary between farming systems, and which management practices can reduce emissions while maintaining productive and profitable farms.

The purpose of this project was to provide practical, region-specific evidence to support lower-emissions dairy farming under Australian conditions. It was designed to inform practical, whole-farm approaches to reducing greenhouse gas emissions. It also aimed to support evidence-based decision-making by farmers, industry and policymakers working toward carbon-neutral dairy systems.

Why carbon research?

The Australian dairy industry is working to reduce greenhouse gas emissions while maintaining productive and profitable farming systems. Dairy Australia has pledged to reduce the emissions intensity of Australian dairy production to 30 per cent below 2015 levels by 2030, contributing to Australia's broader net zero commitments under the Paris Agreement.

Multipronged approach

This Dairy UP project investigated multiple aspects of carbon and greenhouse gas emissions on NSW dairy farms, including:

- Whole-farm greenhouse gas emissions
- Feeding strategies and methane monitoring
- Soil carbon and nutrient management
- Digital soil mapping
- Pasture carbon dynamics.

Carbon on NSW Dairy Farms

Unlocking the potential of Kikuyu

Dairy UP's P1 project aimed to unlock the potential of Kikuyu pastures used by NSW dairy farmers. P1 was a suite of five projects that collectively explored new management options to grow and utilise more Kikuyu over summer and increase the productivity of Kikuyu-based pastures.

P1a – Kikuyu: Remote monitoring

P1b – Kikuyu Investigating toxicity

P1c – Kikuyu: Developing new varieties

P1d – Kikuyu: Carbon on NSW dairy farms

P1e – Kikuyu: Nutritional value

This document is the final update on P1d – Kikuyu: Carbon on dairy farms.

Key findings

Our findings show there is no single pathway to carbon-neutral dairy production. Effective emissions reduction requires integrated, system-specific strategies combining methane reduction, feeding, fertiliser and manure management, and soil stewardship. One-size-fits-all approaches are unlikely to deliver optimal outcomes.

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DairyUP
Unlocking potential

Whole-farm emissions

- Emissions intensity was similar between pasture-based and confinement (housed) systems, but the main emission sources differed between systems.
- Methane produced by cows during digestion (enteric methane) was the largest source of emissions in both systems, accounting for more than half of total farm emissions.
- Confinement systems produced higher manure-related emissions, while pasture-based systems produced higher fertiliser-related and off-farm input emissions.

Feeding and methane monitoring

- Feeding 2–3 t DM/cow/year concentrate increased milk production and reduced emissions intensity by about 12%. However, the impact on profitability depends on each farm's management and market conditions.
- Low-cost methane sensors showed promise for future on-farm methane monitoring but require further development before commercial adoption.

Soil carbon and nutrient management

- Pasture-based systems stored substantially more soil organic carbon and nitrogen than confinement systems, especially under permanent pasture and tree cover. However, confinement farms were in lower rainfall zones than pasture-based farms.
- Confinement systems accumulated much higher soil phosphorus levels, reinforcing the need for targeted nutrient management.
- The Soil and Landscape Grid of Australia (SLGA) was useful for broad whole-farm assessments but not accurate enough for paddock-scale management or carbon crediting.

Pasture carbon dynamics

- At the study site, ryegrass pastures acted as a carbon sink during regrowth. In contrast, kikuyu pastures became a net greenhouse gas source when surplus pasture required slashing and a net sink after the litter decomposed.

More info

P1d Final report

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Research and results

Research aims

1. Quantify greenhouse gas emissions from pasture-based and confinement (housed) dairy systems and identify the main emission sources.
2. Assess the effect of concentrate feeding on milk production, emissions intensity and farm profitability.
3. Evaluate low-cost methane sensors for practical, on-farm methane monitoring.
4. Measure soil carbon and nutrient status across different dairy land-uses and farming systems.
5. Assess the accuracy of the Soil and Landscape Grid of Australia (SLGA) for dairy farm soils.
6. Measure greenhouse gas fluxes from ryegrass and kikuyu pastures and assess how management influences carbon dynamics.

1. Whole-farm GHG emissions

Dairy Australia's Australian Dairy Carbon Calculator was used to analyse greenhouse gas emissions from 10 commercial NSW dairy farms: five pasture-based and five confinement (housed) systems.

Key findings

Pasture-based and confinement (housed) dairy systems in NSW produced similar greenhouse gas emissions intensity — meaning a similar amount of emissions per kilogram of milk produced:

- pasture-based: 1.07 kg CO_2-eq/kg fat- and protein-corrected milk.
- confinement: 1.02 kg CO_2-eq/kg fat- and protein-corrected milk.

However, the main sources of emissions differed

between systems, highlighting different opportunities to reduce emissions (see graph over page).

In both systems, enteric methane (produced by cows during digestion) was the largest source of emissions accounting for more than half of total farm emissions.

Manure emissions were substantially higher in confinement systems (31% of total emissions compared with 13% in pasture-based systems), largely due to stored manure under low-oxygen conditions.

Fertiliser and pre-farm emissions were higher in pasture-based systems, reflecting greater reliance on nitrogen fertiliser.

Tree cover provided modest carbon offsets:

- Pasture-based systems: about 6% of total emissions.
- Confinement systems: about 1%

Implications for farmers

While both systems require methane reduction strategies, the priority for emissions reduction differs between systems:

- Pasture-based: improve fertiliser efficiency and reduce reliance on external inputs.
- Confinement: improve manure management.

2. Feeding strategies

This study examined how concentrate feeding affects milk production, greenhouse gas emissions intensity and farm profitability in pasture-based dairy systems.

The analysis used Dairy Australia's Dairy Farm Monitor Project data from 120 commercial pasture-based dairy farms across New South Wales, Victoria, Tasmania and South Australia. Farms were grouped according to annual concentrate feeding rate:

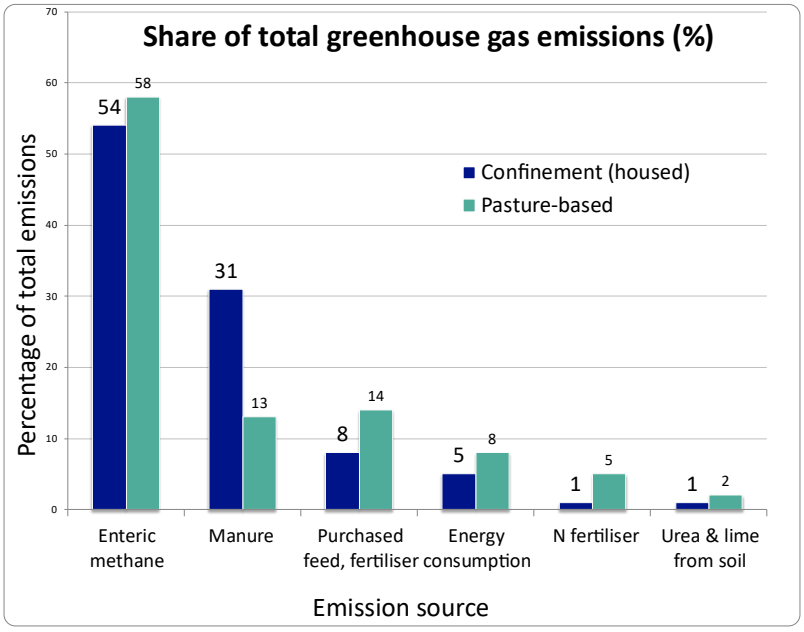
- low: less than 1 t DM/cow/year.
- moderate: 1–2 t DM/cow/year.
- high: 2–3 t DM/cow/year.
- very high: more than 3 t DM/cow/year.

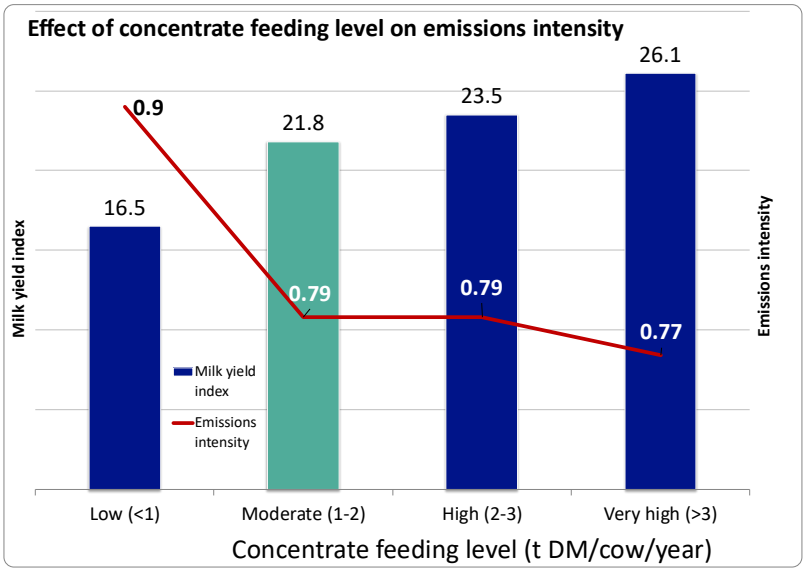
Greenhouse gas emissions were estimated using Dairy Australia's Australian Dairy Carbon Calculator. Economic performance was assessed using gross margin and Earnings Before Interest and Tax (EBIT).

Key findings

Farms feeding concentrates at 2–3 t DM/cow/year generally achieved the strongest balance between productivity, emissions efficiency and profitability. Productivity gains were the main driver of lower emissions intensity.

Feeding 2–3 t DM/cow/year of concentrate increased milk production and reduced greenhouse gas emissions intensity by about 12% compared with farms feeding less than 1 t DM/cow/year (see graph above).





Feeding more than 3 t DM/cow/year delivered diminishing returns, with higher feed costs and increased manure-related emissions reducing the overall benefit.

Table: Profitability of concentrate feeding levels

	Low	Moderate	High	Very high
EBIT $/cow/year	347	371	532	282
Net income $/cow/yr	58.5	135	222	-44.5
EBIT $/kg milk solids	1.07	0.77	1.05	0.54
Net income $/kg ms	0.03	0.23	0.43	-0.12

Implications for farmers

The findings suggest feeding 2–3 t DM concentrates/cow/year can be a practical strategy to improve milk production while reducing emissions intensity in pasture-based dairy systems. However, higher feeding rates do not necessarily improve profitability or emissions outcomes because of the trade-off associated with manure emissions. Feeding strategies should be balanced with feed costs, manure management and overall farm system efficiency.

3. Methane monitoring

This study evaluated whether low-cost methane sensors could provide a practical alternative to expensive methane monitoring systems currently used in research.

A 45-day trial involving 28 heifers compared a low-cost methane sensor with the established GreenFeed methane monitoring system. Researchers assessed how accurately the low-cost sensor measured methane patterns and how consistently it performed under commercial field conditions.

Key findings

While the sensor is not yet suitable as a replacement for established methane monitoring systems, the results demonstrated potential for future development of low-cost methane monitoring technologies for dairy farms.

The low-cost sensor showed moderate agreement with the GreenFeed system, meaning it was able to detect general methane emission patterns but was less accurate for precise measurement.

The low-cost sensor was also less consistent than the GreenFeed system, showing greater variability in measurements.

Implications for farmers

Affordable methane monitoring technologies could eventually help farmers:

- track methane trends within herds for applications such as animal breeding for low emissions
- assess the impact of feeding or management changes
- support emissions benchmarking and reporting.

Further development and validation are still required before low-cost methane sensors are ready for widespread commercial use.

4. Soil carbon and nutrient status

This study measured soil carbon and nutrient status across different dairy farming systems and land uses in NSW.

More than 800 soil samples were collected from nine dairy farms representing pasture-based and confinement (housed) systems. Soil samples were taken from a range of land uses including natural pasture, improved pasture, mixed pasture/forage cropping paddocks and tree-covered areas.

Soil organic carbon, nitrogen, phosphorus and other soil physicochemical properties were analysed to better understand how dairy farming systems and land management practices influence soil nutrient reserves and carbon storage.

Key findings

Pasture-based dairy systems stored substantially more soil carbon and nitrogen than confinement systems, particularly under permanent pasture and tree-covered areas (see table). However, pasture-based farms in this study were located in drier regions.

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Table: Soil organic carbon, nitrogen and phosphorus status		
	Pasture-based systems	**Confinement systems**
	(Mg ha^{-1})	
Soil organic carbon	105	60
Total nitrogen	8.73	5.3
Total phosphorus	0.15	0.56

In the pasture-based systems, cropping paddocks generally had lower soil carbon levels than permanent pastures. This highlights the importance of long-term ground cover and pasture systems for maintaining soil carbon reserves in high rainfall zones.

Confinement systems had much higher phosphorus levels, reflecting gcumulative (legacy) phosphorus from historical inputs and/or inherent differences in parent material and mineralogy, rather than contemporary annual applications alone.

The study also showed substantial variation in soil carbon and nutrient status within farms, reinforcing the importance of direct soil measurement rather than relying solely on regional averages or maps.

Implications for farmers
The findings suggest pasture management, land use and nutrient management practices can strongly influence soil carbon storage and nutrient accumulation on dairy farms.

Maintaining permanent pasture and tree cover may help support soil carbon reserves, while regular soil testing can help identify nutrient imbalances and guide fertiliser management.

5. Digital soil mapping

This study evaluated how accurately the Soil and Landscape Grid of Australia (SLGA) predicts soil carbon and nutrient levels on dairy farms.

The SLGA predictions were compared with laboratory analysis of 810 soil samples collected across nine NSW dairy farms. The study assessed how well the SLGA estimated soil carbon, nitrogen and phosphorus at both whole-farm and paddock scale.

Key findings
The findings suggest digital soil mapping tools can provide useful broad-scale insights for dairy farms where direct sampling is limited. However, the SLGA was not accurate enough for paddock-scale management or carbon crediting, despite being useful for broad whole-farm assessments of soil carbon and nitrogen

The digital mapping system showed moderate accuracy for soil carbon and nitrogen, capturing broad differences between farming systems. However, it was not accurate for phosphorus and was unable to reliably detect fine-scale variation between paddocks.

The findings highlight the importance of validating digital soil maps with direct soil sampling, particularly where farmers are making nutrient management or carbon accounting decisions.

Implications for farmers
Digital soil mapping tools such as the SLGA can be a useful starting point for understanding soil carbon and nutrient status. However, it is not accurate for paddock-level management decisions and accurate carbon accounting.

6. Pasture and carbon dynamics

This study investigated how pasture species and management practices influence greenhouse gas emissions and carbon cycling in dairy systems.

It was conducted at the University of Sydney's Corstorphine research farm at Camden.

Greenhouse gas chambers and gas analysers measured daily greenhouse gas fluxes from ryegrass and kikuyu pastures before, during and after grazing.

The study assessed how grazing, pasture regrowth, slashing and nitrogen fertiliser application influenced short-term carbon exchange between the pasture and atmosphere.

Key findings
The study showed well-utilised pastures can potentially act as a net carbon sink by capturing more carbon dioxide through photosynthesis than they release through greenhouse gas emissions under the conditions measured.

At the study site, ryegrass pastures acted as a carbon sink during active regrowth. Kikuyu pastures also captured carbon during active growth. However, when surplus pasture required slashing, emissions from decomposing plant material reduced the overall carbon benefit. Under the conditions measured, this sometimes shifted the pasture system from a net carbon sink to a net greenhouse gas emitter.

Pasture growth stage, grazing management, and seasonal conditions all influenced greenhouse gas fluxes. Dry conditions reduced photosynthesis and limited the ability of pastures to capture carbon.

The study also highlighted the importance of measurement timing and methodology when assessing pasture greenhouse gas emissions and carbon sequestration.

Implications for farmers
The findings suggest pasture management practices can influence the greenhouse gas balance of dairy systems over short timeframes. Maintaining actively growing pastures and grazing management to avoid the need to slash excessive surplus pasture may help improve carbon capture and reduce emissions from pasture systems.

Limitations
The findings were based on measurements from a single research site and specific seasonal conditions, highlighting the need for further research across multiple environments and years.

PhD student

Mulisa Dida, University of Sydney

Collaborators

The scope of this Dairy UP project was expanded thanks to additional funding from the NSW Environmental Protection Authority (EPA).

Journal articles

Dida et al., 2026 Environmental impacts of dairy farming intensification and land use on soil organic carbon stocks and physicochemical properties. Environmental Impact Assessment Review, Vol 120.

Carbon terminology

Carbon calculator: a tool for farmers to measure their farm's carbon footprint and understand the sources of emissions and explore options for reducing emissions.

Carbon credit (carbon offset): permits the owner to emit a certain amount of carbon dioxide or other greenhouse gases.

Carbon dioxide equivalents: a measure used to compare emissions from various greenhouse gasses on the basis of their global warming potential by converting amounts of other gasses to the equivalent amount of carbon dioxide with the same global warming potential.

Carbon sequestration: the process of capturing and storing carbon dioxide from the atmosphere.

Fat and protein corrected milk **emissions intensity**: kilograms of carbon dioxide equivalents per kilogram of fat and protein corrected milk.

Enteric methane: produced by cows during digestion.

Fluxes: the kilograms of greenhouse gas emissions or uptake per hectare per day.

Dida et al., 2025 Potential applications of a low cost gas sensor to monitor enteric methane emission from ruminant animals. Smart Agricultural Technology, Volume 10.

Dida et al., 2025 Greenhouse gas emissions of confinement and pasture-based dairy farms: Implications for mitigation. Journal of Dairy Science, 2025; 108, 11026-11040

Dida M. F., et al (2024), Dietary Concentrate Supplementation Increases Milk Production and Reduces Predicted Greenhouse Gas Emissions Intensity in Pasture-based Commercial Dairy Farms. Journal of Dairy Science March 22, 2024

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Delivery organisations

The University of Sydney Dairy Research Foundation

NSW Government Department of Primary Industries and Regional Development

Scibus

Dairy Australia

Partner organisations

NSW Farmers Growing the best

eastAUSmilk Advocacy · Unity · Service

Dairy NSW | Dairy Australia

AFMH

Leppington Pastoral Company Pty Ltd

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How reliable are national digital soil maps for farm-scale baseline assessment? A multi-scale assessment using dairy farm data

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ARTICLE INFO

Keywords:
Carbon
Dairy farm
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Nitrogen
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ABSTRACT

Digital soil mapping (DSM) provides a scalable approach for monitoring soil health indicators to support sustainable land management and precision agriculture. However, uncertainties associated with input data and fine-scale management factors including land-use type, fertiliser history, and grazing intensity can limit predictive reliability at operational scales. Evaluating the performance of DSM products under real farm conditions is therefore critical to determine their suitability for baseline assessment. This study assessed the accuracy of the Soil and Landscape Grid of Australia (SLGA), a national-scale DSM product, in predicting topsoil (0–30 cm) soil organic carbon (SOC), total nitrogen (TN), and total phosphorus (TP) using more than 800 independently collected topsoil samples from nine Australian dairy farms. The SLGA prediction performance was evaluated at sub-farm (individual Carbon Estimation Areas, CEAs, i.e., paddocks or groups of paddocks with similar land use and management within a farm) and farm scales (aggregated across all CEAs within a farm) using R^2, Lin's concordance correlation coefficient (CCC), mean absolute error (MAE), and mean bias error (MBE). In addition, linear mixed-effects models tested SLGA's ability to detect differences due to land use and farming systems (pasture-based vs. confinement). The SLGA showed moderate accuracy for SOC and TN at the farm scale (SOC: $R^2 = 0.79$, CCC = 0.88; TN: $R^2 = 0.87$, CCC = 0.79), but substantially weaker agreement at the sub-farm scale, and very poor accuracy for TP across both scales ($R^2 \leq 0.01$; CCC $\leq$ 0.02). Standardized errors were moderate for SOC and TN, and highest for TP. The SLGA successfully detected differences in SOC and TN between pasture-based and confinement systems, demonstrating value for broad farm-scale baseline assessments, but showed limited sensitivity to within-farm land-use variation. Overall, SLGA provides useful farm-scale baseline assessments of SOC and TN in dairy systems; however, improvements are required to enhance sub-farm precision, TP prediction, and the representation of local management effects.

1. Introduction

Climate change is intensifying pressures on global food systems, water resources, and biodiversity, highlighting the urgent need for land-management practices that enhance soil resilience and reduce greenhouse gas (GHG) emissions (IPCC, 2023). Soils play a key role in climate mitigation through carbon (C) storage, nutrient cycling, water regulation, and biodiversity preservation; however, soils can also contribute substantially to GHG emissions when poorly managed (Evangelista et al., 2024; Keesstra et al., 2016; Kopittke et al.,

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Environmental impacts of dairy farming intensification and land use on soil organic carbon stocks and physicochemical properties

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ARTICLE INFO

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Confinement
Cropping
Pasture
Pasture-based
Tree

ABSTRACT

The intensification of livestock farming systems can pose significant environmental challenges, including impacts on soil physicochemical properties, carbon sequestration, and biodiversity. This study is the first to quantify the combined effects of dairy system intensification and land-use allocation on soil organic carbon (SOC) stocks and physicochemical properties at the commercial farm scale. We compared confinement and pasture-based systems across five land-use types: improved pasture (IP), mixed pasture-cropping (MPC), cropping, tree areas (TA), and natural pasture (NP, exclusive to pasture-based systems). A total of 810 soil samples were collected from the top 30 cm across nine farms in New South Wales, Australia. Pasture-based farms stored 75% more SOC and 65% more total nitrogen (TN) per hectare than confinement farms, although differences were partly influenced by regional climate and parent material. Interactions between farming system and land use showed that SOC and TN stocks were highest in IP and TA within pasture-based systems. Conversely, total phosphorus (TP) stocks were 3.7 times higher in confinement systems, independent of SOC patterns. Within pasture-based farms, NP consistently had the highest SOC and TN stocks. These results highlight that dairy system intensification interacts with land-use and environmental conditions to shape soil organic carbon and nutrient stocks. The findings provide policy-relevant benchmarks for soil-carbon accounting and emphasize the need for regionally adapted land-management strategies and future work quantifying annual SOC sequestration rates under commercial dairy conditions.

1. Introduction

Grasslands occupy 67% of global agricultural land and 24% of the total land surface, storing about 20% of the world's soil organic carbon (SOC) stocks to 1 m depth (FAO, 2021; Puche et al., 2019). These systems are vital for regulating the global carbon (C) and water cycles, as soils hold far more C (4000 Pg) than the atmosphere (740 Pg CO_2) (Camacho et al., 2023). Agricultural practices shape SOC dynamics through shifts in management, nutrient cycling, and land use intensity (Camacho et al., 2023; Dondini et al., 2023; FAO, I, 2015). Enhancing SOC in grasslands via targeted grazing and legume integration could offset 2.3–7.3 Pg CO_2eq annually (Bai and Cotrufo, 2022). However, land conversion and intensive farming can significantly affect SOC stocks. Strategic grazing in pastures systems can builds SOC by favouring deep-rooted perennials, root exudates, legume N inputs, and surface litter (Chen et al., 2015).

Enhancing agricultural land intensification for animal products can disrupt ecosystems, altering C stocks, biodiversity, and water/air quality based on practices (Erisman et al., 2013; Hertel, 2015; Silveira et al., 2026). Confinement dairy systems where cows are housed year-round on total mixed rations rely on cultivated forages, leading to SOC declines from tillage, mineralization, and erosion (Lal, 1997; Ritchie, 2020). Additionally, such systems amplify N fertilizer use, increasing reactive N losses and environmental risks (de Klein and Monaghan, 2011; Galloway et al., 2008). Confinement systems maintain livestock at high density in small areas with required manure management practices and use high inputs to produce and harvest feed, which can significantly affect soil organic matter and nutrient cycling (Dondini et al., 2023). On the other hand, pasture-based systems maintain livestock grazing in larger areas with animals depositing manure on pastures, requiring lesser manure management and lower soil disturbance, which may provide more opportunities for SOC sequestration (Adewale et al., 2018;

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Greenhouse gas emissions of confinement and pasture-based dairy farms: Implications for mitigation

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ABSTRACT

Despite extensive research on the environmental effects of dairy farming, comparative GHG emissions from confinement and pasture-based systems remain unclear due to inconsistent findings from prior studies, which were often specific to the local conditions of each system and overlooked carbon sequestration by trees. The present study aimed to compare the GHG emissions of 2 Australian milk production systems (confinement and pasture-based) using a life cycle assessment approach that incorporates C sequestration by trees. The confinement system used a TMR, whereas the grass-based system primarily relied on grazed forage with concentrate supplementation. The Australian Dairy Carbon Calculator, a Tier 3 tool, predicted emission intensity using the National Greenhouse Gas Inventory and Intergovernmental Panel on Climate Change methods, as reported to the United Nations Framework Convention on Climate Change. Emission intensity was calculated as net GHG exchange in CO_2 equivalents (CO_{2eq}), allocated to milk and meat. Animal emissions dominated: 85% of total emissions in confinement systems (54% enteric CH_4, 31% manure) and 71% in pasture-based systems (58% enteric CH_4, 13% manure). The confinement system showed 13% lower enteric CH_4 intensity and 88% lower prefarm embedded intensity (kg CO_{2eq}/kg fat- and protein-corrected milk [FPCM]) but 129% higher manure-related GHG intensity than the pasture-based system. Emission intensities for milk (1.02 ± 0.038 vs. 1.07 ± 0.069 kg CO_{2eq}/kg FPCM) and meat (5.51 ± 0.779 vs. 6.76 ± 0.868 kg CO_{2eq}/kg liveweight) were similar between systems. The emission offset by tree C sequestration (kg CO_{2eq}/kg FPCM) was relatively low in both systems, contributing about 1% of total CO_{2eq} emissions in confinement systems and up to 6% in pasture-based systems. Targeted mitigation should address manure emissions in confinement systems, prefarm embedded, and fertilizer emissions in pasture-based systems, and enteric CH_4 in both.
Key words: dairy, manure, methane, milk yield

INTRODUCTION

The livestock sector faces the dual challenge of increasing productivity while reducing GHG emissions and adapting to a changing climate (Gerber et al., 2013). Livestock currently contributes 34% of global protein intake and 17% of calorie consumption, with demand continuing to rise due to population growth, urbanization, and increasing incomes (FAO, 2022). Dairy products alone contribute 11% of global protein and 5% of calorie intake (FAO, 2022). However, livestock is responsible for 14.5% of global anthropogenic GHG emissions, with ruminants accounting for 75% of these (IPCC, 2019). Dairy farming plays a substantial role in these emissions, contributing 4.0% of global GHG emissions (2.7% from milk production and 1.3% from meat from dairy cattle), accounting for 20% of total global livestock emissions (Gerber et al., 2013). In Australia, the dairy sector contributes 2% of total national and 14% of agricultural emissions (Australian Government, 2023). Addressing the effect of livestock production on climate change is urgent because rising temperatures, variable precipitation, and increased CO_2 concentration negatively affect livestock performance and feed supply (FAO, 2019). In addition, the global demand for both animal- and plant-based foods is projected to double by 2050 (FAO and GDP, 2018; Enahoro et al., 2021; van Dijk et al., 2021) and sustainable intensification of food production may be required (Muscat et al., 2021).

In dairy farming, confinement is one form of intensification, but one key debate concerns its environmental effect compared with pasture-based systems. Confinement systems, which include any type of contained housing such as freestall, loose housing, compost barns, and dairy dry lots (Dairy Australia, 2024), are often considered more efficient in terms of emissions per unit of output due to improved feed conversion and animal productivity

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The list of standard abbreviations for JDS is available at adsa.org/jds-abbreviations-25. Nonstandard abbreviations are available in the Notes.

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Potential applications of a low-cost gas sensor to monitor enteric methane emission from ruminant animals

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ARTICLE INFO

Keywords:
Greenfeed
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ABSTRACT

Enteric methane (CH_4) emissions are a significant concern in ruminant production due to its high global warming potential of 28 times that of CO_2. Research is focused on mitigating these emissions through animal and feed management. However, accurately and economically measuring CH_4 emissions from individual animals remains challenging due to current methods' high cost, slow speed, and labor intensity. Therefore, the primary objective of the present study was to evaluate low-cost gas sensors, named MQ-4, MQ-8, MQ-7, by comparing their data with a well-established GreenFeed system (GF). The MQ-4, MQ-8, and MQ-7are sensitive to CH_4, H_2, and CO, respectively. Sensors were assembled on a single board and placed at various points on the GF system (airflow outlet, pump outlet, and feed tray) to determine optimal placement. The pump outlet showed the highest correlation ($r = 0.51$ for CH_4 and $r = 0.65$ for H_2) with GF data. Following this, the gas sensors were placed in a pump outlet for 45 days, and enteric CH_4 and H_2 were measured simultaneously using both devices on 28 heifers, resulting in a total of 3.88×10^6 paired measurements. Correlation between CH_4 and H_2 data from the GF and gas sensors was assessed using Spearman correlation coefficients (SCC), repeatability, and peak measurement algorithms. The SCC values for the CH_4 concentration from the MQ-4 gas sensor and GF averaged weekly, daily, hourly, and minute were 0.62, 0.56, 0.49, and 0.47, respectively. The repeated measures correlation between (from the per second record) the devices was relatively low (0.30). The repeatability of CH_4 concentration was greater for the GF (0.31) than for the MQ-4 gas sensor (0.13). Additionally, there were significant differences in the detected number of peaks, peak duration, and width between the two devices. The peak detection algorithm indicated that the GF detected more peaks and processed data four times faster than the MQ-4 gas sensor. Although results indicate moderate and significant correlations between H_2 and CH_4 voltages measured by the GF and gas sensors, lower repeatability and discrepancies in peak characteristics observed for the gas sensor suggest caution in interchangeable use without further methodological refinement.

1. Introduction

Enteric methane (CH_4) production by ruminants poses a threat to climate change because it is a powerful greenhouse gas (GHG) with 28-fold greater global warming potential than carbon dioxide (CO_2) [1]. Enteric CH_4 accounts for 39 % of livestock sector emissions, making it a major concern worldwide [2]. Reducing such emissions from livestock can be addressed by combining feeding, genetic improvement, vaccines against methanogens, and management strategies [3–5]. However, accurate and large-scale measurements of GHG emissions from individual animals in their production environment are vital to develop and evaluate mitigation strategies and provide realistic GHG emission figures.

Numerous methods for quantifying enteric CH_4 emissions have been developed, each with its advantages and disadvantages [6–9]. One of the most accurate methods is the (closed-circuit) respiration chamber (RC), but it comes with limitations such as animal confinement, cost, and labor intensity, raising questions about its applicability to commercial or grazing systems [9]. In contrast, the (open-circuit) Greenfeed system (GF) offers higher throughput and shorter measurement periods [10]. However, GF's limitation lies in measuring gas exchange while animals

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Dietary concentrate supplementation increases milk production and reduces predicted greenhouse gas emission intensity in pasture-based commercial dairy farms

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ABSTRACT

Controlled studies have extensively documented that concentrate supplements typically increase enteric CH_4 emissions and milk yield and reduce emissions per unit of milk produced and dry matter intake. However, no studies have been conducted to determine the effect of concentrate on predicted greenhouse gas emissions from dairy farms representing the Australian pasture-based farming system. Thus, this study sought to determine how dietary concentrate supplementation affects enteric and manure CH_4, and N_2O of Australian pasture-based dairy farms. The Australian Dairy Carbon Calculator was used, which incorporates emission factors and methodologies used in the National Greenhouse Gas Inventory as reported to the Intergovernmental Panel on Climate Change. Primary data were collected and analyzed from 120 commercial farms in Australia's major dairy regions. Then the farms were divided into 4 groups based on their dietary concentrate supplementation: ≤1 (low; 15 farms), 1 to 2 (moderate; 35 farms), 2 to 3 (high; 35 farms), and ≥3 (very high; 35 farms) t of concentrate dry matter (tDM) per cow per year. Sources of greenhouse gas emissions were CO_2 from concentrate production, enteric CH_4, and manure CH_4 and N_2O. Total dry matter intake, milk yield, and daily enteric CH_4 production (g/d) quadratically increased with concentrate level, whereas greenhouse gas emission intensity of milk production (kg of CO_2 equivalent per kg of fat- and protein-corrected milk) decreased by 14% for farms supplementing with ≥3 tDM/cow per year compared with those supplementing with ≤1 tDM/cow per year of dietary concentrate. The N_2O and CH_4 emissions from manure increased quadratically and linearly, respectively, with the increasing supplementation of concentrate. Farms supplementing 2 to 3 tDM/cow per year showed substantial increases in gross income, gross margin, earnings before interest and tax, and net income ($/cow per year) compared with those supplementing of ≤1, 1 to 2, and ≥3 tDM/cow per year. Overall, increasing dietary concentrate supplementation for dairy cows resulted in increased milk production per cow, reduced greenhouse gas emissions per unit of milk produced, and increased income and profit. However, a comprehensive life cycle assessment study is needed to account for carbon sequestration by other farm components, such as pastures and trees, which were not considered in the present study. In addition, the present study was based on modeling and did not gather ground truth information for DMI, digestibility, crude protein, and urinary and fecal N excretion. Therefore, data should be interpreted with caution, and studies gathering such information are encouraged.

Key words: milk yield, methane, nitrous oxide

INTRODUCTION

Methane and N_2O are the most important greenhouse gases (**GHG**) from dairy farms, with 28 and 265 times the global warming potential of CO_2, respectively, (IPCC, 2018). Methane is a short-lived climate pollutant, with an atmospheric perturbation lifetime of around 12.5 years (IPCC, 2014). In 2021, CH_4 concentrations were at 1,908 ppb, and N_2O concentrations were at 334.5 ppb in the atmosphere, representing 262% and 124% of pre-industrial levels, respectively (Citaristi, 2022). Methane and N_2O account for 16% and 6% of total global GHG emissions, respectively (Hockstad and Hanel, 2018). Methane from livestock contributes 5% of global GHG and 30% of anthropogenic CH_4 emissions (Jackson et al., 2020; Crippa et al., 2021). Enteric fermentation accounts for 88% of global livestock CH_4 emissions (FAO, 2019). The most significant source of GHG emissions from dairy production systems is enteric CH_4 (>70%), followed by N_2O (13%) and CH_4 (12%) from the animal waste of total on-farm non-CO_2 emissions (Eckard and Clark, 2018). The Australian dairy industry emits 10.0 Mt of CO_2 equivalent ($\mathbf{CO_{2eq}}$), which accounts for 2% of total national emissions and 14% of agricultural emis-

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The list of standard abbreviations for JDS is available at adsa.org/jds-abbreviations-24. Nonstandard abbreviations are available in the Notes.

DairyUP
Unlocking potential

Final Report

P1e – Kikuyu: Nutritional Value

Universidad Nacional de Mar del Plata

This project was led by Prof. Juan Insua,
The University of Mar del Plata

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

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1. Executive Summary

The P1e subproject addressed the rapid decline in Kikuyu pasture nutritive value during regrowth. The project aimed to a) predict, in real-time, rapid, short-term (daily) changes in the nutritive value of kikuyu pastures through modelling of morphophysiological aspects of plant tissue; b) quantify the rate and main drivers of nutritive value decline in Kikuyu pastures during regrowth; and c) generate quantitative benchmarks (e.g. leaf lifespan, rate of fibre digestibility decline) to support improved grazing management decisions. To achieve these goals, the project involved a literature review, a field experiment in summer-autumn repeated across two consecutive years (2023-2024) that included non-destructive and destructive sampling, detailed nutrient analysis at leaf-level and the testing of the Morphogenetic and Digestibility of Pasture (MDP) model to predict quality changes.

The project quantified the main drivers of digestibility decline and calibrated a mechanistic modelling framework to predict short-term changes in pasture nutritive value. Results demonstrated that leaf ageing was the primary determinant of nutritive value decline, with neutral detergent fibre digestibility (NDFD) and dry matter digestibility (DMD) decreasing consistently with thermal time (growing degree days, GDD) of regrowth. Leaf lifespan represents the thermal time of regrowth corresponding to the onset of senescence of the first leaf that appeared after defoliation, and the ageing process explained approximately 70% of the variation in NDFD and more than 60% of the variation in DMD. In this context, changes in neutral detergent fibre (NDF) content were relatively small, indicating that fibre digestibility (NDFD) rather than fibre concentration (NDF) was the main driver of quality decline. leaf lifespan was quantified at approximately 400 GDD, providing a new quantitative reference for this species. The MDP model accurately predicted digestibility dynamics ($R^2 \approx 0.94$), demonstrating strong potential for real-time prediction of Kikuyu pasture nutritive value. The project also identified practical grazing thresholds (~300–350 GDD or 3–4 leaves per tiller) to support improved grazing management and milk production efficiency. In addition, the work establishes a strong foundation for future integration of nutritive value predictions with remote sensing technologies, pasture monitoring systems, and digital agriculture platforms.

2. Project Overview

Item	Description
Project Title	Kikuyu: Nutritional Value
Funding Body	Dairy UP
Dairy UP Project	P1e
Project Duration	2021-2026
Lead Organisation	The University of Sydney's Dairy Research Foundation
Project Lead	Prof. Juan Insua, The University of Mar del Plata
Key Collaborators	Unidad Integrada Balcarce, The University of Mar del Plata NSW DPIRD Local Land Services (Hunter)

3. Abbreviations

ADSA — American Dairy Science Association
ADSA — American Dairy Science Association
ANOVA — Analysis of Variance
DMD — Dry Matter Digestibility
DM — Dry Matter
GDD — Growing Degree Days
JDS — Journal of Dairy Science
MDP — Morphogenetic and Digestibility of Pasture
NDF — Neutral Detergent Fibre
NDFD — Neutral Detergent Fibre Digestibility
NDVI — Normalized Difference Vegetation Index
NS — Not Significant
NSW — New South Wales
RMSE — Root Mean Square Error
R^2 — Coefficient of Determination
USYD — The University of Sydney

4. Project Background and Rationale

Kikuyu grass (*Cenchrus clandestinus*; formerly *Pennisetum clandestinum*) is a tropical grass adapted to both tropical and temperate climates. It is a fast-growing species producing dry matter higher than most tropical pastures (Fulkerson et al., 2007), but of moderate nutritive value overall (García et al., 2014; Marais, 2001). Kikuyu's leaves lose digestibility very rapidly if not grazed at the right time (Fulkerson et al., 2010; Reeves et al., 1996), with the window of opportunity being generally smaller than for temperate grasses (Fulkerson et al., 1999), at times making it a somewhat challenging species to manage.

Kikuyu is limited by nutritive value rather than forage yield, with nutritive value being related to leaf:stem ratio (Marais, 2001; Reeves et al., 1996). Previous studies have shown that as regrowth progresses, Kikuyu pastures rapidly accumulate stem and dead material, resulting in declines in crude protein (CP), dry matter digestibility (DMD), and animal intake potential (Fulkerson et al., 1999; Fulkerson et al., 2010). Reeves et al. (1996) reported a significant decline in dry matter digestibility (DMD) after reaching the 4.5 leaf stage during regrowth (i.e. when each tiller had grown 4.5 leaves or more). This was due to a marked change in the proportion of leaf to stem and dead material content when the sward was harvested to ground level.

For temperate species like perennial ryegrass (*Lolium perenne* L.) and tall fescue (*Festuca arundinacea* Schreb.), changes in nutritive value of leaves have been strongly associated with both leaf age and leaf length (Insua et al., 2017; 2020). Leaf ageing progressively reduces fibre digestibility due to cell wall lignification, while longer leaves generally contain a greater proportion of structural tissues with lower digestibility. These relationships allowed changes in nutritive value to be successfully modelled and predicted (Insua et al., 2019c) using the Morphogenetic and Digestibility of Pasture (MDP) model (Insua et al., 2019a). However, for Kikuyu, the effects of leaf age and leaf length on the digestibility of the cell wall (neutral detergent fibre, NDF) component have not been elucidated yet.

5. Project Objectives

- To predict, in real-time, rapid, short-term (daily) changes in the nutritive value of kikuyu pastures through modelling of morphophysiological aspects of plant tissue.
- Quantify the rate and main drivers of nutritive value decline in Kikuyu pastures during regrowth.
- Generate quantitative benchmarks (e.g. leaf lifespan, rate of NDFD decline) to support improved grazing management decisions.

6. Materials and Methods

- **Literature review**: A comprehensive review was conducted to identify and quantify the main drivers of nutritive value decline in Kikuyu, supporting model development.
- **Experimental design:** A field experiment was conducted at the University of Sydney's Corstorphine dairy farm (Camden, NSW, Australia) across three independent summer regrowth periods (P1, P2 and P3) during 2022–2024 on a Kikuyu pasture using small ($\sim$10 m^2), replicated plots ($n = 2$).
- **Pasture management and sampling:** During regrowth, destructive harvests (pasture cuts) were conducted at successive leaf stages (L1–L6), corresponding to the appearance of new leaves per tiller. At each stage, a randomly selected 1 m^2 area per plot was harvested by cutting the pasture at ground level. Prior to cutting, herbage mass on the randomly selected area was estimated using a calibrated rising plate meter.
- **Morphogenetic measurements:** Leaf appearance, elongation, lifespan, final leaf length, and live leaf number were measured every 2–3 days on permanently marked tillers. Thermal time (GDD) was used to describe regrowth dynamics.
- **Nutritive value analysis:** Leaf blade samples were freeze-dried and analysed for NDF, 24-h NDFD, and DMD.
- **Model parameterisation and evaluation:** The MDP model was parameterised using data from P1 and P2 and independently evaluated using P3 data.
- **Statistical analysis:** Data were analysed using ANOVA and regression approaches to assess relationships between regrowth, morphology, and nutritive value variables. Model performance was evaluated using R^2 and RMSE.

7. Key Findings

- Changes in NDF content were relatively small, indicating that digestibility rather than fibre concentration was the main driver of quality decline (Table 1).
- Kikuyu pastures showed a rapid and consistent decline in NDFD during regrowth (Figure 1, Table 1), primarily driven by leaf tissue ageing (Figure 2).
- The nutritive value changes are better explained by thermal time than by herbage mass.
- The ageing process explained approximately 70% of the variation in NDFD and more than 60% of the variation in DMD.
- Leaf lifespan was quantified at approximately 400 GDD, providing a new quantitative reference for this species (Table 2).
- The MDP model accurately predicted NDFD dynamics ($R^2 \approx 0.94$; RMSE ≈ 1.9%), demonstrating strong capacity to simulate short-term changes (Figure 1).

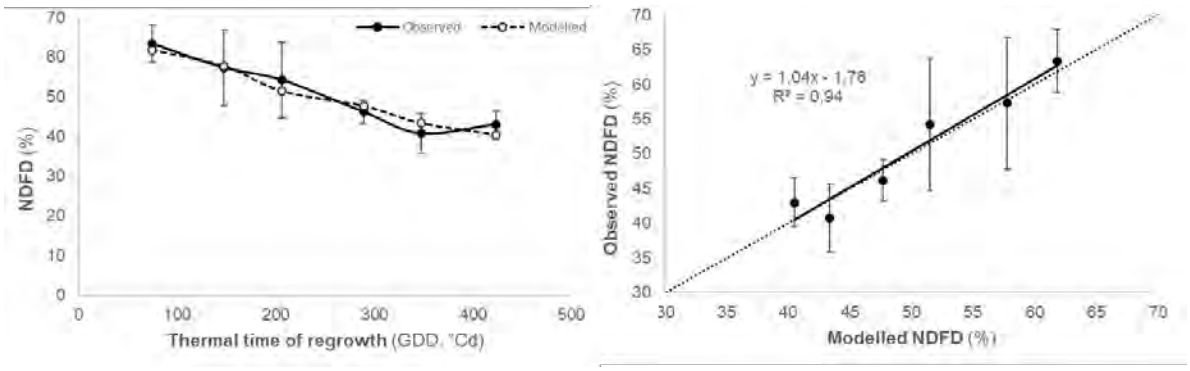

Figure 1. *Evaluation of nutritive value sub-model performance with observed and predicted leaf blade NDFD for Period 3 at leaf stages L1–L6 (left), and the relationship between observed and simulated NDFD compared to the 1:1 line (right) in Kikuyu pastures. Dotted lines show the 1:1 reference (y = x).*

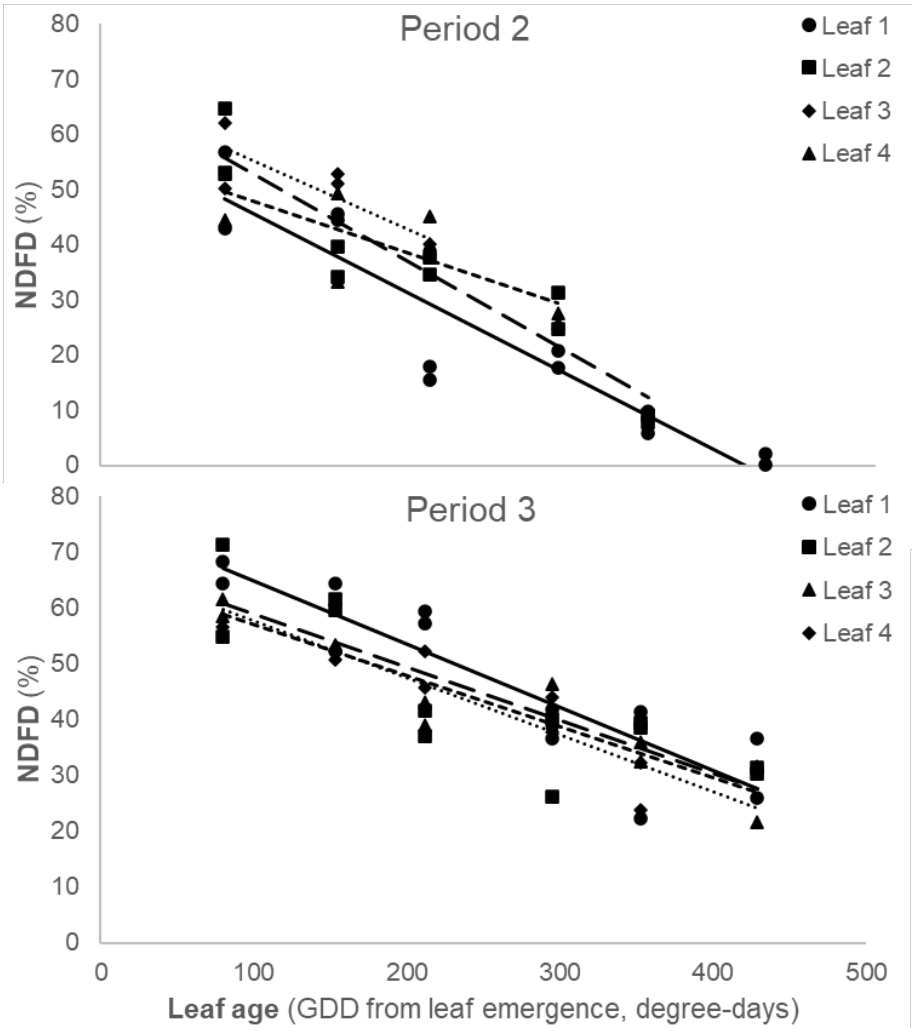

Figure 2. *Relationship between leaf age and digestibility of leaf NDF (NDFD). Leaf age (expressed in growing degree-days, GDD, from emergence) is for the first four leaves produced following cutting of Kikuyu pasture of Period 2 and Period 3. Symbols denote successive leaves: L1 (circles); L2 (squares); L3 (triangles); L4 (diamonds). Lines describe the linear functions for each successive leaf. P1 data are omitted due to no replication.*

Table 1. *Whole-sward nutritive value of leaf blades in different leaf stage of tillers of Kikuyu regrowths of three experimental periods regrowths (P1, P2 and P3). SE: standard error.*

Leaf	NDF (%)			NDFD (%)			DMD (%)		
Stage*	P1	P2	P3	P1	P2	P3	P1	P2	P3
L1	49.8	57.8	60.9	54.8	49.9	66.3	65.6	59.2	60.3
L2	49.5	49.3	57.8	57.9	52.0	60.8	67.2	64.5	62.9
L3	51.3	47.5	61.4	60.2	37.3	59.7	67.7	58.1	61.0
L4	52.6	54.2	63.6	59.1	41.6	45.4	66.6	56.5	48.5
L5	55.1	49.4	64.1	52.6	42.0	40.5	62.0	59.6	45.7
L6	55.7	52.3	63.0	52.2	37.4	45.7	61.4	55.4	49.4
Mean	52.3	51.8	61.8	56.1	43.4	53.1	65.1	58.9	54.6
DS	2.6	3.8	2.3	3.4	6.2	10.5	2.7	3.2	7.6

Table 2. *Morphogenesis characterisation of Kikuyu regrowth for three experimental periods (P1, P2 and P3).*

Variable	P1	P2	P3	Mean
Regrowth period (days)	31	35	35	34 ± 2
Regrowth period (GDD, °Cd)	622	601	721	648 ± 64
Mean daily temperature (°C)	23.2	21.0	23.8	23 ± 1
Herbage mass accumulation (kg DM ha^{-1})	NS	3506 ± 133	3472 ± 62	3489 ± 24
Accumulated leaves (leaves $tiller^{-1}$)	7.5 ± 1.3	8.1 ± 0.8	8.5 ± 1.0	8 ± 1
Growing leaves (leaves $tiller^{-1}$)	1.8 ± 0.6	2.5 ± 0.9	1.7 ± 0.6	2.0 ± 0.4
Leaf lifespan (leaves $tiller^{-1}$)	4.4 ± 0.6	3.5 ± 0.6	4.5 ± 0.9	4.1 ± 0.6
Leaf lifespan (days)	20.7 ± 3.1	15.1 ± 3.0	20.3 ± 4.1	19 ± 3
Leaf lifespan (GDD, °Cd)	396 ± 60	342 ± 75	401 ± 80	380 ± 33
Leaf appearance interval (days)	3.9 ± 0.8	3.0 ± 0.7	3.7 ± 0.8	3.5 ± 0.5
Leaf appearance interval (GDD, °Cd)	76 ± 15	63 ± 16	73 ± 16	71 ± 7
Leaf elongation rate (cm $leaf^{-1}$ day^{-1})	1.16 ± 0.4	0.87 ± 0.2	0.53 ± 0.3	0.90 ± 0.30
Senescence rate (cm $leaf^{-1}$ day^{-1})	0.58 ± 0.3	0.63 ± 0.3	0.45 ± 0.3	0.60 ± 0.10

8. Outputs

- A calibrated MDP model that can simulate leaf digestibility dynamic of kikuyu during pasture regrowth.
- Quantification of the main drivers of nutritive value decline in Kikuyu pastures during regrowth, identifying leaf ageing as the primary factor.
- First quantitative estimates for Kikuyu of key morphogenetic parameters, including leaf lifespan (~400 GDD) and rate of decline in NDFD (~0.17% per GDD).
- Development of a mechanistic framework linking leaf morphogenesis with pasture nutritive value dynamics.
- Better understanding of the nutritive value decline of kikuyu during regrowth for the evaluation of elite candidate lines of kikuyu.
- Generation of quantitative thresholds (e.g. 300–350 GDD regrowth) to support improved grazing management decisions.
- Peer-reviewed manuscript submitted to the Journal of Dairy Science.
- Presentation of results at the 2025 American Dairy Science Association (ADSA) annual meeting.

9. Applications and Impacts

At the cow and farm level, this project quantified the rapid decline in Kikuyu nutritive value during regrowth and identified practical grazing thresholds to minimise reductions in dry matter intake and milk production. Results indicated that maintaining grazing intervals shorter than 300–350 GDD (approximately 3–4 leaves per tiller) is critical to avoid major declines in digestibility. Previous studies indicate that a 10-unit reduction in NDFD may reduce milk production by approximately 2.5 kg per cow per day, highlighting the importance of timely grazing management.

At the industry and advisory level, improved management of Kikuyu pasture quality has the potential to enhance feed utilisation efficiency and reduce variability in milk production associated with poor pasture digestibility. The quantitative thresholds and modelling framework developed in this project provide new information that can support advisory services, digital agriculture platforms, and decision-support tools for pasture-based dairy systems.

In addition, the relationships identified between leaf morphology and digestibility provide a useful framework for future breeding and selection of Kikuyu genotypes with improved nutritive value characteristics.

10. Future Research Opportunities and Actions

Future research should validate these findings under commercial dairy farm conditions across a wider range of environments, management strategies, and seasonal conditions. Additional work is also needed to incorporate other drivers of pasture growth and nutritive value, such as nitrogen status and water availability, into the modelling framework to improve predictive capacity.

There is strong potential to integrate the model with real-time weather, remote sensing, NDVI, and on-farm pasture monitoring technologies to support the development of practical decision-support tools for grazing management. Current work is also focused on transitioning the model from an Excel platform to Python-based applications to facilitate scalability and future integration into digital agriculture systems.

Further opportunities include evaluating whether this modelling approach can support improved management of Kikuyu silage systems and exploring the integration of pasture biomass accumulation with predicted changes in nutritive value to better guide grazing and harvest decisions.

11. Project-wide Dissemination

Findings from the project were presented at the 2025 American Dairy Science Association (ADSA) Annual Meeting in the USA, reaching an international audience of researchers and dairy industry professionals. Results were also communicated internally through Dairy UP presentations and updates to project collaborators and stakeholders.

The project generated one peer-reviewed manuscript submitted to the *Journal of Dairy Science* (JDS), contributing to the dissemination of new knowledge on the nutritive value dynamics of Kikuyu pastures and pasture quality modelling.

In addition, project outcomes were disseminated through Dairy UP newsletter articles and webinars, supporting engagement with industry advisors, farmers, and the broader Dairy UP network.

Table 2. *Scientific publications from this project*

Author	Title	Journal	Year Published
Insua et al.	Understanding and predicting changes in nutritional value of Kikuyu	JDS	Submitted

12. Conclusions and Recommendations

The P1e project successfully quantified the short-term dynamics of nutritive value decline in Kikuyu pastures and demonstrated that these changes can be accurately predicted using a morphogenetic–digestibility modelling approach. Across experimental periods, consistent declines in NDFD and DMD during regrowth confirmed that leaf ageing is the primary driver of nutritive value loss, while leaf morphology influenced the rate and extent of this decline.

The project also demonstrated that thermal time is a more robust predictor of nutritive value dynamics than herbage mass alone, highlighting the importance of considering growth conditions and leaf developmental stage when interpreting pasture nutritive value. The MDP model accurately reproduced observed patterns of digestibility decline, providing a mechanistic framework capable of predicting short-term changes in Kikuyu nutritive value from temperature-driven leaf development processes.

These findings have direct implications for grazing management, as they provide quantitative thresholds and predictive tools to support better timing of grazing and improved utilization of Kikuyu pastures. In addition, the work establishes a strong foundation for future integration of nutritive value predictions with remote sensing technologies, pasture monitoring systems, and digital agriculture platforms.

Across the project, results were disseminated through international scientific conferences, Dairy UP activities, newsletter articles, and a peer-reviewed manuscript submitted to the *Journal of Dairy Science*, contributing to industry and scientific engagement.

13. Annexes

Table 3. *Conference presentations, abstracts and other meetings*

Authors	Title	Presentation Type	Conference/ Event	Location	Year	Audience
Insua et al.	Understanding and predicting changes in nutritional value of Kikuyu	Refereed abstract/Oral presentation	ADSA Annual Meeting	Louisville, USA	**2025**	40

Table 4. Technical reports, pamphlets, published material and other media engagements

Authors	Title	Place Published	Year
Insua, J., García S.	Kikuyu nutritional value	Dairy UP Website	2024
Insua, J., Correa-Luna, M.	P1e – Understanding and predicting changes in nutritional value of Kikuyu	Dairy UP Webinar	2024
Insua, J.	P1e Nutritional Value of Kikuyu-based pasture systems	Dairy UP Webinar	2022

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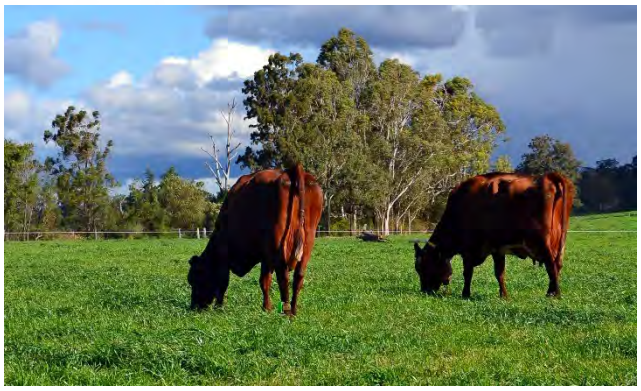
Final Project Summary

P1e – Kikuyu: Nutritional value

May 2026



In Australia's grazing-based dairy systems, pasture utilisation is closely linked to profitability. Kikuyu is well-adapted to subtropical areas. It is fast growing but of moderate quality and generally perceived as a grass of poor nutritive value for dairy cattle.

The Dairy UP team wanted to identify ways Australian dairy farmers could manage kikuyu pasture to improve quality and utilisation.

Timing of grazing

The timing of grazing has a big impact on kikuyu's nutritive value, which affects pasture utilisation.

Kikuyu leaves lose quality very rapidly if not consumed at the right time and the window of opportunity is generally smaller than for temperate grasses.

Grazing intervals also affect stem content. Stems are more fibrous so the digestibility and nutritive value of kikuyu pasture is reduced if there's higher stem content.

Dairy UP research

This project was designed to better understand and predict the rapid, short-term daily changes in the nutritive quality of kikuyu. The key aims were to:

- Predict short-term changes in kikuyu pasture quality in real time by modelling how the plant grows and ages.
- Measure how quickly pasture quality declines during kikuyu regrowth and identify the main factors driving that decline.
- Develop practical benchmarks, such as leaf lifespan and the rate of fibre quality decline, to support better grazing management decisions.

Unlocking the potential of Kikuyu

Dairy UP's P1 project aimed to unlock the potential of Kikuyu pastures used by NSW dairy farmers. P1 was a suite of five projects that collectively explored new management options to grow and utilise more Kikuyu over summer and increase the productivity of Kikuyu-based pastures.

P1a – Kikuyu: Remote monitoring

P1b – Kikuyu Investigating toxicity

P1c – Kikuyu: Developing New varieties

P1d – Kikuyu: Carbon on NSW dairy farms

P1e – Kikuyu: Nutritional value

This document is the final update on P1e – Kikuyu: Nutritional value.

Key findings

Findings from this work highlight the importance of grazing kikuyu earlier to maximise pasture quality and milk production. The project identified practical grazing targets of about 300–350 growing degree days (GDD), or 3–4 leaves per tiller, to help avoid rapid declines in digestibility during pasture regrowth.

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P1e final report

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Research and results

The Dairy UP team modelled changes in the nutritive value of kikuyu plants at different stages of regrowth.

They adapted a model originally developed for tall fescue and ryegrass. Plot-scale, field experiments were conducted in 2023 and 2024 to provide data to calibrate the model for kikuyu.

The model simulated changes in leaf digestibility during regrowth at different leaf stages. It accounted for leaf growth, leaf age, leaf size and nutritive value (fibre content, fibre digestibility and metabolizable energy).

Field experiments

The first stage involved three controlled field studies with small, replicated plots of kikuyu under different growing conditions in 2022, 2023 and 2024.

This enabled detailed assessment of plant morphology (structure) and nutritive value traits including age and leaf length, leaf number per tiller, fibre (NDF, NDFi) and digestibility (DMD and NDFD). This data was used to quantify, model and predict the main factors affecting the rapid decline in kikuyu nutritive value during regrowth.

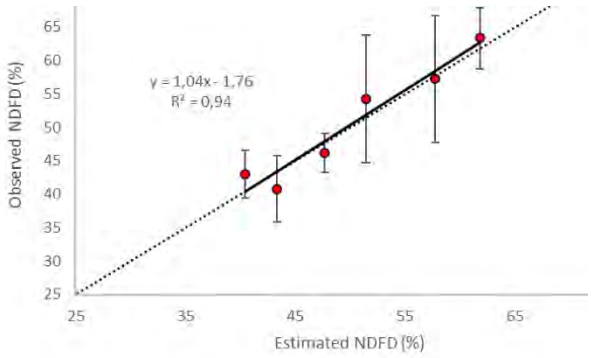
Results from the first two field trials were used for model development and calibration. The last one was used to validate the model by comparing model predictions with independently observed data.

Key terms

- **DMD** — dry matter digestibility (overall digestibility of the pasture).
- **GDD** — growing degree days, a measure of accumulated thermal time (heat over time) used to describe pasture development.
- **MDP model** — Morphogenetic and Digestibility of Pasture model, used to predict changes in pasture quality during regrowth.
- **NDF** — neutral detergent fibre (the fibre component of pasture; almost equivalent to the whole 'cell wall' component of the plant).
- **NDFD** — neutral detergent fibre digestibility (how digestible the fibre is for the animal).
- **NDFi** — neutral detergent fibre indigestibility (a measure of the undigestible fibre fraction in pasture; higher NDFi is associated with lower feed quality and intake).

Model

The model was calibrated using data from the field trials. The quality sub-model was validated and is very accurate (see graph). The model accurately



predicted the decline in digestibility associated with leaf stage.

Growing degree days

To track pasture growth, the Dairy UP team used growing degree days (GDD), a measure of accumulated thermal time.

The GDD approach involves adding up daily mean temperatures above the minimum temperature needed for plants to grow (usually 0°C for C3 plants like ryegrass and 10°C for C4 plants like kikuyu). Being a daily measurement, it combines temperature and age effects on plant maturity and is also used to quantify measures of other things such as leaf stage interval and reproduction stimuluses.

Findings

Key findings

- Kikuyu pastures showed a rapid and consistent decline in fibre digestibility (NDFD)

during regrowth, primarily driven by leaf tissue ageing.

- Changes in pasture quality were more closely associated with accumulated thermal time (GDD) than with pasture biomass accumulation.
- Leaf lifespan was quantified at about 400 growing degree days (GDD), providing a new benchmark for kikuyu pasture management.
- Leaf ageing explained about 70% of the variation in fibre digestibility (NDFD) and more than 60% of the variation in overall pasture digestibility (DMD).
- Changes in fibre concentration (NDF) were relatively small, indicating that declining digestibility of fibre, rather than fibre concentration itself, was the main driver of pasture quality decline.
- The pasture growth and digestibility (MDP) model accurately predicted changes in fibre digestibility (NDFD), demonstrating strong capacity to simulate short-term changes in kikuyu pasture quality during regrowth.

Applications and implications

On farm

The project identified practical grazing targets to help farmers maintain kikuyu pasture quality and milk production.

Findings indicated that grazing before 300–350 growing degree days (GDD), or about 3–4 leaves per tiller, is important to avoid major declines in digestibility.

New kikuyu leaves appeared about every 75 GDD, or every 2–4 days under NSW summer conditions. This suggests leaf stage may provide a practical guide for grazing timing, similar to approaches commonly used in ryegrass systems.

Previous research suggests that a 10-unit reduction in fibre digestibility (NDFD) may reduce milk production by about 2.5 kg/cow/day, highlighting the importance of timely grazing management.

Industry applications

The thresholds and modelling framework developed through this project provide new information that could support advisory services, pasture monitoring technologies and future decision-support tools for pasture-based dairy systems. Improved management of kikuyu pasture quality has the potential to improve feed utilisation and reduce variability in milk production associated with poor pasture digestibility.

Future priorities

- Test the findings under a wider range of commercial dairy farm conditions, seasons and management systems.
- Improve the model by incorporating additional factors affecting pasture growth and quality, such as nitrogen and water availability.
- Further develop the model for integration into future digital agriculture and decision-support tools using weather data, remote sensing and on-farm pasture monitoring technologies.
- Investigate the potential application of the model in kikuyu silage systems.
- Combine pasture growth and quality predictions to better guide grazing and harvest timing decisions.

Related projects

Project P1e used data from the Dairy UP's Remote Farm Monitoring Network (P1a) and from P1c: Exploring the genetic variability of kikuyu.

The modelling work was based on earlier work that modelled changes in pasture fibre digestibility (Insua et al., 2019a) and integrated those changes with remote sensor-derived data of pasture biomass and growth (Insua et al 2019b).

Collaborators

The P1e project was a collaboration between the University of Sydney and the UIB, Argentina.

Read more

Insua J., et al. (2025) Understanding and predicting changes in nutritional value of Kikuyu. ADSA Annual Meeting 2025, p175.

Insua J. R., et al. (2019a) Modeling the nutritive value of defoliated tall fescue pastures based on leaf morphogenesis. Agronomy Journal, 111, 714-724.

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DairyUP
Unlocking potential

FINAL OUTPUT JOURNAL

P2
Unlocking the potential of cows

P2a - Metabolic investigations into dairy cow longevity
P2b - Early alerts
P2c - Milk as a diagnostic tool
P2d - Diet and heat load
P2e - Calf husbandry
P2f - Infectious diseases
P2g - Heifers early calving

DairyUP
Unlocking potential

Final Report

P2 Unlocking the potential of the cow

This project was led by Ian Lean from Scibus

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

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Delivery organisations

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Executive Summary

Dairy UP Project 2 was a broad, integrated research program designed to improve the health, productivity, welfare and longevity of Australian dairy cattle. The project addressed these outcomes across the life course of the animal, from calf and heifer development through to mature cow survival, reproduction, disease risk and the impact of production systems. Across seven subprojects, that each focused on improving dairy cattle health, productivity, welfare, longevity and resilience. The work combined large-scale farm data, prospective field studies, retrospective analyses, diagnostic reviews, molecular technologies and nutrition research.

A central achievement of Project 2 was the development and use of the Dairy UP database, which provided a high-integrity data platform for research, farm reporting and future industry decision-making. The database integrated herd records, milk production, health, reproduction, cow movement, environmental and herd test information, and now contains approximately 200,000 lactation records across 30 farms. This data infrastructure supported several studies within the cow longevity, facility design and heifer rearing programs and demonstrated the value of well-audited, farm-derived data for both research and practical farm management.

The cow longevity studies on body condition score, body weight, blood metabolites and lipidomic profiles provided new insights into the biological changes that occur as cows age. These findings highlighted potential pathways, including omega fatty acid biology, that may contribute to differences in health, reproduction and survival among cows.

Project 2 also addressed system-level changes in the Australian dairy industry. Comparisons between pasture-based and housed or confinement systems indicated that both systems can support strong health and reproductive outcomes, although differences in diet, housing, management and lipid profiles may create different biological risk profiles. The project also examined the effects of nutrition and heat load on milk production, showing that dietary changes may have delayed effects on milk yield and that heat stress remains an important constraint on productivity.

Other subprojects extended the program beyond mature cow longevity. Work on milk as a diagnostic fluid reviewed the growing potential for milk-based monitoring of nutrition, udder health and infectious diseases. Early alert systems were explored as a mechanism to improve preparedness for animal, plant and environmental health risks. Calf and heifer studies examined early-life nutrition, calf performance, rumen adaptation, feedlot outcomes and age at first breeding. The infectious disease project used advanced sequencing technologies to improve understanding of respiratory and enteric pathogens in dairy calves and to strengthen future disease surveillance.

Specific outcomes for the projects include;

- A clear understanding of the increased risks of disease and metabolic disorder with increased parity of cows. This will provide farmers and advisors with clarity around the increased risks with increased parity of cows.
- Identifying, that there are marked differences in lipid profiles with age of cows and that cows in housed facilities have older profiles than those at pasture.
- These differences suggest means by which the problems of aging might be modified with lipid supplementation.
- Cows lose body condition but gain weight as they age, indicating a pathway to reducing risks of problems in older cows.

- We identified differences in herd removals between housed and pastured herds which will help farmers and advisors understand the benefits and risks from different systems.
- We identified methods to improve bedding management in compost bedded pack barns.
- An early alert system for bovine ephemeral fever was established.
- Milk has great value as a diagnostic fluid and is under-utilised by the Australian industry, Milk urea nitrogen will be very valuable to routinely report.
- An important finding was to quantify the lagged influence of diet on milk production highlighting that nutritional responses to diet occur over days to weeks.
- Diets differing in formulation provided the potential to integrate dairy calves into the beef supply chain but with different rumen lifetime adaptations.
- Greater milk consumption in early life (up to day 5) had substantial effects on weaning weight of calves.
- Holstein cattle achieved excellent feedlot performance compared to beef breeds. High performing cattle at feedlot induction were higher performing overall compared to those with lesser early performance.
- Metagenomic and amplicon sequencing methods will change understandings and improve accuracy of veterinary diagnostic testing.
- Calf diarrhoea is rarely caused by a single pathogen and is substantially explained by the farm environment. This reinforces the importance of good husbandry for farmers and advisors.
- Kobuvirus was identified as an emerging pathogen associated with diarrhoea.
- Pestivirus, while of moderate prevalence, was a substantial pathogen. This can be controlled with vaccination.
- Insights were gained on host responses to potential pathogens providing avenues for future control of disease without using antimicrobial treatments.
- Similarly respiratory disease risks were dominated by farm environment. Rhinitis-viruses and Torovirus were identified as pathogens. Again, disease occurs as result of interactions among potential pathogens.
- Host responses to pathogens were an important part of the pathogenesis of the disease.
- Reducing the age at first mating of heifers weighing >330 kg to 10 to 13 months as opposed to mating at >13 months resulted in > 800 L greater milk production in the first lactation as a result of more heifers entering and remaining in the herd. This has implications for costs of rearing and green-house gas footprint.
- Heifers with higher blood free fatty acids had a lesser risk of being actively cycling indicating the importance of a positive nutritional plane near mating.

Collectively, Project 2 delivered new knowledge, research infrastructure and practical insights to support a more resilient Australian dairy industry. The findings provide evidence to guide farm management, veterinary advisory services, future research priorities and industry decision-making as dairy systems become larger, more data-rich and more diverse in feeding and housing practices.

Project Overview

Item	Description
Project Title	Unlocking the potential of the cow
Funding Body	Dairy UP
Dairy UP Project	Project 2
Project Duration	4 years
Subprojects	P2a: Cow longevity P2b: Early alert systems P2c: Milk as a diagnostic fluid P2d: Heat stress P2e: Calves/Heifers P2f: Infectious disease P2g: Heifers early calving
Lead Organisation	Scibus
Project Lead	Ian Lean
Key Collaborators	University of Sydney, DPIRD, DataGene, Charles Sturt University, AgriBio, MLA

Abbreviations

100DICR — 100-day in-calf rate;
ACF — Automated calf feeders;
ADG — Average daily gain;
AFB — Age at first breeding;
AFC — Age at first calving;
ALA — α-linolenic acid;
AMR — Antimicrobial resistance;
ANOVA — Analysis of variance;
ATGL — Adipose triglyceride lipase;
BCS — Body condition score;
BHB — β-hydroxybutyrate;
BHG — British high-bodyweight gain;
BHV-1 — Bovine alphaherpesvirus-1;
BLG — British low-bodyweight gain;
BoRVA — bovine rotavirus A;
BRD — Bovine respiratory disease;
BSI — Bloodstream infection;
BTM — Bulk tank milk;
BW — Body weight;
BWT — Birth weight;
CBP — Compost-bedded pack;
CDP — Cytidine diphosphate;
CDS — Phosphatidate cytidylyltransferase;
CEPT — Choline/ethanolamine phosphotransferase;
CI — Confidence interval;
CL — Corpus luteum;
CON — Control;
CP — Crude protein;
CTP — Cytidylyltransferase;
DAGK — Diacylglycerol kinase;
DGAT — Diacylglycerol acyltransferases;
DHA — Docosahexaenoic acid;
DIM — Days in milk;
DM — Dry matter;
DPIRD — Department of Primary Industries and Regional Development;
ECC — Chromogenic E. coli/coliform media;
EHG — European high-bodyweight gain;
ELG — European low-bodyweight gain;
ELISA — Enzyme-linked immunosorbent assay;
EMAI — Elizabeth Macarthur Agricultural Institute;

EPA — Eicosapentaenoic acid;
ESBL — Extended-spectrum β-lactamase;
FFA — Free fatty acids;
FT-MIR — Fourier-transform mid-infrared;
GC — Gain category;
GPAT — Glycerol-3-phosphate acyltransferase;
HCW — Hot carcase weight;
HHG — Holstein high-bodyweight gain;
HLG — Holstein low-bodyweight gain;
HNBRED — Hazard of not being bred;
HOUSED — Housed or housing-based dairy system;
HPREG — Hazard of pregnancy;
HR — Hazard ratio;
IGF — Insulin-like growth factor;
IGFBP-6 — Insulin-like growth factor-binding protein 6;
LA — Linoleic acid;
LAMP — Loop-mediated isothermal amplification;
LASSO — Least absolute shrinkage and selection operator;
LI — Lactational incidence;
LPAAT — Lysophosphatidic acid acyltransferases;
LPC — Lysophosphatidylcholine;
MALDI-TOF — Matrix-assisted laser desorption/ionisation time of flight;
MLA — Meat & Livestock Australia;
MS — Mass spectrometry;
NCBI — National Center for Biotechnology Information;
NDF — Neutral detergent fibre;
NDICP — Neutral detergent insoluble crude protein;
NEFA — Nonesterified fatty acids;
NFC — Non-fibre carbohydrates;
NSW — New South Wales;
OPAL — Odds of becoming pregnant in a lactation;
O-PLS — Orthogonal partial least squares;
PAP — Phosphatidate phosphatase;
PASTURE — Pasture-based cows or pasture-based system;
PC — Phosphatidylcholine or Principal component;
PCR — Polymerase chain reaction;
PE — Phosphatidylethanolamine;
PEMT — Phosphatidylethanolamine-N-methyltransferase;
PI — Phosphatidylinositol;
PIS — Phosphatidylinositol synthase;
PL — Phospholipids / glycerophospholipids;
PLD — Phospholipase D;

PMT — Phosphoethanolamine methyltransferases;
PREG1 — Pregnancy to first breeding;
PS — Phosphatidylserine;
PSD — Phosphatidylserine decarboxylase;
PSS — Phosphatidylserine synthase;
RNA — Ribonucleic acid;
RVA — Rotavirus A;
SA — South Australia;
SCC — Somatic cell count;
SD — Standard deviation;
SE — Standard error;
SM — Sphingomyelin;
ST — Sequence type;
THI — Temperature-humidity index;
TMR — Total mixed ration;
TRT — Treatment;
UTI — Urinary tract infection;
VFA — Volatile fatty acid;
VIC — Victoria;
VLDL — Very-low-density lipoproteins;
VP — Viral protein;
VP7 — Viral protein 7;
WWT — Weaning weight.

Project Background and Rationale

The Australian dairy industry is changing rapidly. Herds are larger, production systems are becoming more diverse, and many farms are increasingly reliant on digital records, and automated technologies. At the same time, the industry faces persistent challenges including reproductive inefficiency, disease, cow removal, heat stress, calf health, and the need to maintain social licence across both pasture-based and more intensive dairy systems.

Dairy farms now generate large amounts of information, but much of this information remains under-used. Data may be inaccessible, inconsistently recorded, fragmented across software platforms, or insufficiently audited for research and decision-making. Project 2 responded to this challenge by developing a high-integrity database that could integrate data from herd management software, milk processors, weather stations, herd testing and farm records. This provided a platform for research-grade analyses and for returning useful information to producers and advisors.

Cow longevity was a central focus because improving lifetime productivity requires cows to remain healthy, fertile and productive across multiple lactations. Previous industry and research data have consistently indicated that older cows experience greater risks of disease, reproductive failure and removal. However, the biological reasons for these changes remain incompletely understood. Project 2 therefore examined parity-associated changes in reproduction, health, body condition, body weight, blood biochemistry and lipidomic profiles, with the aim of identifying practical management opportunities and biological pathways that may support improved longevity.

The project also recognised that longevity and productivity are influenced by the broader system in which cattle are managed. Australian dairy farming includes both pasture-based and increasingly housed or confinement-based systems. These systems differ in diet, housing, climate exposure, labour structure and management opportunities. Understanding the strengths, limitations and biological consequences of these systems is essential for supporting industry adaptation and maintaining public confidence in dairy production.

Beyond mature cow performance, Project 2 addressed early-life development, milk-based diagnostics, infectious disease surveillance, early warning systems and age at first breeding. These areas are linked by a common need for earlier, more accurate and more practical information to guide decisions.

Project Objectives

This report presents each subproject individually, including the background rationale, objectives, methods, key findings and implications for industry. While each subproject has distinct aims, the combined program addressed the common goals of:

- Building on the strong links between Scibus, NSW DPIRD, the University of Sydney, private and other research providers to produce a program of research designed to unlock the potential of Australian cattle and to enhance their well-being, health, welfare and 'happiness', while reducing the environmental footprint: that is, to enhance our social licence.
- To generate evidence that helps Australian dairy farms improve animal health and productivity while adapting to changes in herd size, data availability, climate risk, disease pressure and production system design.
- Develop new data analytical strategies to determine the effects of novel interventions on health, reproduction and production.
- Use a network of monitored individual farms in NSW, VIC and SA to develop 'co-operative' models for data- and knowledge sharing and innovative, farmer-orientated, extension activities.
- Demonstrate and quantify the potential to increase productivity in commercial farms through advanced systematic, data-based management of cattle through nutrition, genetics, environmental and health management.

Subproject P2a Cow Longevity – i. Dairy UP Database

Summary

A core component that facilitated much of the work was the development of a database that would provide farm data of high integrity. We recognized that the current industry databases had substantial flaws and that a different structure was required to provide data suitable for decision making and evaluation. This required the integration of disparate sources of data including herd records, data from herd recording, environmental data and milk production and health records recorded on farm-based software often associated with milk meters. This innovative database developed by Drs Golder, Sheedy and Lean with the assistance of DataGene. This development was highly regarded by the panel conducting the mid-term review of DairyUP.

There are currently 200,000 lactational records on the database across 30 farms. The database has contributed substantially to all prospective studies within Project 2a, production of highly regarded farm reports and, subject to ongoing development and funding, plans to expand and support future projects (*Figure 5.a*).

Background

The Australian dairy industry generates an enormous amount of data. Most of these data are under-utilised. If these can be accessed and presented in usable, farmer-friendly forms then they can be capitalised on to improve the resilience of the dairy industry.

Missed opportunities for data driven decisions occur because (1) some data are never accessed from the source ie farm computers, (2) data are of too poor quality to be of use, (3) data are collated and held in disaggregated institutions, (4) data are not in an easily usable form, and (5) useful integrations and interpretations of data do not make it back to the source farm. Therefore, practice-change does not occur.

These reasons are exacerbated by Australian dairy farms using a variety of herd management software programs; some farms use more than one software program. The number of software programs available to assist with specific farm management practices such as total mixed ration feeding and fertility, as well as the core herd management software program are increasing. These generate large amounts of data that does not always integrate into the main herd management software, further stalling effective data utilisation. Data entry and data definitions are different for each of the herd management software programs. The Dairy UP database acquires data from both opportunistic and targeted sources to provide a platform to offer new and important insights.

Methods

In brief, data is collected from multiple sources, including herd management software, milk factory, weather station data and herd test centres. Data includes treatment and health, reproductive, cow movements, cow exits, daily milk or feed, local weather, herd test data and herd level descriptors. To help ensure high data quality, the Dairy UP database is dictated by a comprehensive list of rules and requirements outlined in Business Requirements Documents. Errors or unusual data are flagged for manual examination and correction as required. This auditing process often involves clarification through direct consultation with farm managers.

A specific goal of the Dairy UP P2a project was to investigate longevity of dairy cattle, which necessitates the clear understanding of cow exit reasons. The national database was not fit for this specific purpose due to limited culling definitions and extensive missing data. A report by Workie et. al. (2021) indicated that the most common reasons for Australian dairy cow culling was 'other reasons not reported' (37.4%). The Dairy UP database requires each exited cow to have up to three culling or mortality reasons recorded, using an expanded list of 50 exit reasons. Currently there are 29,542 sold animals with only 29 (0.1%) listed as 'sold – unknown reason'. Without the detailed culling data, data on reproduction and health will be less valid.

Implications

The value of an Australian database with research-grade data integrity is demonstrated through the Project P2a studies discussed later in this document. Comparisons between pasture-based dairy production and intensively housed systems indicate that both approaches can achieve similar health and reproductive outcomes. These findings provide important evidence to support the social licence of intensification within the Australian dairy industry. Outcomes from Dairy UP 1.0 have been widely disseminated through consultants, media releases, social media, domestic and international conference presentations, scientific publications, roadshows, industry field days, summary documents,

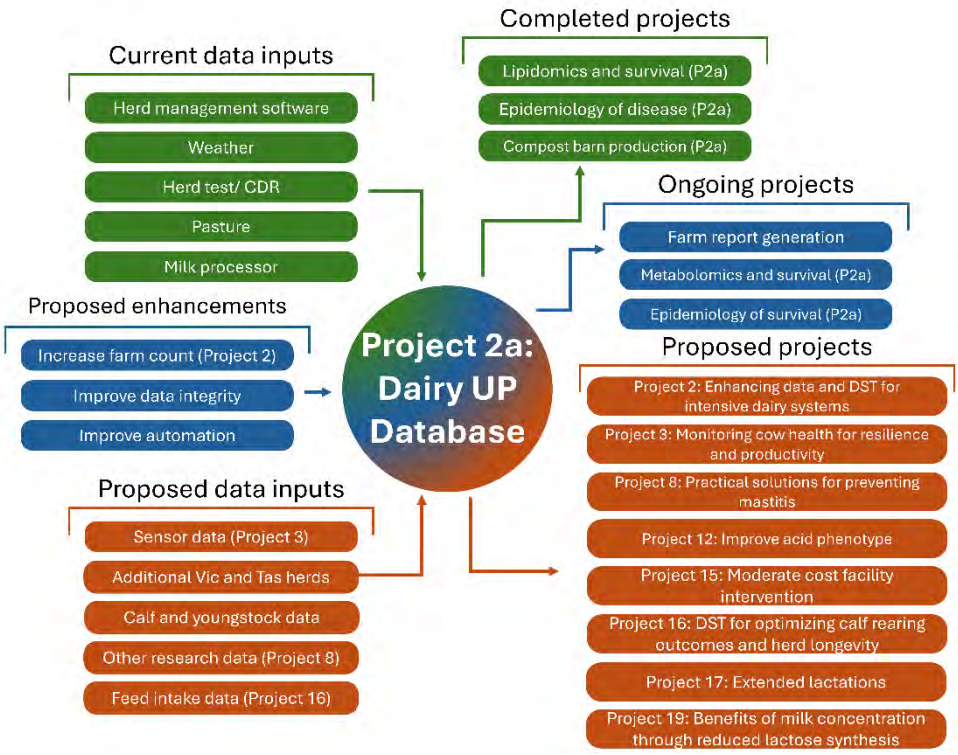

Figure 5.a: Schematic of the current and proposed data inputs, proposed enhancements, completed, ongoing and proposed projects linked to Project 2a – I Dairy UP database

and direct email communication.

Subproject P2a Cow Longevity – ii. Retrospective Study Series

Background

There is a need to ensure that we unlock the potential of the cows to produce by providing interventions to allow them to be productive, healthy and reproductively successful. Further, there is a need to understand the implications of changes in production systems to identify strengths and weaknesses of different systems and to provide social license for different production systems.

Increased parity is negatively associated with survival and reproduction in different production systems

doi.org/10.3168/jds.2021-21672

Authors

I.J. Lean, H.M. Golder, S.J. LeBlanc, T. Duffield, J E.P. Santos

Background

We conducted a retrospective meta-analysis based on individual cow data to assess the associations of parity, level of production, and pasture-based or intensively fed systems with fertility. Our goal was to provide understandings of the role of parity in risks for removal and reproductive failure. We hypothesized that risks of reproductive failure would increase with parity but not differ with production system.

Methods

Multilevel models were used to evaluate the fixed effects of parity, milk, milk solids, milk fat and protein percentage and yield, and production system [intensively fed (n = 28,675) or predominantly pasture fed (n = 4,108)] on reproductive outcomes.

Findings

The outcomes were the hazard of not being bred (**HNBRED**), hazard of pregnancy (**HPREG**), pregnancy to first breeding (**PREG1**), and odds of becoming pregnant in a lactation (**OPAL**). The 32,783 cows were in 13 studies conducted in Australia (14.6% of cows), Canada (2.4% of cows), and the United States (83.0% of cows). There were 38.5% of cows in the sample in parity 1, 27.3% in parity 2, 16.7% in parity 3, 9.0% in parity 4, and 8.6% in parity ≥5. Compared with cows of parity 1, parity ≥5 cows had a greater HNBRED [hazard ratio (**HR**) = 2.45], lesser HPREG (HR = 0.73), and reduced OPAL (odds ratio = 0.36). However, the parity ≥5 cows had similar PREG1 to other parities except for parity 1. This suggests the possibility of a higher proportion of subfertile parity ≥5 cows than for other parities. Associations between parity and reproductive measures were influenced by the different milk production measures, indicating that milk yield and milk component percentages and yields modified the odds or hazards of successful reproduction. All milk production measures had quadratic associations with OPAL, indicating that either low or high production or concentration of solids within a cohort reduced OPAL. This reduced OPAL reflected a greater HNBRED for lower milk yield and milk protein and fat yielding cows. Both milk yield and milk protein percentage had quadratic associations with HPREG. When centered milk yield was categorized into quartiles, small differences in HPREG existed. A more marked association of milk protein percentage occurred with HPREG, with optimal HPREG at approximately 0.5% above group mean milk protein percentage. Milk fat percentage (HR = 0.901), fat yield (kg/d; HR = 0.78), protein yield (kg/d; HR = 0.71), and milk solids yield (kg/d; HR = 0.84) were all linearly associated with reduced HPREG.

Difference in production systems did not have substantive effects on PREG1 but did for HNBRED, HPREG, and OPAL. Estimates of associations of parity with reproductive outcomes HNBRED, HPREG, and OPAL were influenced by milk and milk solids yield; older cows had markedly lower reproductive outcomes. Interestingly, for PREG1, there were few differences among parities and differences were less influenced by milk yield and constituent measures.

Implications

The marked associations of parity with removal for all reasons, deaths and culling, and reductions in HNBRED, HPREG, and OPAL indicate a need to focus on the physiological changes with parity to produce better strategies to support optimal longevity of cows.

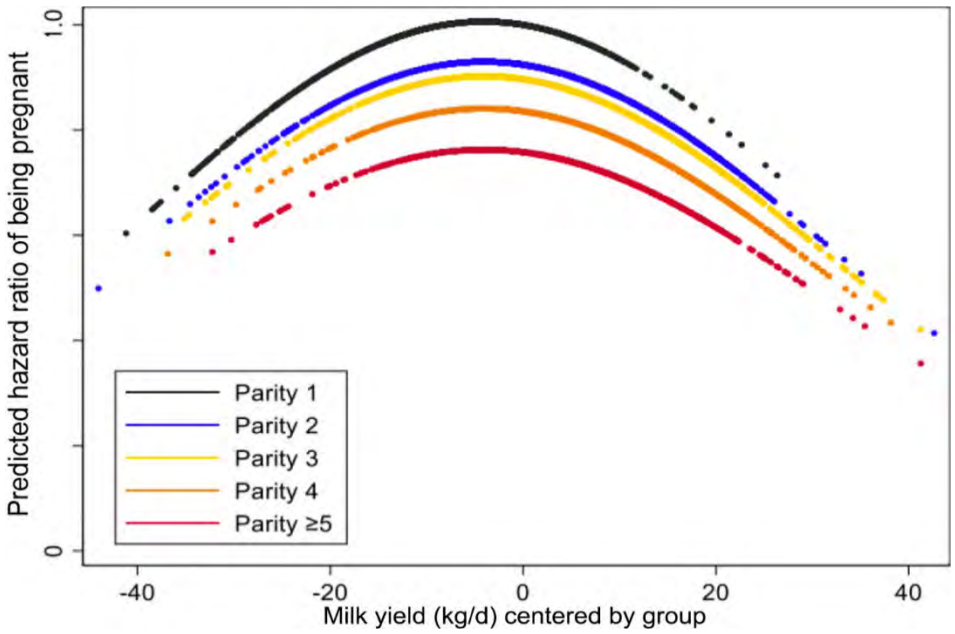

Figure 6.a: The association of milk yield (kg/d) centered in group within herd at approximately 70 DIM on the x-axis with the relative predicted hazard of being pregnant on the y-axis. The data are from 20,071 cows. Based on the model that contains parity, study year, and production system, all parity groups differed in probability or hazard of pregnancy (HPREG). Cows in parity 1 had the greatest daily probability or HPREG, and parity ≥5 had the least probability of pregnancy.

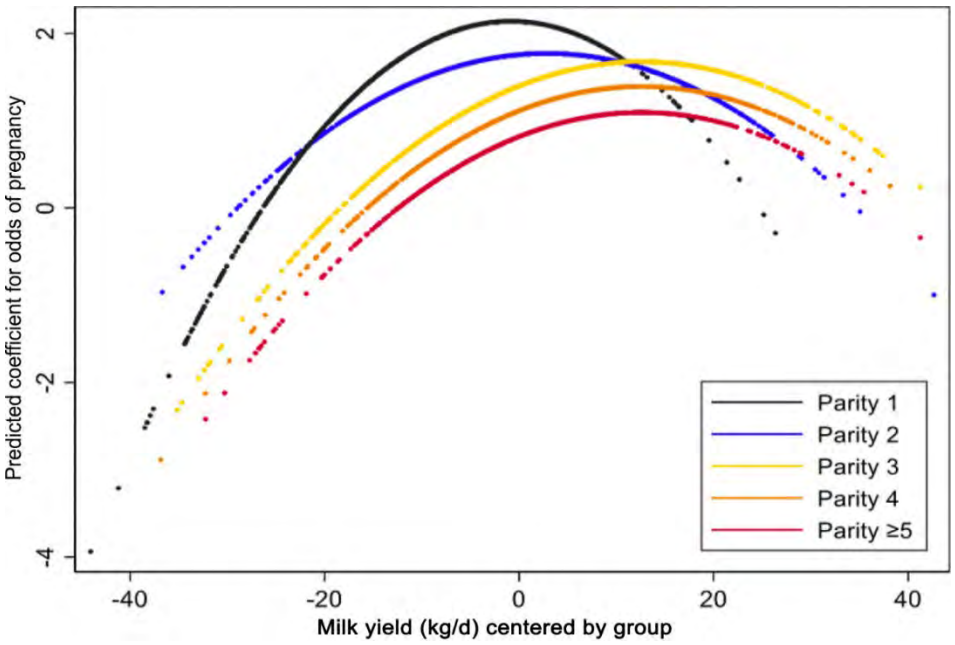

Figure 6.b: The association of milk yield (kg/d) centered on group within herd at approximately 70 DIM with the predicted coefficient of odds of becoming pregnant in a lactation (OPAL). The data are from 28,071 cows. The x-axis is milk yield centered by group, and the y-axis is the predicted coefficient of odds of pregnancy at first insemination. Based on the model that contains parity and production system and the interaction of parity with production system, all parities differed, with cows in parity 1 having the greatest odds and parity ≥5 having the least odds of pregnancy in the lactation.

Associations of parity with health disorders and blood metabolite concentrations in Holstein cows in different production systems

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Authors

I.J. Lean, S.J. LeBlanc, D.B. Sheedy, T. Duffield, J.E.P. Santos, H.M. Golder

Background

The associations among diseases and parity are important, there is a need to identify potential risk factors that may influence these associations. The pathogenesis of the increased risk of removal for reproductive failure and differences in odds or risk of disease with parity, as well as differences in blood metabolites with parity, may indicate aspects of metabolism that alter with parity

Methods

Data were obtained from studies in Australia, Canada, and the United States using individual cow data from 28,230 Holstein cows to evaluate associations between parity and disease. Our goal was to develop understanding of disease risks for cows of differing parity.

Findings

Parity ≥5 represented 2,533 cows or 9.0%, parity 4 was 9.8% (2,778), parity 3 as 19.0% (5,355), parity 2 as 28.1% (7,925), and parity 1 was 34.1% (9,639) of the sample. Of these cows, 15.5% were in Australia, 14.7% in Canada, and 69.8% in the United States. Lactational incidence (**LI**) risk of clinical hypocalcemia increased with parity from 0.1% for parity 1 to 13% for parity ≥5 cows. The marked increase suggests profound differences in metabolism with increased parity. The LI of clinical mastitis was 17.4%. The odds of mastitis increased with parity to 2.5 times greater in parity ≥5 than in parity 1. The LI of lameness increased with parity; specifically, the odds of lameness was 5.6 times greater for parity ≥5 than parity 1. Dystocia incidence was 8.7% and greatest for parity 1 cows. The LI of retained placenta was 7.4% and increased with parity, with the odds for parity ≥5 2.3 times greater than for parity 1. The LI of metritis was 10% and of endometritis 14%, with the greatest odds in parity 1. The LI of clinical ketosis was 3.3% with a marked increase in odds with parity. The prevalence of subclinical ketosis was 26.8% with only cows in parity 1 having lower odds than other parities. Parity ≥5 cows had greater odds (odds ratio = 1.7) of respiratory disease than parity 1 cows, which were lesser than other parities.

Metabolite concentrations were evaluated in 5,154 Holstein cows in the precalving, calving, and immediate postcalving data sets. Metabolic measures near peak lactation provided 1,906 observations. Concentrations of β-hydroxybutyrate (**BHB**) and nonesterified fatty acids increased with parity on d 1 to 3 of lactation and at peak lactation. On d 1 to 3 after calving differences in glucose, nonesterified fatty acids, and BHB indicated a greater reliance on mobilized lipid to export energy to peripheral tissues as BHB for greater parity cows. Differences in concentrations among parity groups were marked at times, for example >0.20 mM in Ca for parity 1 and 2 to parity ≥5 and >0.33 mM for all older parities compared with parity 1 for P on the day of calving. The marked increase suggests profound differences in metabolism with increased parity are probably influenced, in part, by increased production. We found marked differences in concentrations of metabolites with parity that are consistent with reduced reproduction, health, and body condition for higher parity cows. These unfavourable differences in metabolism in Ca, P, glucose, and cholesterol concentrations for higher parity cows also complement the often-substantial differences in disease risk with parity and suggest a need to carefully consider the parity structure in study design.

Implications

Managers and advisors will need to consider methods to reduce risk of health disorders tailored to cows of different ages.

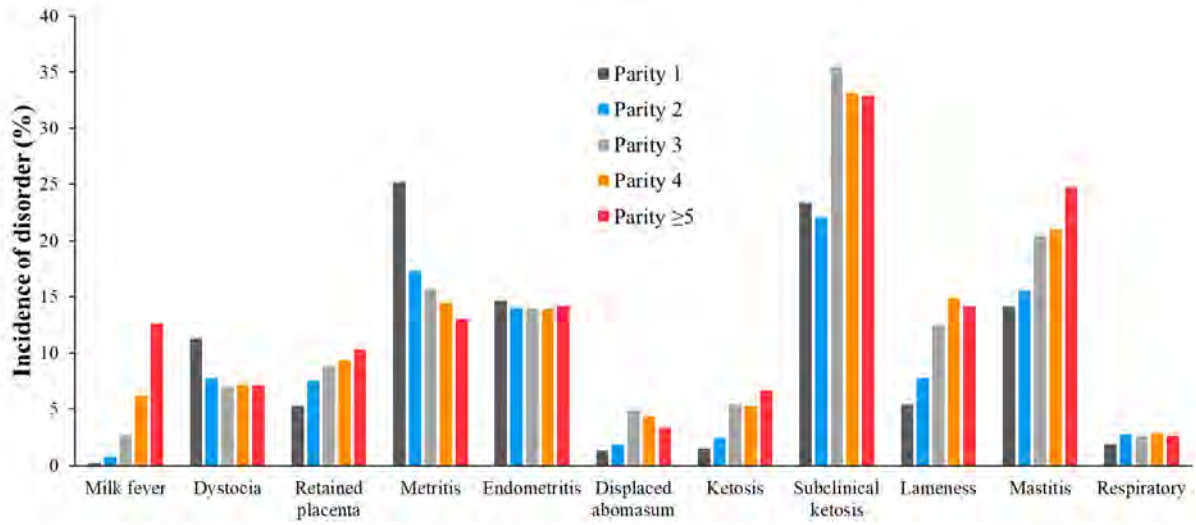

Figure 6.c: Incidence of clinical and subclinical disease by parity

Subproject P2a Cow Longevity – iii. Body Condition Score Series

Background

The accessible metrics of body condition score (**BCS**) and body weight (**BW**) are important in assessing the health of dairy cows. During the transition from late-pregnancy to peak-milk production, cows mobilise their stored energy and protein reserves to meet the homeorhetic demands of milk production. However, excessive mobilisation has been associated with increased disease incidence, reproductive inefficiencies, poor cow welfare, and is observed more often in high parity cows. Although BCS and BW are the most commonly used metrics to assess body tissue reserves, their correlation is considered only low to moderate, and is influenced by differences in visceral adipose tissue, stage of lactation or pregnancy, water intake, gut fill, and parity. To address this limitation a novel combined BCS – BW classification scheme is introduced and explored.

Holstein dairy cows lose body condition score and gain body weight with increasing parity in both pasture-based and total mixed ration herds

doi.org/10.3168/jdsc.2022-0246

Authors

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Background

Body condition scoring (BCS) and body weight (BW) are observations associated with labile tissue reserves, health, and reproduction efficiency of dairy cows.

Methods

The effect of parity (1 through to ≥5) and feeding system (pasture-based and total mixed ration [**TMR**]) on BCS and BW were evaluated utilizing raw data sets from 16 retrospective studies that totalled 24,807 Holstein cows across 3 nations (Australia, Canada, and the United States). Linear regression models were used to investigate the 5 outcome variables of precalving BCS, peak milk BCS, change in BCS from precalving to peak milk, and peak milk BW and their respective associations with parity and feeding system. To help control for the influence of calendar time, study treatment protocols when applicable, and genetic change, all outcome variables were center-transformed around each study group mean.

Findings

Including feeding system as a covariate improved model fit for most outcome variables; however, the relative effect size of parity was generally much greater than feeding system effect size. Parity 2 cows had the lowest precalving BCS of −0.087 [95% confidence interval (**CI**): −0.107, −0.065] less than the mean, whereas parity 1 cows had the greatest, 0.068 (95% CI: 0.043, 0.092) above mean, regardless of feeding system. Peak milk BCS overall decreased with increasing parity (parity 1 to parity ≥5: −0.13, 95% CI: −0.19, −0.08) and BCS change during the transition period monotonically decreased with increasing parity (parity 1 to parity ≥5: −0.22, 95% CI: −0.26, −0.17). Peak milk BW monotonically increased with increased parity (parity 1 to parity ≥5: 114 kg, 95% CI: 104, 125). A waffle plot was used to present the proportions of cows, by parity, that were partitioned into “low BCS and low BW,” “low BCS and high BW,” “high BCS and low BW,” or “high BCS and high BW” groups. Cows were assigned either a high or low status by being above or below their specific centered study group means, respectively. Considering a null hypothesis of 25% per

BCS-BW category, there was a striking change in category from parity 1 cows that were predominantly in the "high BCS and low BW" category (61.2%) to parity ≥5 cows that were predominantly in the "low BCS and high BW" category (55.5%).

Implications

The study supports studies showing increased weight and change in BCS with increased parity. We highlight the associations among production system, BCS, BW, and parity.

Associations among body condition score, body weight, and serum biochemistry in dairy cows

doi.org/10.3168/jds.2024-25425

Background

Body condition score and BW yield insights into body tissue reserves and diet, and serum biochemical measures reflect the metabolic status of cows. Associations between body composition measures and biochemistry are unclear and investigation may reveal important information on the metabolic and physiological status of cattle with varying levels of labile tissue reserves.

Methods

Cohorts of 739 nonlactating, late-pregnancy, dry cows (26.9 d prepartum, standard deviation [**SD**] = 12.4) and 690 peak-milk cows (58.0 DIM, SD = 14.5) were selected by stratified (parity: 1, 2, 3, >3) random sampling from 30 farms (15 pasture, 15 TMR) in this cross-sectional study. Eleven analytes were collected, analyzed, and standardized within group (cohort/breed per farm). Mixed linear models for BCS and BW were specified, with the random effect of group. A 6-point, unordered, categorical body-group classification that combined BCS (greater, equal to, or less than group median; as high, median, or low BCS) and BW (greater or less than group median; as high or low BW) was

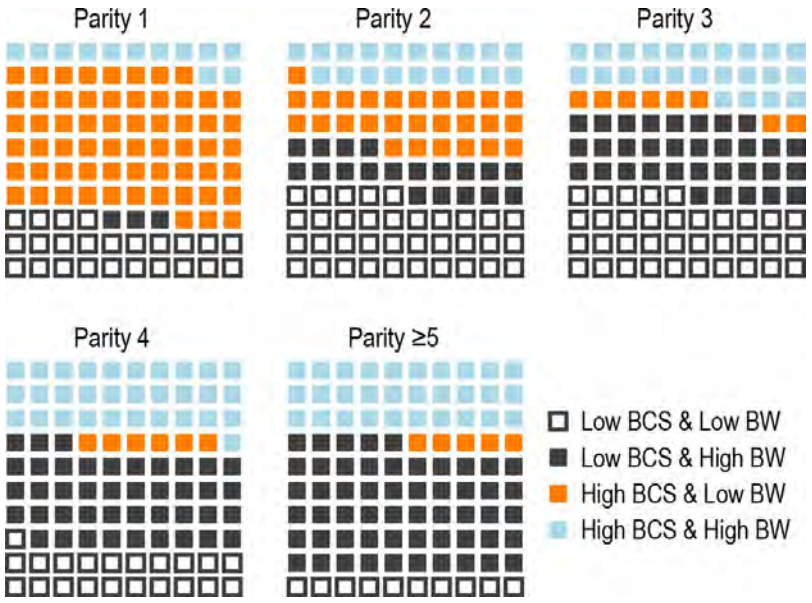

Figure 7.a: Proportion of cows in each category of BCS (low BCS ≤ group mean, high > group mean) and BW (low BW ≤ group mean, high > group mean) by parity at peak milk. Indicating a shift from High BCS and Low BW (orange square) in young cattle to Low BCS and High BW in older cattle (black square).

analyzed by polytomous logistic regression. Effect sizes are listed for a 1 SD increase in the specified analyte, keeping other covariables at their mean value.

Findings

Dry BCS was positively associated with albumin (0.075 BCS ± 0.014 standard error [**SE**]), urea (0.038 BCS ± 0.014 SE), and glucose (0.052 BCS ± 0.014 SE), and negatively with the interaction between cholesterol and days precalving. Dry BW positively associated with albumin (11.03 kg ± 2.48 SE) and negatively with cholesterol (−8.47 kg ± 2.57 SE). Peak-milk BCS was positively associated with albumin (0.47 BCS ± 0.015 SE), BHB (0.048 BCS ± 0.015 SE), and glucose (0.051 BCS ± 0.015 SE). Peak-milk BW was positively associated with albumin (6.94 kg ± 2.35 SE) and negatively with Ca (−7.02 kg ± 2.33 SE). Increasing BW and decreasing BCS was associated with increasing parity, except in dry second-parity cows that had low BCS. The dry polytomous model associated a 1 SD increase in albumin with a 4.89% ± 1.56 SE decreased risk of being low BCS/low BW and 5.87% ± 1.46 SE increased risk of high BCS/high BW. Risk change associated with 1 SD of glucose was −5.61% ± 1.58 SE for low BCS/high BW and 3.17% ± 1.58 SE for high BCS/high BW. For the peak-milk cohort, change in risk was associated with albumin for low BCS/low BW −3.67% ± 1.56 SE, low BCS/high BW −3.22% ± 1.53 SE. Risk change with 1 SD of BHB was −3.36% ± 1.47 SE for median BCS/low BW, 2.86% ± 1.44 SE for high BCS/low BW, and 2.69% ± 1.37 SE for high BCS/high BW. Risk of low BCS/low BW was greatest in second-parity cows, and high BCS/high BW was greatest in dry cows with greater than third parity and third-parity cows in peak milk. There were no interactions between parity and analytes.

Implications

Albumin was consistently and positively associated with body measurements and milk production parameters, and milk production was negatively associated with BCS. We speculate that the observed associations between metabolites that reflect protein and energy metabolism, including albumin, glucose, urea, cholesterol, BHB, and calcium, within a cohort and body tissue reserve measures (BCS and BW) may indicate that protein metabolism importantly differs among cattle. Further investigation into albumin and protein metabolism may identify biological pathways that support high milk production while reducing BCS-related diseases and loss.

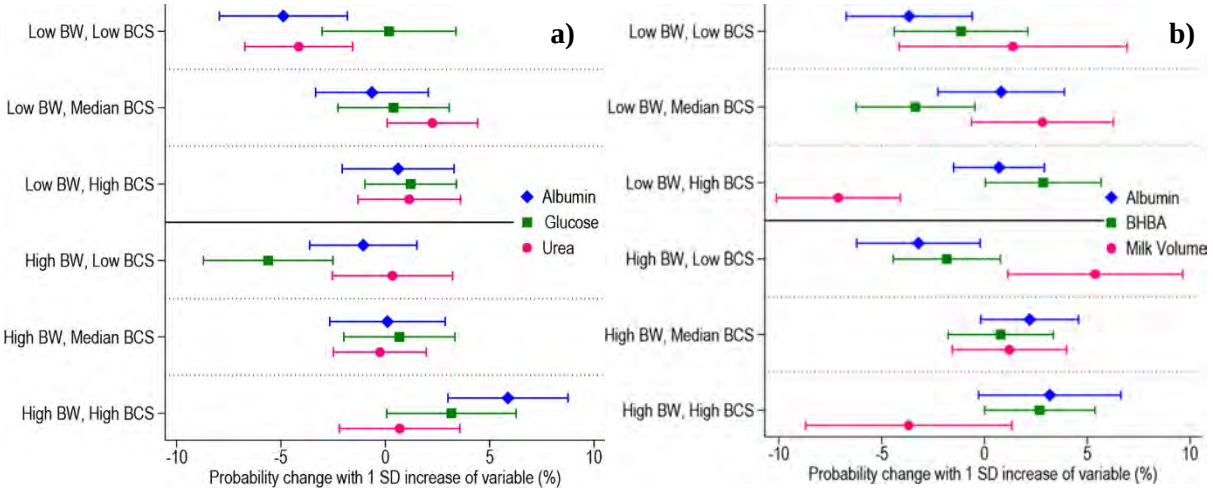

Figure 7.b: The change in probability of being in a specific body condition group associated with a 1-SD change in the shown variable for cows in a) the dry-cow cohort and b) the peak-milk cohort, with all other variables in the multivariable polytomous model held constant at their mean values. The error bars are 95% CI

Integration of body condition score and weight

These two papers introduced and refined a novel metric of considerable importance. We identified only weak associations between BCS and BW and noted that the labile reserves of tissue may differ greatly between cows with high BCS but low BW and those of similar BCS and high BW. Given that the novel metric was associated strongly with parity, metabolite concentrations and milk production, this metric will be used in future research and to more precisely understand body tissue reserves.

Subproject P2a Cow Longevity – iv. Lipidomic series

Background

Reducing the risk of adverse health and reproductive events as cows age could provide, producers with options to shift herd management strategies towards a lower replacement rate and extended longevity. However, while substantial collective research efforts have focused on individual diseases and reproductive challenges, there have been no similar effects into investigating the broader biological changes that occur with the aging process in dairy cattle. This lack of research interest is somewhat surprising given the consistent associations between parity and incidence of diseases and reproductive failure. Understanding the biological underpinnings of aging in cows may reveal novel targets for intervention that allow cows to maintain their health and productivity as they age.

A compelling platform for investigating the biology of dairy cow aging is lipidomics, the large-scale study of lipids. Continued advances in laboratory analytical technology now allow precise quantification of very-low abundance, polar and non-polar lipids that facilitate the identification of clinically relevant bioactive lipids. With lipidomics now an accessible research tool, there have been an increase in cow lipid metabolism studies, reflected by recent reviews on topics of sphingomyelin and ceramide metabolism, omega fatty acids, and the endocannabinoid system. Lipidomics is also being used to study healthy aging and age-related diseases in humans. Given these developments, lipidomics was considered an appropriate analytical platform to investigate the biological changes associated with cattle aging and longevity.

Glycerophospholipids in dairy cow health and longevity: a review

doi.org/10.1017/S0022029926102222

Authors

D.B. Sheedy, H.M. Golder, S.C. Garcia, I.J. Lean

Summary

This review examines the role glycerophospholipids (**PL**) in dairy cow health, with specific focus on phosphatidylcholine (**PC**), phosphatidylethanolamine (**PE**), phosphatidylinositol (**PI**) and phosphatidylserine (**PS**). Increasing parity of cows is associated with lower concentrations of plasma PL that contain very long-chain omega-3 fatty acids, including docosahexaenoic acid and eicosapentaenoic acid, which are precursors for prostaglandin synthesis, and have anti-inflammatory roles. Low concentrations of these PL could plausibly contribute to the increased risk of disease, reproductive failure and mortality in older cows. The bioavailability and metabolism of fatty acids may differ among supplements that are predominately neutral lipids, such as triacylglycerol-rich oils, and those bound to PL including pasture, whole or ground oilseeds and fish meal. Hepatic lipidosis can occur during the transition period if there is insufficient very-low density lipoproteins (**VLDL**) production in the liver to transport lipids into blood circulation. The PC are the primary PL of VLDL and are produced by two main pathways in the liver, the cytidine diphosphate-choline pathway that uses choline as a substrate, and the PE N-methyltransferase pathway that uses PE and methyl-donors as substrates. Co-supplementation strategies that target both pathways may increase PC production over a one-pathway supplementation strategy. The PIs are phosphoinositides precursors, which have broad physiological roles including regulating inflammatory processes and may offer targets for novel treatment and management of disease. Both the PI and PE are precursors to endocannabinoids, important regulators of energy metabolism, immune function and reproduction in mammals. Early findings on the endocannabinoid system in transition dairy cows yielded results that diverge from non-ruminant models. The PS expression on cytoplasmic membranes signals apoptosis, coagulation and contributes to sperm–oocyte recognition. As lipidomic diagnostics become increasingly available, understanding the metabolism of PL will continue to develop and promises to offer novel strategies for optimising cattle health and longevity.

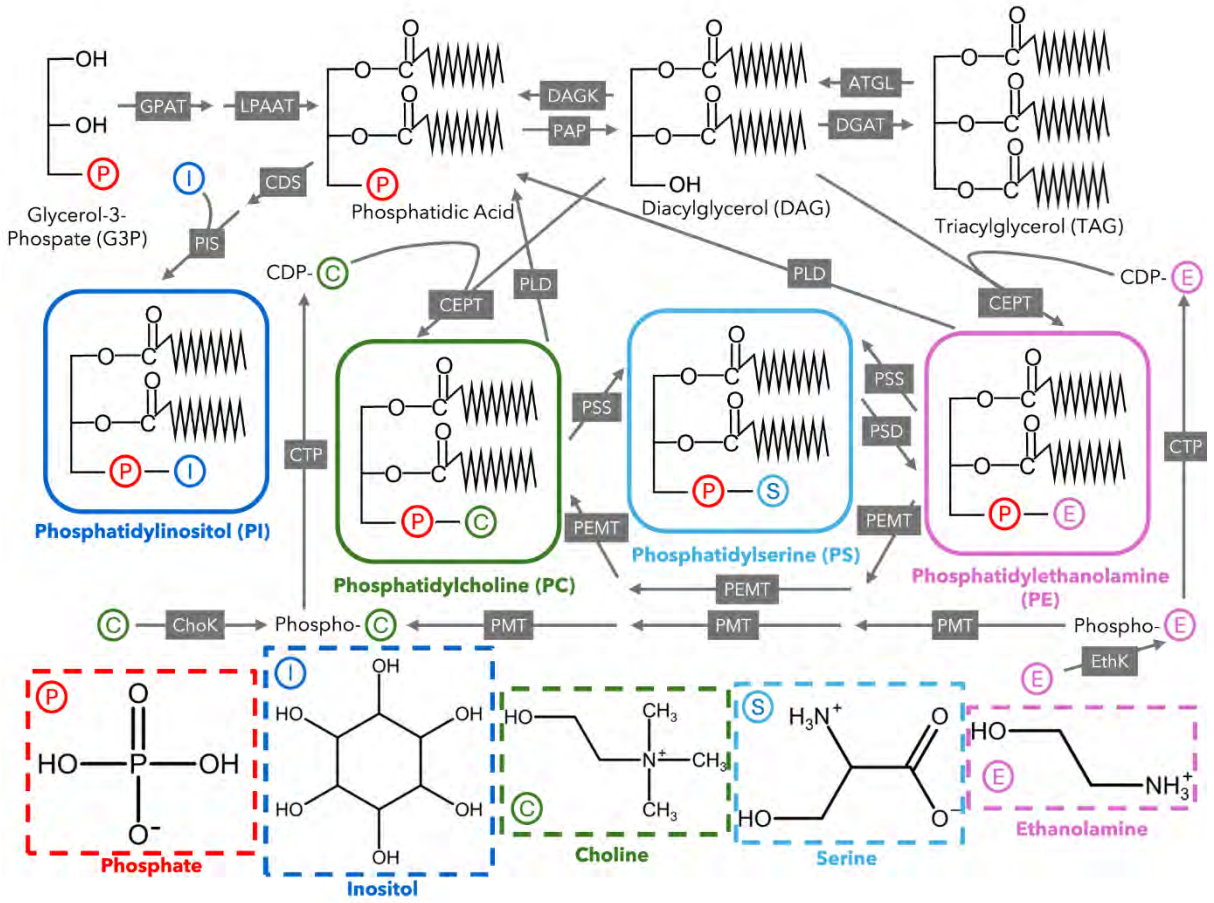

Figure 8.a: Glycerophospholipid metabolism depicting the most common pathways for production of phosphatidic acid, phosphatidylinositol (PI), phosphatidylcholine (PC), phosphatidylserine (PS) and phosphatidylethanolamine (PE). Enzymes are in grey boxes; fatty acyl chains are simplified as wavy chains esterified to the glycerol backbone. Phosphate and the specific head groups are depicted by the circled colored letter described in the bottom of the figure. Intracellular locations and lysophospholipids are not shown. ATGL – adipose triglyceride lipase; CDP – cytidine diphosphate; CDS – phosphatidate cytidylyltransferase; CEPT – choline/ethanolamine phosphotransferase; ChoK – choline kinase; CTP – cytidylyltransferase; DAGK – diacylglycerol kinase; DGAT – Diacylglycerol acyltransferases; EthK – ethanolamine kinase; GPAT – glycerol-3-phosphate acyltransferase; LPAAT – lysophosphatidic acid acyltransferases; PAP – phosphatidate phosphatase; PEMT – phosphatidylethanolamine-N-methyltransferase; PIS – phosphatidylinositol synthase; PLD – phospholipase D; PMT – Phosphoethanolamine methyltransferases; PSD – phosphatidylserine decarboxylase; PSS – phosphatidylserine synthase.

A large multisite lipidomic investigation of parity and aging in dairy cows

doi.org/10.3168/jds.2024-25578

Background

Efforts to optimize the longevity of dairy cows are hindered by the increased risk of adverse health events, culling, or dying on farm with increased parity. Lipidomics provides a platform to help identify important biomarkers and biological pathways associated with increased parity and associated aging.

Methods

A large multisite (15 pasture-based, 15 TMR farms) cross-sectional study collected plasma samples from nonlactating, late pregnant, dry cows (n = 696, ~27 d prepartum) and peak milk cows (n = 796, ~58 DIM) in a disproportionate stratified random sampling frame (parity: 0, 1, 2, >2 for dry cows; 1, 2, 3, >3 for peak milk cows). A total of 185 lipid species, comprising the lipids classes of phospholipids, sphingomyelins (SM) and triacylglycerols, were quantified in a targeted, liquid chromatography-MS approach. Dry and peak milk cohorts were analyzed separately throughout.

Findings

Variation in lipid profiles were mostly attributed to farm of origin (36%–41% of variation), with feeding system explaining 13% to 21% and parity explaining 6% to 9%, according to analysis of variance (**ANOVA**) simultaneous component analysis modelling. Multiple linear regression and orthogonal partial least squares (O-PLS) investigated the association of the lipid profile with age (d), whereas discriminant analysis compared first parity with >3 parity cows in O-PLS discriminant analysis, random forest, and support vector machine models. Rankings of the most important lipid species for each model type were compared. Phospholipids with 40 carbon atoms and 6 double bond equivalents (40:6) were consistently decreased with increasing parity and age across both dry and peak milk cohorts. These lipids most likely contained stearate (18:0) and docosahexaenoic acid (**DHA**, C22:6n-3), an n-3 fatty acid. Additionally, phospholipids with 40:5 and 38:6, lysophosphatidylcholine (17:0), SM(35:1), and SM(35:2) were commonly identified lipids that decreased in concentration with parity and age.

Implications

Docosahexaenoic acid has been associated with improved cattle health, reproduction, and milk production and quality. This study raises the hypothesis that reduced DHA levels in older cows may be an important factor increasing susceptibility to adverse health events, reduced reproductive performance, and herd removal. Studies that supplement DHA or its precursors can test this hypothesis and may be important in optimizing longevity of cows.

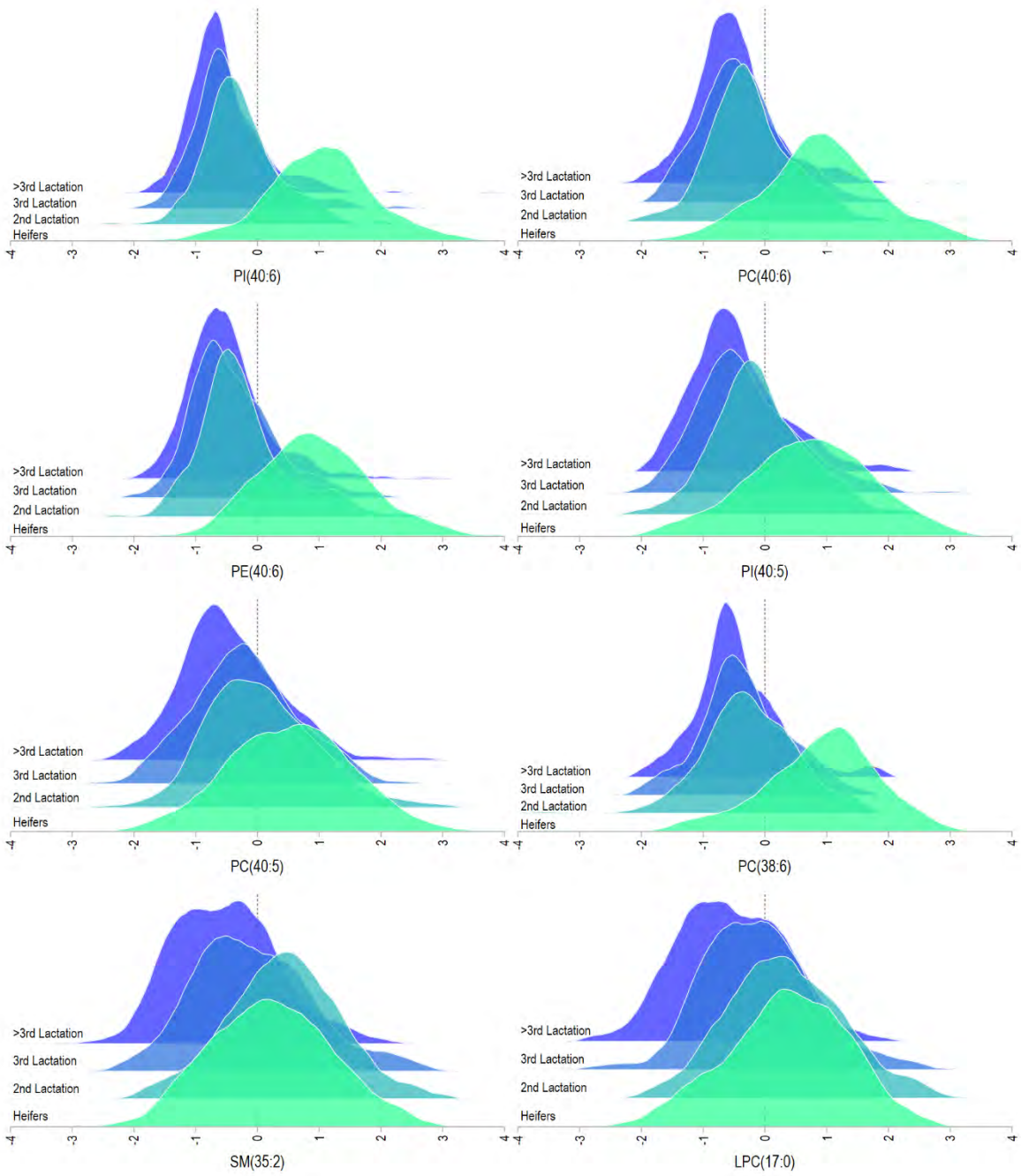

Figure 8.b: Specific lipid species concentrations stratified by parity groups. Presented lipids were identified as influential in differentiating parity and age. Data were centered and normalized on breed within farm (group) and cohorts of dry cows and peak milk cows. All figures contain both lactating and dry cow cohorts except PC(38:6) in the dry cohort only and LPC(17:0) and SM(35:2) in the peak milk cohort only.

Confinement and pasture-based dairy herds differ in plasma lipid profiles

doi.org/10.3168/jds.2025-26782

Authors

D.B. Sheedy, H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Hemsworth, D.E. Vincent, J.E. Pryce, I.J. Lean

Background

Dairy cow housing and management can be broadly described as either intensive confinement-based (**CONFINE**) or extensive pasture-based (**PASTURE**) systems. The diets between systems typically differ in their forage base, with CONFINE farms often utilizing maize silage in a TMR. Consequently, the lipid composition of diets differs between systems. The influence of housing system on blood lipidomics is currently unknown, but due to the bioactive role of lipids in influencing overall health and productivity, differences in diet may have consequences for reproduction, health, and aging of cows. The objective of this cross-sectional, multisite study was to investigate blood lipids and metabolites from cows in PASTURE and CONFINE systems, in the dry period (~27 d prepartum) and at peak milk (~58 DIM).

Methods

After exclusions, blood samples from 303 PASTURE and 398 CONFINE dry-period cows and 350 PAST and 431 CONFINE peak-milk cows from 15 PASTURE and 15 CONFINE farms were analyzed. A total of 185 lipid species (including glycerophospholipids, sphingomyelins, and triacylglycerols) were evaluated using targeted liquid chromatography-MS, as were 11 routinely measured metabolites. Dry and peak-milk cohorts were analyzed separately throughout. Lipids and metabolites associated with housing system were selected using a variable stabilization approach that was achieved by calculating the frequency of inclusion in categorical (housing system) penalized models using bootstrapping. Variables were retained if inclusion frequency exceeded a false-positive threshold. Five different statistical models were used with variable stabilization.

Findings

Dry cows in CONFINE systems had decreased globulin, urea, and glycerophospholipids associated with n-3 fatty acids. The highest total inclusion rates in the dry cohort were phosphatidylcholine (**PC**; 36:5), which mostly comprises palmitic acid (C16:0) and eicosapentaenoic acid (**EPA**; 20:5n-3), then phosphatidylethanolamine (**PE**; 38:5, 16:0/22:5n-3 or 18:0/EPA) and PC(34:3; 16:0/18:3 α-linolenic acid [**ALA**]). No lipids were increased in more than one stabilized model in CONFINE dry cows. Peak-milk CONFINE cows had increased glycerophospholipids associated with n-6 fatty acids. The highest total inclusion-rate lipids in the peak-milk cohort were phosphatidylinositol (PI; 38:3; 18:0/20:3n-6 dihomo-γ-linolenic acid), PC(34:2; 16:0/18:2 linoleic acid [**LA**]), PC(40:7; 18:2/22:5n-6), PC(34:1; 16:0/18:1), and PE(34:2; 16:0/LA). The CONFINE peak-milk cows also had decreased PC(34:3; 16:0/ALA).

Implications

This study identified specific lipids that were strongly associated with housing systems, findings that have not been reported elsewhere. Given the important biological functions of omega fatty acids, the pattern of glycerophospholipids with increased n-6 and decreased n-3 in CONFINE cows may indicate housing systems create different risk profiles for reproduction, health, and aging.

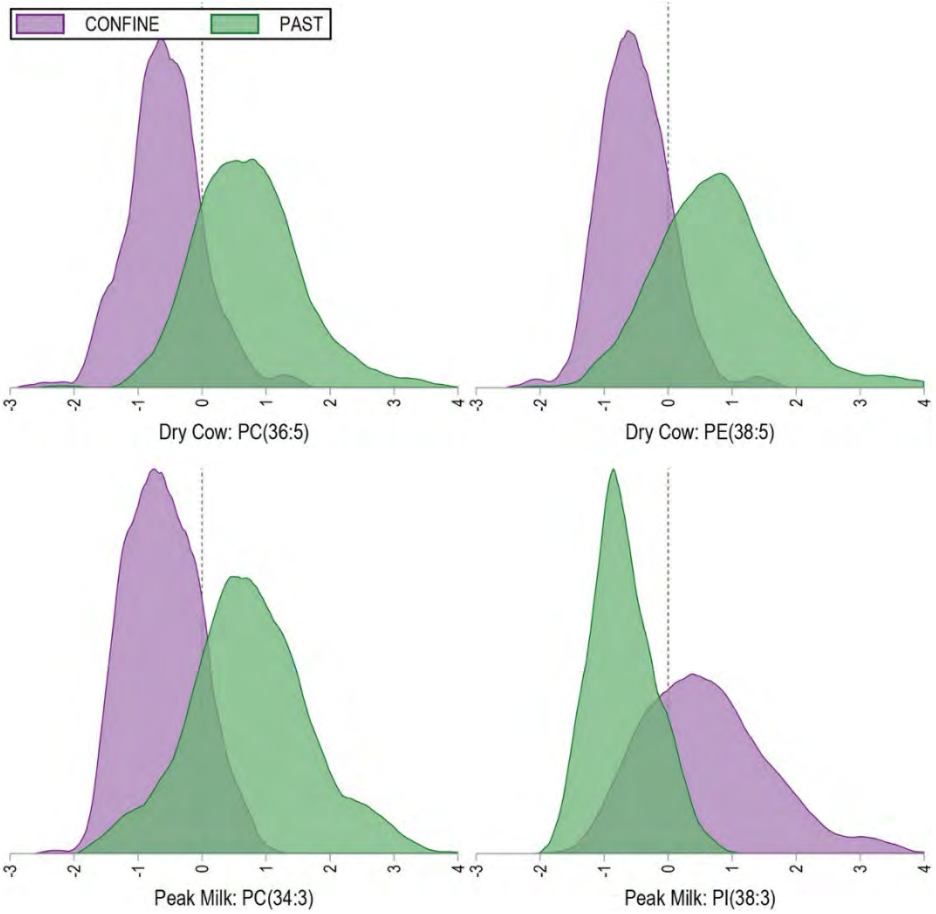

Figure 8.c: Kernel density estimation functions for the 2 most selected compounds associated with production system, from each cohort. PC(36:5), PE(38:5) and PC(34:3) are phospholipids associated with omega-3 fatty acids and PI(38:3) is associated with an omega-6 fatty acid. Compounds are standardized with a mean zero and SD of 1. CONFINE = confinement-based dairy system, according to lactating herd housing; PASTURE = pasture-based dairy system, according to lactating herd. Vertical dashed lines indicate the standardized mean value of zero.

A large, multisite investigation into the lipidomics of survival in dairy cows

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Authors

D.B. Sheedy, H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Hemsworth, D.E. Vincent, J.E. Pryce, I.J. Lean

Background

Identifying physiological determinants of dairy cow survival and their potential modulation by parity may reveal opportunities to improve herd health and longevity. This multisite, prospective, observational study investigated culling and mortality hazards using targeted lipidomic and standard metabolite assays.

Methods

Blood samples, stratified by parity, were collected from 2 cow cohorts (1) dry and (2) peak-milk, from across 29 commercial Australian farms (14 pasture-based, 15 confinement-based). There were 717 nonlactating, late-pregnant, dry cows (~27 d prepartum) and 794 peak-milk cows (~58 DIM) sampled. A total of 186 lipid species (including glycerophospholipids, sphingomyelin, and triacylglycerols) and 11 routinely measured metabolites were evaluated. Sample cows were followed for an average of 693 d and exit reasons recorded. Competing risk survival models were used to estimate the cumulative incidence of culling and mortality by parity and cohort. Blood analytes were autoscaled within cohort and farm, controlling for farm-level effect on metabolites. Survival analysis was performed using an adaptive least absolute shrinkage and selection operator (**LASSO**) Cox full likelihood model that explored associations among blood analytes and hazards of removal, with a shared frailty of farm (accounting for farm-level baseline hazards), and 2 removal outcomes considered: culling (censoring: death, cull from farm accident, end of follow-up) and mortality (censoring: cull, death from farm accident, end of follow-up). Separate models were used to estimate survival outcome by cohort and parity groupings (first, second and third, and greater than third) or with parity as a categorical covariate. Due to high correlations, analyte data were reduced to 25 clusters using Ward's hierarchical clustering criterion. Bootstrapping of the LASSO variable selection procedure identified clusters with high selection frequency for use in the final model. The hazards and cumulative incidence of culling and mortality increased with parity.

Findings

Glycerophospholipids with very-long-chain n-3 fatty acids were associated with reduced hazards of culling in parity 1 peak-milk cows (0.39 hazard ratio [**HR**]), whereas glycerophospholipids with n-3 α-linolenic acid were associated with reduced hazards of culling in parity >3 peak-milk cows (0.18 HR). Sphingomyelin with >C18 fatty acyl chains was associated with increased hazards of culling in parity >3 peak-milk cows (2.02 HR). Clusters containing the routinely evaluated analytes albumin, globulin, urea, magnesium, glucose, triglycerides, β-hydroxybutyric acid, bilirubin, and nonesterified fatty acid were associated with culling and mortality, consistent with their roles in health and reproduction. Lipids collected from dry cows were poor predictors of survival.

Implications

Many novel plasma lipid targets for future research into survival of cattle were identified, with proinflammatory lipid profiles associated with increased risk of culling and mortality.

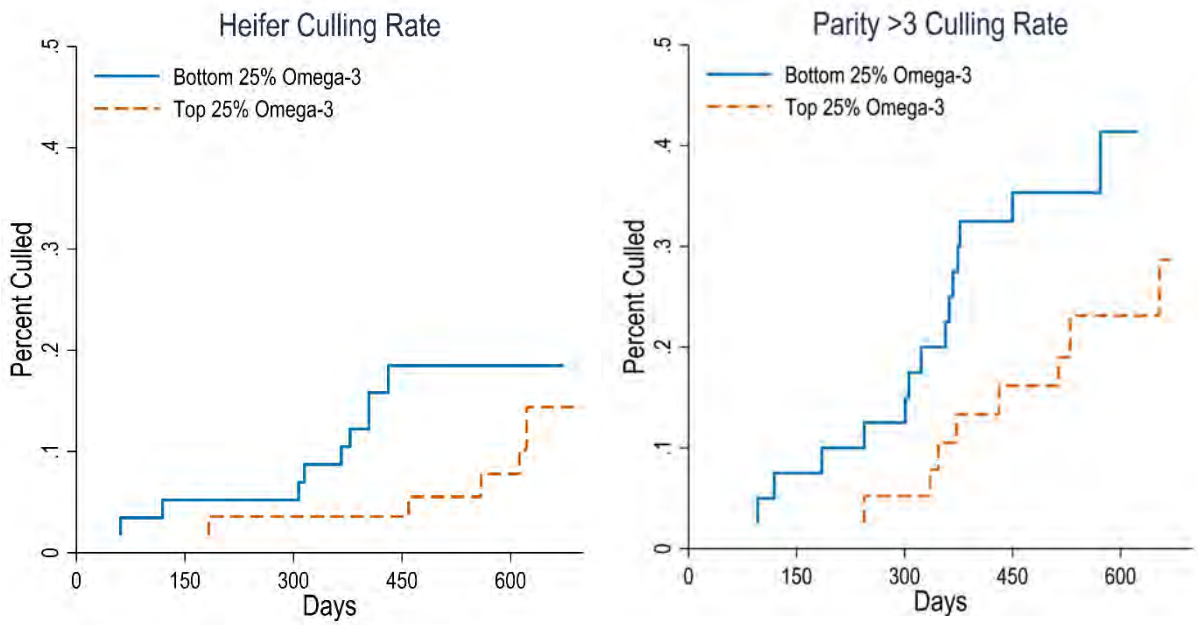

Figure 8.d: Predicted survival of heifers (left) and parity >3 (right) with low (blue, solid line) or high (orange, dashed line) plasma concentrations of phospholipids associated with omega-3 fatty acids. Cows with high omega-3 blood concentrations were predicted to survive in the herd by around 300 days compared to cows with low levels.

Do housed cows have greater metabolic-age than pasture-based cows based on lipid profiles?

Submitted to the Journal of Dairy Science Communications

Authors

D.B. Sheedy, H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Pryce, I.J. Lean

Background

Metabolic clock models use biomarkers to predict the metabolic-age of a subject and may infer information regarding their health. Plasma glycerophospholipids that contain omega-3 fatty acids are reduced with increased age of dairy cows and reduced in cows from TMR-fed, housed systems (**HOUSED**) compared to cows from extensive, pasture-based systems (**PASTURE**). We hypothesized that a lipid-biomarker metabolic clock model that was trained exclusively on either HOUSED or PASTURE cows would produce systematically biased estimates on test datasets comprised of cows from the alternate feeding system. If this were true, the results could infer different metabolic status between feeding systems.

Methods

Data from a previous study of 794 peak-milk cows (~58 DIM), stratified by parity, were sampled from 29 Australian farms (14 PASTURE, 15 HOUSED). A total of 186 lipid species (including glycerophospholipids, sphingomyelin, and triacylglycerols) were quantified. A prediction-oriented, 'cubist' machine learning algorithm was applied using training:test data splits of HOUSED:PASTURE and PASTURE:HOUSED.

Findings

The model trained on HOUSED cows had no bias in the training set (0.0 d, 95% CI: -2.3, 2.3), but the predicted age of PASTURE cows was 627 d younger than their true age (95%CI: -683, -571). The model trained on PASTURE-cows had no bias in the training set (0.0 d, 95%CI: -22.1, 22.1), but the predicted age of HOUSED cows was 175 d greater than their true age (95%CI: 142, 208). The direction and magnitude of systematic bias was consistent when models were restricted to 10 pre-screened predictor lipids that were mutually associated with age in both systems. Implications

Implications

Collectively, results indicated that HOUSED cows had greater metabolic-age than PASTURE cows of the same chronological age. A secondary conclusion reinforced caution regarding the extrapolation of machine-learning models: predictions may be highly precise within the training domain (e.g. Root mean square error = 24 d for HOUSED-trained models), yet substantially biased when applied to external populations. Conversely, consistent directional bias across models provided inferential evidence of biologically meaningful differences between production systems. The difference in metabolic-age may have implications for cow health and longevity across systems of dairying but has yet to be explored.

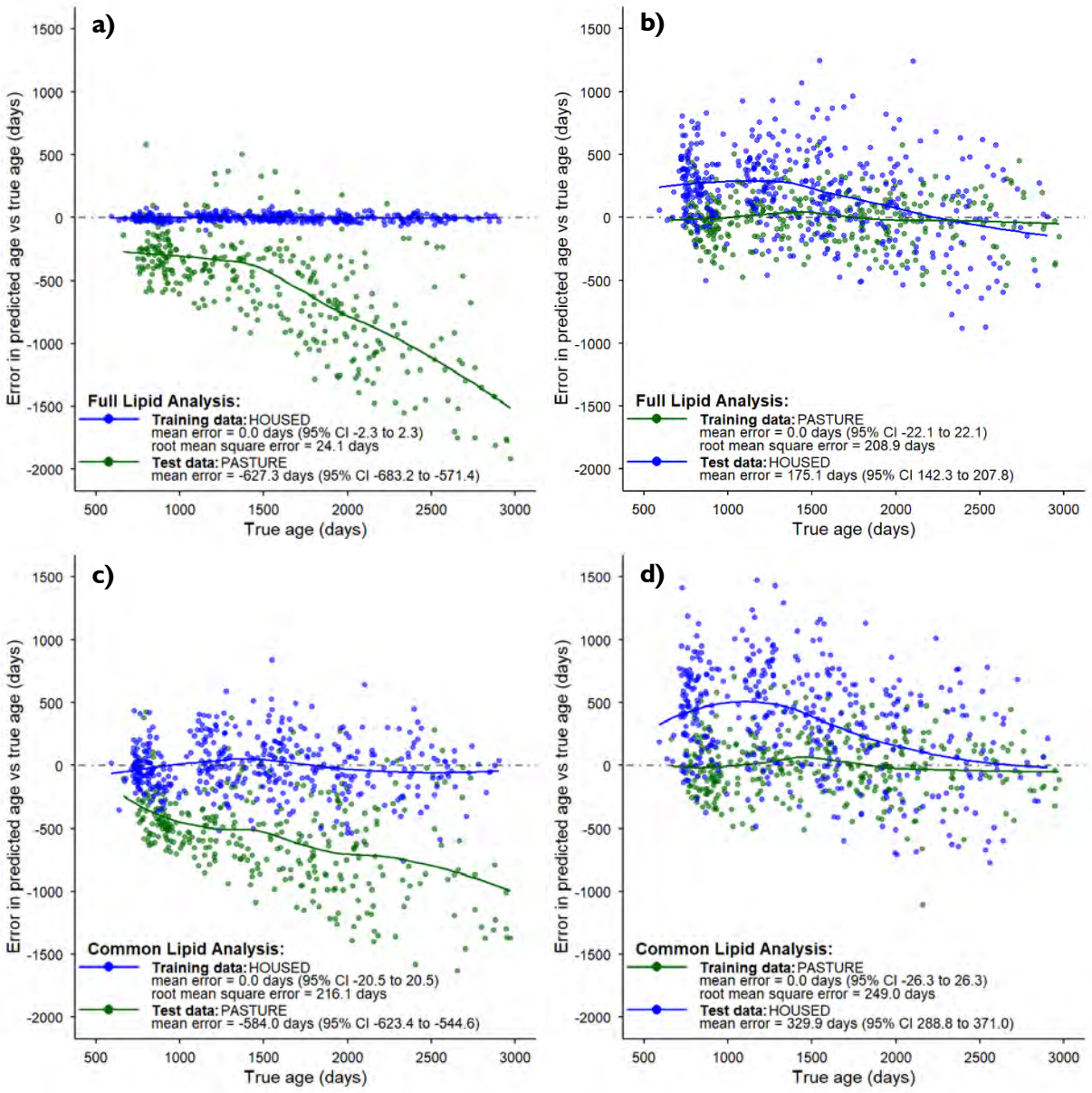

Figure 8.e: Plots of the residual error in the prediction of cow age (d) against their chronological (true) age (d) following a machine-learning metabolic clock model that utilized plasma lipid data. Models were trained exclusively on cows from either TMR, housed-based (HOUSED) or pasture-based (PASTURE) systems and tested on cows from the other system. Data were either a full lipidomic panel of 186 lipids (full lipid analysis) or models were first trained separately on cows from either system with 10 mutually important lipids selected for model training (common lipid analysis). The mean error is an indication of systematic bias in prediction.. Lines are included to indicate the average predication error at a given true age. Full lipid analysis; a) HOUSED:PASTURE, b) PASTURE:HOUSED; Common lipid analysis; c) HOUSED:PASTURE, d) PASTURE:HOUSED.

Subproject P2a Cow Longevity – v. Housing System Series

Background

An important consideration for cow longevity is the structural differences of the farming systems in which cows are managed. There are two broad dairy production housing systems: extensive, pasture-based systems and intensive, housing-based systems. The Australian dairy industry is currently undergoing a rapid transition from milk being produced almost exclusively from pasture-based farms to confinement-based systems now contributing approximately 20% of the national milk production, with expectations that 35-40% of milk will be produced from cows in confinement farms by 2030. This transition is being driven largely by motivations to expand business enterprises, an improved ability to mitigate extreme climatic events, and the decreasing availability of irrigation water. As the transition from pasture to confinement systems is recent and ongoing, there was a unique research opportunity to evaluate longevity-associated performance outcomes and biological differences between the systems of dairying in a contemporary setting.

Reproduction, mastitis and lameness in housed and pasture-based systems: associations with parity

Accepted to the Journal of Dairy Science

Authors

D.B. Sheedy, H.M. Golder, S.C. Garcia, A.K.G. Lean, I.J. Lean

Background

Dairy farming systems can be broadly classified as either housing-based dairy (**HOUSED**) or pasture-based dairy (**PASTURE**) systems. System-level differences in diet, housing, management, and milk production may influence health and reproduction of cows and the interaction of parity with these outcomes.

Methods

This multi-site prospective, observational study investigated risks of pregnancy, mastitis, and lameness for different parities and housing systems (HOUSED and PASTURE). There were 15 HOUSED and 14 PASTURE farms enrolled, with health, reproduction, and production data collected from each farm between Feb 2022 to Jan 2026. Linear regression, logistic regression, and Weibull parametric survival models were used to assess the effects of parity, production system, and season on measures of pregnancy, mastitis and lameness. Robust sandwich estimators controlled within-farm correlation.

Findings

The 100 day in-calf rate (**100DICR**, logistic regression) was not significantly different between PASTURE (34.0%; 95%CI: 28.0, 40.0) and HOUSED (32.9%; 95%CI: 28.9, 36.9) farms. The hazards of pregnancy (**HPREG**) assessed with a Weibull model were not different between PASTURE (1.04 HR, 95%CI: 0.82, 1.25) and HOUSED [1.08 hazards ratio (**HR**); 95%CI: 0.70, 1.45] farms. There was a monotonic decrease in the 100DICR and HPREG with increasing parity regardless of system. Cows breeding during Summer or Spring in PASTURE farms and Spring in HOUSED farms had reduced reproductive efficiency compared to other seasons. The effect of season was greater in PASTURE compared to HOUSED. There were 34,890 mastitis events across 129,039 lactation records. The crude incident rate of mastitis in HOUSED was 42.6 per 100 cows per 365 cow-days (95%CI: 41.9, 43.3) and 34.9 (95%CI: 33.7, 36.3) in PASTURE. The hazard of was numerically

greater in HOUSED (2.85 HR, 95%CI: 1.10, 4.61) compared to PASTURE (2.13 HR, 95%CI: 1.62, 2.64) farms. The hazard of mastitis increased monotonically with parity. There were 9,495 cases of lameness across 125,362 lactation records. The crude incident rate was 8.75 per 100 cows per 365 cow-days (95%CI: 8.43, 9.07) in HOUSED and 10.7 (95%CI: 9.97, 11.4) in PASTURE. The hazard of lameness was similar in HOUSED (1.04 HR, 95%CI: 0.24, 1.84) and PASTURE (1.19 HR, 95%CI: 0.92, 1.44) farms. The hazard of lameness were unchanged in the first three (PASTURE) and two (HOUSED) parities but increased with parity thereafter.

Implications

The association of increasing parity was far greater on health and reproductive outcomes than that of housing system. These results suggest that to support cow health and longevity we must address the impact of increased parity on health and reproduction of cows.

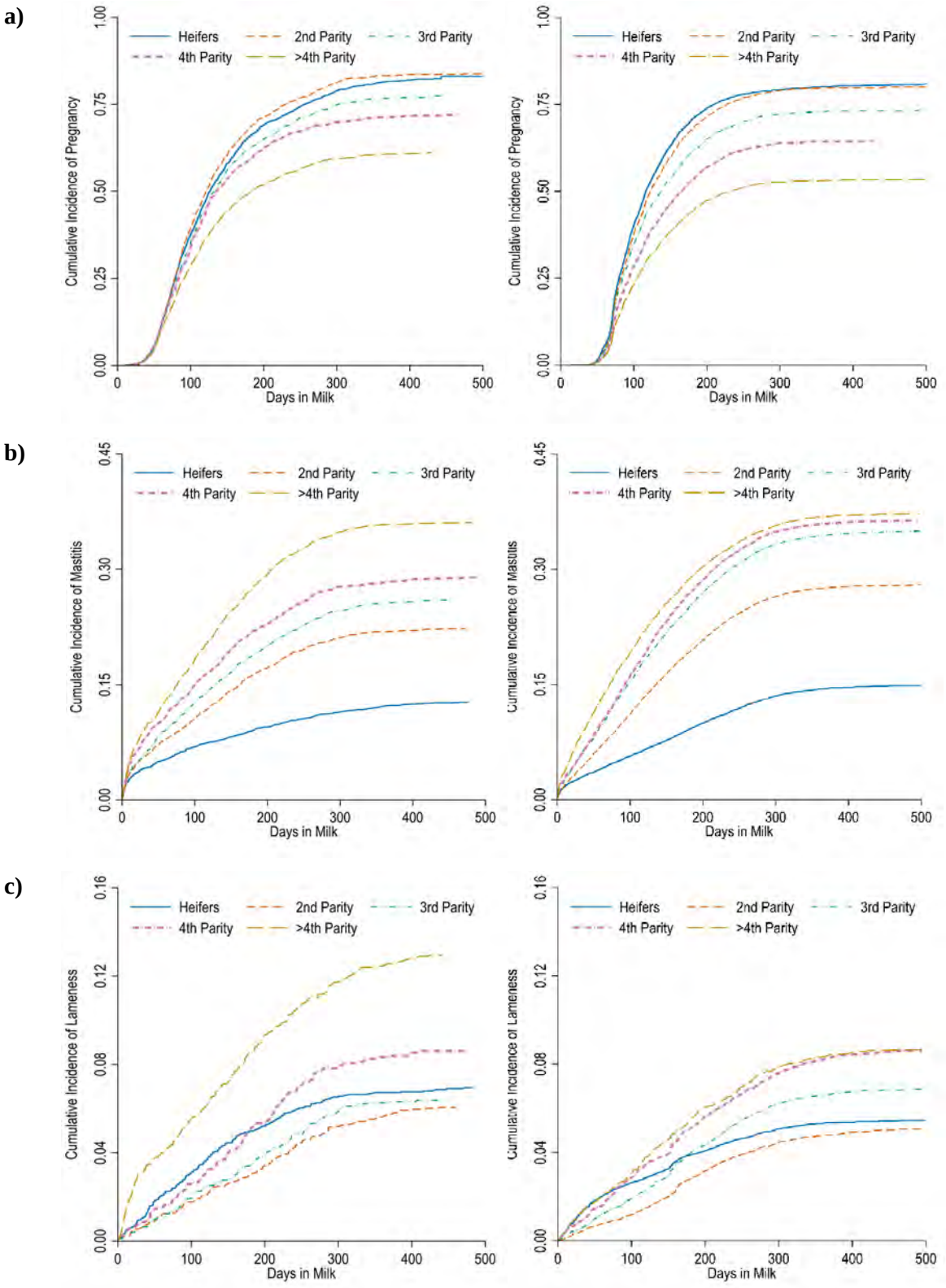

Figure 9.a: Competitive risk models showing the cumulative incidence of a) pregnancy, b) mastitis, and c) lameness by parity in pasture-based systems (left) and housing-based systems (right).

Effects of a bacterial and enzymatic product on physical and chemical properties of bedding and cow preference in compost bedded pack barns

In review

Authors

A.K.G. Lean, H.M. Golder, J.C. Quinn, I.J. Lean, D.B. Sheedy, A.J. Gunn

Background

Compost-bedded pack (**CBP**) barns are increasingly used to house cows worldwide. These are loose housing facilities where an organic bedding base is mixed daily to remove faeces and urine from the bedding surface and oxygenate the pack, allowing composting to occur. These facilities face challenges in maintaining low moisture percentages within the bedding to ensure cow comfort and good udder health. We aimed to investigate effective methods of managing CBP barns to maintain compost moisture and temperature at the recommended levels and assess the effectiveness of a bacterial and enzymatic treatment on bedding characteristics and cows' preference.

Methods

A five-week multisite replicated exposure study was conducted on 40 areas of five commercial CBP barns in Australia from late autumn to early spring in 2023 and 2024. The packs were randomly divided into alternating control (n = 20) and treatment (n = 20) areas. All areas were monitored over one week to obtain a baseline before the treatment was applied in the 20 treatment areas at the start of weeks two and four, as per manufacturer recommendations. Compost temperature (**°C**), area per cow per study area (log m^2/cow), and aerial ammonia concentrations (**ppm**) were assessed as outcome variables every 2 ± 1 day across all five farms. Three farms had additional outcome variables of compost moisture percentage and carbon to nitrogen ratio assessed weekly. Local weather conditions were measured daily, while the cultivation rate of the pack (m^2/min) was recorded as a covariable for three farms. Multilevel mixed models were used to analyse the effects of treatment, covariables, and the interaction of treatment over time.

Findings

There was no effect of treatment on bedding conditions. Aerial ammonia concentrations were only detected on six days. Slowing the cultivation rate (m^2/min) of the pack was effective at increasing compost temperature.

Implications

The product tested was not effective at improving bedding conditions nor did it change cow preference in the packs studied. Management of cultivation rate is important for maintaining bedding conditions. Minimal ammonia detection is a positive finding for human and cattle health in these barns.

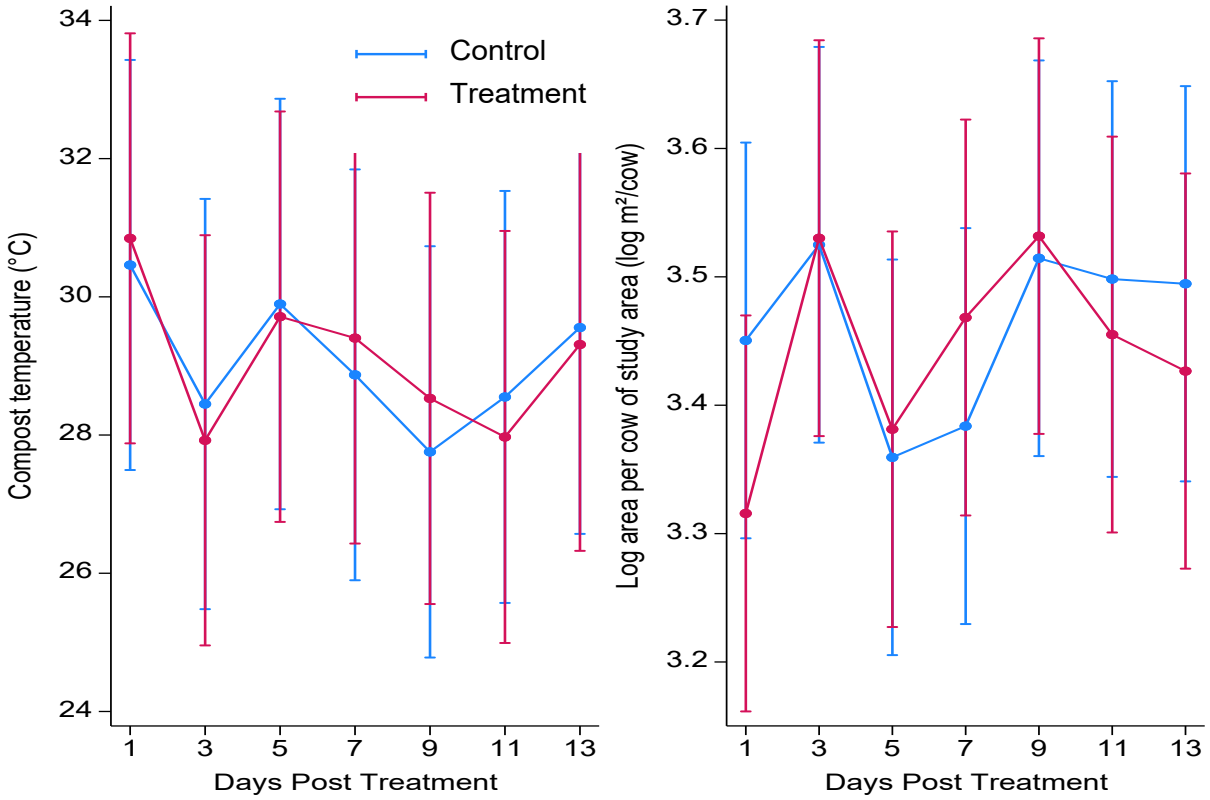

Figure 9.b: Margins plots of the 5-farm models of (A) compost temperature (°C) over time and (B) log area per cow per study area (log_{10} m^2/cow) with standard error of the mean bars

Subproject P2b – Early Alert Systems

Background

We identified a number of information streams from existing activities at Elizabeth Macarthur Agricultural Institute (**EMAI**), Department of Primary Industries and Regional Development (**DPIRD**), University of Sydney and Scibus that can be directed towards farms and their advisors in real time to ensure that diseases and risks, whether these be animal, plant or environmental can be addressed in more timely and effective manners. The use of targeted alerts, coupled with timely advice links will enable farms to more effectively reduce risks to pastures and crops, and cattle to ensure greater productivity, sustainability and health.

Outcomes

The following have been identified as targets for early alert systems; Bovine ephemeral fever, fall army worm and facial eczema. There is now a proposal for Terms of Reference for an industry panel to work on means to provide a rigorous structure to deliver early alerts (NSW DPI July 2025) and agreement from Dairy NSW committee (30/10/2024) to assist in dissemination of alerts.

For ephemeral fever the following web site has been developed as a direct outcome of the Dairy UP initiative to provide information on cases in order to assist producers to make decisions around vaccination, treatments and risk of the disease. https://www.dpi.nsw.gov.au/about-us/services/laboratory-services/veterinary/bovine-ephemeral-fever

Implications

A lack of specific budget for this project has impeded its progress to some degree. However, good will resulting in considerable in -kind contributions from NSW DPIRD and Scibus have resulted in outcomes. There is potential to develop this project further and a communications strategy would enhance the potential for delivery of benefits to animals. farmers and communities.

Subproject P2c – Milk as a Diagnostic Fluid

Background

There are a large number of diagnostic tests currently available through Government and commercial laboratories or through advanced milking platforms. The objective of the review will be to evaluate the diagnostic value of tests at an individual cow and herd level. Costs of diagnostic tools and the value of these to producers will be assessed in order to enhance existing herd recording systems or to develop novel packages for herd monitoring.

Editorial: Milk as a Diagnostic Fluid

doi.org/10.1111/avj.13299

Authors

I.J. Lean, R. Zadocks, B. Brito, H.M. Golder

Editorial

Over 40 years there have been profound changes to the Australian dairy production environment. The number of farms decreased from 21,989 in 1980 to 5055 in 2020, milk production per cow increased from 2888 L/cow per year or 1.9 million cows producing 5.49 million L per year, to 6311 L/cow per year or 8.8 million L from 1.4 million cows. Many dairy farms represent assets valued in the $10 to $100 million or more. The average herd has increased from 85 to 274 cows. Consequently, farm management has less time to engage with the individual cow. These changes influence the delivery of veterinary services as the individual cow now represents a much lower proportion of the enterprise asset value. However, herd health and productivity are critical to an enterprise and farmers are committed to stewardship of their cattle. The challenge for the veterinary profession is to deliver cost-effective services that identify, monitor, and mitigate risks to herd health and productivity. Such services must be designed to deliver better outcomes with greater labour efficiency. In this series of reviews, we evaluate the value of bulk tank milk which provides a readily available and contemporary indicator of herd status of health and production and, where appropriate, compare the value of bulk milk testing to that of individual cow testing, to determine the mastitis, viral, and metabolic status of herds. We provide quantitative and qualitative reviews of tests that may do this. We also note two coincident and valuable scoping reviews of this area, one of which includes data on the value of bulk tank milk for parasite evaluation.

Bulk milk somatic cell counts are routinely utilised by processors and veterinary advisors to assess milk quality and udder health. Because this assessment does not capture cows with clinical mastitis, diagnostics at the cow level may also be needed to manage udder health. Additional markers of inflammation or the humoral immune response are primarily available at cow level, except for antibody testing for *Mycoplasma bovis,* which can be conducted at bulk milk level to support biosecurity efforts. Elevated somatic cell counts are primarily due to intramammary infections, and its causative agents, including those with antimicrobial resistance, can be detected through culture or polymerase chain reaction (**PCR**). Specificity of PCR for contagious pathogens (*Streptococcus agalactiae, Mycoplasma bovis*) is high (0.90) but sensitivity is variable (0.15–0.99) unless repeated bulk milk testing or cow-level testing is used.

For pathogens that may be cow-derived as well as environmental (*Staphylococcus aureus, Streptococcus dysgalactiae, Streptococcus uberis*), sensitivity of bulk milk testing is low (<30%). New technologies such as matrix-assisted laser desorption/ionization time of flight (**MALDI-TOF**) or loop-

mediated isothermal amplification (**LAMP**) offer new insights into intramammary infection but are not applied at the bulk milk level yet.

Bulk tank milk was very effectively used in Australia to evaluate herd status with an enzyme-linked immunosorbent assay (**ELISA**) test to eradicate bovine leukaemia virus. Interpreting results from bulk milk tests requires careful consideration of complexities, including the test target, prior virus exposure including vaccination, duration of antibodies in milk or virus shedding, and other relevant factors. For direct detection of pathogens or the immune or metabolic responses to pathogens, increased herd size and prevalence of organisms or response has significant testing implications. Specifically, as herd size increases the probability of the assay detecting the presence decreases due to potential for dilution.

Studies identified the limits of detection for different assays and estimates for herd sensitivity (0.44–0.97) and herd specificity (0.42–1.00) for pestivirus ELISA. The use of PCR to detect persistently infected animals detected up to 1 animal in 1000, however, other studies indicated a lower sensitivity. Bulk tank milk can also be used to detect bovine alphaherpesvirus-1 (**BHV-1**) which is prevalent in Australia, while the use during a foreign animal disease outbreak is arguable.

There are valuable bulk tank milk assays that can monitor the herd nutritional status. The bulk tank milk urea and protein content provide useful indications of herd nutrition using routine milk testing from milk processors. These tests provide indicators that encourage further investigations of nutritional influences on herd fertility but are unlikely to provide strong diagnostic value. The fat to protein ratio has a high specificity, but poor sensitivity for the detection of fibre insufficiency and acidosis on an individual cow basis and is useful as an alert to evaluate other indicators of acidosis in a herd.

Integrating herd recording demographic information with Fourier transformed mid-infrared spectra (**FT-MIR**) can provide tests that are useful to identify cows with metabolic disorders and these tests will become more available in Australia. Selenium, zinc, β-carotene, and vitamin E status of the herd can be determined using bulk tank milk and could be combined to monitor herds.

Bulk tank milk is an under-utilised resource that can be used to improve the health and productivity of dairy herds. There is considerable potential to increase the availability and adoption of bulk tank milk as part of an efficient integrated farm service. Some of the tests readily available and reported on a daily basis include milk volume, somatic cell counts, fat, protein, and milk urea. More can be done with these data to evaluate herds. However, there appears to be increased scope for testing for milk minerals, vitamins, and assays for mastitis and viral pathogens. These assays can provide cost-effective rapid means of monitoring herds and have the potential to be integrated with statistical monitoring methods to automate detection of changes in herd status and equip veterinary services with new tools to assist farms.

Milk as an indicator of dietary imbalance

doi.org/10.1111/avj.13294

Authors

I.J. Lean, H.M. Golder

Background

Milk provides a readily available diagnostic fluid collected daily or more frequently on an individual animal or herd basis. Milk, as an aggregated sample in bulk tank milk (**BTM**) represents the status of a herd instead of a single animal. In this review, we examine the potential for milk to predict risks to efficient production, reproductive success, and health on the individual cow and herd level.

Findings

For many conditions related to disorders of metabolism including hyperlipdaemia and ketonaemia, improved individual cow milk testing may allow a temporally useful detection of metabolic disorder that can target intervention. However, the extension of these tests to the BTM is made more difficult by the tight temporal clustering of disorder to early lactation and the consequent mixing of cows at even moderately different stages of lactation. Integrating herd recording demographic information with Fourier-transformed mid-infrared spectra (FT-MIR) can provide tests that are useful to identify cows with metabolic disorders. The interpretation of BTM urea and protein content provides useful indications of herd nutrition. These may provide indicators that encourage further investigations of nutritional influences on herd fertility but are unlikely to provide strong diagnostic value. The fat-to-protein ratio has a high specificity, but poor sensitivity for detection of fibre insufficiency and acidosis on an individual cow basis. Selenium, zinc, β-carotene, and vitamin E status of the herd can be determined using BTM.

Implications

There appears to be increasing potential for the use of milk as a diagnostic fluid as more in-parlour tests become available for individual cows. However, the BTM appears to have under-utilised potential for herd monitoring.

Table 11.a: Details tests that are available to evaluate metabolic states including energy deficiency, and fibre sufficiency. If available, information on the diagnostic accuracy, sensitivity and specificity of the tests is provided.

Condition	Test	Index test	Test accuracy	Sensitivity	Specificity
Hyperlipidemia	Blood FFA < 0.6 mmol/L	Milk C18:1 cis-9	0.79	0.75	0.79
Hyperketonemia	Blood BHB (≥ 1.2 mmol/L)	Milk fat C18:1 cis-9-to C15:0 ratio of ≤40	0.82	0.38	0.95
Hyperlipdaemia	Blood FFA ≥ 0.95	Milk fatty acids (C17:0 to 15:0)	0.84	0.72	0.90
Hyperlipdaemia	Blood BHB mmol/L ≥ 0.7	Milk fatty acids, days in milk, lactation number, milk production	77.4	77.1	77.4
Hyperketonemia	Blood BHB (1.0 to 1.4 mmol/L)	Ketotest	0.80	0.86	0.72
Hyperketolactia	Milk acetoacetate (>0.15 mmol/L)	Fourier transformed infra-red spectrometry	0.80	0.86	0.72
Hyperketonemia	Blood BHB (≥ 1.2 mmol/L)	Fourier-transformed infra-red	77.9	84.8	77.1
Fibre sufficiency	Milk fat: Protein ratio < 1.02		0.72	0.54	0.81
Metabolic state	Balanced, unbalanced at 9 days in milk	Demographic data, milk yield and components, FT-MIR for BHB and milk fatty acids	0.85		

Milk as diagnostic fluid for udder health management

doi.org/10.1111/avj.13290

Authors

S. Rowe, J.K. House, R.N. Zadoks

Background

Mastitis is the major disease affecting milk production of dairy cattle, and milk is an obvious substrate for the detection of both the inflammation and its causative infectious agents at quarter, cow, or herd levels. In this review, we examine the use of milk to detect inflammation based on somatic cell count (**SCC**) and other biomarkers, and for the detection of mastitis pathogens through culture-based and culture-free methods.

Findings

The use of SCC at a cow or bulk milk level to guide udder health management in lactation is well-established, and SCC is increasingly used to guide selective dry cow treatment. Other markers of inflammation include electrical conductivity, which is used commercially, and markers of disease severity such as acute phase proteins but are not pathogen-specific. Some pathogen-specific markers based on humoral immune responses are available, but their value in udder health management is largely untested. Commercial pathogen detection is based on culture or polymerase chain reaction, with other tests, for example, loop-mediated isothermal amplification or 16S ribosomal ribonucleic acid microbiome analysis still at the research or development stage. Matrix-assisted laser desorption ionisation time of flight (**MALDI-TOF**) is increasingly used for the identification of cultured organisms whilst application directly to milk needs further development. Details of test sensitivity, specificity, and use of the various technologies may differ between quarter, cow, and bulk milk applications.

Implications

There is a growing array of diagnostic assays that can be used to detect markers of inflammation or infection in milk. The value of some of these methods in on-farm udder health improvement programs is yet to be demonstrated whilst methods with proven value may be underutilised.

Milk as a diagnostic fluid to monitor viral diseases in dairy cattle -

doi.org/10.1111/avj.13293

Authors

B. Brito, P. Hick

Background

Infectious viral diseases in dairy cattle have substantial implications for milk production, quality and overall animal health. Diagnostic tools providing reliable results are crucial for effective disease control at the farm and industry level. Pooled or bulk tank milk (BTM) can be used as a cost-effective aggregate sample to assess herd disease status in dairy farms.

Findings

Detection of pathogens or specific antibodies in milk can be used for monitoring endemic diseases within-farm, region or country-level disease surveillance and to make informed decisions on farm management. The suitability of assays applied to pooled milk samples relies on validation data of fit-for-purpose tests to design an optimal testing strategy. Diverse approaches and variable scope of studies determining test accuracy need to be critically appraised before sourcing the parameters to design sampling strategies and interpreting surveys. Determining if BTM or pooled milk is the best approach for a disease management programme should carefully consider several aspects that will impact the accuracy and interpretation, for example, the size of the lactating herd, the risk of infection in the lactating and non-lactating groups, the expected within-herd prevalence, the duration of infection, the duration and concentration of antibodies in milk and use of vaccination.

Implications

There are examples of tests on BTM samples providing efficient assessments of the herd disease status and supporting disease control programmes for viral diseases. However, challenges arise in pooled milk testing due to the need for accurate estimates of the imperfect sensitivity and specificity of the assays. Integration of new biotechnologies could enhance multiplexing and data interpretation for comprehensive surveillance. The development of highly sensitive assays is necessary to meet the demands of larger dairy herds and improve disease detection and assessment.

Subproject P2d Heat Stress

Effects of changes in total mixed ration nutrition and maximum temperature humidity index (THI) on dairy cow milk yield

Authors

A.K.G. Lean, A.J. Gunn, H.M. Golder, J.C. Quinn, D.B. Sheedy, I.J. Lean, P.C.Thomson

Background

There is a general understanding that changes in the nutrient concentrations of dairy cows' diet have delayed effects on production. Current nutrition models do not take these into account. There have been limited attempts to quantify these effects on milk yield. This study investigates the effects of changes in nutrition on the milk yield of housed dairy cows and the effects of changes in the weekly average maximum temperature-humidity index (**THI**). Methods

Methods

A multisite time series investigated effects of nutritional changes in TMR diets and THI on milk yield of dairy cows. Weekly diet samples were collected and tested from 8 farms for a total of 9 strings or herds for an average of 58 weeks. Nutrient components of the diet examined for effects on milk yield were neutral detergent fibre (**NDF**), lignin, crude fat, crude protein (**CP**), neutral detergent insoluble crude protein (**NDICP**), starch and non-fiber carbohydrates (**NFC**). Milk yield was measured from daily shipped milk or individual milk volumes. Weather data were measured using on-farm or nearby publicly available weather stations. The groups of cows on the diets monitored had mean milk yields between 31 L/cow and 52 L/cow. Univariable regression spline models were used to detrend the data in Stata, and residual and smooth values were generated for each variable, as well as Principal Component (**PC**)1, PC2 and PC3. Lagged variables up to -5 weeks were generated for each covariable's residual within their farm diet. A Pearson correlation matrix of the milk yield residual and the covariable residuals at each week's lag was created to determine the lag of greatest effect. These were included in univariable mixed models before a multivariable mixed model was created through backward stepping with farm diet as a random effect.

Findings

Changes in milk yield were positively associated with changes in PC1 (carbohydrates) 4 weeks before and PC3 (Crude fat/NDICP) 1 week before in univariable and multivariable analysis. In the all-diet nutrient univariable model, changes in NDF 4 weeks earlier and NDICP at the time were positively associated, NFC and starch 4 weeks earlier, and weekly average maximum THI at the time were negatively associated with milk yield changes. In a multilevel multivariable model, NFC 4 weeks earlier and weekly average maximum THI at the time were negatively associated, crude fat 1 week earlier and NDICP at the time were positively associated with milk yield changes. The diets were grouped into low-starch ($n = 3$) and high-starch diets ($n = 6$) with a cut point of 24% mean starch in the diet. The cows on high-starch diets had higher milk yield on average (high-starch = 41.8 L/cow; low-starch 34.8 L/cow). For the low-starch diets, the multivariable model showed a positive association between changes in milk yield and starch 1 week earlier and NDICP at the time. For the high-starch diets, the multivariable model found positive associations for CP 3 weeks earlier, crude fat 4 weeks earlier, while starch 2 weeks earlier and maximum average weekly THI 1 week earlier were negatively associated with milk yield changes.

Implications

This study highlighted that the effects of dietary changes take up to 4 weeks to have their greatest effect on milk yield. The carbohydrate component (PC1) and NFC had their greatest effect on milk yield 4 weeks later, while crude fat and NDICP had effects on milk yield 0-1 weeks later. The farms exceeding a mean starch of 24% likely were inducing some level of rumen dysfunction, possibly subclinical acidosis, as increased starch led to decreased milk yield. In low-starch farms, changes in starch concentration positively impacted milk yield. This study uniquely quantifies the delayed effects of diet on milk yield using time series methodology.

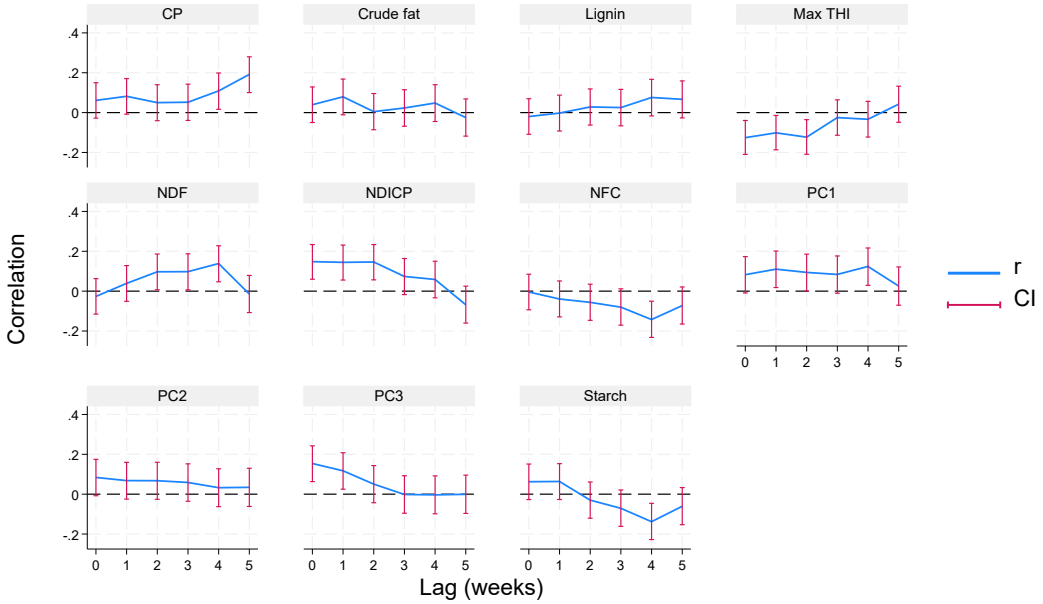

Figure 3: Pearson correlation of the milk yield residuals plotted against the different nutrition components and maximum THI residuals at lags 0 to 5 weeks. "r" is the correlation, "CI" is the 95% confidence interval.

Subproject P2e Calves/Heifers - i. Lifetime Different Diet Effects in Steers

Different lifetime dietary strategies affect carcass characteristics and rumen function in Holstein steers - doi.org/10.1071/AN25153

Authors

H.M. Golder, J. Rehberger, A.H. Smith, E. Block, R. Polkinghorne, H.E. Cuthbertson, M.A. Campbell, V. Vicic, G. Tarr, J.C. Quinn, J. Tong, S.M. Rowe, I.J. Lean

Background

The Australian dairy industry has a considerable surplus of male dairy calves for which there are limited markets and animal welfare concerns; a challenge also faced by other countries. There is a need to improve how "dairy-beef", the term for meat that originated directly or indirectly from dairy herds is integrated into red meat supply chains. Consequently, the challenge that exists with dairy-beef is how to efficiently utilise the opportunity that these calves provide. Our aim was to evaluate the effects of two diets differing primarily in starch, antimicrobial content, and milk replacer volume on (i) short- and long-term rumen adaptation; and (ii) lifetime production in Holstein steers.

Methods

Holstein males, 3-7 days old (n = 72; 36 steers/treatment; 6 per replicate) were randomised to Control (**CON**; 6 L milk replacer/calf.day; 38.2% of dry matter [**DM**] lifetime dietary starch; 50 ppm monensin, 20 ppm flavophospholipol) or Treatment (**TRT**; 4 L milk replacer/calf.day; 47.5% of DM lifetime starch diets with yeast products) strategies. Calves were fed milk replacer twice daily for 42 days, but different pre-starter (days 0-24), starter (days 25-99), and finisher diets (days 100-452). Ruminal fluid was collected at 104, 200, and 438 days old (14 days pre-slaughter) from 24 steers (2 per replicate). Fermentation, production, and carcass measures were analysed by mixed models; ruminal bacterial genera were centre log transformed and subjected to redundancy analysis.

Findings

The CON had higher risk of subclinical ruminal acidosis at day 438, than TRT ($P < 0.001$). Liver abnormalities were 17.1% (CON) and 31.3% (TRT). Controls had greater fermentation with 138.4 ± 5.6 m*M* of total volatile fatty acids vs 111.6 ± 5.6 m*M* (TRT) with 8.4 m*M* higher acetate and 18.1 m*M* higher propionate, but pH was 0.31 units less ($P < 0.050$). Shannon diversity increased over time ($P < 0.001$) and was greater for the TRT at day 200, compared to CON ($P = 0.013$). Bacterial composition differed at each treatment by time comparison ($P \leq 0.01$), with variation increasing over time from 8.6 to 19.2%, suggesting the different diets lead to different microbial successions. The CON steers finished with 12 kg heavier carcasses than the TRT, with 0.8% greater dressing percentage, 1.5 mm more fat at P8, and 1.7 mm more rib fat ($P < 0.050$).

Implications

Control diets produced better carcass weights, P8 and rib fat, and had more fermentability than the TRT diets, likely reflecting better long-term adaptation. Both diets enabled integration of dairy calves into the red meat supply chain, but with differing lifetime rumen adaptations.

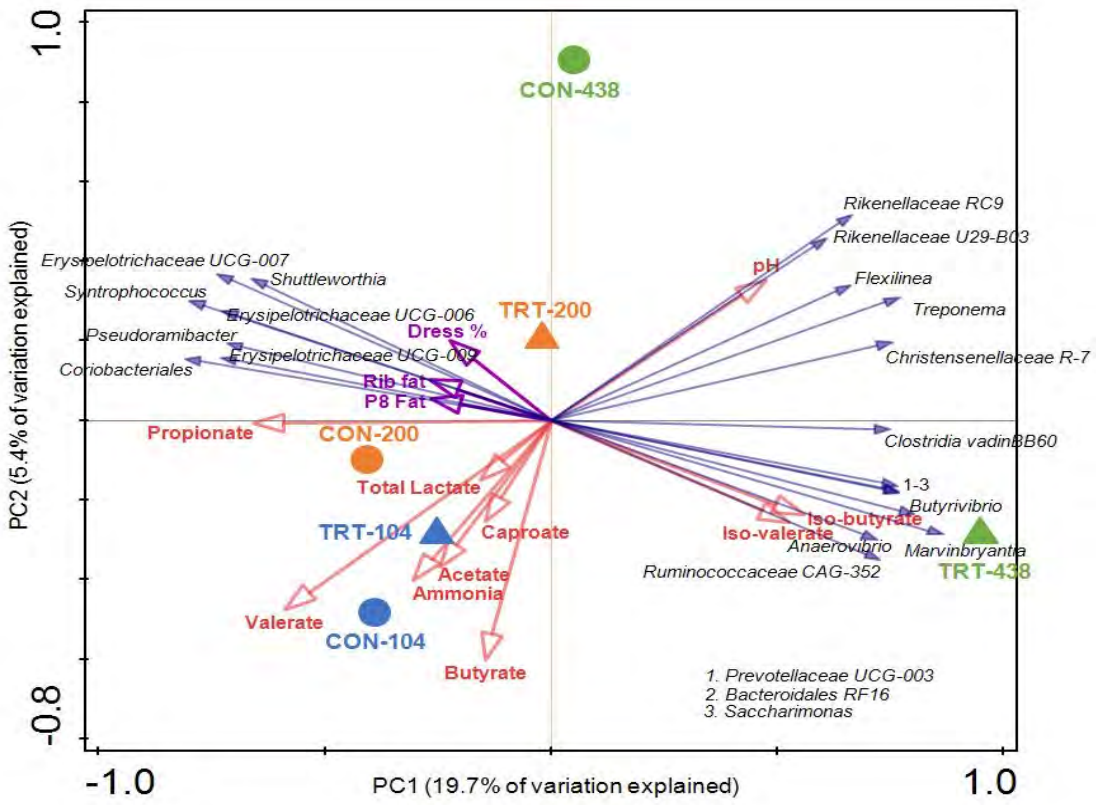

*Figure 13.a: Correlation bi-plot of the redundancy analysis of bacterial genera with respect to treatment and time combinations and ruminal fermentation explanatory variables plus carcass explanatory variables: rib fat, P8 fat, and dressing percentage (Dress %). The 20 genera with the best fit to the explanatory variables are displayed where the length of the arrows are approximate correlation coefficients between the variables and treatment-time combinations. The total variation explained by the treatment-time combinations and explanatory variables is 46.8% and the eigenvalues for the first two axes are 0.20 and 0.05. All pairwise treatment and time combinations differed ($P \leq 0.01$), with the variation in microbiota increasing over time; 8.6, 10.9, and 19.2%, for 104-, 200-, and 438-day samples respectively. Treatment and time combinations were associated with the following variations: TRT-438 = 15.6%, CON-438 = 4.5%, CON-104 = 4.5%, CON-200 = 3.9%, TRT-104 = 3.0%, TRT-200 = 2.5%. Concentrations of propionate and valerate were associated with the most variation of the ruminal fluid measures, 9.3 and 8.5%, respectively ($P = 0.002$). Less variance was associated with iso-butyrate (7.3%; $P = 0.002$) and iso-valerate concentrations (7.0%; $P = 0.002$), rumen pH (5.9%; $P = 0.002$), and concentrations of total volatile fatty acids (**VFA**; 5.8%; $P = 0.002$), ammonia (4.7%; $P = 0.004$), butyrate (3.2%; $P = 0.012$), acetate (3.0%; $P = 0.024$), total lactate (1.7%; $P = 0.204$), and caproate (1.7%; $P = 0.224$). P8 fat, rib fat, and dressing percentage were poorly associated with variance, associated with 2.8% ($P = 0.012$), 2.7% ($P = 0.020$), and 2.2% ($P = 0.072$), respectively. Categorical data is represented by a triangle or circle (blue = samples collected at a mean age of 104 days; orange = 200 days; and green = 438 days) and numerical data is represented with an arrow indicating the direction in which the variable increases (blue = bacterial genera; pink = ruminal fermentation markers, and purple = carcass measures). Blue categorical CON = Control (6 L milk replacer/calf.day for 42 days; 38.2% of DM lifetime starch diets; 50 ppm monensin, 20 ppm flavophospholipol); TRT = Treatment (4 L milk replacer/calf.day for 42 days; 47.5% lifetime starch diets with yeast products); Ruminal fluid samples were collected at a mean of 104, 200, and 438 days old; n = 12 steers/treatment; 2 steers/replicate; total n = 72 ruminal fluid samples.*

Subproject P2e Calves/Heifers - iii. Calf Performance

Milk consumption and behavior of calves in automated calf feeders as early indicators of weaning liveweight

doi.org/10.3168/jdsc.2023-0488

Authors

S.W.J. Legge, P.C. Thomson, C.E.F. Clark, S.C. Garcia

Background

Modern intensive dairy farming relies on data to aid and prioritize management decisions made on farm. Decisions made early in an animal's life can have lasting effects on welfare, productivity, longevity, and profitability. Precision technology such as automated calf feeders (**ACF**) allow the customization of feeding programs, but despite this, weaning weights (**WWT**) vary substantially between calves.

Methods

This observational study used a 3-yr dataset comprising 1,440 female Holstein Friesian calves at a single intensive commercial dairy farm (Dairy Australia feeding system 5; indoor, total mixed ration) using ACF to (1) determine the variability in WWT (as a proxy of animal performance) of calves within this system; (2) identify the contributing factors responsible for the variation in WWT; and (3) identify potential early management intervention points that could be indicative of the performance of calves at weaning within the system. Calves entered the ACF at 10 d of age with 12 calves per ACF; calves were weighed at birth and weaning using weigh scales.

Findings

We discovered a large range of calf WWT (41–118 kg/head) at ~60 d of age despite the application of strict uniform management protocols. Our results from modeling showed that WWT was significantly and positively associated with birth weight (**BWT**), with low BWT calves (<36 kg) achieving an average of 70 kg weight at weaning. In contrast, heavier BWT (>36 kg) calves achieved an average of 82 kg at weaning. Based on calf feeder data, cumulative milk consumption and cumulative unrewarded visits to the feeder, as well as BWT, were identified as indicators of greater WWT as all these were highly significant terms in the model for WWT. Results suggest that quantifying consumption and number of visitations to the ACF at d 5 may allow farmers to identify, with time to intervene, calves underperforming within the feeder or system, therefore increasing their potential for growth.

Implications

this demonstrated that greater milk consumption (>30 kg) and interaction with the feeder up to d 5 in the feeder is more likely to yield a WWT >75 kg, identifying a potential point for management intervention for calves below consumption and interaction thresholds (e.g., by developing alarm systems based on consumption or visitation number).

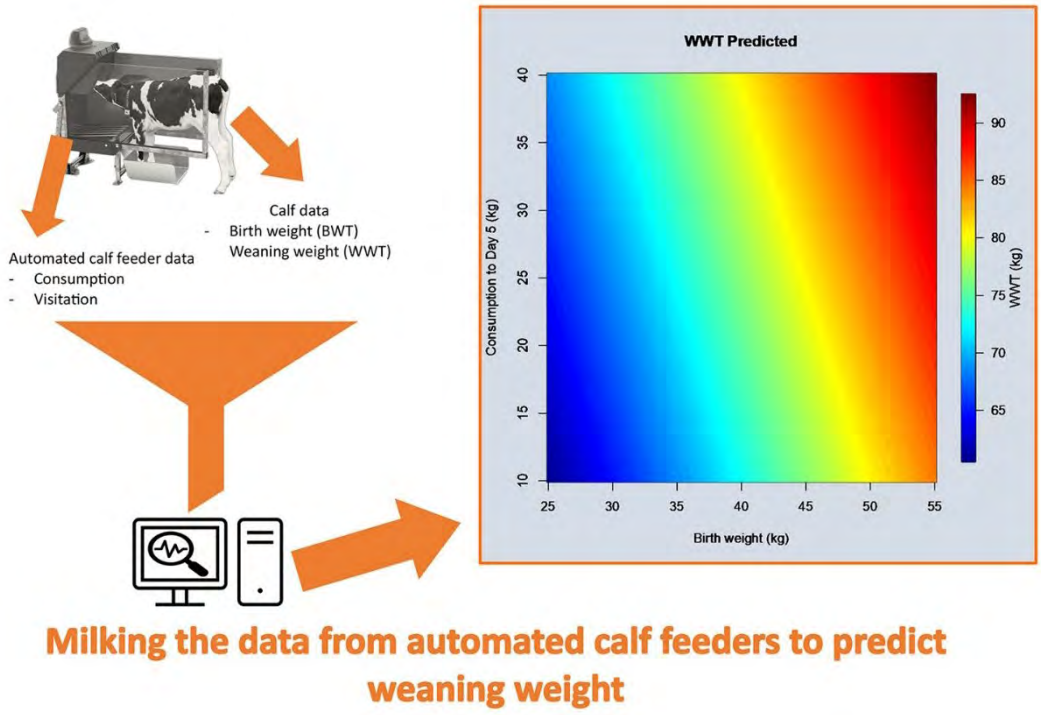

Figure 14.a: Predicted response surface showing the model-based mean weaning weight (WWT) for varying birth weights and cumulative consumption up to day 5. A scale is indicated on the right side of the plot.

Subproject P2e Calves/Heifers - iv. Feedlot Adaptation

Feedlot adaptation: characterising rumen fermentation profiles that determine success or failure

Authors

M. Zhang, I.J. Lean, R. Polkinghorne, H.E. Cuthbertson, M.A. Campbell, G. Tarr, J.C. Quinn, S.M. Rowe, H.M. Golder

Background

Ruminal acidosis is a major digestive disorder of feedlot cattle that leads to significant reductions in cattle health, welfare, and performance, and accounts for 3.7% of mortalities related to digestive disorders in feedlot cattle. We propose that identifying the rumen fermentation characteristics of cattle that adapt well to feedlot induction based on bodyweight gain over the induction period or those that fail to adapt well to feedlot induction may provide vital clues for future interventions to control acidosis, and further knowledge on the aetiology of acidosis. The objective of this study was to evaluate changes in the gastrointestinal tract function of steers introduced to feedlot diets and identify differences between steers with high-weight gain (performing) and low-weight gain (non-performing) during the feedlot induction period (first 3 wk in the feedlot).

Methods

A total of 94 steers of Holstein, British, and European origin were enrolled in the study at entry to the feedlot in 2 batches. After 3 wk, 10 steers from each breed were allocated to 1 of 6 treatment groups based on weight gain between feedlot entry and the end of feedlot induction: Holstein low-bodyweight gain (**HLG**), Holstein high-bodyweight gain (**HHG**), British low-bodyweight gain (**BLG**), British high-bodyweight gain (**BHG**), European low-bodyweight gain (**ELG**), and European high-bodyweight gain (**EHG**). Performance indicators included bodyweight which was measured at feedlot arrival, 3 wk post-feedlot entry (sampling 1; S1), monthly, and 2 wk before slaughter (sampling 2; S2), and average daily gain (**ADG**) which was calculated between each weighing session. Rumen samples were obtained using a stomach tube at S1 and S2 and analysed for pH, volatile fatty acid (**VFA**), ammonia, and lactic acid concentrations. These rumen fermentation measures were used in a validated model based on discriminant analysis and K-means clustering to determine acidosis risk. Following slaughter, carcases, organs and the rumen were inspected for abnormalities.

Findings

No differences in acidosis risk between gain categories (**GC**) or breed were observed ($P = 0.125$ and 0.494, respectively), although HHG steers had a higher risk of acidosis compared to HLG steers ($P = 0.024$). A lack of significant differences in rumen fermentation measures were observed between GC and treatment groups, with the exceptions being isovalerate ($P = 0.001$), acetate to propionate (A:P; $P = 0.010$), isobutyrate ($P = 0.001$), and isovalerate ($P = 0.036$). Steers in the high GC had greater monthly bodyweights ($P < 0.001$) and ADG ($P = 0.003$), final feedlot ADG ($P = 0.003$), hot carcase weight (**HCW**; $P = 0.008$), P8 fat depth ($P = 0.045$), and eye muscle area ($P = 0.039$) compared to low GC steers. There was an overall low incidence of carcase, organ, and rumen abnormalities and no rumen lesions were observed.

Implications

It was concluded that changes in rumen fermentation measures did not influence weight gain, pathology, or meat quality, but rather that the effects of high performance during feedlot induction may translate to more desirable carcase traits at slaughter. The carcass traits demonstrated by Holstein breeds also provide support for the emergence of a dairy beef industry in Australia to reduce male bobby calf wastage.

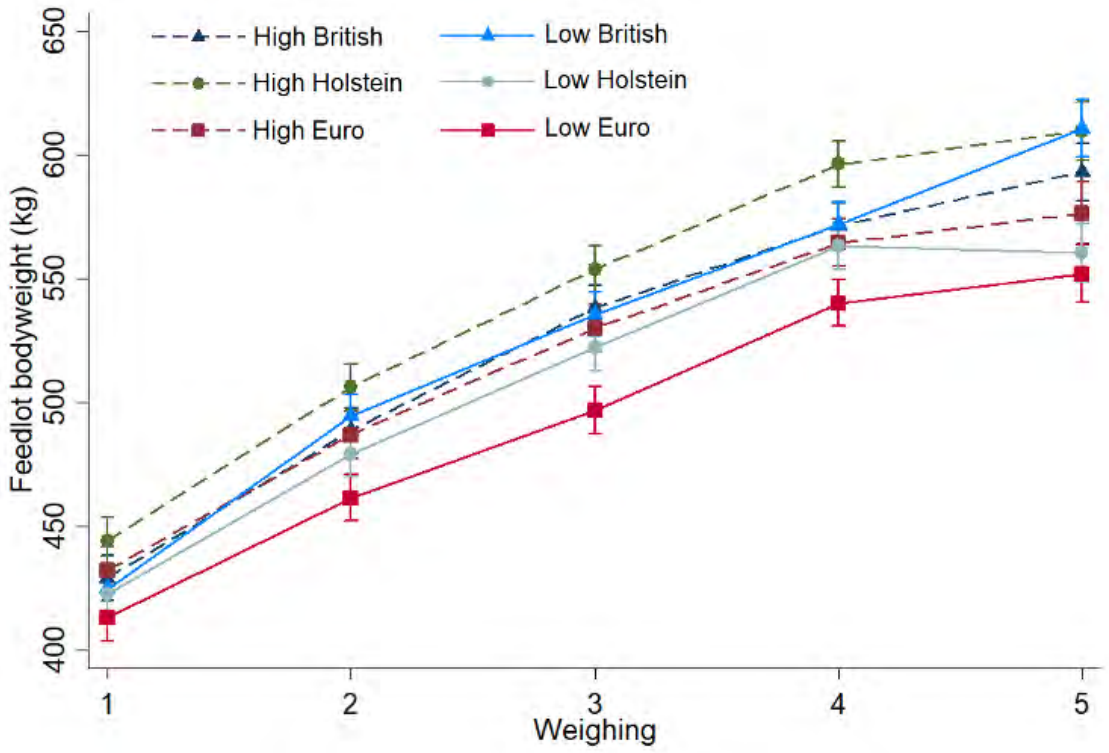

Figure 15.a: Average feedlot bodyweight of Holstein, British and European steers in high and low gain categories at each monthly weighing. High = high gain category; Low = low gain category.

Subproject P2f The Infectome

Background

Infectious diseases are the main cause of disease and mortality in calves. The knowledge of the prevalence and diversity of infectious disease-causing agents in NSW dairy cattle has not been comprehensively assessed. Thus, the immediate goal of this project was to redress this knowledge gap using untargeted microbial genomic sequencing to characterise and identify known and emerging enteric and respiratory pathogens in dairy calves. We determined the occurrence and distribution of their microbial species across all NSW dairy regions. This enables the Australian dairy industry to improve animal health and productivity, and diagnostic capacity, which will allow farmers to make informed management decisions about disease control strategies.

The main objectives of the project were:

To identify the respiratory and enteric infectome diversity in NSW dairy farms: we determined the calves' respiratory and enteric viral, bacterial and eukaryotic species taxonomic diversity in farms across NSW using meta-transcriptomics.

To describe the occurrence and spread of selected ribonucleic acid (**RNA**) viruses and different genotypes through shotgun sequencing and phylogenomics analyses.

To obtain representative isolates and samples from *E. coli* across dairy farms in NSW and characterise the whole genome by the study of *E. coli*'s molecular epidemiology and the study of antimicrobial resistance profile and mechanisms of antimicrobial resistance spread.

Next-generation detection in bovine respiratory and enteric diseases: metagenomic and amplicon sequencing insights into microbial diversity

doi.org/10.3389/fvets.2026.1788101

Authors

Z.U. Abedien, I.J. Lean, S.P. Djordjevic, P.M. Hick, M.E. Westman, J. Mckay-Demeler, J. Webster, B.P. Brito

Summary

Respiratory and enteric diseases are major contributors to morbidity, mortality, and economic loss in cattle production, with significant implications for animal welfare, particularly in calves. Traditional diagnostic approaches have laid the foundation for pathogen detection in cattle, providing essential tools for disease surveillance and control. However, their targeted nature limits the capacity to identify unexpected, novel, or polymicrobial infections that often underlie complex respiratory and enteric syndromes. Recent advances in molecular technologies, particularly amplicon sequencing (metataxonomics), metagenomics, and metatranscriptomics, enable untargeted, high-resolution profiling of microbial communities directly from clinical samples, offering transformative potential for research and diagnostics. This review synthesises current applications of these approaches in bovine respiratory and enteric disease research, highlighting key findings across virology, bacteriology, and parasitology. Collectively, these studies have expanded the catalogue of the microbial diversity, yet their interpretation remains challenged by the still-evolving understanding of microbial contributions to pathogenesis. Progress toward clinical integration is further hindered by the need for methodological standardisation, validation, and improved interpretive frameworks. Looking ahead, advancing these technologies will require harmonised protocols, integration of multi-omics datasets,

and robust experimental and epidemiological studies to establish causal links between microbial signatures and disease outcomes. By bridging discovery and application, these approaches hold the potential to enhance diagnostic accuracy, strengthen surveillance, and support sustainable cattle production systems. As these technologies continue to evolve, they are likely to play an increasingly central role in bovine disease research and diagnostics.

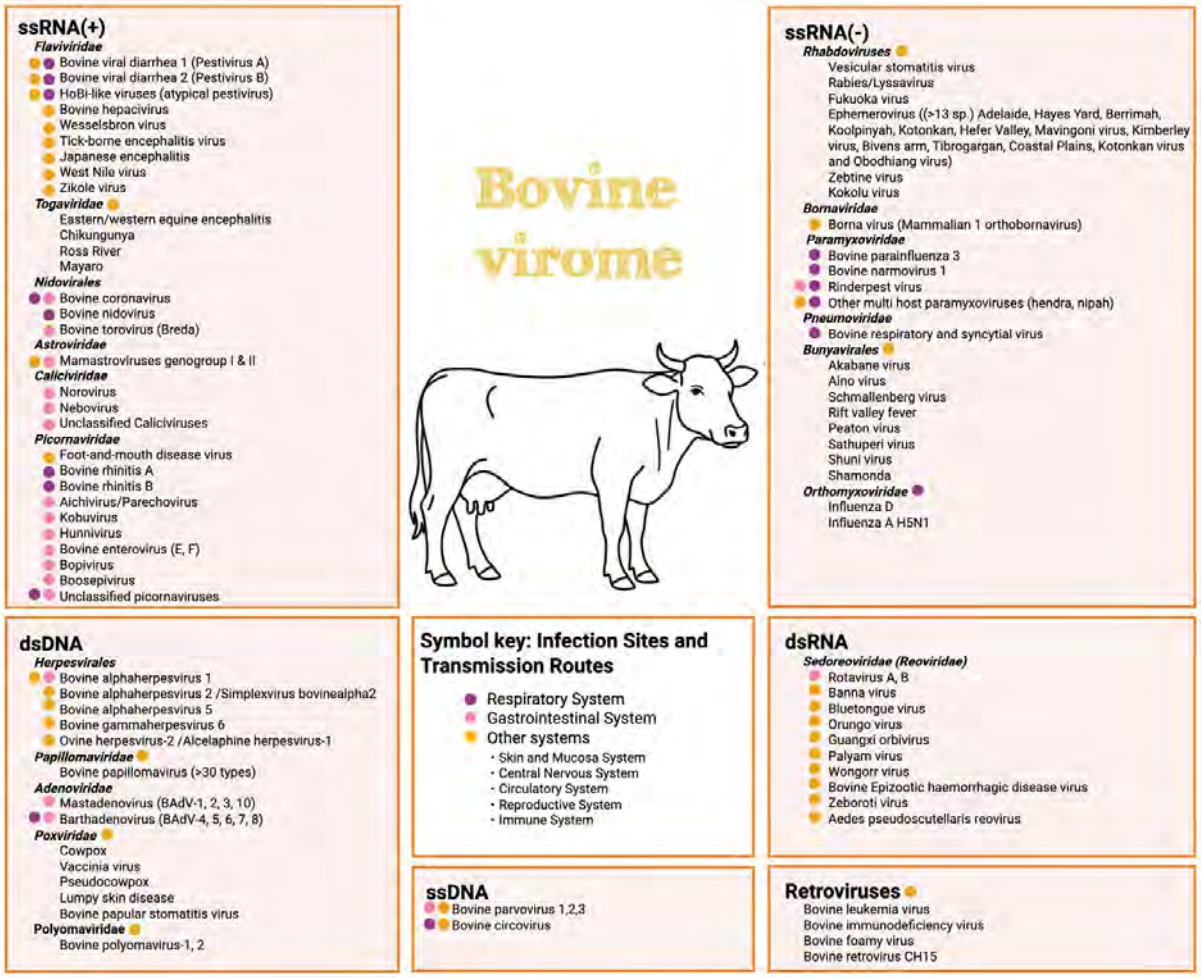

Figure 16.a:Summarises viruses detected in cattle, organised by genome type and taxonomic group. Coloured dots indicate the primary infection sites or transmission routes. This overview highlights both viruses known to cause infection in cattle and those more recently detected in cattle, whose pathogenic potential or effects remain to be explored.

Integrated Metatranscriptomic Profiling of Enteric Pathogens in Bovine Neonatal Diarrhea

Authors

B. Brito, Z.U. Abedien, J. Webster, S. Djordjevic, E.C. Holmes, I.J. Lean

Project Background

Diarrhea in newborn calves is a significant health challenges. It is a leading cause of death and illness in pre-weaned calves, and the costs to producers extend well beyond animal loss, encompassing veterinary treatment, labour, and the long-term impact on growth and milk production later in life. Despite decades of research, the precise combination of infectious agents responsible for diarrhea in calves under real farm conditions has not been investigated using untargeted detection methods. Standard diagnostic tests are limited in scope, typically checking for only a handful of known pathogens at a time, while missing many others that may be contributing to disease.

This project set out to address that gap by applying metatranscriptomics, a cutting-edge genomic sequencing technology, to characterise the full range of actively infectious agents present in calves across New South Wales dairy farms. Unlike conventional diagnostics, metatranscriptomics captures the genetic expression of bacteria and can simultaneously, detect viruses and parasites, providing an unprecedented window into what is actually happening inside a sick or healthy calf's gut. Importantly, it detects organisms that are actively replicating or expressing virulence traits, rather than simply present. This project represents the most comprehensive field-scale investigation of calf enteric disease in Australia to date, spanning 72 farms across all major dairy regions of NSW.

Methods

Between November 2022 and December 2023, the research team visited dairy farms across all major dairy-producing regions of New South Wales, including the Far North Coast, Central West, Riverina, Hunter, Mid Coast, South Coast, Far South Coast. Farms were selected using stratified random sampling to ensure the study was representative of the NSW dairy industry. In total, rectal swabs were collected from 918 pre-weaned calves (under seven weeks of age) from 72 farms, with each farm contributing samples from both healthy and diarrheic calves wherever possible. Calves were grouped into four age brackets to allow comparisons at different life stages.

Swabs were preserved immediately using a nucleic acid stabilisation medium and stored at -80°C at the Elizabeth Macarthur Agricultural Institute (**EMAI**). In the laboratory, 593 samples were selected for metatranscriptomic sequencing. The RNA was extracted and sequenced using the Illumina NovaSeq X platform, generating an average of over 60 million read pairs per sample. Bioinformatic analysis involved removal of host and background sequences, followed by detection and quantification of viral, parasitic, and bacterial virulence gene transcripts against curated reference databases. Multiple statistical frameworks were applied to identify associations between specific pathogens and diarrheal disease, including machine-learning models, mixed-effects regression, and differential abundance analysis, all accounting for farm-level differences and calf age.

Region	Healthy	Respiratory signs	Respiratory + diarrhea	Diarrhea	Unspecific signs	Total
Central West	51	5	1	8	5	70
Far South Coast	121	8	2	34	5	170
Far North Coast	51	4	0	18	3	76
Hunter	49	1	0	13	0	63
Mid Coast	165	13	5	29	7	219
Riverina	68	31	3	26	12	140
South Coast	74	14	1	42	8	139
Wagga & Inland	31	6	0	4	0	41
Total	610	82	12	174	30	918

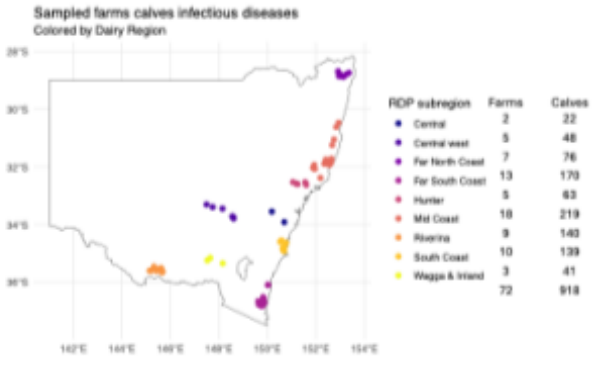

Figure 17-b Geographic distribution and health status of 918 pre-weaned calves sampled across major dairy regions in New South Wales, Australia. The table shows calf counts by clinical status and region, while the map visualizes farm locations coloured by dairy region.

Key Findings

Viruses: Astrovirus was the most abundant viral transcript detected across all samples, followed by Rotavirus A and Kobuvirus. Rotavirus A, long recognised as a primary cause of calf diarrhea, was confirmed as a significant early-life pathogen. Bovine Kobuvirus emerged as a particularly strong predictor of diarrhea in calves under three weeks of age, with disease risk linked to the amount of virus present rather than its mere detection. Strikingly, many viruses, including astroviruses and picornaviruses, were detected at high levels in clinically healthy calves, demonstrating that viral presence alone does not equal disease.

Parasites: Cryptosporidium was detected on 83% of farms and was the only parasite significantly associated with diarrhea. Giardia, although widespread, showed a negative association with diarrhea, potentially reflecting disruption of its intestinal niche during disease rather than a protective effect. Substantial shedding of all parasites was observed in healthy calves, underscoring that farm-wide exposure is the norm rather than the exception.

Bacteria: Rather than simply identifying which bacteria were present, this study measured the expression of bacterial virulence genes, the tools bacteria use to cause disease. Genes putatively associated with *Escherichia coli* adhesion, invasion, and iron acquisition were significantly enriched in diarrheic calves during the first weeks of life. Campylobacter-associated motility and colonisation genes were also strongly linked to diarrhea, particularly their abundance in positive samples. Farm of origin was the strongest determinant of bacterial virulence profiles, explaining over 45% of variation, highlighting how farm management, hygiene, and environment shape the bacterial disease landscape far more than any single pathogen.

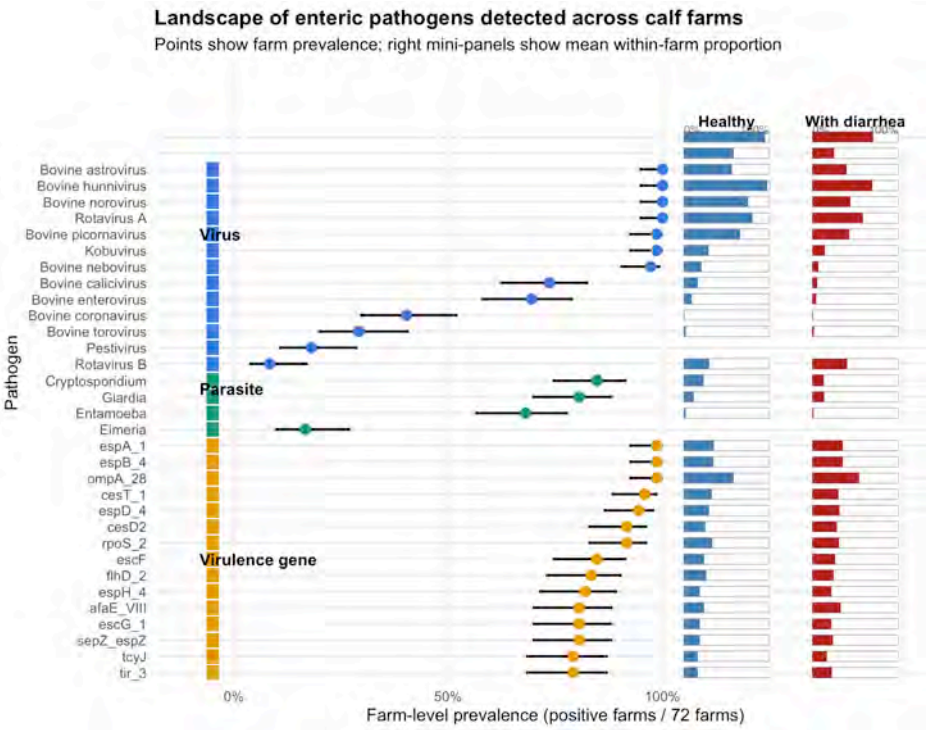

Figure 17-c: Landscape of enteric pathogens detected across calf farms. Points represent the farm level prevalence (n = 72 farms) with 95% Wilson confidence intervals. Pathogens are grouped by category (viruses, parasites, and bacterial virulence genes), indicated by the coloured sidebar. Right mini-panels show the mean within-farm proportion of positive calves in healthy animals (blue) and calves with diarrhea (red), averaged across farms.

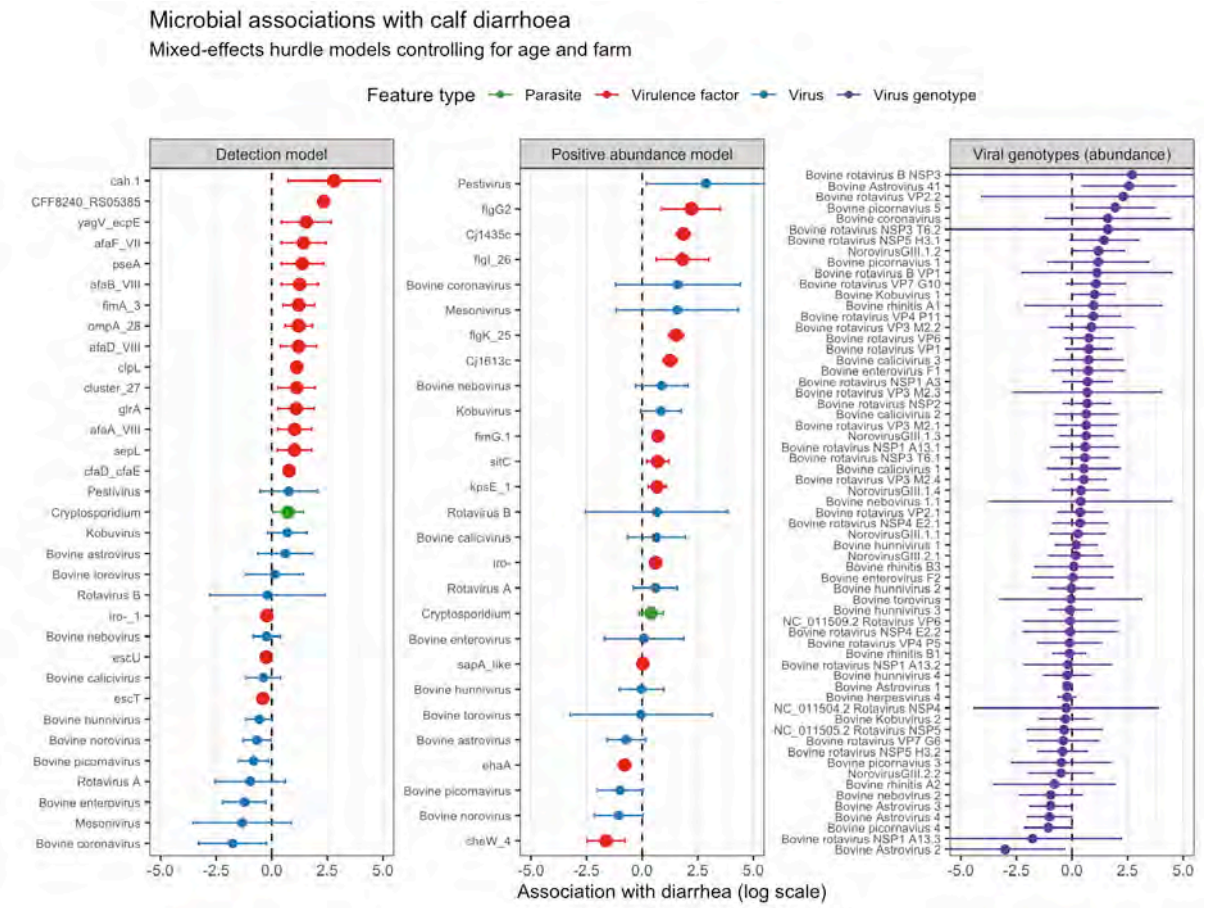

Figure 17-d: Microbial features (taxa) associated with calf diarrhoea identified using mixed-effects hurdle models. Detection models (left) show associations with the probability of presence (log-odds), while positive abundance models (middle) show associations with abundance among positive samples (log fold change). The right panel shows genotype-level viral associations based on the abundance component of the model. Points represent model coefficients and horizontal bars indicate 95% confidence intervals. Positive values indicate higher detection probability or abundance in diarrheic calves, whereas negative values indicate higher detection or abundance in healthy calves. Colours denote microbial feature type: parasites (green), virulence factors (red), viruses (blue), and viral genotypes (purple). All models include farm as a random effect and adjust for calf age.

Implications for the Dairy Industry

These findings have practical implications for diagnostics of enteric disease in dairy calves. The study demonstrates that calf diarrhea is rarely caused by a single pathogen, and the microbial variation is shaped mostly by the farm environment. Current diagnostic tests, which typically screen for only four or five pathogens, likely miss a significant proportion of the microbial complexity driving disease. This study identifies bovine Kobuvirus as a potential emerging relevant pathogen of particular concern for young calves that is not currently included in standard diagnostic panels or commercial vaccines in Australia.

A major practical takeaway is the understanding of the high viral or bacterial load carried by some healthy calves can frequently shed high levels of pathogens without becoming ill, meaning a positive test result alone provides limited guidance for treatment or intervention. Future diagnostic frameworks should incorporate pathogen load and age-specific context, and potentially host biomarkers to better guide producer and veterinary decision-making. The strong farm-level effect on bacterial virulence profiles also points to the central role of on-farm management practices, namely colostrum delivery, hygiene, stocking density, and antibiotic stewardship, in determining disease risk, reinforcing the value of farm-specific advisory services.

From a biosecurity and public health perspective, the identification of *Campylobacter* virulence activity in calves with diarrhea has further implications. Understanding the conditions under which virulence genes are expressed in calves can inform strategies to reduce contamination risk at the farm level. More broadly, this project has generated a rich, publicly available dataset, including 593 metatranscriptomes deposited in the National Center for Biotechnology Information (**NCBI**) Sequence Read Archive, that will serve as a foundational resource for future research into bovine enteric disease, antimicrobial resistance, and emerging infectious agents relevant to Australian dairy production.

Bovine Respiratory Disease in NSW Dairy Calves- Understanding Respiratory Viruses, Bacteria, and Calf Immune Responses Across NSW Dairy Farms

Authors

B. Brito, Z.U. Abedien, S. Djordjevic, E.C. Holmes, I.J. Lean

Project Background

Bovine respiratory disease (**BRD**), commonly one of the costliest and welfare-significant infectious disease affecting cattle worldwide. In pre-weaned calves, it is a significant cause of illness, death, and long-term production losses, contributing to reduced growth, impaired future milk yields, and increased antibiotic use. Despite its economic and animal welfare importance, several questions remain at a fundamental level: we know that multiple viruses and bacteria are involved, but we have had a limited ability to determine which specific pathogens matter most, how they interact with each other, and how the calf's own immune system responds to infection. In New South Wales (NSW), there was no systematic, state-wide data on which respiratory viruses and bacteria are circulating in dairy calf populations, or how prevalent they are. This information gap has hampered the design of effective prevention strategies and has left the industry relying on diagnostic tools and vaccines originally developed for overseas or feedlot systems, contexts that differ substantially from dairy farming. This project was designed to fill these gaps. Using a cutting-edge genomic technique called metatranscriptomics, which allows us to simultaneously detect all active viruses, bacteria, and the calf's own immune gene activity from a single nasal swab, we conducted the first comprehensive, state-wide survey of respiratory pathogens in NSW dairy calves. The study enrolled 72 farms and 908 pre-weaned calves across all major dairy regions of NSW, generating what is now the largest dataset of its kind for Australian dairy cattle, and one of the most comprehensive globally.

Methods

Between November 2022 and December 2023, the research team visited dairy farms across NSW and collected nasal swabs from calves under 60 days of age. Farms were selected randomly from a state registry to ensure results were representative of the NSW dairy industry. Each calf was assessed clinically and classified as healthy, showing non-specific signs, having respiratory disease, having diarrhoea, or a combination.

The nasal swabs were processed using a technique called metatranscriptomics, a form of genetic sequencing that reads all the active biological "messages" present in a sample at the time of collection. This captures a snapshot of everything that is actively happening in the calf's nasal cavity: which viruses are replicating, which bacteria are active, and which of the calf's own defence genes are switched on or off. This approach is far more comprehensive than conventional diagnostic tests such as polymerase chain reaction (PCR) panels, which only detect a pre-selected list of known targets and can miss unexpected or newly recognised pathogens.

Each swab generated approximately 75 million genetic sequence reads. These were processed through a computational pipeline that separated calf RNA from microbial RNA, then matched sequences to databases of known viruses, bacteria, and calf immune genes. Statistical analyses were used to determine which pathogens were most prevalent across NSW farms, which were most strongly associated with disease, and how the calf's immune system responded differently depending on which pathogen was present. The farm of origin was also factored into all analyses to account for the natural variation between properties.

Table 17-a: Number of nasal swabs metatranscriptomes successfully sequenced per health status, and age category.

Age strata	Healthy	Respiratory	Respiratory & diarrhea	Diarrhea	Unspecific	Total
<25	104	23	3	33	12	175
≥25	102	42	4	15	13	176
Unknown	31	10	4	6	4	55
Total	237	75	11	54	29	406

Findings

The study generated several important discoveries. First, and most strikingly, the farm a calf was born on was the single biggest factor shaping which viruses and bacteria were present in its nasal secretion, and how its immune system was responding, far outweighing the effect of whether the calf was sick or healthy. This tells us that respiratory disease in dairy calves is fundamentally a farm-environment problem, driven by the microbial world each herd harbours.

Second, the pathogens most commonly found in NSW dairy calves were different to the ones currently targeted by most commercial vaccines and diagnostic tests. Bovine rhinitis viruses A and B (never routinely tested in Australia), bovine torovirus, and bovine adenovirus 3 were the most prevalent respiratory viruses detected, found on 35–52% of farms. In contrast, viruses such as bovine respiratory syncytial virus and infectious bovine rhinotracheitis virus were detected rarely or not at all in this age group.

Third, the bacterial genera most consistently associated with disease were Moraxella, Mannheimia, and Pasteurella, supporting their role as key drivers of clinical pneumonia. Critically, when these bacteria were abundant, the calf's immune system showed signs of tissue damage and inflammation.

Finally, the calf's immune response followed a clear pattern: early in infection, antiviral defence genes were activated; in calves with established disease, the response shifted toward damaging inflammation. This transition appears to be a key feature of disease progression.

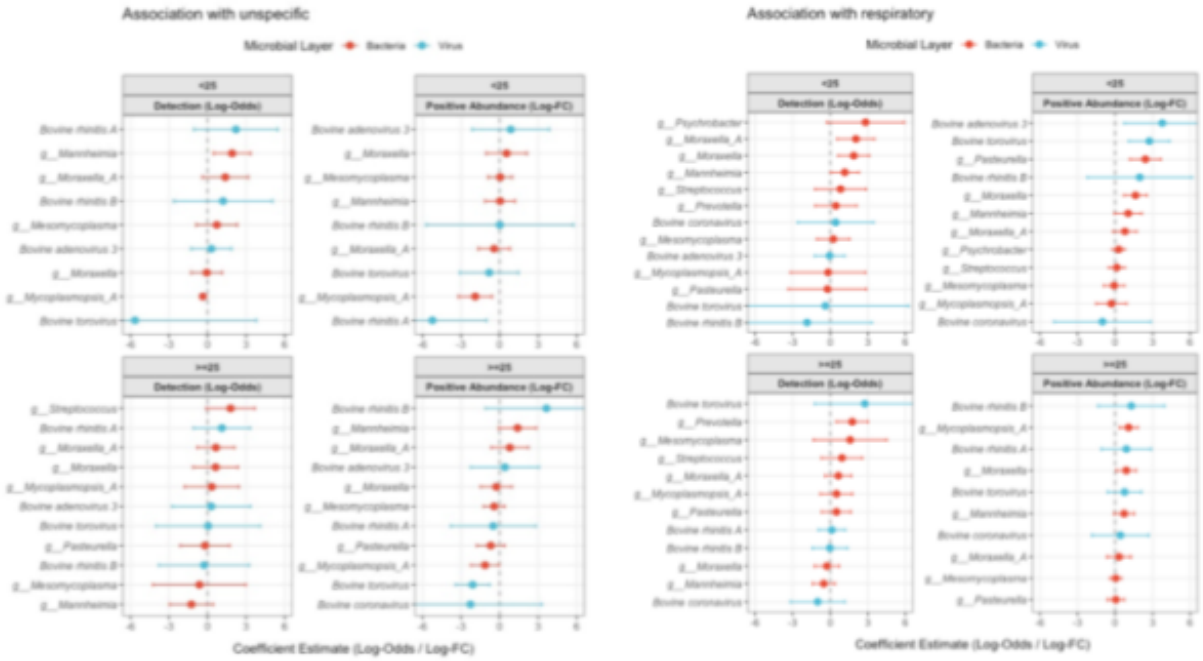

Figure 17-e: Microbial associations with animals showing respiratory or unspecific signs across age groups. Associations between microbial features (viruses and bacteria) and clinical status were assessed using mixed-effects hurdle models, separating detection (presence/absence; log-odds) and positive abundance (log fold change). Results are shown for two comparisons: unspecific vs. healthy (left) and respiratory vs. healthy (right). Within each comparison, analyses were stratified by age (<25 days and ≥25 days), and results are presented separately for detection and positive abundance components. Points represent model estimates, and horizontal lines indicate 95% confidence intervals. Positive estimates indicate higher prevalence or abundance in diseased animals relative to healthy ones, while negative estimates indicate association with healthy status. Models included clinical status as a fixed effect and farm as a random intercept to account for clustering. Only features meeting predefined quality control criteria (minimum sample size and sufficient positive observations per group) were included in each model component; therefore, not all features are present in all panels. Colours indicate microbial layer (viruses in blue; bacteria in red).

Implications

These findings have direct and significant implications for how BRD is managed in dairy calves. The dominance of the farm environment as a driver of disease risk means that the most effective strategies for reducing calf respiratory disease lie in improving management at the herd level. This supports a shift from reactive to preventive farm health planning, and highlights that consistent implementation of existing best practices could substantially reduce disease burden across the NSW industry. The identification of rhinitis viruses and bovine torovirus as the most prevalent respiratory viruses in Australian dairy calves is a pivotal finding for the vaccine and diagnostics industry. While with this investigation we identified the microbial populations present in the upper respiratory tract of the calves and its association with disease, the next step is to determine their association with lower respiratory tract infection and pneumonia. Despite this limitation, this study helps guide the development of relevant diagnostic panels for respiratory disease investigations. Internationally, these findings add to a growing body of evidence that BRD vaccine portfolios need updating to reflect the actual pathogens circulating in dairy calf populations. The host immune data provide a roadmap for future therapeutic development. By identifying the specific immune pathways that are protective

versus damaging, researchers and industry can begin to explore targeted interventions, such as immune modulators or mucosal vaccines, that strengthen early defence without triggering the excessive inflammation that causes lung damage. Together, these findings provide a science-based foundation for rethinking calf respiratory health strategy at the farm, national, and global level.

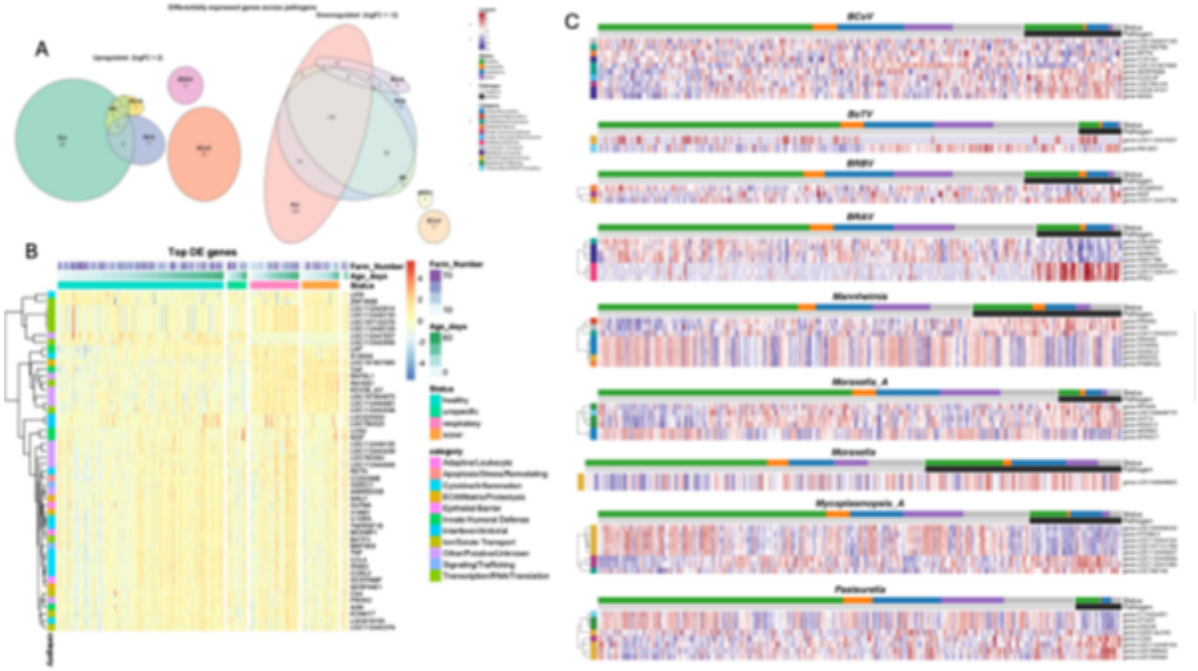

Figure 17-f: A: Euler diagram of differentially expressed genes in animals with unspecific symptoms or respiratory symptoms compared healthy animals based on pathogen positivity. B: Global host transcriptional response in bovine nasal swab metatranscriptomes stratified by clinical status. Heatmap of z-score-scaled expression values for the top differentially expressed host genes across clinical status.

The molecular epidemiology of rotaviruses in NSW dairy calves, Australia

Authors

Y. Kida, Z.U. Abedien, C.M. Donato, S. Djordjevic, M.J. Frost, A. Read, P. Hick, E.C. Holmes, I.J. Lean, B. Brito

Project overview

Neonatal calf diarrhea is a leading cause of mortality in young calves worldwide, with bovine rotavirus A (BoRVA) recognized as a major etiological agent. Genomic variation among circulating strains can influence antigenicity, virulence, transmissibility, and potentially vaccine effectiveness. To comprehensively characterize BoRVA diversity and associated host responses in NSW, Australia, we performed a large-scale metatranscriptomic study of pre-weaned calves, analyzing 593 rectal and 408 nasal transcriptomes.

Rotavirus genomes were frequently detected in both diarrheic and clinically healthy calves, with substantial viral loads observed in asymptomatic animals, indicating widespread subclinical shedding. Genotyping revealed that dominant bovine genotypes: G6 or G10 in combination with P[5] or P[11], were consistently distributed across NSW. Rare genotypes, including G24 and G8, as well rotavirus B, were also identified.

Genotype-specific age patterns were evident. Rotavirus G10P[11] genotypes were detected across a broader age range and maintained high abundance over an extended window compared to G6P[5], which exhibited a narrower peak in early life.

Host transcriptome analysis revealed that high-burden infections were associated with coordinated downregulation of epithelial barrier, transport, and metabolic pathways, alongside upregulation of epithelial stress and remodeling signatures, consistent with functional mucosal disruption rather than isolated inflammatory amplification.

Together, these findings highlight the local genetic diversity, the extensive subclinical BoRVA circulation, genotype-specific age dynamics, and distinct host transcriptional responses, underscoring the importance of genomic surveillance to inform vaccine and control strategies.

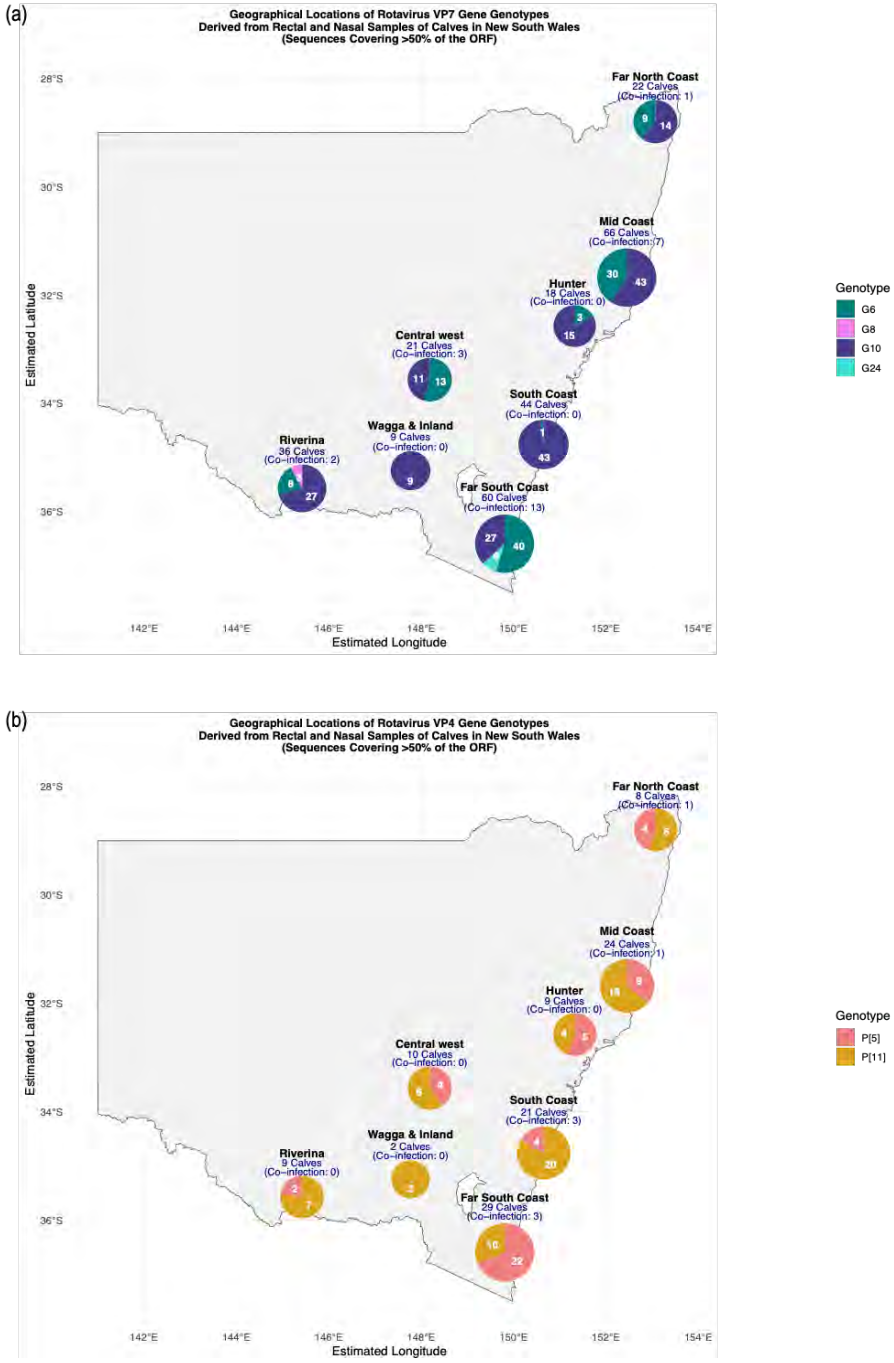

*Figure 17-g: Geographical distribution of rotavirus genotypes in calves across NSW. (a) Geographical locations of each genotype of viral protein (**VP**)7 of rotavirus A (**RVA**). Pie charts show the proportion of VP7 genotypes detected in all rectal and nasal samples combined for each area. The size of each pie reflects the total number of calves in which rotavirus was detected. Numbers inside pies indicate the number of calves carrying each genotype. Areas with co-infected calves are highlighted. Colours represent genotypes: G6 (teal), G8 (violet), G10 (dark slate blue), and G24 (turquoise). (b) Geographical locations of each genotype of VP4 of the detected RVA. Pie charts represent the proportion of VP4 genotypes detected in rectal (R) or nasal (N) samples for each geographic area. The size of each pie chart is proportional to the number of unique calves sampled. Numbers inside each pie indicate the number of calves*

carrying each genotype. Areas with co-infected calves are indicated separately. Colours correspond to genotypes as follows: P[5] (light coral), and P[11] (golden).

Bovine Respiratory Disease Study – Mesonivirus Findings

Authors

B. Brito, E. Holmes, I.J. Lean.

Project background

Respiratory disease is a leading cause of illness in young dairy calves, yet many cases have no identified cause even after extensive testing. This project used advanced genetic sequencing to characterize all microorganisms present in nasal samples from calves across New South Wales farms, with the goal of uncovering previously undetected pathogens.

Methods

Nasal swabs were collected from 406 pre-weaned calves, both healthy and showing signs of respiratory illness, across 72 dairy farms. Samples were analyzed using broad-spectrum RNA sequencing capable of detecting any actively replicating microorganism, without needing to know in advance what to look for.

Findings

The objective of this study was to investigate an unexpected finding consisting of the detection of Alphamesonivirus-1 in calves from 12 farms. This virus belongs to the family Mesoniviridae within the order Nidovirales and has previously been considered insect-specific, with reported associations limited primarily to mosquitoes. Notably, the five calves with the highest viral abundances were all affected by respiratory disease.

Importantly, no mosquito-derived genetic material was detected in these samples, reducing the likelihood that the viral sequences originated from an accidentally sequenced mosquito. Instead, these findings support the possibility that Alphamesonivirus-1 was present within the upper respiratory tract of the calves themselves. While the biological significance and host range of this virus remain unclear, its repeated detection across multiple farms and association with respiratory disease warrant further investigation to determine whether it may represent a previously unrecognised bovine-associated virus or an opportunistic respiratory agent.

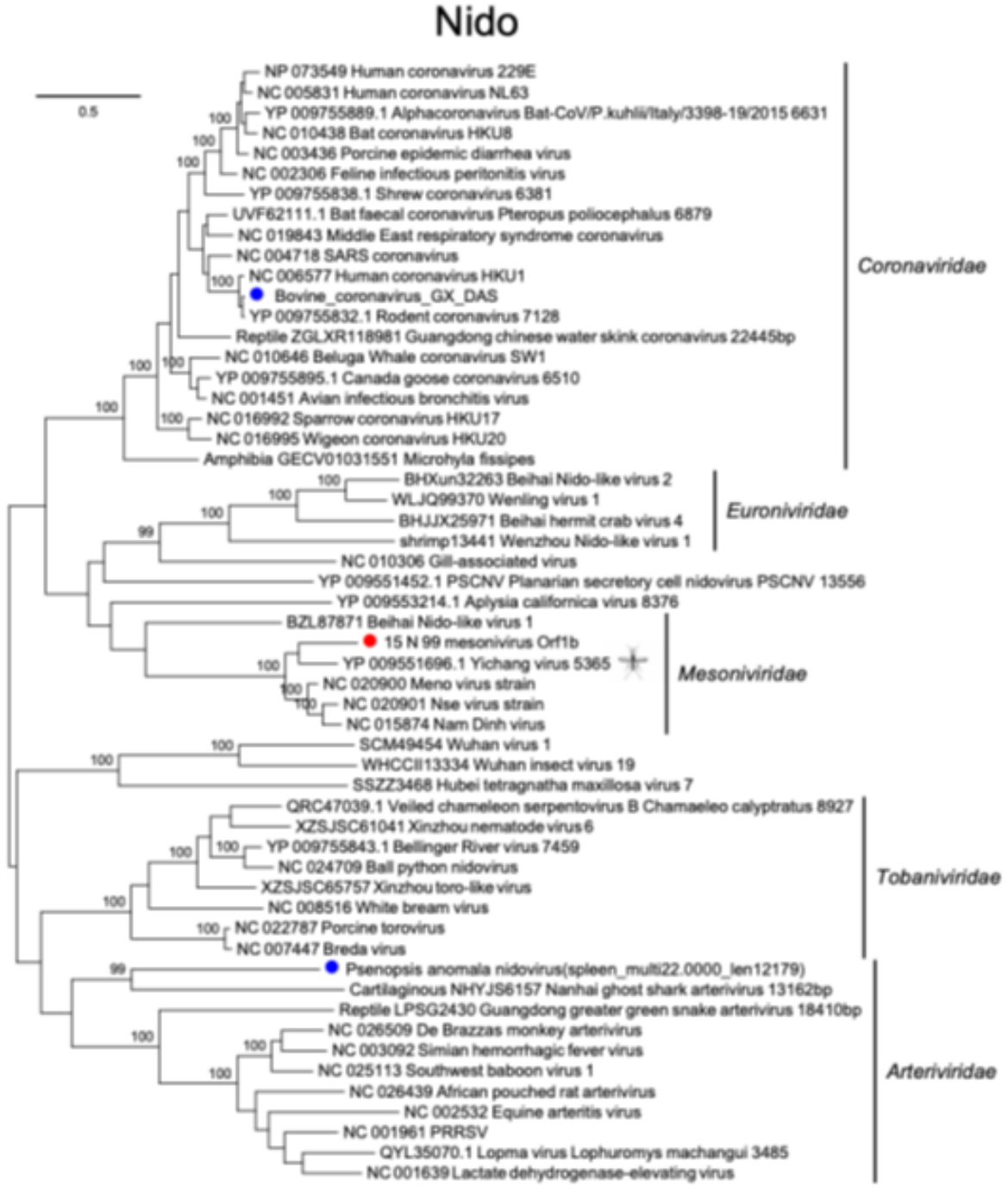

Figure 17-h: Phylogenetic analysis of the amino acid sequence of ORF1b from Nidovirales representative species. The virus found in nasal swabs in this study is highlighted in red (15 N 99 mesonivirus Orf1b).

Implications

This is one the first reports of this virus in cattle and only the second time it has been detected in any land animal (previous detection was from a respiratory case in a horse). While it remains unclear whether this virus causes disease, the finding suggests it may have a broader host range than previously recognized and warrants further investigation as a potential contributor to unexplained respiratory illness in cattle.

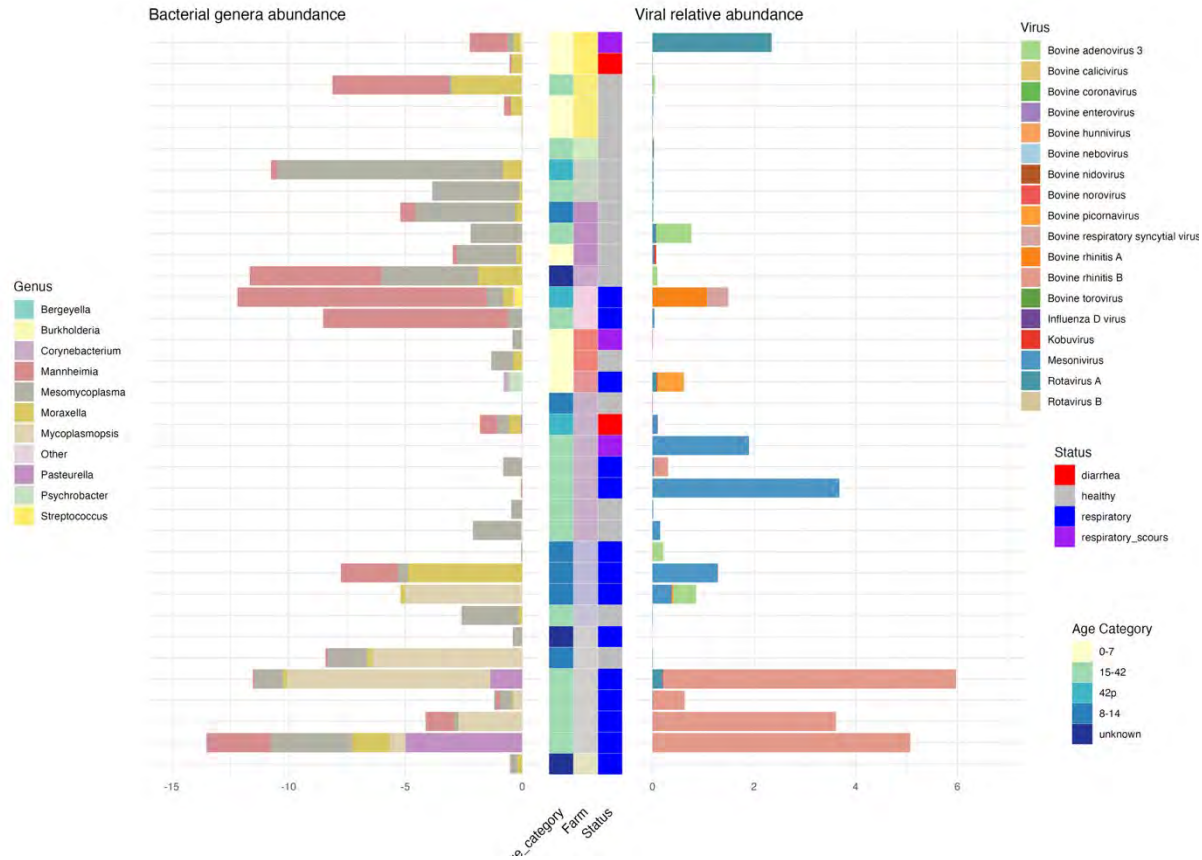

Figure 17-i: A diverse range of bacteria and viruses was present in nasal samples where mesonivirus was detected. Among the Mesonivirus-positive animals, 14 animals with respiratory disease (as indicated by blue annotation tiles) showed low Mesonivirus abundance and instead had high levels of Bovine rhinitis A and B viruses, along with Mannheimia, Mycoplasma, and Pasteurella, all common respiratory bacteria.

Whole-genome surveillance of Escherichia coli from healthy and scouring dairy calves across New South Wales: a matched case-control study

Authors

A.M. Stanczak, V.M. Jarocki, E.R. Wyrsch, I.J. Lean, B. Brito, S. Djordjevic

Background

Neonatal calf diarrhoea ("scours") is the leading cause of morbidity and mortality in pre-weaned dairy calves worldwide, imposing substantial economic and welfare costs on the dairy industry and driving early-life antimicrobial use. E. coli, particularly enterotoxigenic (ETEC) and other diarrhoeagenic pathotypes, is a key contributor, yet the population structure, virulence repertoire and resistance gene burden of E. coli circulating in healthy versus scouring calves on Australian dairy farms remain poorly characterised, limiting evidence-based diagnostics, intervention design and antimicrobial stewardship.

Methods

A matched case-control study was conducted across 40 commercial dairy farms spanning eight major dairy regions of New South Wales. Faecal samples were collected from 126 calves (63 scour cases, 63 clinically healthy controls) stratified by age (0–7, 8–14, 15–42 and >42 days). Multiple colonies per animal were recovered on chromogenic (ECC) and ESBL-selective media to capture within-host diversity and ESBL-selection-context strains, then subjected to whole-genome sequencing on the Illumina short-read platform. Following quality control, 451 high-quality E. coli genomes (352 ECC, 99 ESBL-selection) were retained for downstream genomic analysis.

Results

In silico multilocus sequence typing resolved 91 distinct STs across the case-control cohort, with the assemblage dominated by globally distributed lineages, ST10 (n=74), ST58 (n=49), ST88 (n=34) and ST362 (n=22), alongside 20 isolates with novel STs. Twenty-eight STs were shared between healthy and scouring calves, while 37 were recovered exclusively from healthy controls and 26 exclusively from scour cases; most exclusive STs were singletons from single farms, but several were supported by multiple animals and farms. ST10 and ST69 were over-represented among scouring calves, and ST602, ST56, ST711 and ST75 were recovered exclusively from scour cases across multiple animals and farms, marking them as priority candidates for pathotype characterisation. Conversely, ST3268, ST3018, ST38 and ST1613 were recovered exclusively from healthy controls, alongside lineages such as ST117, ST1434 and ST3268 that were strongly skewed to healthy carriage. Comprehensive downstream analyses will include in silico pathotyping and virulence-gene screening (Enterotoxigenic E. coli/Enteropathogenic E. coli/Shiga toxin-producing E. coli determinants, fimbrial adhesins, toxins, iron-acquisition systems), antimicrobial resistance gene and plasmid replicon profiling, mobile genetic element (insertion sequence, integron, transposon, prophage) characterisation, and pangenome analysis to identify accessory gene content and lineage-specific signatures associated with disease status.

Implications

This dataset provides one of the most comprehensive genomic baselines of E. coli from Australian dairy calves to date and establishes a framework for disentangling commensal carriage from disease-associated lineages. Identification of ST- and gene-level markers linked to scours will inform targeted on-farm diagnostics, evidence-based antimicrobial use, and One Health surveillance of high-risk E. coli lineages with zoonotic and food-chain relevance.

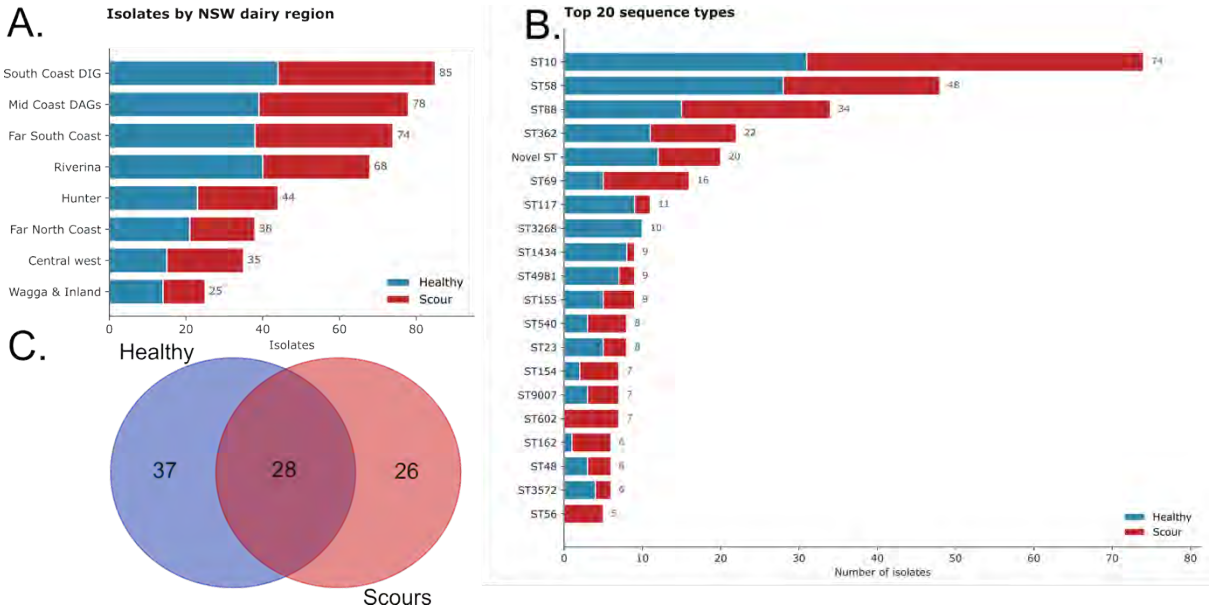

Figure 17-j: E. coli diversity in healthy and scouring dairy calves A) cohort overview B) Stacked bars showing healthy/scour split for each of the top 20 most abundant lineages C) Venn diagram illustrating ST overlap between E. coli derived from healthy and scouring calves.

Detection of blaCTX-M genes in Escherichia coli in dairy calves in Australia.

Authors

A.M. Stanczak, V.M. Jarocki, E.R. Wyrsch, I.J. Lean, B. Brito, S. Djordjevic

Background

Escherichia coli is a leading cause of antibiotic-resistant urinary tract (UTI) and blood stream (BSI) infections, ventilator-associated pneumonia, meningitis and wound infections. E. coli is also a major pathogen across intensive animal production systems causing gastrointestinal and extraintestinal disease. From a genomic perspective, E. coli has been intensively studied as a human pathogen but remains understudied in livestock, fresh produce, retail meats, waste streams, and wildlife, hindering One Health considerations of antimicrobial resistance (AMR) and pathogen evolution.

Methods

A comprehensive E. coli sampling regime devised to examine phylogeny, AMR and virulence gene carriage across different dairy production systems generated 1300 rectal samples from 67 farms across NSW. E. coli was isolated from calves <7 weeks old in 67 farms using both non-selective (Chrome-Agar) and selective (Chrome-agar with an extended spectrum β-lactam (ESBL) antibiotic) approaches.

Results

Phenotypic resistance to ESBLs was observed on 59 farms (88%) with ESBL non-susceptible E. coli isolates collected from 52 farms. After quality control analysis of whole genome sequencing data, 117 isolates representing 49 farms comprised the final collection. Among these, 23 sequence types (ST) were identified with ST10 (n=22), ST88 (n=15), ST9007 (n=10), ST155 (n=10) and ST117 (n=9) dominant. All E. coli phylogroups except B2 were represented with A (n=48), B1 (n=31) and C (n=15) common. Phenotypically resistant isolates had blaCTX-M-14 (n=59), blaCTX-M-15 (n=45), blaCTX-M-27 (n=1), blaCTX-M-1 (n=1), blaDHA-1 (n=1), and blaCMY-2 (n=4) present; we could account for ESBL resistance in six isolates. Genes encoding resistance to aminoglycosides, sulphonamides, tetracyclines, β-lactams, trimethoprim, and macrolides were also prevalent on a herd basis and most isolates (n=75; 64%) carried the class 1 integrase gene intI1. Virulence gene carriage analysis of the genome sequences indicated that ColV (n=36; 31%) plasmids and the Yersinia High Pathogenicity Island (n=42; 36%), both high priority ExPEC virulence factors found in E. coli that cause UTI and BSI, were frequently detected in these E. coli genomes.

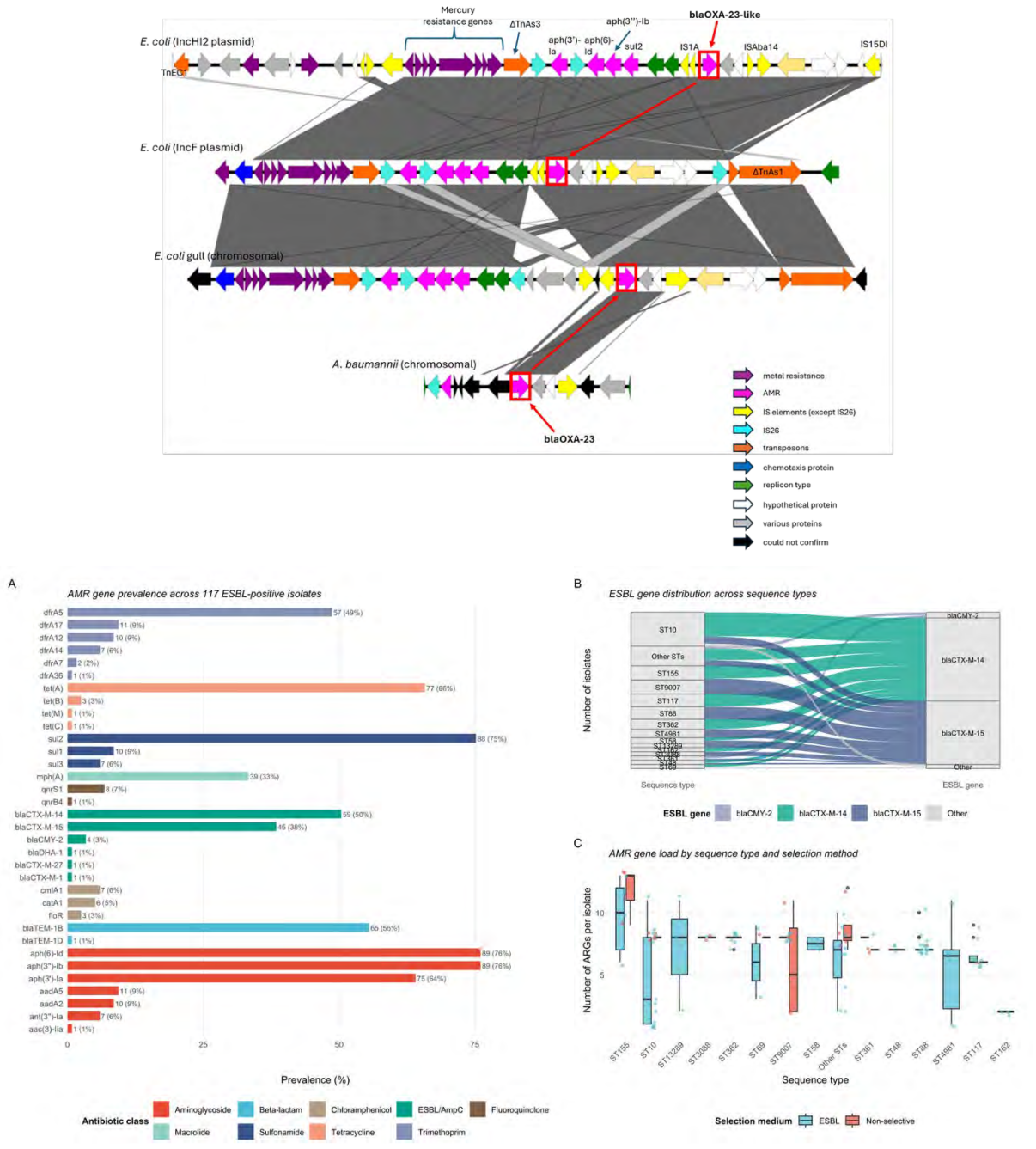

Figure 17-k: Top panel: Genetic context of blaOXA-23 gene in an E. coli plasmid and the comparison with the genetic context of a related gene found in gull. Lower panels: A. The resistome found in rectal swabs from NSW healthy calves. B. ESBL gene distribution across different E. coli ST.

Conclusions/implications:

This study represents the most comprehensive whole-genome sequencing analysis of third-generation cephalosporin–resistant E. coli subpopulations in calves from Australian dairy operations. Although a high prevalence of ESBL-producing E. coli was observed on a herd basis in this early-life cohort, previous studies indicate that such colonization is typically transient in young calves.

Our findings provide important genomic context for interpreting early-life AMR prevalence. Collectively, these data establish a critical genomic baseline for ongoing AMR surveillance and support sustainable antimicrobial stewardship within the dairy sector.

Subproject P2g Heifers Early Calving

Background

We identified in P2a marked declines in health and reproductive performance with increased parity. Further, we found marked differences in metabolite and lipids with increased parity and changes in BCS and BW. In this series of studies we are investigating whether reducing the age at calving can reduce the risks of removal and reproductive failure while maintaining milk production. We also identified that there was considerable potential to reduce the age at calving in some pasture-based herds as these had a much older age for heifers at calving.

Meta-analysis of the effects of age at first calving on production outcomes, calving difficulty, and reproduction in dairy heifers

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Authors

A.K.G. Lean, A.J. Gunn, J.C. Quinn, I.J. Lean, K. Breinhild, H.M. Golder

Background

This study aims to determine the effects of feeding, management, and reproductive strategies used to reduce age at first calving (**AFC**) on production and reproductive performance through meta-analytic methods.

Methods

A literature search using three search engines was conducted to identify prospective studies that evaluated AFC for data extraction. Inclusion criteria included specific dairy breeds, a treatment group with an AFC below 26 mo of age, and prospective enrolment of heifers. There were few prospective studies suitable for meta-analysis, with 16 studies containing 35 appropriate experiments. There was substantial heterogeneity likely due to differences in study design and interventions used to induce an AFC difference. Outcomes were assessed using classical multilevel random effects meta-analytic models with standard mean difference, effect sizes, or risk ratios being evaluated where appropriate. Meta-regression was performed using the difference in age at first calving.

Findings

There was a loss of production for first lactation milk (-2.06 L/d of earlier calving), fat (-0.12 kg/d of earlier calving), and protein (-0.08 of kg/d earlier calving) yield for heifers calving at a younger age than their peers. There were no differences in second and third lactation milk yield, calving difficulty, heifer reproductive performance, survival to calving, and survival to the end of first lactation.

Implications

While the production in first lactation is decreased, this may be offset by fewer days to first calving without compromising the heifers' ability to calve, become pregnant for the first time, or survive to the end of first lactation. Given the lack of prospective studies, more randomized controlled experiments over multiple lactations, including health, production, and reproductive data, are needed to evaluate this topic further.

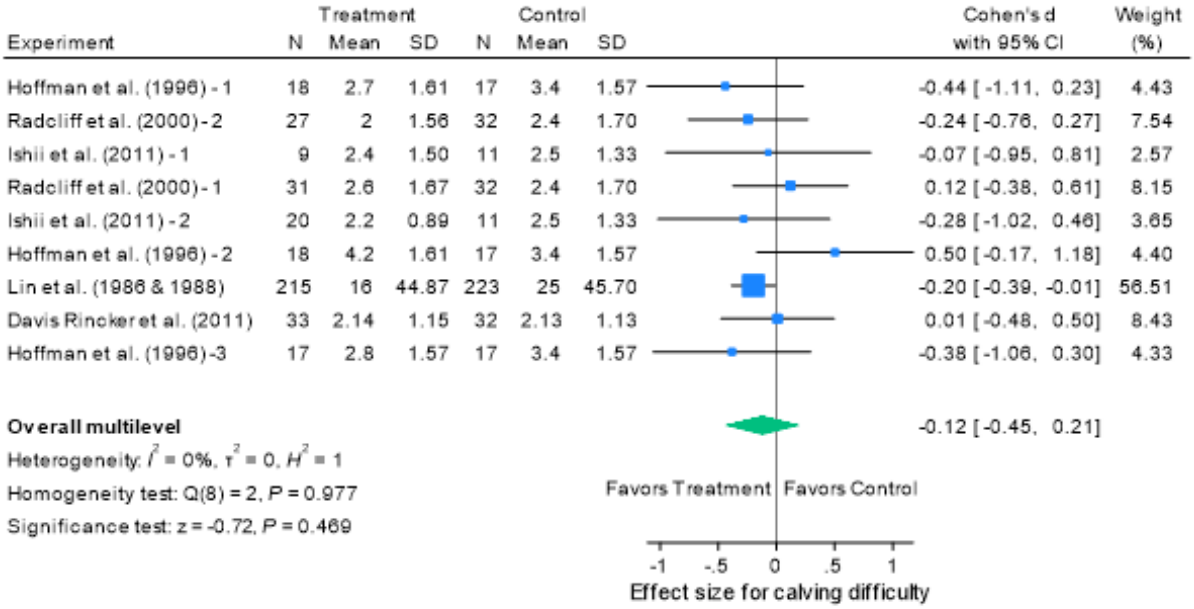


Figure 17.a: A forest plot representing the effect size of calving difficulty. The solid vertical line represents no effect or zero. Points to the left of the line indicate a reduction in calving difficulty, while points to the right indicate increased calving difficulty. Each square on the graph represents the mean effect size for comparison at the univariate level, with the lines extending from the square being the 95% CI of that experiment. The size of the square indicates the weighting of the experiment at a univariate level, the value of which is recorded in the right-hand column. The diamond at the base represents the results from the overall multilevel model, which was non-significant (P = 0.469). The forest plot is ordered by ascending age at first calving (AFC) of the treatment group from the top. There is little heterogeneity between the experiments, as can be seen by the low values of I2 and τ2 and high H2

Age at first breeding in Holstein dairy cattle: Effects on age at first calving, production, dystocia, health and reproduction in primiparous heifers

In Review

Authors

A.K.G. Lean, H.M. Golder, J.C. Quinn, I.J. Lean, A.J. Gunn

Background

Although lowering the age at first calving (**AFC**) for dairy heifers has been investigated over the last 60 years, recent prospective studies are lacking (Lean et al., 2026). Age at first breeding (**AFB**), a driver of AFC, has been less well investigated as a mechanism for production gain. The objective of this study was to investigate AFB and AFC in well-grown modern Holstein dairy heifers and their effects on production, reproduction, and health, with heifers bred from the age of > 10 mo.

Methods

Age at first breeding was investigated by breeding heifers from 5 commercial farms at > 330 kg at younger or older than 13 months of age. Heifers were weighed, and body condition scored at recruitment. Where the farmer elected to breed heifers < 330kg at the same sites, these were included in the study population based on their AFB. A total of 489 heifers were recruited, with heifers entering the normal reproductive programs of the farms; of these, 477 were bred. A second weight was recorded at the transition or periparturient period. Heifers were monitored for reproductive performance, abortions, and survival to calving (< 13 months, N = 249, or > 13 months, N = 228) and AFC group (< 22 months, N = 131, 22-24 months, N = 180 or > 24 months, N = 128). In primiparous lactation, all outcomes were investigated for the effects of AFB, including 305 d milk production per lactating heifer and enrolled heifer, milk fat and protein percentages, days of medical treatment, time to first mating and pregnancy, dystocia, stillbirths, and survival in primiparous lactation and over the whole trial. All outcomes were investigated using multilevel, multivariable models either in the nulliparous or primiparous year with farm location as a random effect. Mixed linear, logistic regression, Poisson regression, and Weibull survival models were used.

Findings

Heifers with an AFB > 13 months had a higher BW at enrolment and increased odds of abortion in their nulliparous year compared with the heifers with an AFB < 13 months. In primiparous lactation, heifers with an AFB > 13 months an increased hazard of exit from the herd, a decreased risk for medical treatment and produced 814 L less milk over 305 d per enrolled heifer when compared with the heifers with an AFB < 13 months. Each kg of extra BW at enrollment was associated with 13.0 L of milk over a 305 d lactation. There was no difference in milk production per lactating heifer between the AFB groups.

When the data were analyzed for AFC groups, there was no difference between groups for milk production per lactating or enrolled heifer. There was a decrease in fat percentage in older AFC heifers when compared to the < 22 mo AFC group. The 22-24 mo AFC group had a decrease of 0.18 % milk fat, while the > 24 mo AFC group had a decrease of 0.28 % fat in the milk. There was a decreased risk of medical treatment for heifers having a female calf and an AFC of > 24 mo when compared to those with an AFC of < 22 mo. Odds of stillbirth were decreased with an AFC of > 24 mo compared to < 22 mo.

Implications

Lowering the recommended AFB for well-grown (> 330kg) heifers to 10-13 months of age may therefore be of benefit as a management strategy, though risks of increased medical treatment and stillbirth must be considered.

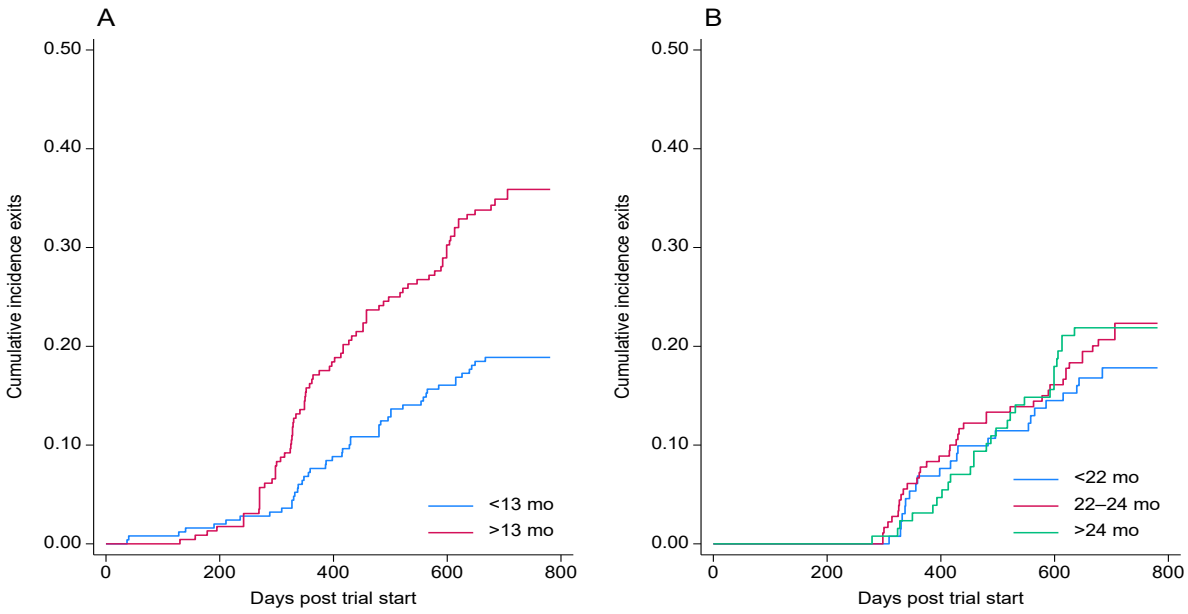


Figure 17.b: Kaplan Meier failure graphs illustrating the cumulative incidence of exits by age at first breeding (Figure A) and age at first calving (Figure B). As heifers exit the proportion of heifers exiting increases illustrating exits over time. Heifers can only have an age at first calving after reaching calving, resulting in fewer heifers and a later start time in Figure B. In Figure A the groups are age at first breeding < 13 mo and > 13 mo. In Figure B the groups are age at first calving < 22 mo, 22-24 mo, and > 24 mo.

Age at first breeding in Holstein dairy cattle: Associations of hormone, biochemical and protein measures - *In Review*

Authors

A.K.G. Lean, H.M. Golder, J.C. Quinn, I.J. Lean, A.J. Gunn

Background

Age at first breeding (**AFB**) has traditionally been targeted to 15 mo of age to allow for calving before the spring flush in seasonal calving systems. Despite this being an industry norm for many decades, few studies have evaluated metabolism and AFB and insights are needed to help optimize management strategies. The objective of this study is to investigate heifer reproductive and growth performance by assessing the BW, BCS, presence of a corpus luteum (**CL**), time to pregnancy and survival to first lactation in relation to hormonal, biochemical, and protein metabolism as well as AFB. Understanding these interactions may enable the selection of heifers for mating at earlier ages based on metabolite status and BW, markers that could be readily tracked for performance traits in dairy systems.

Methods

Holstein dairy heifers (N = 411) sourced from 4 commercial farms were aged from 11 to 18 mo of age at first breeding. Heifers were sourced from 4 commercial farms that fed total mixed rations. Most utilized pasture feeding for the heifers at times. Heifers were weighed, BCS scored and enrolled for immediate breeding if > 330 kg in BW and assigned to mating groups of < 13 mo or > 13 mo at first breeding. Of 411 heifers enrolled into the trial, a subset of 149 heifers were systematically blood sampled at the time of weighing. A metabolic profile was tested consisting of total protein, albumin, globulin, urea, creatinine, glucose, Ca, Mg, P, Cu, Zn, vitamin B12, glutathione peroxidase, leptin, insulin growth factor (**IGF**)-1, IGF binding protein 6 (**IGFBP-6**), insulin, undercarboxylated osteocalcin, and osteocalcin. A further subset of 76 heifers from this group were tested for the presence of a corpus luteum. Heifers were monitored for time to pregnancy, age at first calving and survival. The effects of AFB and the above metabolites were investigated for associations with the BW, BCS, presence of a corpus luteum, reproductive performance and survival at first calving. Analysis applied multilevel mixed linear, logistic regression and Weibull survival models with farm as a random effect.

Findings

No difference was found between the AFB groups for the BCS, presence of a corpus luteum, time to pregnancy or survival. Older heifers had a heavier BW. Albumin and Mg were positively associated with heifer BW, whereas vitamin B12 and log insulin were negatively associated. There were positive associations for urea, BW, log IGF-1 and P with BCS. Non-esterified fatty acid, Mg and vitamin B12 were associated negatively with Cu, and vitamin B12 were positively associated with the presence of a corpus luteum. Globulin and glutathione peroxidase were negatively associated with time to pregnancy. There were no metabolic associations with survival.

Implications

The negative association of NEFA with the presence of a CL highlights the importance of effective nutrition for growth in heifers so they are ready for mating. Blood Cu may have importance in the onset of puberty and needs further investigation. These results give insights into the metabolic influences on Holstein dairy heifers at first breeding.

Table 17.a: Univariable results from analysis of time to pregnancy, survival, presence of the corpus luteum, BW, and BCS with a random effect of farm. Body weight and blood test results have been centered by farm for the analysis.

Variable	**Time to pregnancy**			**Survival at calving**			**Corpus luteum**		
Statistical model	**Weibull Survival**			**Weibull Survival**			**Logistic regression**		
	Hazard ratio	**SE**	***P*-value**	**Hazard ratio**	**SE**	***P*-value**	**Odds ratio**	**SE**	***P*-value**
Age at first breeding (d)	1.004	1.004	0.038	1.003	0.006	0.631	1.002	0.008	0.820
Breeding group (< 13 mo^{ref} or > 13 mo of age)	1.211	0.244	0.342	2.733	1.784	0.123	0.623	0.05	0.982
BW at blood test (kg)	1.003	0.002	0.118	0.995	0.009	0.598	0.993	0.008	0.374
Total protein (g/L)	0.986	0.012	0.259	0.997	0.044	0.950	1.011	0.042	0.801
Albumin (g/L)	0.990	0.038	0.801	0.838	0.133	0.185	0.936	0.132	0.614
Globulin (g/L)	0.981	0.015	0.198	1.024	0.053	0.652	1.025	0.050	0.623
Urea (mmol/L)	0.971	0.111	0.794	0.717	0.323	0.303	1.300	0.317	0.408
Glucose (mmol/L)	1.032	0.153	0.839	1.018	0.569	0.975	0.847	0.590	0.778
Creatinine (μmol/L)	0.996	0.008	0.558	0.970	0.024	0.203	0.969	0.026	0.218
NEFA (mmol/L)	2.738	0.465	0.030	0.036	2.671	0.213	0.001	2.666	0.012
Leptin (ng/ml)	1.020	0.013	0.143	1.038	0.049	0.439	0.985	0.072	0.827
Undercarboxylated Osteocalcin (ng/ml)	1.242	0.114	0.056	1.139	0.384	0.734	0.943	0.310	0.850
Osteocalcin (ng/ml)	1.007	0.004	0.051	1.003	0.016	0.826	1.003	0.019	0.882
Ca (mmol/L)	0.782	0.659	0.709	2.032	1.974	0.719	0.429	2.003	0.672
Mg (mmol/L)	17.712	1.084	0.008	0.253	3.524	0.696	0.001	3.544	0.043
Ca to Mg ratio	0.459	0.328	0.017	1.724	1.003	0.587	4.749	1.016	0.125
P (mmol/L)	1.096	0.305	0.764	0.938	1.076	0.953	1.626	1.024	0.635
Glutathione peroxidase (U/gHB)	0.996	0.002	0.066	0.993	0.007	0.283	0.976	0.012	0.035
Cu (μmol/L)	1.002	0.044	0.956	1.064	0.146	0.669	1.204	0.134	0.166
Zn (μmol/L)	0.968	0.039	0.410	1.004	0.126	0.974	1.277	0.124	0.048
Vitamin B12 (pmol/L)	0.997	0.002	0.098	1.007	0.006	0.204	0.990	0.006	0.080
Log Insulin (μmol/L)	0.944	0.088	0.511	1.370	0.337	0.351	0.945	0.274	0.838
Log IGF1(ng/L)	1.009	0.086	0.918	0.998	0.302	0.995	1.121	0.260	0.661
Log IGFBP-6 (ng/ml)	0.913	0.089	0.305	0.842	0.335	0.608	0.804	0.387	0.574

Project-wide Dissemination

The project findings have been disseminated through multiple formal research channels, including peer-reviewed publications, conference oral and poster presentations, and PhD theses. Two PhDs have been completed by Sarah Legge and David Sheedy, with Andrew Lean in the final stages of completion. Academic and industry conferences where findings have been presented include the American Dairy Science Association Annual Meeting, the Dairy Research Foundation Symposium, the Australian Cattle Veterinarians Conference, the ANZCVS Vet Science Week, the and International Society for Veterinary Epidemiology and Economics Symposium.

Findings have also been communicated directly to industry through Dairy UP workshops, Dairy UP project update meetings, field-based activities, producer and advisor engagement, and industry events including Raising the Roof, Appetite for Success and Herd '25. International visibility was strengthened through Dairy UP's special symposium, The DairyUp Project: Attacking Major Challenges for Dairy in a Multidisciplinary Project, at the American Dairy Science Association Annual Meeting in 2025, helping showcase Australian dairy research to a global audience.

Table: Peer-reviewed publications.

Authors	Title	Journal	Year Published	Citations (to April 26)
I.J. Lean, H.M. Golder, S.J. LeBlanc, T.F. Duffield, and J.E.P. Santos	Increased parity is negatively associated with survival and reproduction in different production systems	Journal of Dairy Science	2022	35
Lean, I.J., D.B. Sheedy, S.J. LeBlanc, T. Duffield, J.E.P. Santos, and H.M. Golder	Holstein dairy cows lose body condition score and gain body weight with increasing parity in both pasture-based and total mixed ration herds	Journal of Dairy Science Communications	2022	12
Lean, I.J., S.J. LeBlanc, D.B. Sheedy, T. Duffield, J.E.P. Santos, and H.M. Golder	Associations of parity with health disorders and blood metabolite concentrations in Holstein cows in different production systems	Journal of Dairy Science	2023	96
Brito, B., and P. Hick	Milk as a diagnostic fluid to monitor viral diseases in dairy cattle	Australian Veterinary Journal	2024	14

Lean, I.J., and H.M. Golder	Milk as an indicator of dietary imbalance	Australian Veterinary Journal	2024	6
Rowe, S., J.K. House, and R.N. Zadoks	Milk as diagnostic fluid for udder health management	Australian Veterinary Journal	2024	4
Legge, S.W.J., P.C. Thomson, C.E.F. Clark, and S.C. García	Milk consumption and behavior of calves in automated calf feeders as early indicators of weaning liveweight	Journal of Dairy Science Communications	2024	1
Lean, I., R. Zadoks, B. Brito, and H. Golder	Milk as a diagnostic fluid	Australian Veterinary Journal	2024	-
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Moate, P. Reddy, S.J. Rochfort, J.E. Pryce, and I.J. Lean	A large multisite lipidomic investigation of parity and aging in dairy cows	Journal of Dairy Science	2025	6
Sheedy, D.B., H.M. Golder, S.C. Garcia, P. Reddy, J.E. Hemsworth, D.E. Vincent, S.J. Rochfort, J.E. Pryce, and I.J. Lean	Associations among body condition score, body weight, and serum biochemistry in dairy cows	Journal of Dairy Science	2025	3
Golder, H.M., J. Rehberger, A.H. Smith, E. Block, R. Polkinghorne, H.E. Cuthbertson, M.A. Campbell, V. Vicic, G. Tarr, J.C. Quinn, J.K. Tong, S.M. Rowe, and I.J. Lean	Different lifetime dietary strategies affect carcass characteristics and rumen function in Holstein steers	Animal Production Science	2025	-

Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Hemsworth, D.E. Vincent, J.E. Pryce, and I.J. Lean	Confinement and pasture-based dairy herds differ in plasma lipid profiles	Journal of Dairy Science	2025	-
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Hemsworth, D.E. Vincent, J.E. Pryce, and I.J. Lean	A large, multisite investigation into the lipidomics of survival in dairy cows	Journal of Dairy Science	2026	I
Sheedy, D.B., H.M. Golder, S.C. Garcia, and I.J. Lean	Glycerophospholipids in dairy cow health and longevity: a review	Journal of Dairy Research	2026	-
Lean, A.K.G., A.J. Gunn, J.C. Quinn, I.J. Lean, K. Breinhild, and H.M. Golder	Meta-analysis of the effects of age at first calving on production outcomes, calving difficulty, and reproduction in dairy heifers	Journal of Dairy Science	2026	-
Abedien, Z.U., I.J. Lean, S.P. Djordjevic, P.M. Hick, M.E. Westman, J. Mckay-Demeler, J. Webster, and B.P. Brito	Next-generation detection in bovine respiratory and enteric diseases: metagenomic and amplicon sequencing insights into microbial diversity	Frontiers in Veterinary Science	2026	-

Annexes

Table: Conference presentations, abstracts and other meetings

Authors	Title	Presentation Type	Conference/ Event	Location	Year
B. Brito, Z.U. Abedien, Y. Kida, J. Webster, S. Djordjevic, E. Holmes, and I.J. Lean	Epidemiology 2.0: Metagenomic Pathogen Detection in NSW Dairy Calves - High-Resolution Mapping of Respiratory and Enteric Microbial Populations	Oral, conference	Australia and New Zealand College of Veterinary Scientist Vet Science Week	Gold Coast, QLD	2026
Sheedy, D.B., H.M. Golder, S.C. Garcia, A.K.G. Lean, and I.J. Lean	Reproduction, mastitis, and lameness in confinement and pasture-based systems: Associations with parity	Poster, conference	American Dairy Science Association Annual Meeting	Milwaukee, USA	2026
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Hemsworth, D.E. Vincent, J.E. Pryce, and I.J. Lean	Challenges and consequences of dairy cow longevity: Genetics, management, health and assessing the economics	Oral, conference	Australian Cattle Veterinarian Conference	Brisbane, QLD	2026
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Pryce, and I.J. Lean	System-level bias in lipidomic biological clock models imply greater metabolic age in confinement vs pasture-based cows	Poster, conference	American Dairy Science Association Annual Meeting	Milwaukee, USA	2026
Stanczak, A., B. Brito, S. Hem, I.J. Lean, V. Jarocki, and S. Djordjevic	Detection of blaCTX-M genes in Escherichia coli in dairy calves in Australia	Oral, conference	Environmental Dimensions of Antimicrobial Resistance Conference	Brisbane, QLD	2026
Abedien, Z.U.	Cracking the Code of Calf Health: Insights from Enteric Microbes	Oral, conference	Dairy Research Foundation Symposium	Wollongong, NSW	2025
Abedien, Z.U., S. Djordjevic, I.J. Lean, J. Webster, and B. Brito	Microbial diversity and gene expression using meta-transcriptomics study, an insight to untargeted approach in dairy calves	Poster, conference	American Dairy Science Association Annual Meeting	Louisville, USA	2025

Abedien, Z.U., S. Djordjevic, I.J. Lean, J. Webster, and B. Brito	Meta-transcriptomic insight into resistome, virulome, and microbial diversity in calves	Oral, conference	Australia and New Zealand College of Veterinary Scientist Vet Science Week	Gold Coast, QLD	2025
Brito, B.	When bugs go bad- who is attacking your calves?	Oral, conference	Dairy Research Foundation Symposium	Wollongong, NSW	2025
Brito, B.	NSW dairy calves microbial genomics study- preliminary findings	Oral, conference	Elizabeth Macarthur Agricultural Institute Lunch and Learn Seminar	Menangle, NSW	2025
Brito, B., Z.U. Abedien, and I.J. Lean	The enteric and respiratory viral diversity of calves in health and disease—A state-wide metatranscriptomic study.	Oral, conference	American Dairy Science Association Annual Meeting	Louisville, USA	2025
Golder, H.M., and I.J. Lean	Investigating genotype and environment effects on the rumen microbiome	Oral, conference	American Dairy Science Association Annual Meeting	Louisville, USA	2025
Kida, Y., Z.U. Abedien, I.J. Lean, and B. Brito	Molecular epidemiology of Rotavirus in New South Wales dairy calves	Poster, conference	American Dairy Science Association Annual Meeting	Louisville, USA	2025
Kida, Y., Z.U. Abedien, I.J. Lean, and B. Brito	Comprehensive analysis of genetic diversity in bovine rotavirus	Poster, conference	Australia and New Zealand College of Veterinary Scientist Vet Science Week	Gold Coast, QLD	2025
Kida, Y., Z.U. Abedien, I.J. Lean, and B. Brito	Comprehensive analysis of genetic diversity in bovine rotavirus	Oral, conference	Australia and New Zealand College of Veterinary Scientist Vet Science Week	Gold Coast, QLD	2025
Lean, A.K.G, H.M. Golder, J. Quinn, I.J. Lean, and A. Gunn	Meta-analysis of the effects of age at first calving on production outcomes, calving difficulty, and reproduction in dairy heifers	Oral, conference	Australia and New Zealand College of Veterinary Scientist Vet Science Week	Gold Coast, QLD	2025

Lean, A.K.G, H.M. Golder, J. Quinn, I.J. Lean, D.B. Sheedy, and A. Gunn	How early should you calve your heifers?	Oral, conference	Dairy Research Foundation Symposium	Wollongong, NSW	2025
Lean, A.K.G, H.M. Golder, J. Quinn, I.J. Lean, D.B. Sheedy, and A. Gunn	Evaluation of the effects of an enzymatic and biological additive on bedding in compost bedded pack barns in Australia	Oral, conference	American Dairy Science Association Annual Meeting	Louisville, USA	2025
Lean, I.J.	Unlocking the potential of the cow; adaptation to achieve profit	Oral, work shop	Appetite for Success	Sale, Inverloch and Warrnambool, VIC	2025
Lean, I.J.	Calcium metabolism and practical transition management	Oral, conference	New Zealand Veterinary Association	Wellington	2025
Lean, I.J., H.M. Golder, A.K.G. Lean, and D. Sheedy	Harvesting the power of the cow	Oral, conference	Dairy Research Foundation Symposium	Wollongong, NSW	2025
Lean, I.J., H.M. Golder, B. Brito, N. Moss, D. Sheedy, A.K.G. Lean, S. Garcia, C. Clark, A. Gunn, and J. Quinn	You don't eat the coat! How does beef on dairy stack up?	Oral, conference	Herd '25	Bendigo, VIC	2025
Lean, I.J., H.M. Golder, D.B. Sheedy, C. Old, and A.K.G. Lean	Case definition and metabolic disorders: More accurate phenotypes	Oral, conference	American Dairy Science Association Annual Meeting	Louisville, USA	2025
Legge, S.W.J., P.C. Thomson, C.E.F. Clark, and S.C. García	The positive associations between calf and lactation performances in dairy cattle	Oral, conference	American Dairy Science Association Annual Meeting	Louisville, USA	2025
Legge, S.W.J., P.C. Thomson, C.E.F. Clark, and S.C. García	Identifying key factors affecting dairy cow longevity	Oral, conference	Dairy Research Foundation Symposium	Wollongong, NSW	2025
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort,	Do cows in total mixed ration systems have old lipid profiles compared with pasture-based cows?	Oral, conference	American Dairy Science Association Annual Meeting	Louisville, USA	2025

J.E. Hemsworth, D.E. Vincent, J.E. Pryce, and I.J. Lean					
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Hemsworth, D.E. Vincent, J.E. Pryce, and I.J. Lean	Life and death in pasture and TMR farms - the fat?	Oral, conference	Dairy Research Foundation Symposium	Wollongong, NSW	2025
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Pryce, and I.J. Lean	Variable stabilization in adaptive lasso cox frailty models to explore lipidomic associations with survival of dairy cows.	Oral, conference	Animal Science Modellers Special Interest Group	Louisville, USA	2025
Stanczak, A., B. Brito, S. Hem, I.J. Lean, V. Jarocki, and S. Djordjevic	Molecular Characterisation of blaOXA-23 genes identified during a genomic survey of Escherichia coli in Australian dairy cattle.	Oral, conference	MedVetPathogens - FEMS	Prato, Italy	2025
Sheedy, D.B.	P2a Project Updates	Oral, meeting	Dairy UP Teams Meeting	On-line	2023 2024 2025
Brito, B.	P2f Project Updates	Oral, meeting	Dairy UP Teams Meeting	On-line	2022 2023 3x 2025
A.K.G, H.M. Golder, J. Quinn, I.J. Lean, D.B. Sheedy, and A. Gunn	Age at first calving	Oral, conference	Australia and New Zealand College of Veterinary Scientist Vet Science Week	Gold Coast, QLD	2024
Golder, H.M.	P2c Project Updates	Oral, meeting	Dairy UP Teams Meeting	On-line	2024
Lean, A.K.G, H.M. Golder, J. Quinn, I.J. Lean, and A. Gunn	Assessment and management of dairy compost bedded pack barns	Oral, conference	Australia and New Zealand College of Veterinary Scientist Vet Science Week	Gold Coast, QLD	2024

Lean, A.K.G, H.M. Golder, J. Quinn, I.J. Lean, and A. Gunn	Understanding and assessing compost bedded pack dairy barns	Oral, conference	Australian Cattle Veterinarian Conference	Wodonga, VIC	2024
Lean, A.K.G, H.M. Golder, J. Quinn, I.J. Lean, and A. Gunn	Managing and improving loose housing facilities with a compost bedded pack	Oral, conference	Raising the Roof	Cessnock, NSW	2024
Lean, I.J., H.M. Golder, B. Brito, N. Moss, D. Sheedy, A.K.G. Lean, S. Garcia, C. Clark, A. Gunn, and J. Quinn	Unlocking the potential of the cow	Oral, conference	Raising the Roof	Cessnock, NSW	2024
Sheedy, D.B., H.M. Golder, S.C. Garcia, and I.J. Lean	Metabolic investigation into dairy cow longevity	Oral, conference	School of Life & Environmental Sciences Higher Degree Research Showcase	Sydney, NSW	2024
Sheedy, D.B., H.M. Golder, S.C. Garcia, and I.J. Lean	Survival and longevity	Oral, conference	Raising the Roof	Cessnock, NSW	2024
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Pryce, and I.J. Lean	Associations among body condition score, body weight and serum biochemistry in dairy cows	Poster, conference	American Dairy Science Association Annual Meeting	West Palm Beach, USA	2024
Sheedy, D.B., H.M. Golder, S.C. Garcia, Z. Liu, P. Reddy, S.J. Rochfort, J.E. Pryce, and I.J. Lean	Associations among body condition score, body weight and serum biochemistry in dairy cows	Poster, conference	International Society for Veterinary Epidemiology and Economics	Sydney, NSW	2024
Brito, B.	The bugs of calves- a state-wide study of endemic infectious diseases	Oral, work shop	Dairy UP workshops	Bega, Bodalla, Casino	2023
Brito, B., H.M. Golder, and I.J. Lean	The respiratory infectome of dairy calves characterized by a total RNA sequencing approach.	Oral, conference	American Dairy Science Association Annual Meeting	Ottawa, CAN	2023

Brito, B., H.M. Golder, E. Wrysch, S. Djordjevic, and I.J. Lean	Expression of virulence factors and antimicrobial resistant genes in total RNA sequenced from rectal swabs from diarrheic calves	Poster, conference	American Dairy Science Association Annual Meeting	Ottawa, CAN	2023
Brito, B., H.M. Golder, E. Wrysch, S. Djordjevic, J. Rothwell, and I.J. Lean	Untargeted metatranscriptomic methods to characterize the enteric infectome of calves with and without diarrhea	Poster, conference	American Dairy Science Association Annual Meeting	Ottawa, CAN	2023
Lean, A.K.G, H.M. Golder, J. Quinn, I.J. Lean, D.B. Sheedy, D. Lewis, and A. Gunn	Assessment and microbial enhancement of dairy compost-bedded pack barns	Oral, conference	Charles Sturt University Faculty of Science and Health Higher Degree by Research and Honours Symposium	Wagga Wagga, NSW	2023
Lean, I.J, A.K.G. Lean, and D. Sheedy	Cow longevity and metabolomics	Oral, work shop	Dairy UP workshops	Bega, Bodalla, Casino	2023
Sheedy, D.B., H.M. Golder, S.C. Garcia, and I.J. Lean	Insights into exit reasons between intensively housed and pasture-based dairy systems	Poster, conference	School of Life & Environmental Sciences Higher Degree Research Showcase	Sydney, NSW	2023
Sheedy, D.B., H.M. Golder, S.C. Garcia, and I.J. Lean	The metabolomics of dairy cow longevity	Oral, conference	Dairy Research Foundation Symposium	Camden, NSW	2023
Brito, B.	Infectious diseases calves, kikuyu poisoning	Oral, conference	District Veterinarians Conference	Camden, NSW	2022
Sheedy, D.B., H.M. Golder, S.C. Garcia, and I.J. Lean	A metabolomic investigation into Australian dairy cow longevity.	Oral, conference	School of Life & Environmental Sciences Higher Degree Research Showcase	Sydney, NSW	2022

Table: Technical reports, pamphlets, published material and other media engagements

Authors	Title	Place Published	Year
	P2f fact sheets https://dairyup.com.au/project/p2f-factsheets/		

Conclusion and Recommendations

Dairy UP Project 2 has delivered a substantial and integrated body of work addressing dairy cow health, productivity, welfare and longevity across multiple life stages and production systems. The project has generated knowledge through a broad program of database development, retrospective and prospective studies, lipidomics, milk diagnostics, heat stress research, molecular infectious disease investigations, calf and heifer studies, and housing system studies. Collectively, these outputs demonstrate both the breadth and depth of the project and its value to the Australian dairy industry.

The project has achieved strong research and industry dissemination, with outcomes communicated through peer-reviewed publications, industry presentations, conference abstracts, field days and direct engagement with producers and advisors. The national and international relevance of the work is reflected in the breadth of publications and presentations, including Dairy UP's presence as a special symposium, The DairyUp Project: Attacking Major Challenges for Dairy in a Multidisciplinary Project, at the America Dairy Science Association annual meeting in 2025. This visibility has helped place Australian dairy research on the global stage.

Specific outcomes for the projects include;

- A clear understanding of the increased risks of disease and metabolic disorder with increased parity of cows. This will provide farmers and advisors with clarity around the increased risks with increased parity of cows.
- Identifying, that there are marked differences in lipid profiles with age of cows and that cows in housed facilities have older profiles than those at pasture.
- These differences suggest means by which the problems of aging might be modified with lipid supplementation.
- Cows lose body condition but gain weight as they age, indicating a pathway to reducing risks of problems in older cows.
- We identified differences in herd removals between housed and pastured herds which will help farmers and advisors understand the benefits and risks from different systems.
- We identified methods to improve bedding management in compost bedded pack barns.
- An early alert system for bovine ephemeral fever was established.
- Milk has great value as a diagnostic fluid and is under-utilised by the Australian industry, Milk urea nitrogen will be very valuable to routinely report.
- An important finding was to quantify the lagged influence of diet on milk production highlighting that nutritional responses to diet occur over days to weeks.
- Diets differing in formulation provided the potential to integrate dairy calves into the beef supply chain but with different rumen lifetime adaptations.
- Greater milk consumption in early life (up to day 5) had substantial effects on weaning weight of calves.
- Holstein cattle achieved excellent feedlot performance compared to beef breeds. High performing cattle at feedlot induction were higher performing overall compared to those with lesser early performance.

- Metagenomic and amplicon sequencing methods will change understandings and improve accuracy of veterinary diagnostic testing.
- Calf diarrhoea is rarely caused by a single pathogen and is substantially explained by the farm environment. This reinforces the importance of good husbandry for farmers and advisors.
- Kobuvirus was identified as an emerging pathogen associated with diarrhoea.
- Pestivirus, while of moderate prevalence, was a substantial pathogen. This can be controlled with vaccination.
- Insights were gained on host responses to potential pathogens providing avenues for future control of disease without using antimicrobial treatments.
- Similarly respiratory disease risks were dominated by farm environment. Rhinitis-viruses and Torovirus were identified as pathogens. Again, disease occurs as result of interactions among potential pathogens.
- Host responses to pathogens were an important part of the pathogenesis of the disease.
- Reducing the age at first mating of heifers weighing >330 kg to 10 to 13 months as opposed to mating at >13 months resulted in > 800 L greater milk production in the first lactation as a result of more heifers entering and remaining in the herd. This has implications for costs of rearing and green-house gas footprint.
- Heifers with higher blood free fatty acids had a lesser risk of being actively cycling indicating the importance of a positive nutritional plane near mating.

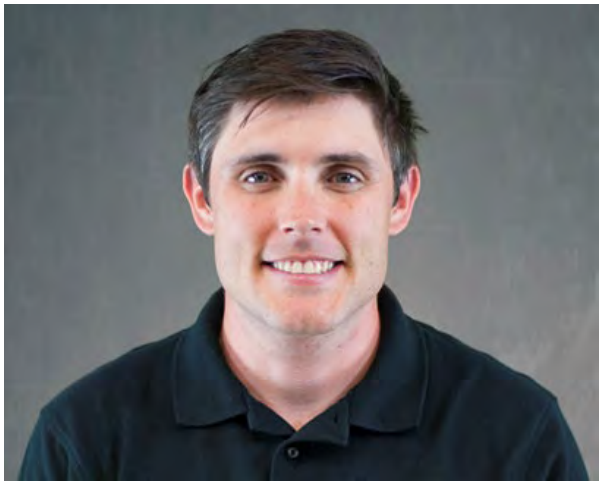
Further investment is recommended to build on the momentum created by Project 2. Support is needed for extension activities that translate findings into practical farm decision-making, and for early- and mid-career researchers who have developed expertise through the program. Continued support would help retain research capacity, strengthen industry relationships and ensure that the knowledge generated through Dairy UP is converted into measurable benefits for farmers, veterinarians, nutritionists and the wider dairy sector.

A key priority for future funding should be to move from exploratory findings to targeted implementation studies. Several projects have identified promising areas for intervention, including feeding strategies, omega fatty acid biology, heat stress mitigation, heifer development, housing design and early warning systems. The risk of inaction is that the collaborations, data infrastructure, research capacity and industry momentum developed through Dairy UP will not be fully realised. Continued investment would allow the program to progress from discovery to practical application, supporting a more productive, resilient and socially sustainable Australian dairy industry.

Final Project Summary

P2a: Metabolic investigations into dairy cow longevity

May 2026



Longevity matters

Cows tend to produce more milk with each lactation: older cows produce about 16% more milk than their younger counterparts.

But cows in Australian dairy herds are young. Only 10% of cows produce milk for five lactations or more and about 60% of the average national milking herd has calved twice or less.

Optimising the productive life of dairy cows ensures dairy farm businesses receive the best return on their investment from rearing the calf; it also increases milk production and reduces the environmental footprint of individual cows.

David Sheedy's PhD investigated metabolic markers that could be associated with longevity in dairy cows.

Key findings

Cows in housed systems were metabolically older than cows of the same age in pasture-based systems. Further work is needed to determine if this has implications for cow health and longevity in different systems.

Collectively, the findings from this work and earlier studies suggest that targeting the fat profile of diets could help optimise longevity in Australian dairy herds. Specifically, supplementary omega-3 warrants further investigation.

Unlocking the potential of cows

Dairy UP's P2 project aimed to unlock the potential of dairy cows to achieve their genetic potential under NSW conditions.

P2 was a suite of seven projects that collectively explore ways to profitably increase both productivity and wellbeing in commercial settings.

P2a: Cattle Longevity
P2b: Early Alerts
P2c: Milk as a Diagnostic Tool
P2d: Diet and Heat Load
P2e: Calf Husbandry
P2f: Infectious Diseases ('Infectome')
P2g: Heifers Early Calving

This document provides a final summary of the findings from research undertaken by David Sheedy for his PhD thesis which contributed to Project P2a.

Benefits

Retaining the best dairy cows in herds for longer is expected to make dairy businesses more profitable and less risky.

Thanks to technology such as sexed semen and fixed time artificial insemination, retaining more older cows could also provide the opportunity for a dairy business to produce more dairy-beef.

Increasing the proportion of older cows in a dairy herd would also ensure that dairy farm businesses receive a return on the investment in rearing these calves. A full return for the cost of rearing a dairy heifer often isn't realised until she calves for the third time.

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From an industry and farm perspective, reducing the "wastage" of older cows is good news for dairy's environmental footprint. The more milk a cow produces over its lifetime, the less its environmental emissions intensity.

In addition, there are expected industry 'social licence' benefits from optimising the longevity of dairy cows.

Research approach

This project established a unique large-scale data set, created by a partnership between DataGene and Scibus. It included herd test data and farm management records from selected Australian dairy herds with excellent quality records. Weather, bulk milk tank and pasture data were also included.

Twenty-nine Australian dairy herds provided farm records for inclusion in the data set. Fourteen of these herds were pasture-based and 15 operated contained housing systems.

The project also involved metabolomics in collaboration with researchers at AgriBio, Victoria. Metabolomics is the large-scale study of small molecules called metabolites within each cow and can provide insights into health and longevity.

Blood samples were taken from about 1700 cows (half at peak milk production and half dry) for metabolomic analysis.

Novel statistical methods were used to gain insights into relationships between blood fats and cow age/parity, feeding system and longevity.

Results

1. Health and parity

The results confirmed findings from earlier studies that used previous health and reproduction data from 13 studies conducted in Australia, Canada, and the USA.

As cows age (especially beyond the second lactation):

- Pregnancy risk decreases (they are harder to get in calf)
- Mastitis risk increases.
- Lameness risk increases.
- Survival (culling or mortality) risk increases.

2. Body condition and body weight

As cows age, they gain weight but lose body condition. This occurs in both pasture-based and contained housing systems. The exception was cows entering their second lactation, who often had the lowest body condition and low body weight.

Albumin, a blood protein, was strongly associated with high body condition and body weight.

This finding paves the way to develop nutritional interventions (protein metabolism) to improve body condition in older cows.

3. Blood lipids (fats) and age

This work investigated 185 blood lipids in cows of different ages.

The results showed that older dairy cows had lower levels of omega 3 fatty acids in their blood.

This is an important finding as omega 3 fatty acids have been associated with improved reproduction, health and possibly survival in cattle.

4. Blood lipids (fats) and survival/longevity

To investigate survival, cows that had blood samples taken were followed for about 600 days and all exit reasons were recorded.

Multiple lipids were associated with the timing of cow removal. Importantly, this included the omega-3 fatty acids that were associated with increased survival time.

5. System differences – lipids and health

Herds in contained housing had higher levels of mastitis compared to pasture-based herds.

However, there was no difference in reproductive performance or lameness. Increasing parity had a larger impact on these metrics than the effect of housing system.

Cows in contained housing systems had lower blood levels of omega 3 fatty acids and higher omega 6 compared with pasture-based cows of the same age.

The differences in omega fatty acid profiles are

likely to be associated with maize silage which is the main forage source in contained housing TMR diets. Maize silage is comparatively low in omega 3 fatty acids and high in omega 6, while fresh pasture has high omega 3 and low omega 6 content. A different lipid profile between housing systems may imply different survival risks between systems.

Cows in housed systems were metabolically older than cows of the same age in pasture-based systems. Further work is needed to determine if this has implications for cow health and longevity in different systems.

Collaborators

The data base used for this work was developed through a collaboration between researchers from Dairy UP, Sc*ibus*, DataGene, AgriBio and the participating commercial farms.

Read more

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Delivery organisations

The University of Sydney – Dairy Research Foundation | NSW Government Department of Primary Industries and Regional Development | Scibus | Dairy Australia

Partner organisations

NSW Farmers Growing the best | eastAUSmilk | Dairy NSW / Dairy Australia | AFMH | Leppington Pastoral Company Pty Ltd | NSW Local Land Services | Norco

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Glycerophospholipids in dairy cow health and longevity: a review

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Review Article

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Abstract

This review examines the role glycerophospholipids (**PL**) in dairy cow health, with specific focus on phosphatidylcholine (**PC**), phosphatidylethanolamine (**PE**), phosphatidylinositol (**PI**) and phosphatidylserine (**PS**). Increasing parity of cows is associated with lower concentrations of plasma PL that contain very long-chain omega-3 fatty acids, including docosahexaenoic acid and eicosapentaenoic acid, which are precursors for prostaglandin synthesis, and have anti-inflammatory roles. Low concentrations of these PL could plausibly contribute to the increased risk of disease, reproductive failure and mortality in older cows. The bioavailability and metabolism of fatty acids may differ among supplements that are predominately neutral lipids, such as triacylglycerol-rich oils, and those bound to PL including pasture, whole or ground oilseeds and fish meal. Hepatic lipidosis can occur during the transition period if there is insufficient very-low density lipoproteins (**VLDL**) production in the liver to transport lipids into blood circulation. The PC are the primary PL of VLDL and are produced by two main pathways in the liver, the cytidine diphosphate-choline pathway that uses choline as a substrate, and the PE *N*-methyltransferase pathway that uses PE and methyl-donors as substrates. Co-supplementation strategies that target both pathways may increase PC production over a one-pathway supplementation strategy. The PIs are phosphoinositides precursors, which have broad physiological roles including regulating inflammatory processes and may offer targets for novel treatment and management of disease. Both the PI and PE are precursors to endocannabinoids, important regulators of energy metabolism, immune function and reproduction in mammals. Early findings on the endocannabinoid system in transition dairy cows yielded results that diverge from non-ruminant models. The PS expression on cytoplasmic membranes signals apoptosis, coagulation and contributes to sperm–oocyte recognition. As lipidomic diagnostics become increasingly available, understanding the metabolism of PL will continue to develop and promises to offer novel strategies for optimising cattle health and longevity.

Lipid metabolism influences all aspects of dairy production and health. Lipids are broadly classified into eight categories; glycerophospholipids (**PL**), fatty acyls (**FA**), glycerolipids, sphingolipids, sterol lipids, phenol lipids, saccharolipids and polyketides (Sud *et al.*, 2007). The PL typically contribute around 41–50% (w/w%) of serum or plasma lipids, with cholesteryl esters (44–50%), unesterified cholesterol (5–9%), free FA (1–2%) and triacylglycerols (trace-3%) also present (Yamdagni and Schultz, 1970; Raphael *et al.*, 1973; Bitman *et al.*, 1984). Though much dairy research has been performed on free FA, especially non-esterified fatty acids, until recently there has been comparatively little emphasis on the PL class. Advances in and availability of bioinformatics and lipidomics are enabling researchers to precisely quantify low-abundance lipids, including the FA composition of specific PL classes (Zehethofer and Pinto, 2008; Li *et al.*, 2014; Han and Gross, 2021; Liu *et al.*, 2025). Subsequently, specific PL and their FA compositions are increasingly recognised as being significantly associated with dairy cows' health and longevity (Humer *et al.*, 2016; Rico *et al.*, 2021; Sheedy *et al.*, 2026). These roles include hepatic lipid export during the transition period, aging processes, reproduction and modulation of inflammatory responses. The purpose of this narrative review was to (1) introduce readers to basic glycerophospholipid biochemistry, (2) consolidate current knowledge on the major PL classes in adult dairy cows, (3) to identify emerging evidence on the role of PL in circulation and tissue in relation to cattle longevity through their associations with age, health and reproduction, and (4) highlight areas for further research. Unless otherwise stated, all references are to adult cattle.

This review should be considered in the context of dietary precursors for fatty acids, especially the essential fatty acids that are not only potent anti-inflammatory agents, antioxidants

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A large multisite lipidomic investigation of parity and aging in dairy cows

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ABSTRACT

Efforts to optimize the longevity of dairy cows are hindered by the increased risk of adverse health events, culling, or dying on farm with increased parity. Lipidomics provides a platform to help identify important biomarkers and biological pathways associated with increased parity and associated aging. A large multisite (15 pasture-based, 15 TMR farms) cross-sectional study collected plasma samples from nonlactating, late pregnant, dry cows (n = 696, ~27 d prepartum) and peak milk cows (n = 796, ~58 DIM) in a disproportionate stratified random sampling frame (parity: 0, 1, 2, >2 for dry cows; 1, 2, 3, >3 for peak milk cows). A total of 185 lipid species, comprising the lipids classes of phospholipids, sphingomyelins (SM) and triacylglycerols, were quantified in a targeted, liquid chromatography-MS approach. Dry and peak milk cohorts were analyzed separately throughout. Variation in lipid profiles were mostly attributed to farm of origin (36%–41% of variation), with feeding system explaining 13% to 21% and parity explaining 6% to 9%, according to ANOVA simultaneous component analysis modeling. Multiple linear regression and orthogonal partial least squares (O-PLS) investigated the association of the lipid profile with age (d), whereas discriminant analysis compared first parity with >3 parity cows in O-PLS discriminant analysis, random forest, and support vector machine models. Rankings of the most important lipid species for each model type were compared. Phospholipids with 40 carbon atoms and 6 double bond equivalents (40:6) were consistently decreased with increasing parity and age across both dry and peak milk cohorts. These lipids most likely contained stearate (18:0) and docosahexaenoic acid (DHA, C22:6n-3), an n-3 fatty acid. Additionally, phospholipids with 40:5 and 38:6, lysophosphatidylcholine (17:0), SM(35:1), and SM(35:2) were commonly identified lipids that decreased in concentration with parity and age. Docosahexaenoic acid has been associated with improved cattle health, reproduction, and milk production and quality. This study raises the hypothesis that reduced DHA levels in older cows may be an important factor increasing susceptibility to adverse health events, reduced reproductive performance, and herd removal. Studies that supplement DHA or its precursors can test this hypothesis and may be important in optimizing longevity of cows.
Key words: lipids, parity, n-3 fatty acids, docosahexaenoic acid

INTRODUCTION

Optimal longevity of dairy cows is critical for farm profitability, cattle welfare, environmental sustainability, and the social license of the industry (Dallago et al., 2021). However, increased parity is associated with increased risk of adverse health events, being culled, or dying on farm (Lean et al., 2023a,b). To optimize longevity, it is critical to understand the underlying biology of increased parity. Lipidomics, the large-scale study of lipids, is a burgeoning and appropriate platform to help identify biomarkers and biological pathways associated with increased parity and associated aging (Wenk, 2005; Almeida et al., 2021).

For example, the phospholipid (**PL**) lipid class, which comprises of a phosphate containing hydrophilic core and 1 or 2 attached fatty acids (**FA**), are well recognized for their structural importance in the bilayers of cellular compartments, but are themselves highly diverse with wide-ranging functions that include associations with many diseases and aging (Harayama and Riezman, 2018; Dai et al., 2021). High levels of PL with PUFA are negatively associated with maximal age in many species, potentially due to high rates of peroxidation of PUFA and subsequent risk of oxidative damage (Hulbert

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Confinement and pasture-based dairy herds differ in plasma lipid profiles

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ABSTRACT

Dairy cow housing and management can be broadly described as either intensive confinement-based (CONFINE) or extensive pasture-based (PAST) systems. The diets between systems typically differ in their forage base, with CONFINE farms often utilizing maize silage in a TMR. Consequently, the lipid composition of diets differs between systems. The influence of housing system on blood lipidomics is currently unknown, but due to the bioactive role of lipids in influencing overall health and productivity, differences in diet may have consequences for reproduction, health, and aging of cows. The objective of this cross-sectional, multisite study was to investigate blood lipids and metabolites from cows in PAST and CONFINE systems, in the dry period (~27 d prepartum) and at peak milk (~58 DIM). After exclusions, blood samples from 303 PAST and 398 CONFINE dry-period cows and 350 PAST and 431 CONFINE peak-milk cows from 15 PAST and 15 CONFINE farms were analyzed. A total of 185 lipid species (including glycerophospholipids, sphingomyelins, and triacylglycerols) were evaluated using targeted liquid chromatography-MS, as were 11 routinely measured metabolites. Dry and peak-milk cohorts were analyzed separately throughout. Lipids and metabolites associated with housing system were selected using a variable stabilization approach that was achieved by calculating the frequency of inclusion in categorical (housing system) penalized models using bootstrapping. Variables were retained if inclusion frequency exceeded a false-positive threshold. Five different statistical models were used with variable stabilization. Dry cows in CONFINE systems had decreased globulin, urea, and glycerophospholipids associated with n-3 fatty acids. The highest total inclusion rates in the dry cohort were phosphatidylcholine (PC; 36:5), which mostly comprises palmitic acid (C16:0) and eicosapentaenoic acid (EPA; 20:5n-3), then phosphatidylethanolamine (PE; 38:5, 16:0/22:5n-3 or 18:0/EPA) and PC(34:3; 16:0/18:3 α-linolenic acid [ALA]). No lipids were increased in more than one stabilized model in CONFINE dry cows. Peak-milk CONFINE cows had increased glycerophospholipids associated with n-6 fatty acids. The highest total inclusion-rate lipids in the peak-milk cohort were phosphatidylinositol (PI; 38:3; 18:0/20:3n-6 dihomo-γ-linolenic acid), PC(34:2; 16:0/18:2 linoleic acid [LA]), PC(40:7; 18:2/22:5n-6), PC(34:1; 16:0/18:1), and PE(34:2; 16:0/LA). The CONFINE peak-milk cows also had decreased PC(34:3; 16:0/ALA). This study identified specific lipids that were strongly associated with housing systems, findings that have not been reported elsewhere. Given the important biological functions of omega fatty acids, the pattern of glycerophospholipids with increased n-6 and decreased n-3 in CONFINE cows may indicate housing systems create different risk profiles for reproduction, health, and aging.

Key words: lipidomics, omega fatty acid, housing, phospholipids

INTRODUCTION

Dairy cow housing systems can broadly be classified as either intensive confinement-based (**CONFINE**) or extensive pasture-based (**PAST**) systems. Beyond the inherent physical and managerial differences between these systems, diet composition typically differs substantially. Maize silage, a high DM yield forage, is commonly the main forage source in CONFINE systems and is provided as a component of a TMR. The distinct differences in lipid compositions of maize silage and fresh pasture may influence health, reproduction, and longevity of cows.

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Associations among body condition score, body weight, and serum biochemistry in dairy cows

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ABSTRACT

Body condition score and BW yield insights into body tissue reserves and diet, and serum biochemical measures reflect the metabolic status of cows. Associations between body composition measures and biochemistry are unclear and investigation may reveal important information on the metabolic and physiological status of cattle with varying levels of labile tissue reserves. Cohorts of 739 nonlactating, late-pregnancy, dry cows (26.9 d prepartum, SD = 12.4) and 690 peak-milk cows (58.0 DIM, SD = 14.5) were selected by stratified (parity: 1, 2, 3, >3) random sampling from 30 farms (15 pasture, 15 TMR) in this cross-sectional study. A single serum, BCS (1–5 scale), BW, and milk-production datum was collected per cow, per cohort between November 2022 and July 2023. Eleven analytes were collected, analyzed, and standardized within group (cohort/breed per farm). Mixed linear models for BCS and BW were specified, with the random effect of group. A 6-point, unordered, categorical body-group classification that combined BCS (greater, equal to, or less than group median; as high, median, or low BCS) and BW (greater or less than group median; as high or low BW) was analyzed by polytomous logistic regression. Effect sizes are listed for a 1 SD increase in the specified analyte, keeping other covariables at their mean value. Dry BCS was positively associated with albumin (0.075 BCS ± 0.014 SE), urea (0.038 BCS ± 0.014 SE), and glucose (0.052 BCS ± 0.014 SE), and negatively with the interaction between cholesterol and days precalving. Dry BW positively associated with albumin (11.03 kg ± 2.48 SE) and negatively with cholesterol (−8.47 kg ± 2.57 SE). Peak-milk BCS was positively associated with albumin (0.47 BCS ± 0.015 SE), BHB (0.048 BCS ± 0.015 SE), and glucose (0.051 BCS ± 0.015 SE). Peak-milk BW was positively associated with albumin (6.94 kg ± 2.35 SE) and negatively with Ca (−7.02 kg ± 2.33 SE). Increasing BW and decreasing BCS was associated with increasing parity, except in dry second-parity cows that had low BCS. The dry polytomous model associated a 1 SD increase in albumin with a 4.89% ± 1.56 SE decreased risk of being low BCS/low BW and 5.87% ± 1.46 SE increased risk of high BCS/high BW. Risk change associated with 1 SD of glucose was −5.61% ± 1.58 SE for low BCS/high BW and 3.17% ± 1.58 SE for high BCS/high BW. For the peak-milk cohort, change in risk was associated with albumin for low BCS/low BW −3.67% ± 1.56 SE, low BCS/high BW −3.22% ± 1.53 SE. Risk change with 1 SD of BHB was −3.36% ± 1.47 SE for median BCS/low BW, 2.86% ± 1.44 SE for high BCS/low BW, and 2.69% ± 1.37 SE for high BCS/high BW. Risk of low BCS/low BW was greatest in second-parity cows, and high BCS/high BW was greatest in dry cows with greater than third parity and third-parity cows in peak milk. There were no interactions between parity and analytes. Albumin was consistently associated with BCS and BW, potentially reflecting innate differences in protein metabolism of cows.

Key words: body condition, parity, albumin, protein, body weight

INTRODUCTION

Cows mobilize body reserves to facilitate the strong homeorhetic drive to produce large quantities of milk in early lactation (Bauman and Currie, 1980). This period of negative nutrient balance typically results in body condition and weight loss that reaches a nadir around 40 to 100 DIM (Westwood et al., 2002; Roche et al., 2009; Hernandez-Gotelli et al., 2023). Cows differ in their ability to partition and mobilize body reserves, with excessive mobilization associated with increased disease incidence, reproductive inefficiencies, and potentially poor cow welfare (Roche et al., 2009). Monitoring BCS

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Optimal longevity of dairy cows is critical for farm profitability, cattle welfare, environmental sustainability, and the social license of the industry (Dallago et al., 2021). However, increased parity is associated with increased risk of adverse health events, being culled, or dying on farm (Lean et al., 2023a,b). To optimize longevity, it is critical to understand the underlying biology of increased parity. Lipidomics, the large-scale study of lipids, is a burgeoning and appropriate platform to help identify biomarkers and biological pathways associated with increased parity and associated aging (Wenk, 2005; Almeida et al., 2021).

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Increased parity is negatively associated with survival and reproduction in different production systems

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ABSTRACT

We conducted a retrospective meta-analysis based on individual cow data to assess the associations of parity, level of production, and pasture-based or intensively fed systems with fertility. Our goal was to provide understandings of the role of parity in risks for removal and reproductive failure. Multilevel models were used to evaluate the fixed effects of parity, milk, milk solids, milk fat and protein percentage and yield, and production system [intensively fed (n = 28,675) or predominantly pasture fed (n = 4,108)] on reproductive outcomes. The outcomes were the hazard of not being bred (HNBRED), hazard of pregnancy (HPREG), pregnancy to first breeding (PREG1), and odds of becoming pregnant in a lactation (OPAL). The 32,783 cows were in 13 studies conducted in Australia (14.6% of cows), Canada (2.4% of cows), and the United States (83.0% of cows). There were 38.5% of cows in the sample in parity 1, 27.3% in parity 2, 16.7% in parity 3, 9.0% in parity 4, and 8.6% in parity ≥5. Compared with cows of parity 1, parity ≥5 cows had a greater HNBRED [hazard ratio (HR) = 2.45], lesser HPREG (HR = 0.73), and reduced OPAL (odds ratio = 0.36). However, the parity ≥5 cows had similar PREG1 to other parities except for parity 1. This suggests the possibility of a higher proportion of subfertile parity ≥5 cows than for other parities. Associations between parity and reproductive measures were influenced by the different milk production measures, indicating that milk yield and milk component percentages and yields modified the odds or hazards of successful reproduction. All milk production measures had quadratic associations with OPAL, indicating that either low or high production or concentration of solids within a cohort reduced OPAL. This reduced OPAL reflected a greater HNBRED for lower milk yield and milk protein and fat yielding cows. Both milk yield and milk protein percentage had quadratic associations with HPREG. When centered milk yield was categorized into quartiles, small differences in HPREG existed. A more marked association of milk protein percentage occurred with HPREG, with optimal HPREG at approximately 0.5% above group mean milk protein percentage. Milk fat percentage (HR = 0.901), fat yield (kg/d; HR = 0.78), protein yield (kg/d; HR = 0.71), and milk solids yield (kg/d; HR = 0.84) were all linearly associated with reduced HPREG. Difference in production systems did not have substantive effects on PREG1 but did for HNBRED, HPREG, and OPAL. Estimates of associations of parity with reproductive outcomes HNBRED, HPREG, and OPAL were influenced by milk and milk solids yield; older cows had markedly lower reproductive outcomes. Interestingly, for PREG1, there were few differences among parities and differences were less influenced by milk yield and constituent measures. The marked associations of parity with removal for all reasons, deaths and culling, and reductions in HNBRED, HPREG, and OPAL indicate a need to focus on the physiological changes with parity to produce better strategies to support optimal longevity of cows.

Key words: production, milk-protein, parity, pregnancy, longevity

INTRODUCTION

It is necessary to ensure that cows economically optimize their genetic potential by providing environments that permit them to be productive, healthy, and reproductively successful. Dallago et al. (2021) noted that the longevity of dairy cattle has decreased in most high milk-producing countries over time and that increasing longevity would have benefits including increased profitability. De Vries and Marcondes (2020) suggested that the economic trade-offs between longevity and profit may be complex given that rapid genetic gain is possible. A study conducted in the United States identified that more than 40% of cows

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Associations of parity with health disorders and blood metabolite concentrations in Holstein cows in different production systems

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ABSTRACT

Data were obtained from studies in Australia, Canada, and the United States using individual cow data from 28,230 Holstein cows to evaluate associations between parity and disease. Our goal was to develop understanding of disease risks for cows of differing parity. We hypothesized that there would be increased risks of disease and changes in metabolite concentrations with increased parity. Parity ≥5 represented 2,533 cows or 9.0%, parity 4 was 9.8% (2,778), parity 3 as 19.0% (5,355), parity 2 as 28.1% (7,925), and parity 1 was 34.1% (9,639) of the sample. Of these cows, 15.5% were in Australia, 14.7% in Canada, and 69.8% in the United States. Lactational incidence (LI) risk of clinical hypocalcemia increased with parity from 0.1% for parity 1 to 13% for parity ≥5 cows. The marked increase suggests profound differences in metabolism with increased parity. The LI of clinical mastitis was 17.4%. The odds of mastitis increased with parity to 2.5 times greater in parity ≥5 than in parity 1. The LI of lameness increased with parity; specifically, the odds of lameness was 5.6 times greater for parity ≥5 than parity 1. Dystocia incidence was 8.7% and greatest for parity 1 cows. The LI of retained placenta was 7.4% and increased with parity, with the odds for parity ≥5 2.3 times greater than for parity 1. The LI of metritis was 10% and of endometritis 14%, with the greatest odds in parity 1. The LI of clinical ketosis was 3.3% with a marked increase in odds with parity. The prevalence of subclinical ketosis was 26.8% with only cows in parity 1 having lower odds than other parities. Parity ≥5 cows had greater odds (odds ratio = 1.7) of respiratory disease than parity 1 cows, which were lesser than other parities. Metabolite concentrations were evaluated in 5,154 Holstein cows in the precalving, calving, and immediate postcalving data sets. Metabolic measures near peak lactation provided 1,906 observations. Concentrations of β-hydroxybutyrate (BHB) and nonesterified fatty acids increased with parity on d 1 to 3 of lactation and at peak lactation. On d 1 to 3 after calving differences in glucose, nonesterified fatty acids, and BHB indicated a greater reliance on mobilized lipid to export energy to peripheral tissues as BHB for greater parity cows. Differences in concentrations among parity groups were marked at times, for example >0.20 m*M* in Ca for parity 1 and 2 to parity ≥5 and >0.33 m*M* for all older parities compared with parity 1 for P on the day of calving. The marked increase suggests profound differences in metabolism with increased parity are probably influenced, in part, by increased production. We found marked differences in concentrations of metabolites with parity that are consistent with reduced reproduction, health, and body condition for higher parity cows. These unfavorable differences in metabolism in Ca, P, glucose, and cholesterol concentrations for higher parity cows also complement the often-substantial differences in disease risk with parity and suggest a need to carefully consider the parity structure in study design. Managers and advisors will need to consider methods to reduce risk of health disorders tailored to cows of different ages.

Key words: parity, disease, longevity, blood metabolites

INTRODUCTION

Many have noted a concern about the longevity of dairy cattle (De Vries and Marcondes, 2020; Dallago et al., 2021). Cows with parity ≥2 in a US-based study (Golder et al., 2019) and in large Australian studies were at 2 times greater risk for many diseases and had reduced odds and hazard of pregnancy than parity 1 cows (Golder et al., 2021). An evaluation of associa-

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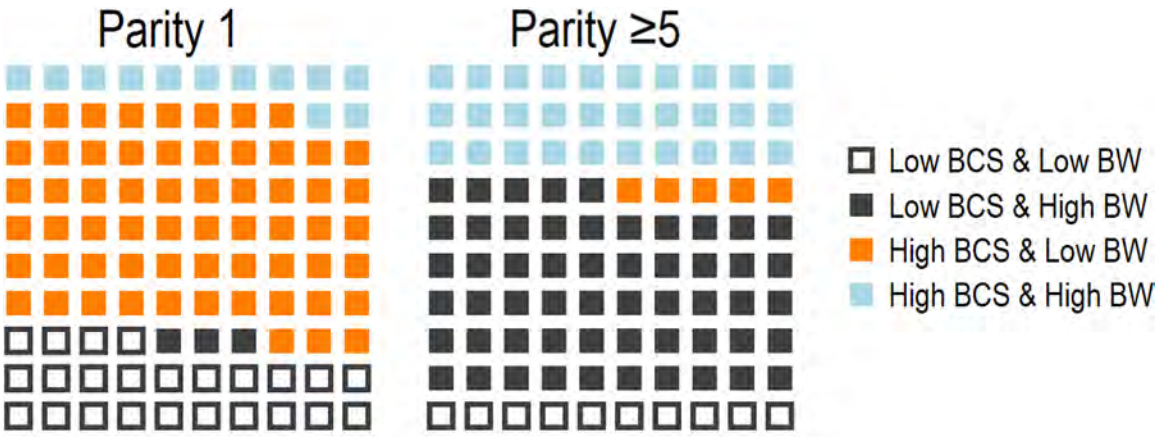
Short Communication
Health, Behavior, and Well-being

Holstein dairy cows lose body condition score and gain body weight with increasing parity in both pasture-based and total mixed ration herds

I. J. Lean,[1,2] D. B. Sheedy,[1,2]* S. J. LeBlanc,[3] T. Duffield,[3] J. E. P. Santos,[4] and H. M. Golder[1,2]

Graphical Abstract



Summary

Body condition scoring (BCS) and body weight (BW) measurements are associated with health and reproductive efficiencies of cows. We used raw data sets from 16 studies to evaluate associations of parity and feeding system (pasture-based or total mixed ration) with BCS at precalving and peak milk, and change in BCS, and BW at peak milk. With increasing parity, there is a general decrease in BCS and increase in BW regardless of feeding system, with most young cows having low BW and high BCS and older cows having high BW but low BCS.

Highlights

- Body condition score decreases and BW increases in older cows.
- Parity had a greater effect on BCS and BW than feeding system.
- Second-parity cows have the lowest pre-calving BCS.

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Short Communication
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Holstein dairy cows lose body condition score and gain body weight with increasing parity in both pasture-based and total mixed ration herds

I. J. Lean,[1,2] D. B. Sheedy,[1,2]* S. J. LeBlanc,[3] T. Duffield,[3] J. E. P. Santos,[4] and H. M. Golder[1,2]

Abstract: Body condition scoring (BCS) and body weight (BW) are observations associated with labile tissue reserves, health, and reproduction efficiency of dairy cows. The effect of parity (1 through to ≥5) and feeding system (pasture-based and TMR) on BCS and BW were evaluated utilizing raw data sets from 16 retrospective studies that totaled 24,807 Holstein cows across 3 nations (Australia, Canada, and the United States). Linear regression models were used to investigate the 5 outcome variables of precalving BCS, peak milk BCS, change in BCS from precalving to peak milk, and peak milk BW and their respective associations with parity and feeding system. To help control for the influence of calendar time, study treatment protocols when applicable, and genetic change, all outcome variables were center-transformed around each study group mean. Including feeding system as a covariate improved model fit for most outcome variables; however, the relative effect size of parity was generally much greater than feeding system effect size. Parity 2 cows had the lowest precalving BCS of −0.087 [95% confidence interval (CI): −0.107, −0.065] less than the mean, whereas parity 1 cows had the greatest, 0.068 (95% CI: 0.043, 0.092) above mean, regardless of feeding system. Peak milk BCS overall decreased with increasing parity (parity 1 to parity ≥5: −0.13, 95% CI: −0.19, −0.08) and BCS change during the transition period monotonically decreased with increasing parity (parity 1 to parity ≥5: −0.22, 95% CI: −0.26, −0.17). Peak milk BW monotonically increased with increased parity (parity 1 to parity ≥5: 114 kg, 95% CI: 104, 125). A waffle plot was used to present the proportions of cows, by parity, that were partitioned into "low BCS and low BW," "low BCS and high BW," "high BCS and low BW," or "high BCS and high BW" groups. Cows were assigned either a high or low status by being above or below their specific centered study group means, respectively. Considering a null hypothesis of 25% per BCS-BW category, there was a striking change in category from parity 1 cows that were predominantly in the "high BCS and low BW" category (61.2%) to parity ≥5 cows that were predominantly in the "low BCS and high BW" category (55.5%). The study supports studies showing increased weight and change in BCS with increased parity. We highlight the associations among production system, BCS, BW, and parity.

Body condition score in dairy cattle is a rapid visual assessment that is highly associated with available energy reserves, primarily mobile fat reserves (Edmondson et al., 1989). Though different rating systems exist, a low score universally reflects low energy stores or emaciation and a high score with excessive energy stores or obesity (Roche et al., 2004). During early lactation, dairy cows are under a strong homeorhetic drive to mobilize energy reserves to produce large quantities of milk (Bauman and Currie, 1980; Baumgard et al., 2017), resulting in a loss of body condition that typically reaches a nadir after the first 40 to 100 DIM (Gallo et al., 1996). Excessive BCS loss during this period is associated with increased disease incidence, reproductive inefficiencies, and potentially poor cow welfare (Roche et al., 2009). Greater parity dairy cows often experience a greater BCS loss compared with nulliparous cows during the early postparturient period and older cows are also associated with an increased risk of removal from the herd due to reproductive failure, disease, and death (Roche et al., 2009; Pinedo et al., 2010).

Body weight also reflects the protein, fat, and energy reserves of cattle (Botts et al., 1979; Gregory et al., 1998; Thorup et al., 2013). There is an imperfect association between BCS and BW that may be influenced by factors such as rumen fill, stage of pregnancy and lactation, and udder fill as well as composition and location of labile tissue pools. Body weight is also associated with parity, with heifers entering the lactation herd typically at around 80% of their mature BW (Berry et al., 2011; Berry and Evans, 2022). Different feeding systems are also reported to influence BCS and BW, with herds from TMR systems typically having higher total BCS and BW than those from pasture-based systems (Washburn et al., 2002; Roche et al., 2007). However, the influence of feeding system on within-herd BCS and BW during the parturition period is less clearly understood and may be similar between systems (Roche et al., 2007). It is important to understand the association of BCS and BW with parity and management systems as this may provide insight into increased risks for disease, reproductive failure, and survival of older cows and how to manage these risks.

In this study we examine associations of parity with precalving and peak milk BCS, changes of BCS between precalving and peak milk observations, and peak milk BW for Holstein cows in different production systems (pasture-based or TMR) across 3 countries using retrospectively collected data. We hypothesized that BCS and BW of dairy cows are associated with cow parity and that BCS

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Final Project Summary
P2b: Early Alerts

May 2026

This project explored opportunities for the future development of early-warning alerts for diseases, pests and seasonal risks affecting dairy farms.

Providing early alerts is important to ensure necessary protective actions are in place as soon as possible or the appropriate investigations can begin promptly. Early alerts have potential benefits for individual herds, animal welfare, the dairy industry and national biosecurity.

The project reviewed existing information streams from industry, government and research activities that could potentially support more timely alerts and advisory information for farmers and advisors.

Targeted alerts, coupled with timely advisory information, would help farms respond more effectively to risks affecting pastures, crops and cattle.

Outcomes

The team reviewed existing activities at the Elizabeth Macarthur Agricultural Institute (EMAI), Department of Primary Industries and Regional Development (DPIRD), University of Sydney and Sci*bus*.

Unlocking the potential of cows

Dairy UP's P2 project aimed to unlock the potential of dairy cows to achieve their genetic potential under NSW conditions.

P2 was a suite of seven projects that collectively explored ways to profitably increase both productivity and wellbeing in commercial settings.

P2a: Cattle Longevity

P2b: Early Alerts

P2c: Milk as a Diagnostic Tool

P2d: Diet and Heat Load

P2e: Calf Husbandry

P2f: Infectious Diseases ('Infectome')

P2g: Heifers Early Calving

This document provides a final update on P2b: Early Alerts.

The following priority targets for early alert systems were identified:

- Bovine ephemeral fever (3-day sickness).
- Fall army worm.
- Facial eczema.

The project also prepared the terms of reference for an industry panel to help guide future development of early alert systems. The concept has been supported by Dairy NSW and NSW DPIRD.

Ephemeral fever

NSW DPIRD has published a website with online resources relating to bovine ephemeral fever to support farmer awareness and decision-making.

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DairyUP
Unlocking potential

Collaborators

The project was a collaboration between Scibus and NSW DPIRD.

More info

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P2 final report

Delivery organisations

The University of Sydney Dairy Research Foundation
NSW Government Department of Primary Industries and Regional Development
Scibus
Dairy Australia

Partner organisations

nsw Farmers Growing the best
eastAUSmilk Advocacy · Unity · Service
Dairy NSW / Dairy Australia
AFMH
Leppington Pastoral Company Pty Ltd
NSW Government Local Land Services
Norco

Additional program supporters, collaborations or partnerships

Charles Sturt University | DairyBio | DataGene | Eagle Direct | Entegra
Macquarie University | NSW EPA | smaXtec | UC Davis | University of Technology Sydney

Final Project Summary
P2c: Milk as a Diagnostic Tool

May 2026

Milk as a monitoring tool

Testing samples from bulk tank milk for somatic cell count (SCC) is a well-established practice. But testing milk samples has the potential for broader uses to monitor, predict and manage risks to dairy cow health, productivity and fertility.

Bulk tank milk samples provide insights at a herd level, while herd testing offers insights at an individual cow level. And the growing adoption of inline metering offers opportunities for more convenient and frequent on-farm sampling to provide insights about individual cows, including specific udder quarters.

Research approach

This project involved a literature review to investigate opportunities to use milk samples as a monitoring and diagnostic tool in Australian dairy herds. It focussed on potential insights about:

- Dietary imbalance
- Udder health (mastitis)
- Viral diseases

Unlocking the potential of cows

Dairy UP's P2 project aimed to unlock the potential of dairy cows to achieve their genetic potential under NSW conditions.

P2 was a suite of seven projects that collectively explored ways to profitably increase productivity and wellbeing in commercial settings.

- P2a: Cattle Longevity
- P2b: Early alerts
- P2c: Milk as a diagnostic tool
- P2d: Diet and Heat Load
- P2e: Calf husbandry
- P2f: Infectious diseases ('Infectome')
- P2g Heifers early calving

This document is the final update on P2c: Milk as a diagnostic tool. This project was closely linked with P2f: Infectious diseases.

Potential applications

While the work was NSW-focussed, the outcomes are potentially applicable to all Australian dairy herds.

Using milk as a monitoring and diagnostic tool has potential benefits at the industry, herd and individual cow level.

Biosecurity

Ultimately, the diagnostic capability of bulk tank milk could help reduce the risk of disease spread within and among herds.

At an industry level this is vital for biosecurity as the detection of pathogens or specific antibodies in milk can be used for monitoring endemic diseases within a farm, region, or country.

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DairyUP
Unlocking potential

Herd/animal health monitoring
Testing bulk tank milk could be a cost-effective and fast way to monitor herd health changes, potentially equipping veterinary services, or milk processors with new tools to assist farms as part of an efficient integrated farm service.

For example, antibody testing for Mycoplasma bovis (which causes pneumonia, mastitis and arthritis in cattle) can be conducted at bulk tank milk level.

Management interventions
Using bulk tank milk as a diagnostic tool on farm would help farmers make informed business management decisions to improve animal well-being, productivity, and profit.

Early and accurate detection of metabolic disorders can guide intervention strategies, while bulk tank milk urea and protein content are useful indicators of herd nutrition.

Bulk tank milk could also provide a cost-effective and efficient assessment of a herd's disease status with this information supporting disease control programs for viral diseases.

At an individual level, the early diagnosis and treatment of infections and nutritional disorders will improve animal health and welfare.

More information

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P2 final report

Key findings: Ways milk can be used as a diagnostic tool

Commercially available applications

- Somatic cell count — udder health monitoring, mastitis management, selective dry cow treatment
- Bulk tank milk protein and urea — herd nutrition and fertility risk
- Herd testing — earlier identification of cows at risk of metabolic disorders such as ketosis
- Conductivity — early indicator of udder inflammation and mastitis
- Polymerase chain reaction (PCR) testing — identification of mastitis-causing pathogens
- Bulk tank milk surveillance — herd-level disease and biosecurity e.g. bovine leukosis, *Mycoplasma bovis*

Tools with potential for greater use

- Bulk tank milk — herd-level monitoring of nutrition, minerals, vitamins and disease risk
- Integration of herd testing and inline milk data — earlier identification of health and metabolic issues
- More proactive use of bulk tank and individual cow milk data — for earlier intervention and herd management decisions

Emerging technologies

- Microbiome analysis — bacterial profiling for mastitis and udder health
- Immune biomarkers — early indicators of inflammation and infection
- MALDI-TOF milk testing — rapid identification of mastitis-causing bacteria
- Multiplex milk testing — multiple diseases from one milk sample
- Advanced metabolic profiling — earlier detection of ketosis and acidosis risk
- Integrated sensor systems — combining milk, behaviour and herd data for earlier alerts

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Research and results

Literature review

The Dairy UP team undertook a literature review of previous work, focusing on the role of milk as an indicator of dietary imbalance, milk as a diagnostic fluid for udder health management and milk as a diagnostic fluid to monitor viral diseases in dairy cattle.

This review also informed further consideration of the development of diagnostic tests using milk, in addition to those already commercially available.

Dietary imbalance

Herd level

The review concluded that bulk tank milk is an under-used resource for monitoring herd health and nutrition.

Bulk tank milk has potential as a low-cost herd monitoring tool, but results can be harder to interpret because it is a mix of milk from cows at different stages of lactation.

Protein and urea levels in bulk milk samples can provide useful indications of herd nutrition and may help flag nutritional issues affecting fertility.

Bulk tank milk can be used to assess herd mineral and vitamin status, including selenium, zinc, beta-carotene and vitamin E.

Individual cows

Testing individual milk samples can help identify metabolic problems in individual cows early enough to support targeted management or treatment.

Herd testing reports can aid earlier identification of cows at risk of metabolic disorders such as ketosis.

The milk fat-to-protein ratio can help identify fibre deficiency and acidosis risk in individual cows, although it is better at confirming problems than detecting all affected cows.

Udder health (mastitis)

The review found a growing range of milk-based tests that are becoming available to detect inflammation and infection associated with mastitis.

The performance and interpretation of diagnostic tests can differ depending on whether they are applied to quarter milk, individual cow milk or bulk milk samples.

Some proven diagnostic tools may still be under-used on farm, while newer technologies require further validation before widespread adoption.

Herd level

The review concluded that the well-established practice of testing somatic cell count (SCC) in bulk tank milk remains an important tool for monitoring herd udder health.

Individual cows or quarter sampling

Testing milk samples from individual cows (or quarters) for somatic cell count is an important tool for detecting udder inflammation and guiding mastitis management, including decisions about selective dry cow treatment.

Electrical conductivity testing is used commercially as an indicator of udder inflammation and mastitis.

Additional markers of inflammation can indicate disease severity, although they do not identify the specific pathogen involved.

Milk samples can be used to identify mastitis-causing pathogens using laboratory culture or polymerase chain reaction (PCR) testing.

A number of emerging technologies show promise for improving the detection and identification of mastitis-causing pathogens. They include loop-mediated isothermal amplification (LAMP), microbiome analysis and advanced organism identification methods such as matrix-assisted laser desorption/ionization time of flight (MALDI-TOF). However, many of these technologies are still in the research or development stage and are not yet used with bulk tank milk testing.

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Some pathogen-specific immune markers are available, but their practical value for on-farm udder health management is still unclear.

Viral diseases

This review focused on the use of bulk tank or pooled milk samples to monitor viral diseases at the herd level. In this context, pooled milk refers to milk combined from groups of cows within a herd rather than individual animals.

Milk testing can detect either viral pathogens or antibodies, supporting herd, regional and national disease surveillance programs. The results can help inform biosecurity decisions and on-farm disease management.

At the herd level, bulk tank milk can be a cost-effective way to monitor exposure to viral diseases.

Bulk tank milk testing has already supported disease control programs for some viral diseases, such as bovine leukosis.

Choosing the most suitable testing approach requires careful consideration of the disease being monitored and the structure of the herd.

The accuracy of pooled milk testing depends on factors such as herd size, infection prevalence, vaccination status, and how long infected cows shed virus or antibodies into milk.

Reliable interpretation of pooled milk results depends on understanding the strengths and limitations of each test, including false positive and false negative risks.

Larger dairy herds increase the need for highly sensitive tests because infected milk can become diluted in the bulk tank.

Technology opportunities

Improved testing systems could strengthen herd surveillance and support faster disease detection in the future.

For example, new technologies may allow multiple diseases to be monitored from a single milk sample.

But testing "pooled" milk does come with challenges. The accuracy of tests relies on achieving both low false positive and false negative estimates together. This can be difficult and influences the value of the tests to industry.

Researchers concluded that the integration of new biotechnologies could enhance multiplexing and data interpretation for comprehensive surveillance.

Related projects

P9a: Producing milk with less lactose

P9b: Adding value to dairy waste

Collaborators

The P2c project was a collaboration between researchers from Dairy UP, Scibus, University of Sydney and NSW DPIRD.

Journal articles

Milk as an indicator of dietary imbalance
Milk as a diagnostic fluid to monitor viral diseases in dairy cattle
Milk as diagnostic fluid for udder health management

Delivery organisations

The University of Sydney Dairy Research Foundation
NSW Government Department of Primary Industries and Regional Development
Scibus
Dairy Australia

Partner organisations

NSW Farmers Growing the best
eastAUSmilk
Dairy NSW
Dairy Australia
AFMH
Leppington Pastoral Company Pty Ltd
NSW Government Local Land Services
Norco

Additional program supporters, collaborations or partnerships

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INVITED REVIEW

Milk as a diagnostic fluid to monitor viral diseases in dairy cattle

B Brito[a,b,c*] and P Hick[a]

Background Infectious viral diseases in dairy cattle have substantial implications for milk production, quality and overall animal health. Diagnostic tools providing reliable results are crucial for effective disease control at the farm and industry level. Pooled or bulk tank milk (BTM) can be used as a cost-effective aggregate sample to assess herd disease status in dairy farms.

Findings Detection of pathogens or specific antibodies in milk can be used for monitoring endemic diseases within-farm, region or country-level disease surveillance and to make informed decisions on farm management. The suitability of assays applied to pooled milk samples relies on validation data of fit-for-purpose tests to design an optimal testing strategy. Diverse approaches and variable scope of studies determining test accuracy need to be critically appraised before sourcing the parameters to design sampling strategies and interpreting surveys. Determining if BTM or pooled milk is the best approach for a disease management programme should carefully consider several aspects that will impact the accuracy and interpretation, for example, the size of the lactating herd, the risk of infection in the lactating and non-lactating groups, the expected within-herd prevalence, the duration of infection, the duration and concentration of antibodies in milk and use of vaccination.

Conclusions There are examples of tests on BTM samples providing efficient assessments of the herd disease status and supporting disease control programmes for viral diseases. However, challenges arise in pooled milk testing due to the need for accurate estimates of the imperfect sensitivity and specificity of the assays. Integration of new biotechnologies could enhance multiplexing and data interpretation for comprehensive surveillance. The development of highly sensitive assays is necessary to meet the demands of larger dairy herds and improve disease detection and assessment.

Keywords aggregate samples; animal disease surveillance; diagnostic; virus

Abbreviations BoHV-1, bovine alphaherpesvirus-1; BLV, bovine leukosis virus; BTM, bulk tank milk; BVDV, bovine viral diarrhea virus; DIVA, differentiating infected from vaccinated animals; DSe, diagnostic sensitivity; DSp, diagnostic specificity; ELISA, enzyme linked immunosorbent assay; HSe, herd sensitivity; HSp, herd specificity; OD, optical density; PI, persistently infected; qRT-PCR, real-time reverse transcription polymerase chain reaction; VI, virus isolation

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Introduction

Infectious diseases in dairy cattle significantly impact milk production, quality and animal well-being. Effective disease management, control and surveillance are paramount to prevent outbreaks, safeguard human health from potential zoonotic infections and maintain sustainable dairy operations. Controlling infectious diseases is essential for preserving international trade opportunities and ensuring the overall health and productivity of dairy cattle. Several viral pathogens can cause subclinical, mild or severe diseases leading to significant economic losses to the industry. The intensification of the dairy industry allows for close monitoring of animals and the optimisation of productivity. However, animals with high physiological requirements and mixing cohorts with varying pathogen exposure create favourable conditions for disease caused by viral pathogens. A requisite to effectively control viral disease at the farm and industry level is the availability of diagnostic tools that provide reliable results for decision-making.

Tests of pooled milk can be a cost-effective tool for assessing disease status in a herd and for monitoring endemic viruses.[1,2] The combined milk collected from a dairy farm contains valuable target analytes that can identify infection or prior exposure to pathogens. Testing bulk milk has been critical in disease eradication programmes to determine herd infection status and to build confidence of freedom from infection for specific agents. However, the success of bulk milk testing depends on the informed use of assays that have been thoroughly validated. With the increasing size of lactating herds, the challenge lies in developing highly sensitive assays to detect diluted target analytes in large milk volumes of bulk tank milk (BTM). The accuracy of the assay must align with the testing objective, whether it is to assess an individual herd or for monitoring as part of an industry or government programme for disease control or eradication. For example, a study evaluating Caprine arthritis and encephalitis virus detection and control in Australian dairy goats found that antibody enzyme-linked immunosorbent assay (ELISA) testing of BTM can be a cost-effective strategy, particularly in herds with low infection prevalence. However, pooling approaches may not be beneficial in herds with higher infection rates.[3]

The viral infection dynamics, the concentration and persistence of antibodies or viruses in milk, as well as the expected herd prevalence, play a crucial role in determining the most effective approach for

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AUSTRALIAN VETERINARY JOURNAL
AUSTRALIA'S PREMIER VETERINARY SCIENCE TEXT
PRODUCTION ANIMALS

Check for updates

INVITED REVIEW

Milk as diagnostic fluid for udder health management

S Rowe,[a,b]* JK House[a,b] and RN Zadoks[a,b]

Background Mastitis is the major disease affecting milk production of dairy cattle, and milk is an obvious substrate for the detection of both the inflammation and its causative infectious agents at quarter, cow, or herd levels. In this review, we examine the use of milk to detect inflammation based on somatic cell count (SCC) and other biomarkers, and for the detection of mastitis pathogens through culture-based and culture-free methods.

Findings The use of SCC at a cow or bulk milk level to guide udder health management in lactation is well-established, and SCC is increasingly used to guide selective dry cow treatment. Other markers of inflammation include electrical conductivity, which is used commercially, and markers of disease severity such as acute phase proteins but are not pathogen-specific. Some pathogen-specific markers based on humoral immune responses are available, but their value in udder health management is largely untested. Commercial pathogen detection is based on culture or polymerase chain reaction, with other tests, for example, loop-mediated isothermal amplification or 16S microbiome analysis still at the research or development stage. Matrix-assisted laser desorption ionisation time of flight (MALDI-ToF) is increasingly used for the identification of cultured organisms whilst application directly to milk needs further development. Details of test sensitivity, specificity, and use of the various technologies may differ between quarter, cow, and bulk milk applications.

Conclusions There is a growing array of diagnostic assays that can be used to detect markers of inflammation or infection in milk. The value of some of these methods in on-farm udder health improvement programs is yet to be demonstrated whilst methods with proven value may be underutilised.

Keywords MALDI-TOF; mastitis; milk ELISA; PCR; somatic cell count

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Critical review

Udder health management is a coherent set of actions at the herd level aimed at preventing or controlling mastitis. Mastitis (inflammation of the mammary gland) generally occurs when microbes (most commonly bacteria, occasionally algae or yeast) infect the mammary gland (intramammary infection; **IMI**) and elicit a host response. It is an important disease of dairy cows because it can negatively impact cow welfare,[1] productivity,[2] milk quality,[3] and economic returns.[4] Effective udder health management requires that farm staff and advisors make interventions in areas that are likely to reduce the number and duration of existing cases and prevent new IMI from occurring. To achieve this, the predominant causative agents and reservoirs of infection need to be identified, which may be the environment that the herd lives in ('environmental mastitis'), or infected cows within the herd ('contagious mastitis'). If the microbial reservoir, exposure routes, and risk factors predisposing animals to IMI can be identified, often requiring integration of findings from herd inspections, records analysis, and milk testing, interventions can be tailored to the farm. The two major ways that milk can be used as a diagnostic fluid to aid this process include (1) surveillance of mastitis using somatic cell count (SCC) and other markers of inflammation or infection; and (2) identification of IMI-causing pathogens with the potential downstream characterisation of pathogen heterogeneity as an indicator of sources and transmission routes.

Using patterns in somatic cell counts

Milk SCC has long been used as a measure of milk quality, an indicator of mastitis and a proxy for IMI. The SCC of healthy mammary glands is low and the 'somatic cells', which originate from the host's body (hence: somatic) rather than from microorganisms, are largely comprised of macrophages (28%–81%), lymphocytes (10%–45%) and polymorphonuclear neutrophils (0%–62%).[5] These cells play a role in non-specific immune defence of the gland. When IMI occurs, neutrophils are recruited from the blood circulation into the gland, causing elevations in SCC in milk.[6] If the IMI is successfully eliminated by the immune system, with or without therapeutic assistance, the SCC will drop to baseline level within weeks,[7] whereas SCC remains elevated if IMI persists.[8] Hence, SCC can be used as an indicator of udder health, with applications at quarter, cow, and bulk-tank level. Because enzymes from neutrophils impact negatively on sensory quality[9] and shelf life,[10] bulk milk SCC is both a health metric and a milk quality metric, and cow health as well as milk quality can be improved through good udder health management.

Bulk milk somatic cell counts. Bulk milk SCC (BMSCC) surveillance is conducted by milk processors in many countries and provides an early detection system for milk quality issues originating from udder health problems as opposed to, for example, cleaning, cooling, or compliance-related factors. As a rule of thumb, BMSCC

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AUSTRALIAN VETERINARY JOURNAL
AUSTRALIA'S PREMIER VETERINARY SCIENCE TEXT
PRODUCTION ANIMALS

INVITED REVIEW

Milk as an indicator of dietary imbalance

IJ Lean[a,b]* and HM Golder[a,b]

Background Milk provides a readily available diagnostic fluid collected daily or more frequently on an individual animal or herd basis. Milk, as an aggregated sample in bulk tank milk (BTM) represents the status of a herd instead of a single animal. In this review, we examine the potential for milk to predict risks to efficient production, reproductive success, and health on the individual cow and herd level.

Findings For many conditions related to disorders of metabolism including hyperlipdaemia and ketonaemia, improved individual cow milk testing may allow a temporally useful detection of metabolic disorder that can target intervention. However, the extension of these tests to the BTM is made more difficult by the tight temporal clustering of disorder to early lactation and the consequent mixing of cows at even moderately different stages of lactation. Integrating herd recording demographic information with Fourier-transformed mid-infrared spectra (FT-MIR) can provide tests that are useful to identify cows with metabolic disorders. The interpretation of BTM urea and protein content provides useful indications of herd nutrition. These may provide indicators that encourage further investigations of nutritional influences on herd fertility but are unlikely to provide strong diagnostic value. The fat-to-protein ratio has a high specificity, but poor sensitivity for detection of fibre insufficiency and acidosis on an individual cow basis. Selenium, zinc, β-carotene, and vitamin E status of the herd can be determined using BTM.

Conclusions There appears to be increasing potential for the use of milk as a diagnostic fluid as more in-parlour tests become available for individual cows. However, the BTM appears to have under-utilised potential for herd monitoring.

Keywords bulk milk tank; dairy cattle; metabolism; nutrition

Abbreviations BTM, bulk tank milk; FFA, free fatty acids; FT-MIR, fourier-transformed mid-infrared spectra; MUN, milk urea nitrogen; NEFA, non-esterified fatty acids

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Milk provides a readily available diagnostic fluid collected daily or more frequently on an individual animal or herd basis. Milk, as an aggregated sample in bulk tank milk (BTM) represents the status of a herd instead of a single animal. In this review, we examine the potential for milk to predict risks to efficient production, reproductive success, and health on the individual cow and herd level. Our goal is to provide detail and evaluate existing tests to identify the potential to develop predictive models for dietary imbalance and metabolic disease including reproductive failure. The challenge of evaluating milk as a diagnostic fluid is substantial as one of the pre-conditions for producing a test that provides both sensitive and specific results is the issue of the definition of disease or disorder or having a rigorous index test. Lean et al.[1] note that definitions of disease often reflect historical understandings of disease (e.g., 'milk fever') or terms that reflect aspects of the perceived aetiology (e.g., 'grass tetany') and that classification of disease should be accurate and reflect standardised and repeatable criteria. They[1] noted that Evans postulates[2] should be applied to infectious disease and suggested a modification of these postulates that can be applied to define the metabolic disease. There are differences in case definitions throughout the literature reviewed for this study, hence overall assessments of test value may be more variable. For example, Tatone et al.[3] noted the variable thresholds in concentrations of ketones used to evaluate hyperketonaemia and evaluated three different ketone index tests against a reference standard of biochemical blood assay of 3-hydroxybutyrate (BHB). The challenge of case definition is far greater if one were to extend this to such a variably defined condition such as clinical ketosis.[1]

Diseases need not be 'clinical' to incur substantial costs on a herd, such as parasitic diseases[4] and there is considerable merit in monitoring the estimated prevalence in a population. There is also considerable merit in tests that identify epidemic diseases and provide alerts to the presence of these to allow timely interventions to prevent the spread.[5] The opportunity to diagnose metabolic disorders in individual cows may improve with better in-line detection systems; however, many of the disorders for which herds are at risk considered in this review may be better diagnosed at the herd level. Consequently, our review is both qualitative and quantitative and evaluates existing assays and those where milk assays may be valuable with a particular focus on BTM.

Energy deficiency and excess

The temporal clustering of metabolic disease to very early lactation limits the potential to use milk samples to diagnose hypocalcemia, hypomagnesemia, clinical ketosis, lipid mobilisation disorders, displaced abomasum, and metritis. Severe nutrient deficiency states for lactating cattle result from inappropriate management, primarily nutritional management, or disease. The genetic and phenotypic potential to produce greater amounts of milk and milk solids

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Final Project Summary

P2d: Diet and Heat Load

May 2026

Managing cows in hot, humid weather

In hot, humid conditions, cows eat less, resulting in reduced milk production, loss of bodyweight and more health, reproductive and welfare challenges.

Australian dairy farmers use a range of strategies to manage their herds in this weather. These include fans, sprinklers and management adjustments to diet, mating and daily routine.

Dairy UP research

This project investigated how heat load and diet interact to influence milk production in intensive dairy systems.

The research examined how milk yield changed over time in response to changes in total mixed ration (TMR) diet composition and hot weather as measured by temperature humidity index (THI).

A time-series analysis was undertaken, using data from eight commercial dairy farms and local weather data. The analysis was used to identify both the immediate and delayed effects of dietary and environmental changes on milk production.

Unlocking the potential of cows

Dairy UP's P2 project aimed to unlock the potential of dairy cows to achieve their genetic potential under NSW conditions.

P2 was a suite of seven projects that collectively explore ways to profitably increase both productivity and wellbeing in commercial settings.

P2a: Cattle Longevity

P2b: Early Alerts

P2c: Milk as a Diagnostic Tool

P2d: Diet and Heat Load

P2e: Calf Husbandry

P2f: Infectious Diseases ('Infectome')

P2g: Heifers Early Calving

This document provides an update on P2d: Diet and Heat Load.

Key findings

Changes in diet can affect milk production for up to four weeks. Increased heat load affects milk production for up to two weeks after the heat event, reinforcing the value of forward planning during hot conditions.

The research also highlighted the importance of balancing starch and fibre in total mixed rations to maintain milk production and reduce the risk of rumen dysfunction.

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Benefits

The findings provide a clearer understanding of the acute and long-term effects of diet and heat load on milk production over time.

The information can better equip farmers to manage and plan for hot weather events, including the days following.

Better heat load management can improve dairy cow health, reproduction, production, and welfare. This contributes to the profitability of NSW dairy businesses and reinforces NSW dairy farmers' social license to operate.

Research approach

Weekly diet samples, milk production records and weather data were analysed from eight commercial dairy farms over an average of 58 weeks. All herds were fed a total mixed ration, with average milk production ranging from 31 to 52 L/cow/day.

The study examined how changes in nutritional components including starch, non-fibre carbohydrates (NFC), crude fat and protein were associated with changes in milk production, and whether these effects occurred immediately or after a delay.

Time series analysis

Time-series analysis was used to investigate how milk production was influenced over time by dietary changes and weather conditions, measured using temperature humidity index (THI).

This approach analysed data collected at consecutive points in time, allowing researchers to identify both immediate and delayed responses in milk production following changes in diet composition or THI.

Results

Delayed effects of diet changes

The study showed that diet changes can affect milk production for up to four weeks after the change. The lag effect depended on dietary components. Carbohydrate-related changes had their greatest effects on milk yield around four weeks later, while crude fat and neutral detergent insoluble crude protein (NDICP) had more immediate effects within one week.

Heat load and milk production

Increases in temperature humidity index (THI) were negatively associated with milk production, confirming the impact of heat load under Australian conditions.

Starch levels and milk yield

Farms feeding higher-starch diets – averaging more than 24% starch – generally had higher milk yields than farms feeding lower-starch diets. However, these diets were more likely to have been sampled from the cows earlier in lactation.

However, within these higher-starch diets, further increases in starch concentration were associated with reduced milk yield, suggesting higher starch levels were leading to rumen dysfunction or subclinical acidosis.

Time-series analysis

The study demonstrated the value of time-series analysis for understanding how nutrition and heat load interact to influence milk production over time.

PhD

Andrew Lean, Charles Sturt University

Journal articles

Lean, A.K.G., et al. 2026 *Effects of changes in total mixed ration nutrition and maximum temperature humidity index (THI) on dairy cow milk yield* (to be submitted to Journal of Dairy Science)

Collaboration

Thank you to the farmers and farm staff who contributed to this research by collecting samples for more than a year.

More info

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P2 final report

DairyUP
Unlocking potential

Final Project Summary

P2e: Calf husbandry

May 2026

The early life performance of calves on dairy farms is strongly associated with lifetime productivity in the herd. Calves reared on dairy farms may include both female calves for replacements and non-replacement calves reared for dairy beef or other markets.

Improving calf care reduces antimicrobial use, lowers mortality rate, improves dairy business profitability and helps maintain social licence.

There were two elements to the project:

- Automated calf rearing.
- Rearing Holstein steers for dairy beef.

Unlocking the potential of cows

Dairy UP's P2 project aimed to unlock the potential of dairy cows to achieve their genetic potential under NSW conditions.

P2 was a suite of seven projects that collectively explore ways to profitably increase both productivity and wellbeing in commercial settings.

P2a: Cattle Longevity

P2b: Early Alerts

P2c: Milk as a Diagnostic Tool

P2d: Diet and Heat Load

P2e: Calf Husbandry

P2f: Infectious Diseases ('Infectome')

P2g: Heifers Early Calving

This document provides a final update on P2e: Calf Husbandry.

Automated calf rearing

The project was undertaken as part of Dr Sarah Legge's PhD thesis, titled '*The lasting impact of calf performance*'.

The study focussed on calves reared with automatic feeders on a large commercial dairy farm in NSW following industry best practices.

Research approach

The aim of this research was to determine the factors affecting calf growth and if there's an opportunity to intervene to improve lifetime performance.

This research was 'farmer-led' in that the project arose from the farmer's observation of a relatively large, unexplained variability in the size of calves at weaning. Despite uniform management and adherence to best practice standards, there was concern that calves with problems were not always identified early enough for intervention or prevention of issues. The impact of this variability on lifetime productivity and profitability was not known.

Real-farm data underpinned the three studies in this research. On-farm records and the automatic feeders provided a rich data set including birth dates, weights at various ages, health indicators and feed intake.

The first and second study followed a cohort of 1,440 calves reared on automatic feeders.

The third study involved modelling, drawing upon a dataset of more than 14,000 calves.

Key findings

The first study looked at factors affecting the growth of calves reared with automated feeders. It found a wide variation (60kg) in weaning weight, despite a very low mortality rate (<0.1%). Birth weight, visitation and

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consumption had the greatest impact on weaning weight.

Underperforming calves on automatic feeders could be identified from day 5 in the feeder system (cumulative consumption), offering the opportunity for early management intervention.

The second study followed the long-term survival and lactation performance of the 1,440 calves in the milking herd. It looked at the association between weaning weight and future lactation performance (yield and number of lactations).

Of the 1,440 calves reared in the first study, only 55 reached their fourth lactation in the herd:

- 406 were removed between weaning and their first lactation (surplus to farm requirements or not in calf).
- 233 were removed after their first lactation.
- 392 were removed after their second lactation.
- 354 were removed after the third lactation.

A heavier weight at weaning translated to higher production (especially in lactation 1 and 2) and a longer life in the herd.

Weaning weight was more strongly associated with survival in the herd than birth weight. For example, calves with a weaning weight of 100kg were more than twice as likely to remain in the herd for three or more lactations than calves with a weaning weight of 50kg (40% compared with 17%).

Weaning weight and cumulative consumption at day 5 had a significant impact on total lifetime milk production.

The third – modelling – study investigated the effect of preweaning factors on removal from the herd, using a dataset from more than 14,000 animals from the same herd.

Birth weight, weaning weight and days in milk significantly influenced the likelihood of a cow being removed from the herd. There were three points in the lactation where cows were more likely to be removed from the herd: early lactation (transition period), around 128 days in milk (not in calf) and after 305 days (long lactation cows). Cows with a lighter weaning weight were more likely to be removed from the herd early.

On-farm implications

This study quantified strong and positive associations between the performance of female dairy calves at weaning and their subsequent performance as lactating animals.

The findings provide the basis to develop early alert systems for calves being reared with automatic calf feeders.

This work is an example of the huge opportunity to use passive data collection from modern technologies such as automated calf feeders to improve farm management.

Further research

Further research is currently being undertaken by masters student, Kimberly Church. This project is using automatic calf feeder data from multiple commercial dairy farms to investigate whether similar patterns occur across different rearing systems and management conditions.

The project aims to:

- Quantify differences in calf weaning weight across commercial dairy systems and identify the main factors influencing these.
- Investigate how early-life calf performance is linked to later productivity, fertility and longevity in the herd.
- Develop early-life risk indicators that may help predict poorer lifetime performance.
- Help identify lower-performing animals earlier to improve management and culling decisions.

This broader dataset may help identify practical early-life indicators that can be used to improve calf-rearing strategies, herd management and long-term productivity outcomes.

Holstein steers for dairy beef

There were two elements to this study:

- Feeding strategies
- Feedlot adjustment

The feeding strategies study tested whether an integrated dairy-beef management strategy developed in Spain could be adapted for rearing Holstein steers under Australian conditions, including systems for the ‘antimicrobial-free’ beef market.

DairyUP
Unlocking potential

Two lifetime feeding strategies were compared for 72 Holstein steers from 3–7 days of age through to slaughter at 452 days:

- A lower starch (38%) diet containing antimicrobials (monensin and flavophospholipol).
- A higher starch (47.5%) diet using yeast products instead of antimicrobials.

The steers were evaluated for rumen function, microbial adaptation, production and carcass characteristics.

The feedlot adaption study investigated the differences between high- and low-performing steers during the first three weeks after feedlot induction. Ninety-four Holstein, British and European breed steers were assessed for rumen fermentation, growth performance, carcass traits and indicators of acidosis risk.

Key findings

Feeding strategies: Both feeding systems successfully produced dairy-beef animals suitable for the Australian red meat supply chain.

Steers fed the lower starch antimicrobial-containing diet:

- Produced heavier carcasses.
- Had higher dressing percentage and greater fat cover.
- Showed greater rumen fermentation activity.
- Had a lower risk of subclinical ruminal acidosis late in the feeding period.

The higher starch antimicrobial-free system also successfully finished steers for beef production, although liver abnormalities were more common and carcass weights were lower.

Different feeding strategies resulted in different patterns of rumen microbial development over time, highlighting the long-term impact of early nutrition and feeding management on rumen function.

The feedlot adaptation study found:

- Steers with higher weight gain during feedlot induction achieved better growth rates and more desirable carcass traits at slaughter.
- Rumen fermentation profiles alone did not explain differences in performance.
- Overall rates of rumen lesions and other abnormalities were low under good management.

Collaborators/funders

The P2e project was a collaboration between SciBus, the University of Sydney and Charles Sturt University, Meat & Livestock Australia, Church and Dwight and participating dairy herds.

Read more

S.W.J. Legge, et al. (2024) Milk consumption and behaviour of calves in automated calf-feeders as early indicators of weaning liveweight, JDS Communications Volume 5, Issue 5, Pages 474-478

Golder HM, et al. (2025) Different lifetime dietary strategies affect carcass characteristics and rumen function in Holstein steers. *Animal Production Science* 65

H.M. Golder, et al. (2024) Shifts in rumen profiles of Holstein steers fed antimicrobial and starch diets. Journal of Dairy Science, Vol 107 (Suppl 1), pg 431 (abstract 2712)

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P2 final report

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RESEARCH PAPER
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ANIMAL PRODUCTION SCIENCE

Different lifetime dietary strategies affect carcass characteristics and rumen function in Holstein steers

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OPEN ACCESS

ABSTRACT

Context. How to improve integration of male dairy calves into red meat supply chains. **Aims.** To evaluate the effects of two diets differing primarily in starch, antimicrobial content and milk replacer volume on: (1) short- and long-term rumen adaptation; and (2) lifetime production in Holstein steers. **Methods.** Holstein males, 3–7 days old (n = 72; 36 steers/treatment; six per replicate), were randomised to control (CON; 6 L milk replacer/calf.day; 38.2% of DM lifetime dietary starch; 50 ppm monensin, 20 ppm flavophospholipol) or treatment (TRT; 4 L milk replacer/calf.day; 47.5% of DM lifetime starch diets with yeast products) strategies. Calves were fed milk replacer twice daily for 42 days, but different pre-starter (Days 0–24), starter (Days 25–99) and finisher diets (Days 100–452). Ruminal fluid was collected at 104, 200 and 438 days old (14 days pre-slaughter) from 24 steers (2 per replicate). Fermentation, production and carcass measures were analysed by mixed models; ruminal bacterial genera were centre log transformed and subjected to redundancy analysis. **Key results.** The CON group had a higher risk of subclinical ruminal acidosis at Day 438 than the TRT group ($P < 0.001$). Liver abnormalities were 17.1% (CON) and 31.3% (TRT). The CON group had greater fermentation, with 138.4 ± 5.6 mM of total volatile fatty acids versus 111.6 ± 5.6 mM (TRT), and 8.4 mM higher acetate and 18.1 mM higher propionate, but pH was 0.31 units less ($P < 0.050$). Shannon diversity increased over time ($P < 0.001$) and was greater for the TRT group at Day 200, compared with the CON group ($P = 0.013$). Bacterial composition differed at each treatment by time comparison ($P \leq 0.01$), with variation increasing over time from 8.6 to 19.2%, suggesting the different diets lead to different microbial successions. The CON steers finished with 12 kg heavier carcasses than the TRT steers, with 0.8% greater dressing percentage, 1.5 mm more fat at P8 and 1.7 mm more rib fat ($P < 0.050$). **Conclusions.** CON diets produced better carcass weights, P8 and rib fat, and had more fermentability than the TRT diets, likely reflecting better long-term adaptation. **Implications.** Both diets enabled integration of dairy calves into the red meat supply chain, but with differing lifetime rumen adaptations.

Keywords: carcass weight, dairy beef, flavophospholipol, liver abscess, monensin, rumen adaption, rumen microbiome, ruminal acidosis.

Introduction

The Australian dairy industry has a considerable surplus of male dairy calves for which there are limited markets and animal welfare concerns (Vicic *et al.* 2022; Bolton *et al.* 2024); a challenge also faced by other countries (Creutzinger *et al.* 2021; Maher *et al.* 2021; Van Selm *et al.* 2021). There is a need to improve how 'dairy beef', the term for meat that originated directly or indirectly from dairy herds (Berry 2021), is integrated into red meat supply chains. Consequently, the challenge that exists with dairy beef is how to efficiently utilise the opportunity that these calves provide.

Diets with a high starch content (Hill *et al.* 2016; Hu *et al.* 2018; Chishti *et al.* 2021) or that use in-feed antimicrobials, such as monensin, tylosin and flavophospholipol, to stabilise the rumen (Pfaller 2006; El-Waziry *et al.* 2022; Asad *et al.* 2023; Firkins and Mitchell 2023)

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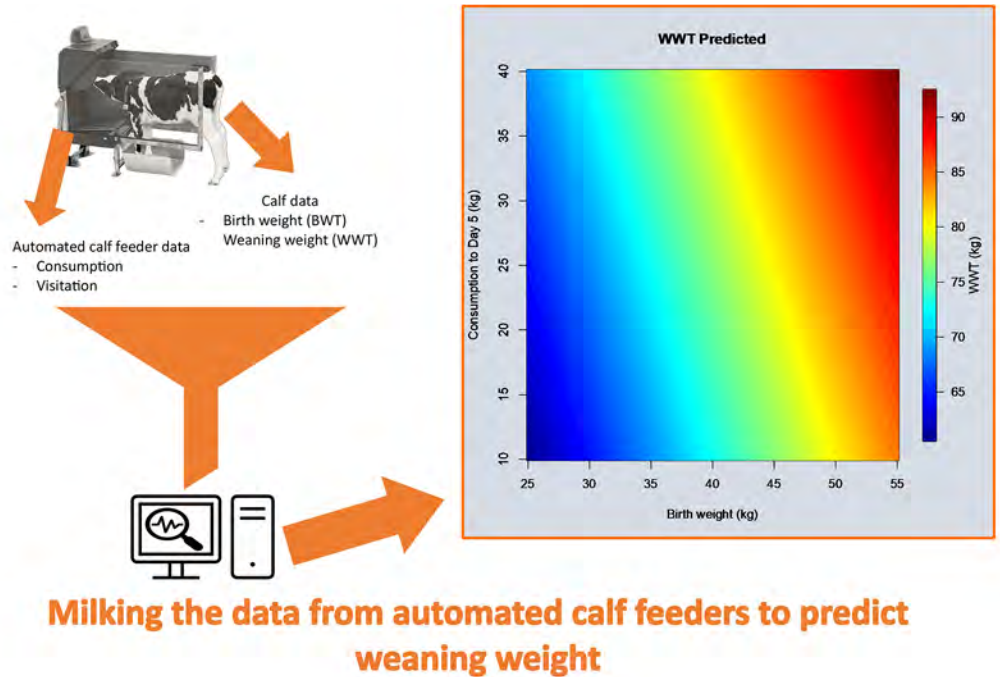
Short Communication
Health, Welfare, and Behavior

Milk consumption and behavior of calves in automated calf feeders as early indicators of weaning liveweight

S. W. J. Legge,[1,2]* P. C. Thomson,[2,3] C. E. F. Clark,[1,2] and S. C. García[1,2]

Graphical Abstract



Summary

This study suggests that by monitoring calves' consumption and visitation frequency using automated calf feeders on day 5, farmers can identify underperforming calves early, allowing for timely intervention to boost their growth potential. Additionally, calves exhibiting higher milk consumption and increased interaction with the feeder during the first 5 days are more likely to achieve a weaning weight exceeding 75 kg. This highlights an opportunity for management intervention in cases where calves fall below these consumption and interaction thresholds.

Highlights

- Weaning weight among calves showed considerable variability, ranging from 41 to 118 kg.
- Milk consumption and number of visits are strong indicators of performance.
- Intervention points could be effective as early as 5 days.

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The list of standard abbreviations for JDSC is available at adsa.org/jdsc-abbreviations-24. Nonstandard abbreviations are available in the Notes.

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Short Communication
Health, Welfare, and Behavior

Milk consumption and behavior of calves in automated calf feeders as early indicators of weaning liveweight

S. W. J. Legge,[1,2]* P. C. Thomson,[2,3] C. E. F. Clark,[1,2] and S. C. García[1,2]

Abstract: Modern intensive dairy farming relies on data to aid and prioritize management decisions made on farm. Decisions made early in an animal's life can have lasting effects on welfare, productivity, longevity, and profitability. Precision technology such as automated calf feeders (ACF) allow the customization of feeding programs, but despite this, weaning weights (WWT) vary substantially between calves. This observational study used a 3-yr dataset comprising 1,440 female Holstein Friesian calves at a single intensive commercial dairy farm (Dairy Australia feeding system 5; indoor, total mixed ration) using ACF to (1) determine the variability in WWT (as a proxy of animal performance) of calves within this system; (2) identify the contributing factors responsible for the variation in WWT; and (3) identify potential early management intervention points that could be indicative of the performance of calves at weaning within the system. Calves entered the ACF at 10 d of age with 12 calves per ACF; calves were weighed at birth and weaning using weigh scales. We discovered a large range of calf WWT (41–118 kg/head) at ~60 d of age despite the application of strict uniform management protocols. Our results from modeling showed that WWT was significantly and positively associated with birth weight (BWT), with low BWT calves (<36 kg) achieving an average of 70 kg weight at weaning. In contrast, heavier BWT (>36 kg) calves achieved an average of 82 kg at weaning. Based on calf feeder data, cumulative milk consumption and cumulative unrewarded visits to the feeder, as well as BWT, were identified as indicators of greater WWT as all these were highly significant terms in the model for WWT. Results suggest that quantifying consumption and number of visitations to the ACF at d 5 may allow farmers to identify, with time to intervene, calves underperforming within the feeder or system, therefore increasing their potential for growth. Additionally, this demonstrated that greater milk consumption (>30 kg) and interaction with the feeder up to d 5 in the feeder is more likely to yield a WWT >75 kg, identifying a potential point for management intervention for calves below consumption and interaction thresholds (e.g., by developing alarm systems based on consumption or visitation number).

Automated calf feeders (**ACF**) are an alternative to manual milk feeding. Computer-controlled systems control the amount of milk offered and typically are associated with housing calves in groups while providing individualized feeding of milk. They provide a method to rear calves in groups, especially in intensive systems, by allowing variable amounts of milk to be fed individually several times a day, in some circumstances resulting in an improved rate of weight gain and welfare (Fujiwara et al., 2014) and reducing health-related issues (Jensen, 2003). Automatic calf feeders record the time and number of visits to the feeder, whether the visit was rewarded with milk, the total time spent feeding and total milk consumption for that visit, and the speed of milk consumption in milliliters per minute. However, these data have been predominantly used for the surveillance of calf health (Lowe et al., 2019) rather than to identify factors affecting individual weaning weight (**WWT**) variability, or to estimate future productivity or key points of intervention for calves that are underperforming in terms of daily live weight gain. The performance and growth of calves are known to have an effect on future productivity (Heinrichs and Heinrichs, 2011). Anecdotal evidence suggests that the variability among individual calves (within a group or pen) in their WWT is considerable, although such variability has not been properly quantified through research. Thus, there is a clear gap in knowledge, and therefore an opportunity to explore the potential of the data generated by ACF to quantify individual animal variability, identify factors affecting it, and assess the value of data as a predictive tool of animal performance. The objectives of this cohort observational study were to (1) determine the variability in WWT of animals within this system; (2) identify the contributing factors responsible for the variation in WWT; and (3) identify potential early management intervention points that could be indicative of the performance of calves at weaning within the system.

Ethical approval was not needed as the study was an observational study and only routine animal husbandry procedures were performed. Preweaning calf data from February 2017 through June 2019 were used from an intensive commercial dairy farm milking 1,200 dairy cows in New South Wales, Australia, which operated under strict uniform management to Australian standards. The farm protocol for managing newborn calves was as follows: the calf was removed from the dam as soon as the calf was found, and navels were dipped with 7% iodine. Calves were fed 4 L of colostrum as soon as possible following birth. A second feeding of colostrum

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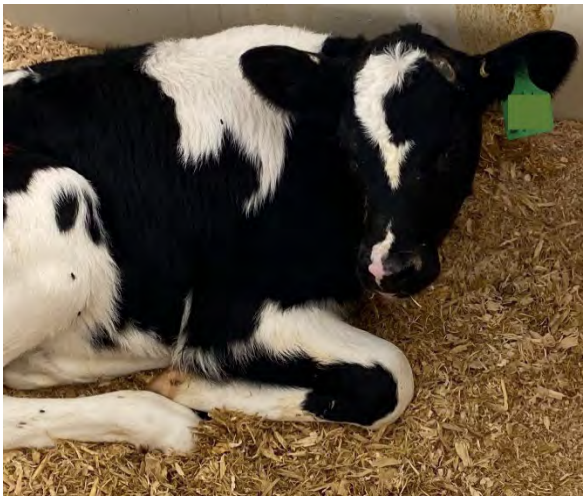
Final Project Summary
P2f: Infectious Diseases

May 2026



Diagnosing scours and respiratory diseases in calves

Scours and respiratory diseases are common causes of death in young dairy calves. On-farm, affected calves are often treated based on symptoms without identifying the microbes involved.

This project investigated the occurrence of viruses, bacteria and parasites in NSW dairy herds. It identified a much wider range of microbes than is routinely included in standard diagnostic tests. Some microbes associated with disease were also commonly present in healthy calves. The results suggest the presence of a microbe alone does not determine disease risk. Pathogen load, calf age and farm management practices also influence whether disease occurs.

The findings provide new insights to support future improvements in diagnosis, treatment and disease prevention strategies. They also provide a foundation for improved disease surveillance, including earlier identification of new or emerging disease-causing strains that may affect the dairy industry.

This project was closely linked with Dairy UP's P2b project – Early Alerts.

Unlocking the potential of cows

Dairy UP's P2 project aimed to unlock the potential of dairy cows to achieve their genetic potential under NSW conditions.

P2 was a suite of seven projects that collectively explored ways to profitably increase both productivity and wellbeing in commercial settings.

- P2a: Cattle Longevity
- P2b: Early Alerts
- P2c: Milk as a Diagnostic Tool
- P2d: Diet and Heat Load
- P2e: Calf husbandry
- P2f: Infectious Diseases ('Infectome')
- P2g: Heifers early calving

This document provides a final update on P2f: Infectious Diseases.

Benefits

Earlier and more accurate identification of the microbes causing disease may help improve treatment decisions, reduce unnecessary antibiotic use and support efforts to minimise antimicrobial resistance.

Although this work focussed on NSW dairy calves, the diagnostic approaches developed may have broader application across Australian dairy regions. This may become increasingly important as extreme weather events and environmental change alter patterns of infectious disease.

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Research approach

The research team used an approach called "metagenomics" to identify bacteria and viruses circulating in NSW dairy calves. Metagenomics analyses all genetic material in a sample and compares it with sequence databases of all known organisms to sort out which organism is present in the samples.

This knowledge may help improve future diagnostic testing and inform future treatment decisions. This work involved visiting farms, collecting samples and laboratory sequencing.

A second part of the study investigated *E. coli,* one of the most common causes of infection in animals and humans, including the different genetic types present in calves and the occurrence of antimicrobial resistance.

In early 2023, the team received an additional $434k in funding from the Australian Research Council's Linkage scheme. This funding expanded work on microbial surveillance and helped build the knowledge base needed to improve future diagnostic tools for calf diseases.

On farm samples

The Dairy UP team visited 72 dairy farms from all NSW dairying regions to collect nasal and faecal samples from calves up to 7 weeks old, including all sick calves and up to 10 healthy calves. In addition, samples were collected from up to 10 cows that had calved within the past 50 days.

Participating farmers were also asked to fill in a survey to gain insights into relevant management practices such as management of colostrum, bedding materials, nutrition, and calf rearing management. Survey responses were received from 52 of the farms.

Sequencing

About 1900 nasal and rectal swabs were collected on the collaborating farms. These samples were sequenced to identify specific 'strains' of the microbes present.

Key results

Many of the microbes detected were also present in healthy calves, showing that the presence of a pathogen alone does not necessarily mean it is causing disease. The findings suggest factors such as pathogen load, calf age and farm management practices also influence whether calves become sick.

Viruses

Rotavirus

An important finding was that Rotavirus A was found on almost all farms including some with vaccination programs.

The dominant genotypes were G6 and G10, which are both included in currently available vaccines. We need to further investigate if there are genetic differences within the genotype that may compromise the vaccine efficacy.

New viruses

The study also identified a range of viruses that were not previously thought to be common on NSW dairy farms. Here are some examples.

- The relatively recently discovered Kobuvirus was present on most of the farms tested.
- Adenovirus (not usually tested by labs) was present on half of all farms.
- Rhinitis A and Rhinitis B were commonly detected in calves with respiratory disease.

In addition, Pestivirus was found on 17 different farms.

The team prepared a series of fact sheets, collating current knowledge about 11 diseases for farmers and vets.

Bacteria

The team often detected genes known to be used by bacteria to cause disease. These genes are commonly found in *E. coli* and *Campylobacter* (associated with scours).

Most farms had *Mycoplasma*, but Mannheimia, Moraxella and Pasteurella were the bacteria most often associated with respiratory disease.

Bacterial disease patterns differed markedly between farms, highlighting the importance of management practices such as hygiene, colostrum management, calf housing and stocking density in disease risk.

Parasites

The study identified several parasites associated with scours in calves, including Cryptosporidium

("Crypto"), Entamoeba, Giardia and Coccidia, demonstrating the diversity of microbes involved in calf scours.

Implications of findings

Rotavirus
While the findings support routine vaccination for rotavirus, logistical issues can compromise its effectiveness and farmer confidence in the value of investing in vaccination.

Vaccinated cows pass antibodies to their calves through colostrum. An annual booster, given about six weeks before calving is required to ensure maximum colostrum antibody levels. This vaccine is sold in multi-dose vials with a limited shelf life once opened. This can pose a challenge in year-round calving herds where only a small number of cows calve each week. The cost per dose becomes prohibitive if the whole vial can't be used before expiry. The alternative is to use all the doses, which involves vaccinating some cows outside of the optimum timing.

This dataset provides a foundation for evaluating whether currently circulating virus strains are sufficiently similar to vaccine strains to provide adequate protection — a key question given that vaccine mismatch can substantially reduce efficacy.

Salmonella
In the past, Salmonella has been considered an important microbe in calf scours and respiratory disease but Salmonella was only identified in one sample. Detection of Salmonella was rare in our study (preweaned calves), and with the methods used.

Pestivirus
Pestivirus A is known to be an important virus in cattle. This virus was found on 17 farms in the study, with most possessing the 1c genotype, some with the 1a and 1b genotypes. The presence of even a single clinically sick animal should not be ignored, as it may signal a persistently infected animal circulating within the herd. A single case carries the risk of significant economic losses due to reproductive failure, calf mortality, immunosuppression leading to increased respiratory disease and overall ill-thrift, resulting in chronic production losses such as reduced milk production and increased culling rates.

Diagnostics and antimicrobial resistance
Scours is recognised as having multiple causes and not all of the potential pathogens are covered by the scours vaccine.

The findings also highlight limitations of current diagnostic approaches that test for only a small number of pathogens. Future diagnostic tools may need to consider pathogen load, calf age and farm-specific management practices when interpreting results.

The project established a baseline for monitoring antimicrobial resistance in disease-causing bacteria on Australian dairy farms, supporting future surveillance and antimicrobial stewardship efforts.

PhD students

Two PhD students enrolled at the University of Technology Sydney (UTS) are working on the project.
Zain Ul Abedien
Aleksandra 'Ola' Stanczak

Collaborators

The P2f project was a collaboration between researchers from Dairy UP, UTS, NSW DPI Elizabeth Macarthur Agricultural Institute, Scibus and the 72 participating dairy farms.

Read more

Fact sheets
Bovine Norovirus factsheet
Bovine Parechovirus factsheet
Bovine Picornaviruses factsheet
Bovine Rhinitis A & B factsheet
Bovine Toravirus Breda factsheet
Bovine Adenovirus factsheet
Bovine Enterovirus factsheet
Bovine Giardiasis factsheet
Bovine Hunnivirus factsheet
Bovine Kobuvirus factsheet
Bovine Nebovirus factsheet

Bovine Pestivirus infection, NSW DPI primefact sheet

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Journal articles

Brito Rodriguez et al., *Next-generation detection in bovine respiratory and enteric diseases: metagenomic and amplicon sequencing insights into microbial diversity*. Frontiers in Veterinary Science, Volume 13 2026.

Abstracts ADSA meeting, 2025

- Brito et al, The enteric and respiratory viral diversity of calves in health and disease – a state-wide study.
- Kida et al, Genetic diversity of Bovine Rotavirus in Dairy Calves with and without diarrhoea in New South Wales, Australia.
- Abedien et al, Microbial diversity in dairy calves: an insight of the resistome and virulome in health and enteric disease.
- Brito et al, Untargeted meta-transcriptomic methods to characterize the enteric infectome of calves with and without diarrhea.
- Brito et al, Expression of virulence factors and antimicrobial resistant genes in total RNA sequenced from rectal swabs from diarrheic calves.
- Brito et al, The respiratory infectome of dairy
- calves characterized by a total RNA sequencing approach.

P2 final report

Project lead

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www.dairyup.com.au

Delivery organisations

The University of Sydney – Dairy Research Foundation
NSW Government – Department of Primary Industries and Regional Development
Scibus
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Fact Sheet: Adenoviruses

P2f Infectious Diseases

August 2025

Bovine Adenoviruses (BAdVs)

Adenoviruses are a group of viruses that infect cattle. They commonly cause scours and respiratory diseases, usually in calves.

Antibodies for BAdVs are commonly found in cattle, indicating widespread exposure, most likely as young animals.

Clinical signs and pathogenesis

Clinical signs of BAdV infection vary depending on the form of disease. The respiratory form is characterised by coughing, nasal and ocular discharge, fever, and laboured breathing.

The enteric form presents as diarrhoea, which may range from watery to haemorrhagic, often accompanied by depression, recumbency, and in severe cases, sudden death.

Systemic involvement, particularly associated with BAdV-7, can lead to weak calf syndrome and multi-organ disease.

Transmission

BAdVs spread through faecal-oral and respiratory routes, often shed by animals without symptoms. Stressors like weaning and transport can trigger outbreaks.

Diagnosis

Diagnosis of BAdV infection relies on a combination of histopathology, molecular testing, and serology. Intranuclear inclusion bodies in endothelial cells are characteristic findings on histopathology.

Treatment

There is no specific antiviral treatment for BAdV infections. Management involves supportive care, and treatment of secondary bacterial infections if present.

Key points

- Different types of BAdVs, cause different symptoms ranging from respiratory to scours (enteric) and systemic disease.
- There is no specific antiviral treatment for BAdV infections. Management involves supportive care, and treatment if secondary bacterial infections are present.
- Infections may be triggered by stress and worsened by co-infections such as Bovine Viral Diarrhea Virus (BVDV).
- There is currently no commercially available vaccine in Australia.
- Many cattle have antibodies for BAdVs indicating widespread exposure, most likely as young animals.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

About Dairy UP

Dairy UP is a research and development program to help NSW farmers unlock the potential of their dairy businesses. Led by the University of Sydney's Dairy Research Foundation, Dairy UP is delivered through NSW DPIRD, Scibus, Dairy Australia, and the University of Sydney.

Control and prevention

Control of BAdV infections relies on good hygiene, strong biosecurity, and reducing stressors such as weaning and mixing calves from different sources.

While a vaccine for BAdV-7 is available in Japan, there are currently no widely used commercial vaccines elsewhere.

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Epidemiological notes

BAdV-3 and BAdV-7 are frequently linked to bovine respiratory disease complex (BRDC), while BAdV-10 is associated with fatal hemorrhagic enteritis in calves. Co-infections, particularly with bovine viral diarrhoea virus (BVDV), can worsen clinical outcomes.

Seroprevalence studies indicate high herd-level exposure, with up to 82% of cattle testing positive for BAdV antibodies, although specific virus types are not always identified.

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More info

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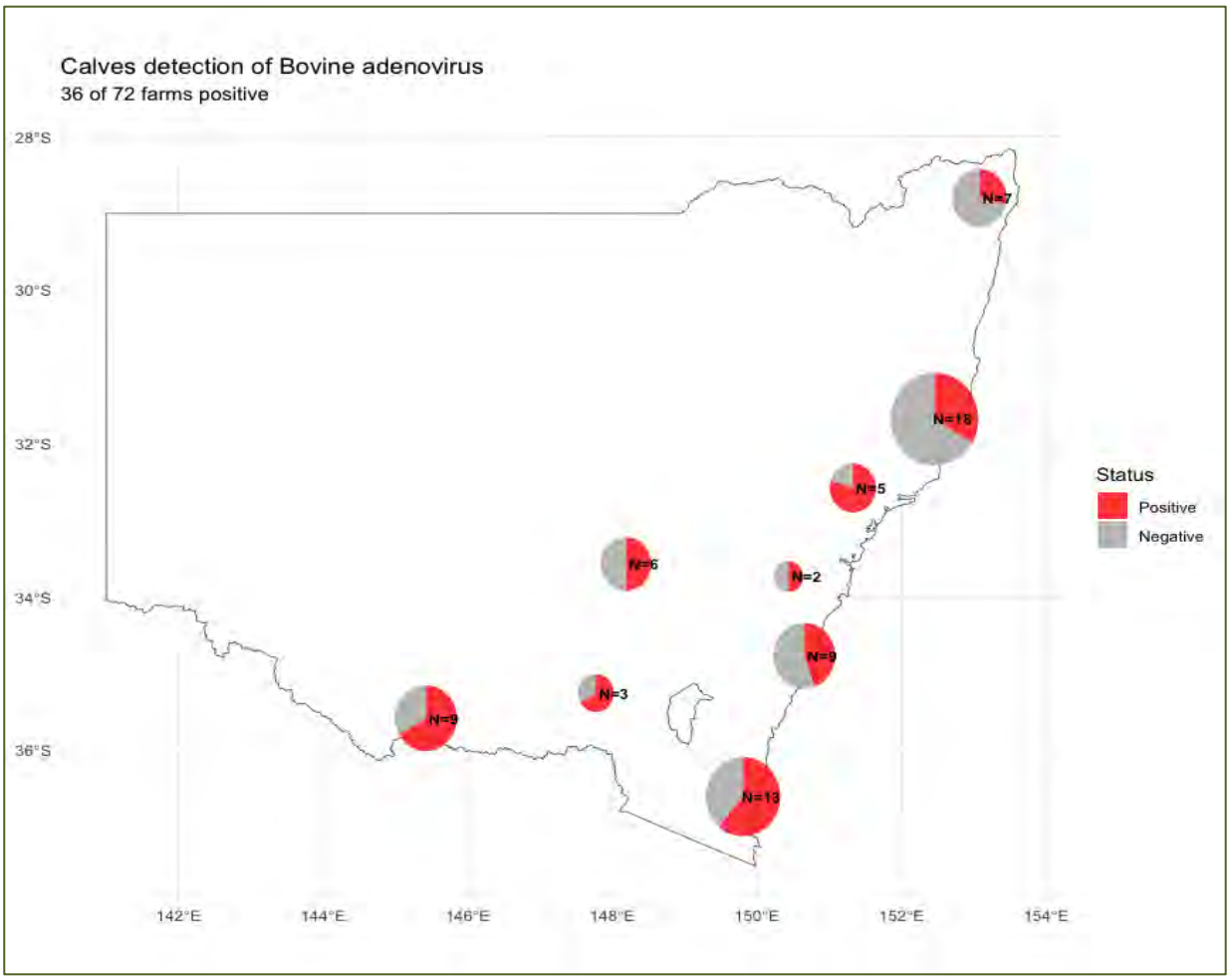

The map shows only BAdV-3, which was the most prevalent. We did not find 7 in samples collected, and only one sample was positive to 6.

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Australian Government – Australian Research Council
UTS

Fact Sheet: Bovine enteroviruses

P2f Infectious Diseases

August 2025

Bovine enteroviruses (BEVs)

Bovine enteroviruses are viruses that live in the gut of cattle. They are common in cattle all over the world and often found in healthy animals.

Most of the time they don't make the animal sick, but sometimes they cause mild scours or other gut problems in calves.

Transmission

Transmission is usually faecal-oral: infected calves shed the virus in their manure and it is spread to other calves when they swallow manure usually from dirty teats, feeders or bedding. Enterovirus can also spread through aerosols, contaminated soil or water, and vertically (from cow to calf before birth).

These viruses are widely distributed in cattle populations worldwide and are generally considered to have low virulence, often detected in healthy animals.

Bovine enteroviruses (BEVs) are small, non-enveloped, positive-sense RNA viruses in the family Picornaviridae, genus Enterovirus. There are two species: Bovine enterovirus E (subtypes E1–E4) and Bovine enterovirus F (F1–F6). These viruses are widely distributed in cattle populations worldwide and are generally considered to have low virulence, often detected in healthy animals.

Clinical signs and pathogenesis

BEVs are usually asymptomatic but can occasionally cause mild diarrhoea, and rarely respiratory, reproductive (abortion, stillbirth), neurological (encephalitis, ataxia), or cardiovascular signs.

Pathogenesis is unclear; experimental infections show inconsistent disease despite viral replication and localization in the terminal ileum, ileocecal and cecocolonic junctions, spiral colon, lungs, and muscle tissue.

Types of bovine enteroviruses

Bovine enteroviruses (BEVs) are small, non-enveloped, positive-sense RNA viruses in the family Picornaviridae, genus Enterovirus. There are two species: Bovine enterovirus E (subtypes E1–E4) and Bovine enterovirus F (F1–F6).

Key points

- Bovine enteroviruses (BEVs) are common in healthy cattle all over the world.
- They occasionally cause mild scours; rarely respiratory, reproductive, neurological, or cardiac signs.
- It spreads mainly via manure but can also spread in the air, contaminated environment, and from cow to calf before birth.
- There is no specific treatment or vaccine. Control relies on hygiene and supportive care.
- It's not clear how this virus leads to illness. Trials show it can grow in parts of the gut, lungs, and muscle, but development of disease is inconsistent.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

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Laboratory detection

Detection relies on RT-PCR targeting the 5' UTR. Virus isolation in cell cultures (MDBK, BHK-21, FBK) and electron microscopy can confirm infection. Serological surveys show antibodies in cattle and humans, though no zoonotic disease is confirmed.

Treatment and control

There is no specific antiviral treatment; care is supportive, focusing on hydration and managing secondary infections. Prevention depends on biosecurity, clean feed and water, as no vaccines are currently available.

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More info

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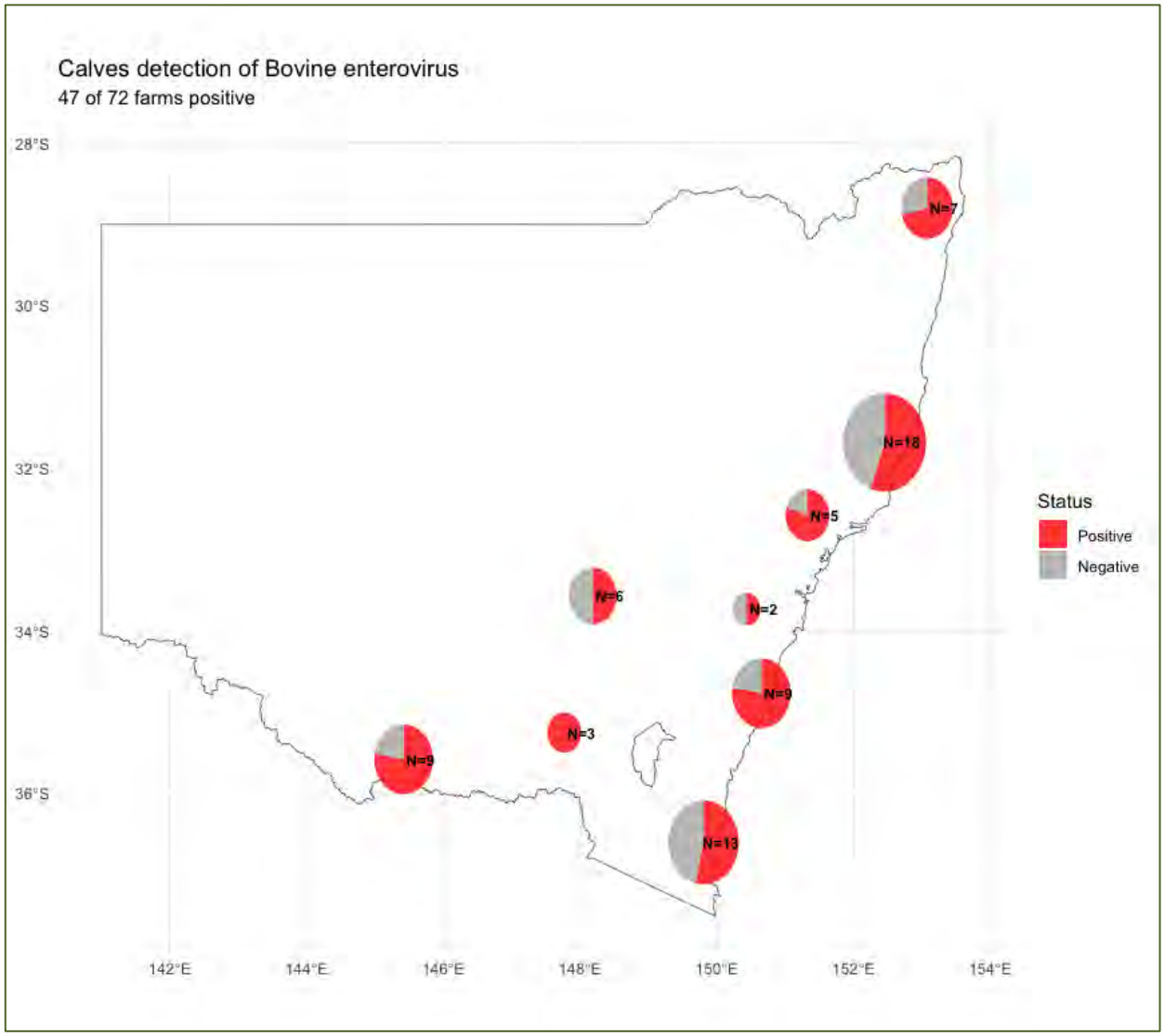

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Fact Sheet: Bovine Giardiasis

P2f Infectious Diseases

August 2025

Bovine Giardiasis

Giardiasis is gut illness that affects calves (especially under 8 weeks), lambs, and humans. Infected calves may show no symptoms, or have mild scours, reduced weight gains or ill thrift.

It is caused by a parasite (protozoa) that infects the small intestine - *Giardia duodenalis* (also known as *G. intestinalis* or *G. lamblia*).

It is spread between animals as the parasite passes out in manure as cysts, which survive in the environment and infect other cattle when swallowed.

Species affected

Calves and lambs are the most likely to be affected by Giardiasis.

Severity

The severity of symptoms varies by age and strain (assemblages A, B, E). Studies show negative effects on growth. Treatment reducing cyst shedding improves weight gain in calves.

Transmission

The parasite is spread via the faecal–oral route through ingestion of infective cysts. Transmission occurs through contaminated water, surfaces, and direct contact with infected animals.

Laboratory detection

Common diagnostic tools include coproantigen ELISA, microscopy, direct immunofluorescence, and PCR.

Treatment and control

Treatment with fenbendazole is effective. Control relies on hygiene: cleaning and disinfecting pens, removing manure, allowing empty periods, and minimizing contact with shedding animals. Coccidiostats are not effective.

Key points

- Bovine Giardiasis is a protozoan parasite that infects the small intestine of calves, lambs, and humans.
- Symptoms are often mild but it can cause scours, poor growth, and ill-thrift.
- It is spread via manure; cysts can persist in the environment.
- Diagnosis is by ELISA, microscopy, immunofluorescence, or PCR.
- Treat with fenbendazole; control through hygiene and biosecurity.
- It is unclear whether *G. duodenalis* can be transmitted from cattle to humans; more research is needed on transmission and prevalence.

Microbial surveillance in dairy cattle

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Zoonotic potential

G. duodenalis is an emerging zoonotic pathogen. Assemblage A poses the greatest risk for human infection.

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Research gaps

Unclear clinical impact in cattle, limited data on assemblage distribution, zoonotic transmission dynamics, and molecular diversity. More studies are needed on epidemiology and risk factors.

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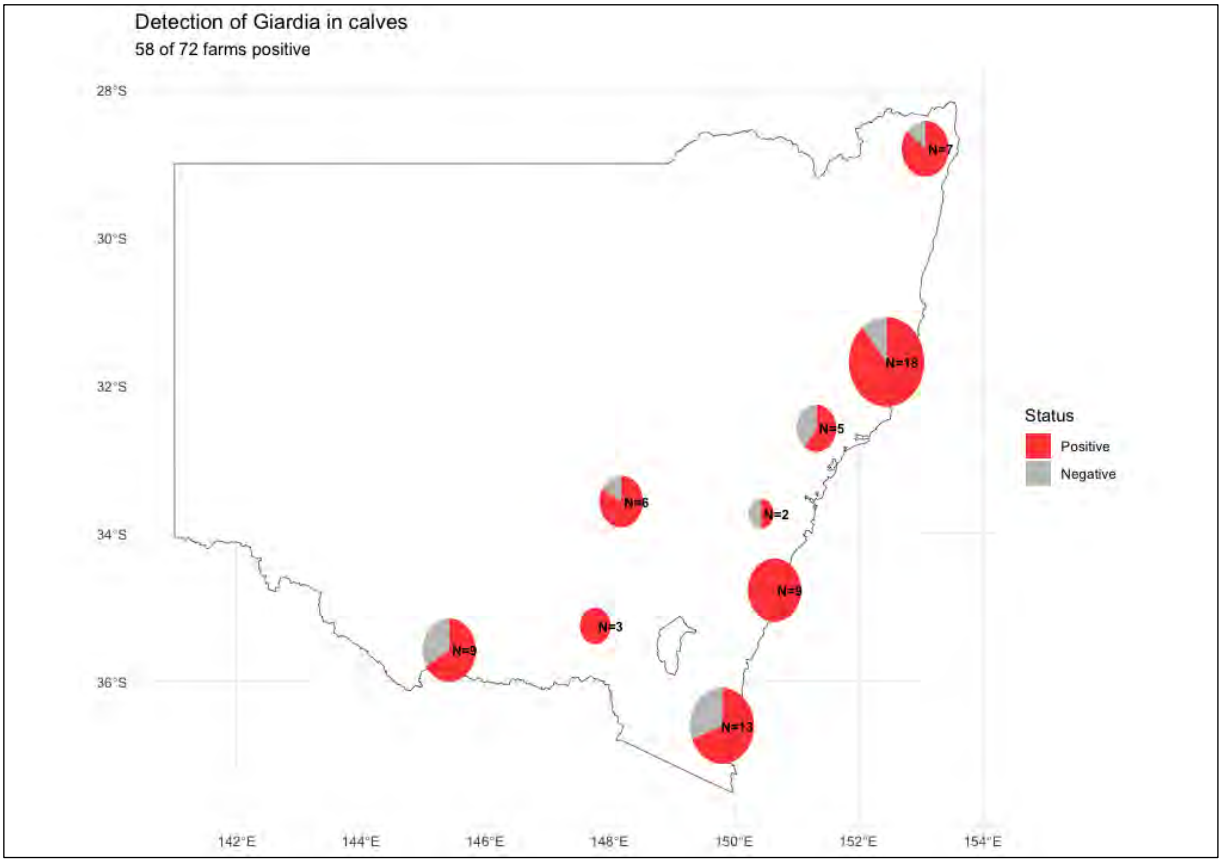

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Fact Sheet: Bovine Hunnivirus P2f Infectious Diseases

August 2025

Bovine Hunnivirus

Bovine hunnivirus (BHuV) has been detected in cattle in multiple countries, but its role in causing disease remains unclear. It was detected on all farms in our study.

There is no consistent pattern of clinical symptoms – it has been detected in calves with scours as well as in healthy animals. There are no reports of respiratory or systemic illness in cattle. It is thought to be spread via the faecal-oral route.

Other species

Laboratory studies show that a related hunnivirus in buffalo can damage kidney and intestinal cell lines after 72 hours, but no tissue damage has been observed in animals. In other species, hunniviruses have been linked to gastrointestinal disease and occasionally to heart, liver, or brain issues. The tissues targeted by BHuV in cattle remain unknown, but in buffalo, the virus may suppress immune responses by blocking interferon production.

Transmission

Transmission is likely faecal–oral, though this has not been experimentally confirmed.

Laboratory detection

Routine testing for BHuV is not widely available. Detection primarily relies on RT-PCR to identify viral RNA in faeces. Virus isolation has been achieved in Madin-Darby bovine kidney (MDBK) cells, and ELISA has been used in seroprevalence studies, revealing low antibody levels in cattle. No standardized diagnostic protocols or culture methods are currently in place.

Notes

Bovine hunnivirus (BHuV) is a non-enveloped, single-stranded RNA virus in the family Picornaviridae (genus Hunnivirus).

The causal role of BHuV in disease has not been established. Although it is frequently detected in diarrheic calves, it is also found in healthy animals, and no definitive link to illness has been proven. Further research is needed to clarify its pathogenic significance.

Key points

- Bovine hunnivirus (BHuV) is a picornavirus detected in cattle worldwide.
- Its role in causing disease is unclear. It is found in calves with and without diarrhoea.
- There is currently no evidence it causes respiratory or systemic disease in cattle.
- More research is needed to find out which tissues the virus affects and how this links to disease.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

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Fact Sheet: Bovine Kobuvirus

P2f Infectious Diseases

August 2025

Bovine Kobuvirus (BKV)

Bovine Kobuvirus (BKV) mainly affects calves, especially newborns and 1–2 months old. It is detected in both healthy and scouring cattle worldwide.

There is no vaccine or antiviral. Prevention relies on biosecurity and hygiene. Treatment involves supportive care for scours (fluids, electrolytes).

Clinical signs

The main reported symptom of BKV infection is diarrhoea. Its role remains unclear due to frequent co-infections and detection in healthy animals. One study linked BKV to necrotizing enteritis in calves that tested negative for other pathogens.

Transmission

Transmission is believed to occur via the faecal-oral route which is typical of gut enteric viruses. The high prevalence suggests widespread exposure in cattle populations.

Control and prevention

There is no specific antiviral treatment for BKV. Supportive care for diarrhoea (fluids, electrolytes) is standard.

No vaccine is available for BKV. Prevention relies on strict biosecurity and good hygiene practices to minimize exposure.

Bovine Kobuvirus (BKV)

Bovine Kobuvirus (BKV) is a non-enveloped, single-stranded, positive-sense RNA virus in the Picornaviridae family, Kobuvirus genus (species Aichivirus B). The name "kobu" is derived from the Japanese word for "bump," reflecting its characteristic appearance under electron microscopy.

Key points

- Diarrhoea is the main reported symptom, but BKV is often found in healthy animals.
- Its role in disease is unclear as BKD is frequently co-detected with other gut pathogens.
- No vaccine is available. Prevention relies on biosecurity and hygiene.
- It is found globally in cattle and small ruminants, suggesting widespread exposure.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

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Laboratory detection

BKV is diagnosed mainly by RT-PCR or real-time RT-PCR targeting the 3D gene. Virus isolation is possible in Vero cells, showing cytopathic effects. It can also be detected in faecal samples using metagenomic sequencing and specific PCR assays.

Epidemiology and research gaps

In some outbreaks, BKV has been detected more frequently than traditional enteric viruses. Its pathogenic role in calves remains uncertain. Only one study has linked BKV to histopathologic changes consistent with primary viral enteritis in pathogen-negative calves. In goats, experimental inoculation with a related kobuvirus (Aichivirus C) confirmed its ability to cause diarrhoea.

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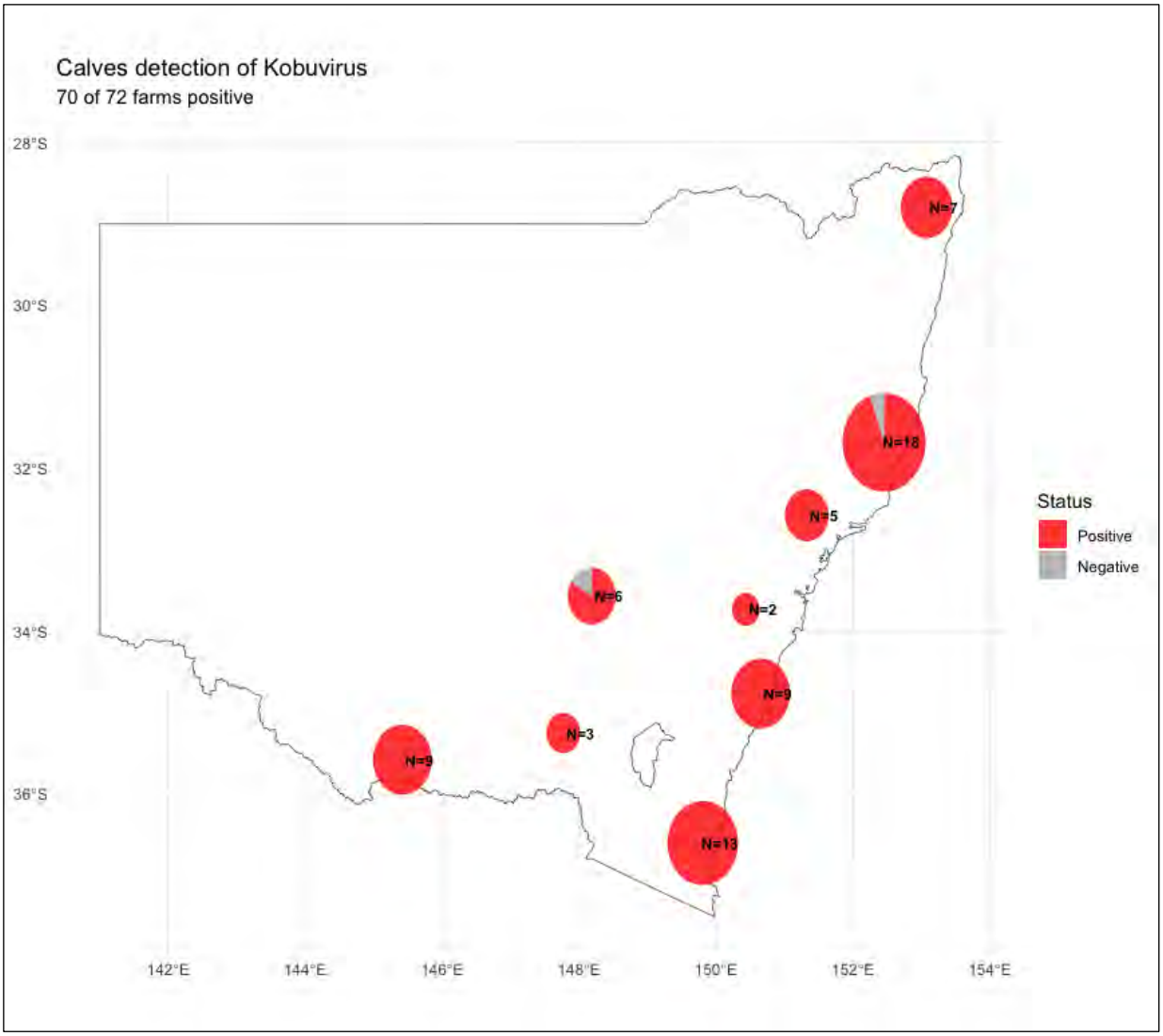

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Fact Sheet: Bovine Nebovirus

P2f Infectious Diseases

August 2025

Bovine Nebovirus (BoNeV)

Bovine nebovirus causes scours in calves with varying severity, often accompanied by loss of appetite and lethargy. No vaccines or specific treatments exist. Control relies on hygiene and supportive care.

Transmission

BoNeV spreads mainly through direct contact between animals and the faecal-oral route: infected calves – even those without symptoms shed the virus in their manure for extended periods. It is spread to other calves when they come in contact with from contaminated feed, water and equipment.

Clinical signs and pathogenesis

Bovine nebovirus infections mainly affect young calves, causing diarrhoea that can range from mild to acute, along with loss of appetite, lethargy, and temporary weight loss. In some cases, malabsorption occurs, meaning calves may not properly digest certain nutrients, which can slow growth and affect overall condition.

Treatment and control

There are no commercial vaccines or specific treatments for BoNeV. Management focuses on supportive fluid therapy for diarrhoea and strict hygiene measures, such as cleaning pens and separating calves, to reduce transmission.

Post-mortem, pathological findings

Bovine nebovirus primarily affects the upper small intestine (duodenum and jejunum), causing villus atrophy, crypt hyperplasia, and mucosal inflammation. Viral replication occurs in the intestinal epithelial cells. These changes impair nutrient absorption, leading to diarrhoea and poor growth. Rarely, severe cases may show necrotizing hepatitis and intestinal haemorrhages.

Key points

- Bovine nebovirus (BoNeV) is an enteric calicivirus affecting mainly young calves.
- It causes scours of varying severity, often with loss of appetite and lethargy; malabsorption may occur.
- The virus targets the upper small intestine, causing villus atrophy and crypt hyperplasia.
- It is spreads via the faecal-oral route; asymptomatic calves can shed the virus.
- No vaccines or specific treatments exist; control relies on hygiene and supportive care.
- Research is limited due to lack of cell culture systems and diagnostic tools, leaving its full pathogenic role unclear.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

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Laboratory detection

RT-PCR is the primary tool for detecting BoNeV. Other methods include electron microscopy, immunofluorescence, and antigen capture ELISA, though commercial diagnostic kits are not widely available.

Genetics

Bovine nebovirus (BoNeV) is an enteric virus belonging to the genus *Nebovirus* in the family *Caliciviridae*. It is genetically distinct from bovine

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noroviruses but shares similar structural features typical of caliciviruses. BoNeV is a recognized enteric pathogen in cattle, especially young calves, and has been linked to diarrhoea outbreaks. It may act alone or in combination with other gut pathogens.

Bovine nebovirus (BoNeV) includes two main genotypes: the Nebraska (NB) strain from the USA and the Newbury1 strain from the UK. A related virus, Kırklareli virus, has been detected in Turkey and is genetically closest to neboviruses, though it is not officially classified as one.

Research challenges

Its exact pathogenic role is unclear, it could be a primary pathogen, co-infective agent, or commensal. No cell culture system, reverse genetics tools, or animal models exist for detailed study. Limited diagnostics hinder epidemiological understanding, and its overall impact on calf health and productivity needs further investigation.

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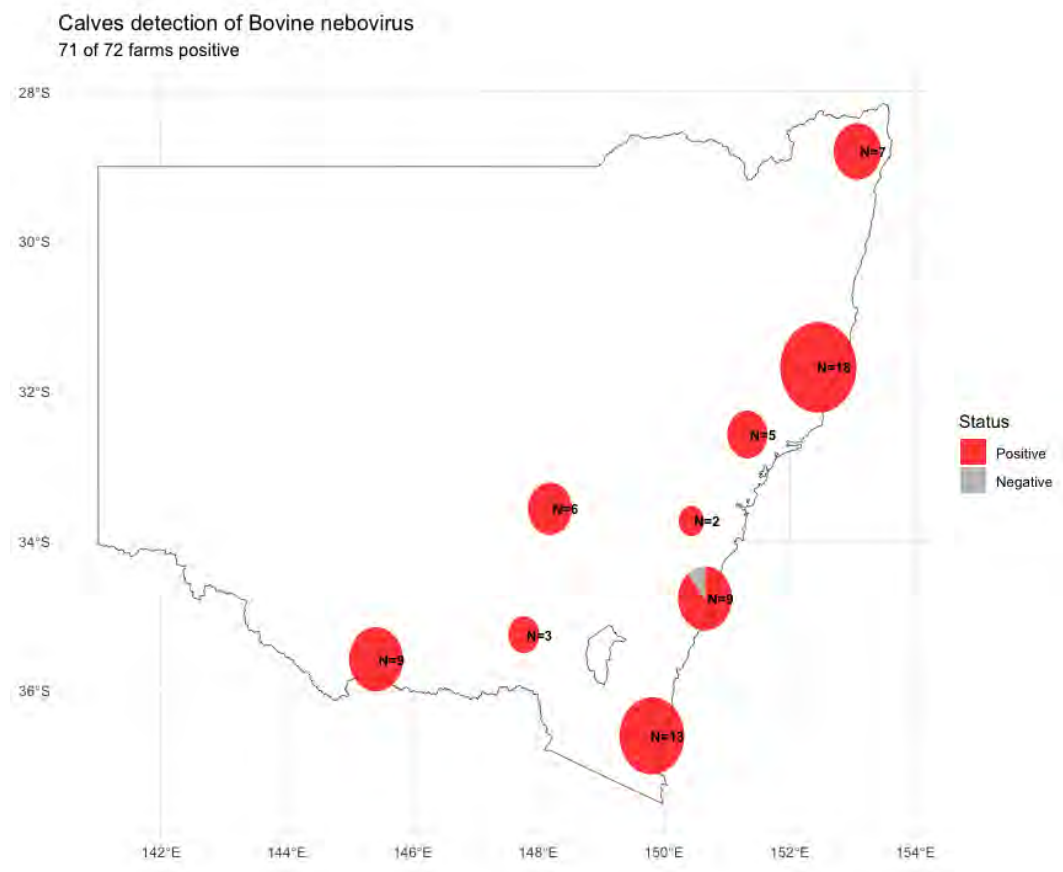

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Fact Sheet: Bovine Norovirus

P2f Infectious Diseases

August 2025

Bovine Norovirus (BoNoV)

Bovine noroviruses (BoNoVs) are found worldwide and mainly affect newborn and pre-weaned calves. Infections are uncommon in calves older than six months and disease is rarely seen in adult cattle. It was found on all farms sampled in our study.

Transmission

Bovine noroviruses spread mainly through the faecal-oral route, meaning calves become infected by ingesting the virus from contaminated manure or surfaces. Healthy-looking calves can carry and shed the virus, so infection can occur even without obvious sick animals in the group.

Treatment and control

There is no specific antiviral treatment for bovine norovirus. Management focuses on supportive care, including oral rehydration and good nutrition. Preventive measures rely on hygiene—keeping calving pens clean, minimizing faecal contamination, and isolating sick calves.

Vaccines are not yet available, but research is exploring virus-like particle (VLP) vaccines for future use.

Genetics

Bovine caliciviruses are part of the *Caliciviridae* family. Different strains can vary in how sick they make calves, with some causing worse symptoms than others. BoNoV belongs to genogroup GIII, which has four genotypes. The most important two are:

- **GIII.1 (Jena virus):** can cause more severe diarrhoea in calves under experimental conditions.
- **GIII.2 (Newbury virus):** most common in the field, usually linked to mild or moderate diarrhoea.

Key points

- Bovine noroviruses (BoNoVs) mainly affect newborn and pre-weaned calves.
- Clinical signs range from mild to severe watery diarrhoea, depending on the strain (GIII.1 more severe, GIII.2 more common but milder).
- Transmission is faecal-oral, and healthy calves can shed the virus without showing symptoms.
- No specific treatment or vaccine exists; control relies on hygiene and supportive care.
- Research is limited due to difficulties growing the virus in cell culture, slowing vaccine development.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

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Clinical signs and pathogenesis

The severity depends on the strain. GIII.1 (Jena virus) can trigger sudden, severe watery diarrhoea within 14–16 hours, lasting about three days.

GIII.2 (Newbury virus) is more common in the field and usually causes milder signs like soft stools, reduced appetite, and tiredness.

The virus mainly targets the small intestine,

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especially the jejunum and ileum. It causes villus atrophy (loss of absorptive structures) and crypt hyperplasia (increased cell growth in intestinal glands), which reduce nutrient absorption and lead to diarrhoea and poor growth in calves.

Laboratory detection

Bovine norovirus is most reliably detected using RT-PCR or qRT-PCR. ELISA tests for antigens or antibodies can be used for research or herd-level screening but are less sensitive than PCR.

Research challenges

Because they don't grow well in standard lab cell cultures, studying their role in disease has been challenging.

Historically, detection relied on electron microscopy or infecting animals. While recent advances have enabled human norovirus growth in special cell systems, these methods don't work for bovine strains yet. This limitation slows progress in understanding the virus and developing vaccines.

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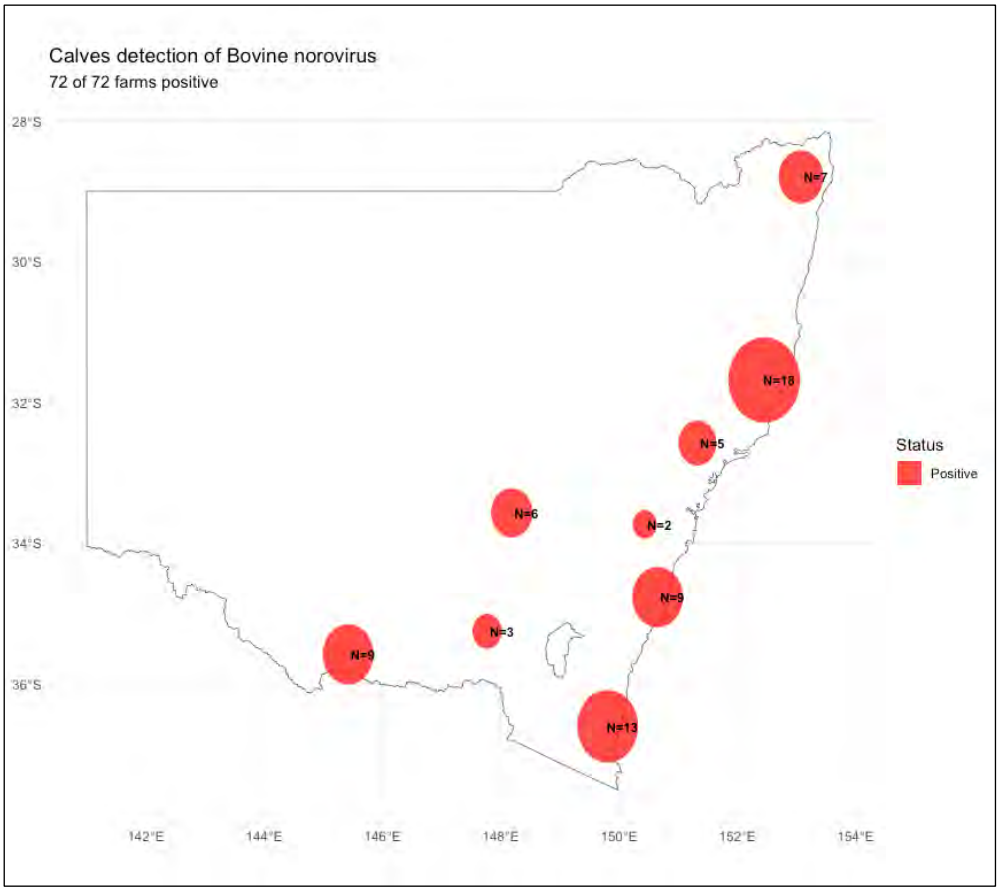

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Fact Sheet: Bovine Parechovirus

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August 2025

Bovine Parechovirus

Bovine parechovirus (BoParV) was discovered fairly recently and its role in disease is currently not clear. Knowledge about its biology and clinical relevance remains limited.

Since its discovery in 2018, BoParV has been detected in both calves and adult cattle. It's been found healthy animals and those with scours, suggesting possible subclinical infections.

The modes of transmission have not been studied but it has been detected in manure so probably spreads via the faecal oral route: infected animals shed the virus in their manure and spreads to other calves when they swallow material from contanimated manure and surfaces.

Current knowledge

Clinical signs, pathogenicity & in vitro evidence

BoParV is an unclassified virus within the genus Parechovirus (family Picornaviridae), a non-enveloped, positive-sense RNA virus. It was first identified through metagenomic sequencing.

No definitive clinical syndrome has been linked to BoParV, though it is often found in faecal samples from diarrheic cattle. Experimental infections have not been performed, and no histopathology data exist. The virus has been isolated from bovine faeces in MA104 and Marc-145 cells, producing cytopathic effects within 2–3 days after inoculation.

Its pathogenic role remains unclear, and it is frequently detected alongside other enteric pathogens.

Laboratory detection

No routine diagnostic tests or serology are available. It can be detected with real-time RT-PCR targeting parechovirus RNA and virus isolation in MA104 cells. Both of these tools are used only in research settings.

Notes

The clinical significance of BoParV is uncertain. More research is needed to clarify its pathogenicity, tissue tropism, and epidemiology.

Key points

- Bovine parechovirus (BoParV) is a picornavirus first detected in 2018.
- Its role in disease is unclear.
- BoParV is found in scouring and healthy cattle; no consistent clinical signs confirmed.
- Isolated in MA104 and Marc-145 cells; causes cytopathic effect in vitro.
- Likely faecal–oral transmission; no experimental infections or histopathology data.
- Diagnosis by RT-PCR or virus isolation (research only); no routine tests available.
- No vaccines or treatments; control relies on hygiene and biosecurity.
- Further research needed on pathogenicity, tissue tropism, and prevalence.

Microbial surveillance in dairy cattle

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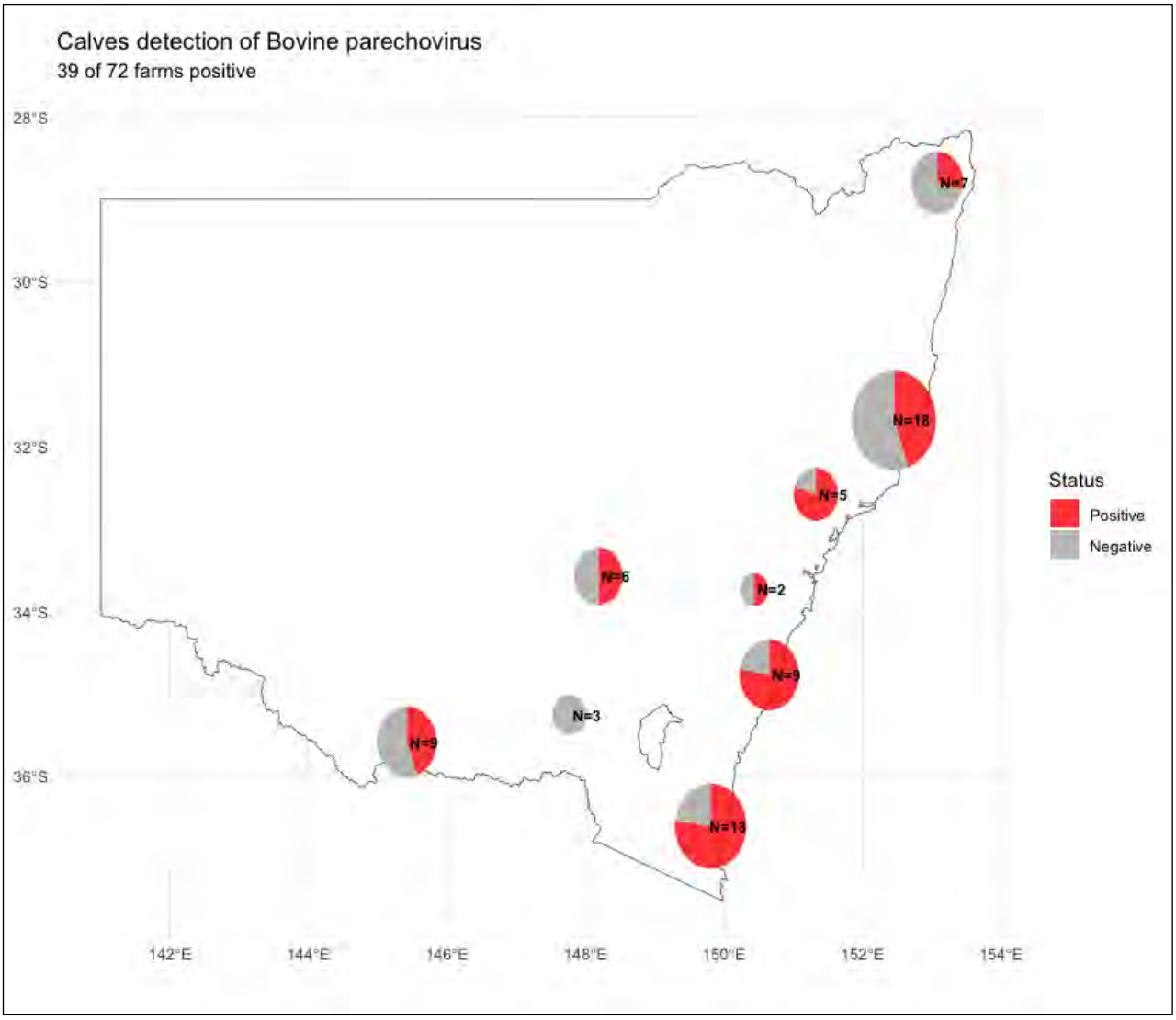

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Fact Sheet: Bovine Picornaviruses

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Bovine Picornaviruses

Boosepivirus and Bopivirus are recently identified members of the *Picornaviridae* family.

They have been detected in young livestock, mainly young cattle under 12 months old.

These viruses are most often found in manure faecal samples but their role in causing disease is unclear.

Bopivirus has also been reported in sheep and goats, though its clinical significance in these species is unknown.

Current knowledge

These viruses have been found in both scouring healthy calves, suggesting possible subclinical infections. No consistent clinical signs, experimental infections, or tissue studies have been reported, and their pathogenic potential is unknown. Virus isolation in cell culture has not been documented.

Laboratory detection

Detection relies on RT-PCR or metagenomic sequencing in research settings. No routine diagnostic tests, serology, or culture methods are currently available in veterinary practice.

Notes

The presence of these viruses in diarrheic calves may be incidental. Further research is needed to determine their relevance to cattle health.

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Key points

- Boosepivirus and Bopivirus are picornaviruses detected in young cattle;
- Their role in disease is uncertain.
- They are found in scouring and healthy calves; no consistent clinical signs confirmed.
- There have been no experimental infections, histopathology, or cell culture isolation reported.
- Diagnosis is not routinely available in labs. It can be detected with RT-PCR or metagenomics.
- The clinical significance of these viruses remains unclear; more research is needed.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

About Dairy UP

Dairy UP is a research and development program to help NSW farmers unlock the potential of their dairy businesses. Led by the University of Sydney's Dairy Research Foundation, Dairy UP is delivered through NSW DPIRD, Scibus, Dairy Australia, and the University of Sydney.

More info

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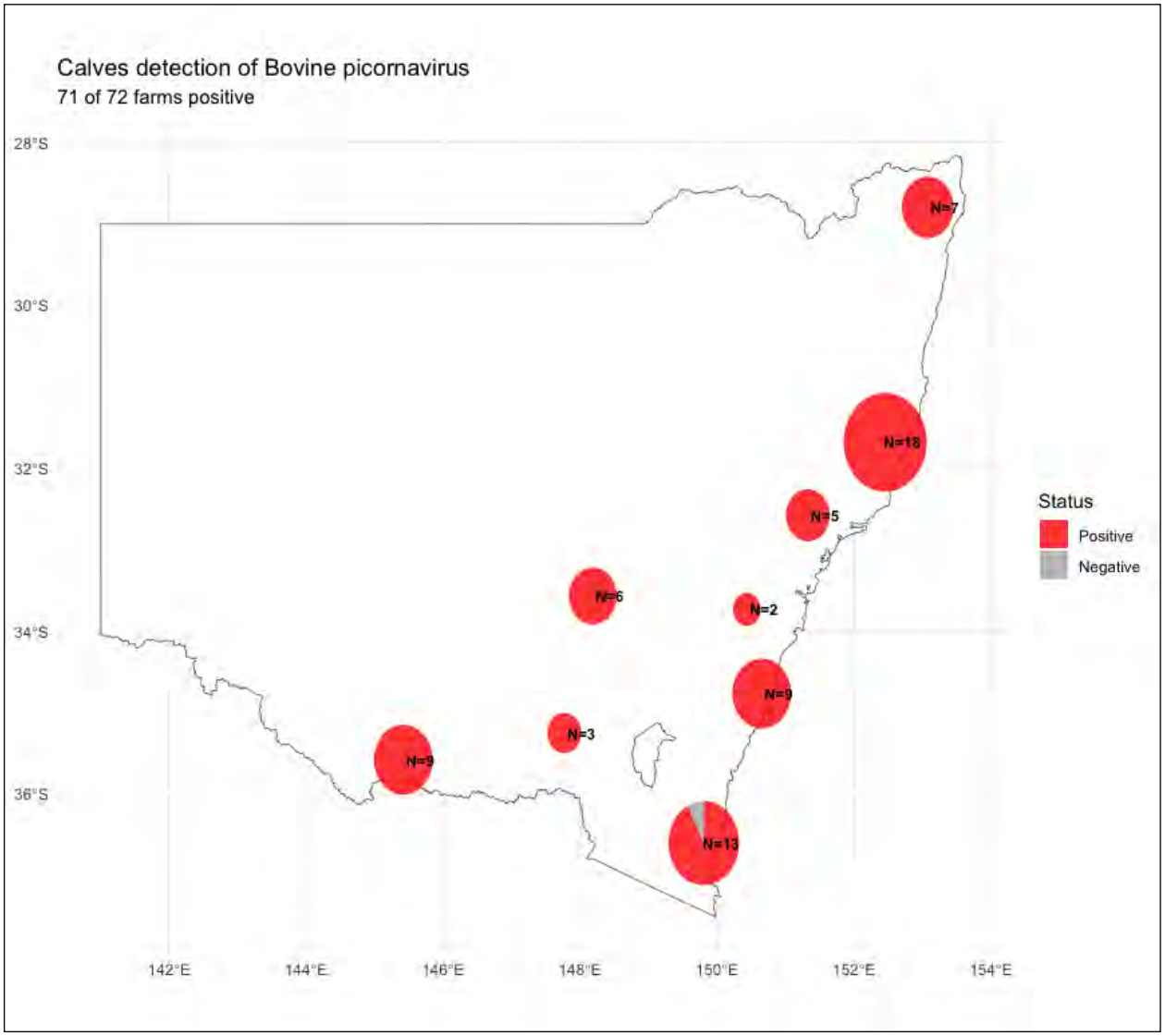

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Fact Sheet: Bovine Rhinitis A & B

P2f Infectious Diseases

August 2025

Bovine Rhinitis A & B viruses

Bovine rhinitis A and B viruses (BRAV, BRBV) contribute to Bovine Respiratory Disease (BRD). They mostly infect cattle, with limited evidence suggesting potential spillover to sheep.

Infections are usually mild but may contribute to outbreaks. No vaccines are currently available.

Transmission

BRAV and BRBV are likely transmitted via aerosol and direct contact, with spread facilitated by stress, crowding, and co-infections, conditions commonly encountered in feedlot environments.

Treatment

There is no specific antiviral therapy available for BRAV or BRBV infections. Management involves supportive care and addressing co-infections.

Control and prevention

Currently, no vaccines are available for BRAV or BRBV. Control relies on broader BRD management strategies, including minimizing transport stress, improving ventilation, quarantining new arrivals, and controlling other respiratory pathogens.

Clinical signs and pathogenesis

Clinical signs are generally mild or subclinical, including nasal discharge, coughing, sneezing, and occasionally fever or upper airway irritation.

However, BRAV and BRBV may act as primary initiators or co-factors in respiratory outbreaks, often in co-infection with other pathogens. Both viruses infect epithelial cells of the nasal cavity and trachea, with BRBV frequently localized to these tissues in experimental and natural infections. Histopathological findings are minimal, with viral RNA detected in mucosal tissues. Both viruses may impair mucosal immunity, facilitating secondary infections by other BRD pathogens.

Key points

- BARAV and BRBV are RNA viruses from the *Aphthovirus* genus, *Picornaviridae* family.
- Both contribute to the Bovine Respiratory Disease Complex (BRDC), often in co-infection with other respiratory pathogens.
- Though typically causing mild signs, they can act as initiators or co-factors in respiratory disease outbreaks.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

About Dairy UP

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Diagnosis

Bovine rhinitis A and B viruses, members of the *Aphthovirus* genus (*Picornaviridae*).

BRAV and BRBV are not available in routine diagnostic panels, yet they are increasingly recognized as contributors to the BRD.

They are primarily diagnosed using RT-PCR, which offers sensitive and specific detection. Viral isolation and sequencing are valuable for monitoring genetic diversity and tracking viral evolution. Although genetically distinct, both viruses exhibit regional variation and recombination, underscoring the importance of molecular surveillance in outbreak investigations.

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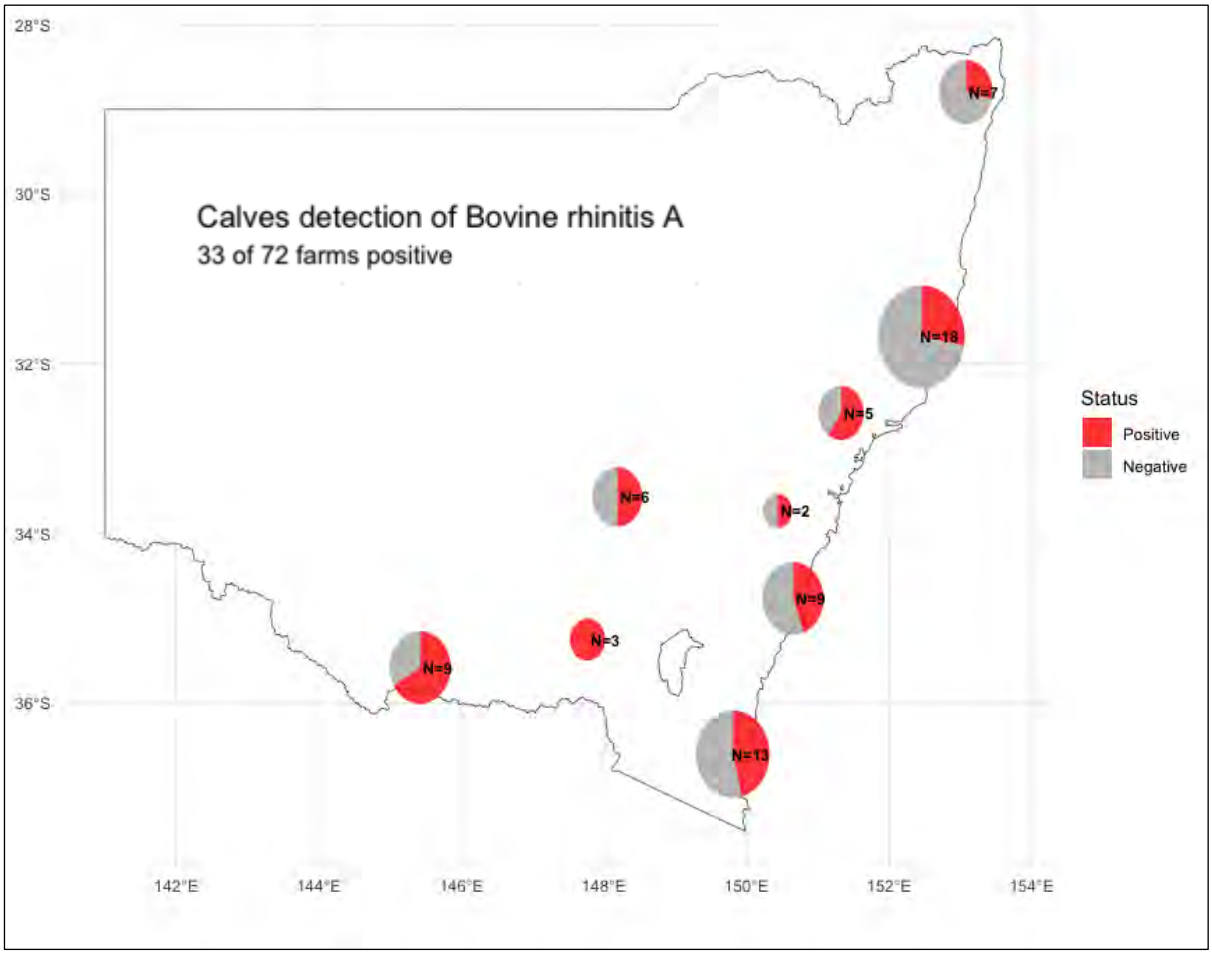

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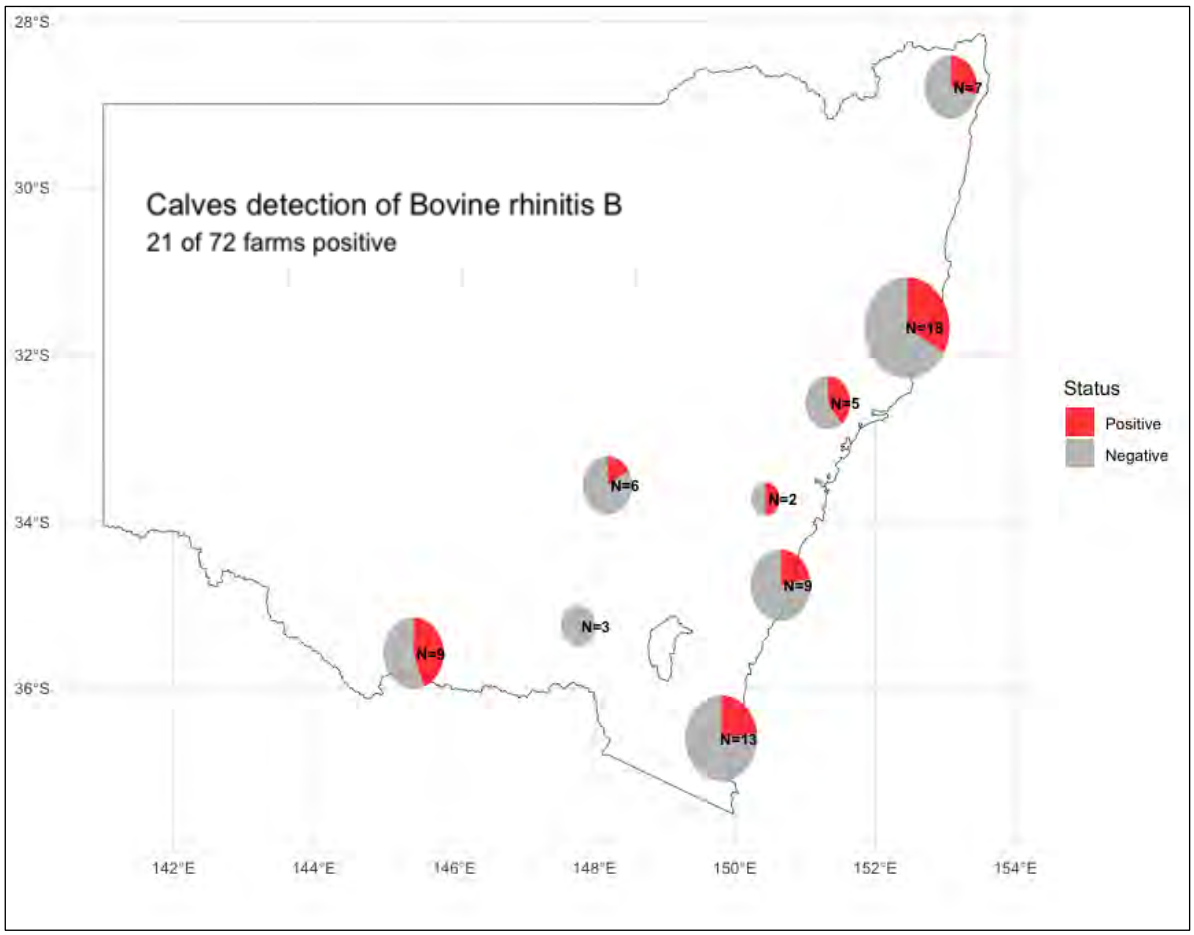

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Fact Sheet: Bovine Torovirus

P2f Infectious Diseases

August 2025

Bovine Torovirus (Breda virus)

Bovine Torovirus (BoTV) – also known as Breda virus – is recognised as a cause of scours in young calves and may also affect the respiratory tract.

It mostly affects calves 1-6 weeks old but there is widespread antibody presence in adult cattle.

This virus is widespread in cattle herds globally.

Clinical signs

The most common signs are diarrhoea, dehydration and weakness, especially in calves under 3 weeks old.

The virus has also been found in nasal secretions of calves with respiratory symptoms.

Transmission

Calves catch BoTV from other infected calves, via faeces, and possibly through the fluids in the nose and mouth.

Treatment

There is no specific antiviral available for Breda virus. Follow standard care practices for calf scours: hydration and supportive therapy.

Control and prevention

As there isn't a vaccine available, control and prevention relies on hygiene, colostrum intake, and minimising stress in calves.

Diagnosis

Bovine Torovirus (BoTV) is an RNA virus in the *Coronaviridae* family. Samples from faecal or nasal swabs can be used for RT-PCR and ELISA tests.

The virus is hard to culture; only one strain (Aichi/2004) has been isolated in lab.

Key points

- BoTV (Breda virus) is detected in both gut and respiratory tracts.
- It is commonly found with other gut and respiratory bugs but can act alone.
- The fact that many adult cattle have antibodies suggests they were exposed to the virus, likely when they were young.
- There is no vaccine available.
- BoTV is still under investigation, with ongoing studies to clarify its full impact on calf health.

Microbial surveillance in dairy cattle

This series of fact sheets has been prepared for cattle vets. It covers a range of microbes that were identified by Dairy UP team in samples collected from cattle on NSW dairy farms in 2023 and 2024. As many of these viruses are new, and knowledge about them is still emerging, we have collated current knowledge as a handy reference.

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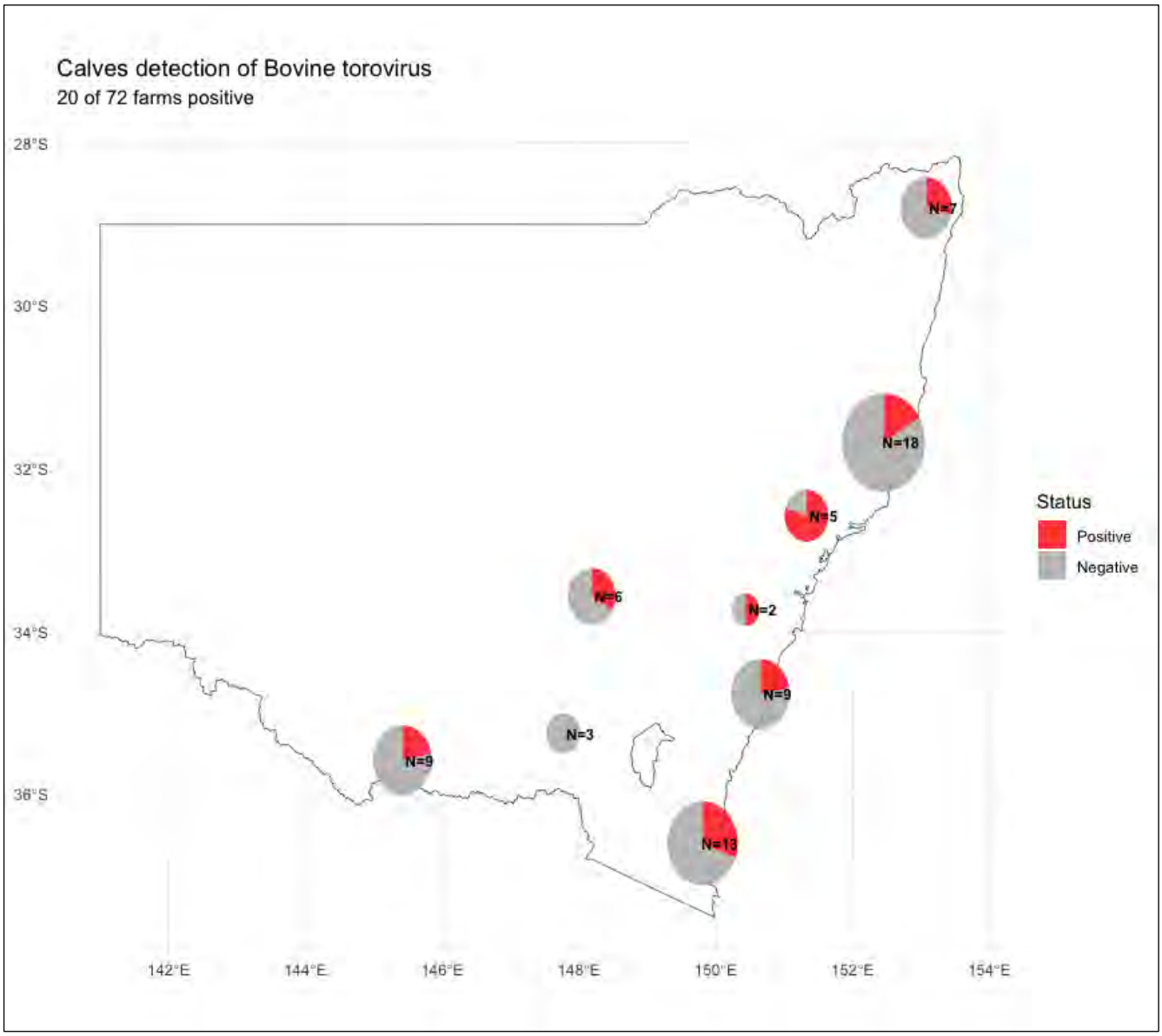

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Final Project Summary

P2g: Heifers Early Calving

May 2026

This project investigated the effects of age at first calving on dairy cow production, health, reproduction and survival.

The findings give farmers greater confidence that earlier joining of well-grown heifers can improve farm efficiency and lifetime productivity while maintaining animal performance.

Unlocking the potential of cows

Dairy UP's P2 project aimed to unlock the potential of dairy cows to achieve their genetic potential under NSW conditions.

P2 was a suite of seven projects that collectively explored ways to profitably increase both productivity and wellbeing in commercial settings.

P2a: Cattle Longevity
P2b: Early Alerts
P2c: Milk as a Diagnostic Tool
P2d: Diet and Heat Load
P2e: Calf Husbandry
P2f: Infectious Diseases ('Infectome')
P2g: Heifers Early Calving

This document provides an update on P2g: Heifers early calving.

Age of first calving

The age that dairy heifers calve for the first time affects their health, welfare and lifetime productivity along with the economics of the dairy farm business. Australian dairy farmers with seasonal/split calving systems generally aim to calve their cows at about 24 months of age. In some Australian dairy herds, the average age of first calving is closer to 36. The main benefits of calving heifers earlier are:

- Earlier return on investment in breeding replacements.
- Improved lifetime production.
- Improved reproductive performance.
- Faster genetic gain through reduced generation interval.
- Fewer unproductive animals on the farm.
- Markedly reduced greenhouse gas (GHG) emissions intensity by producing more milk across an animal's lifetime.

Some dairy farmers with pasture-based, year-round calving systems have been hesitant to join heifers earlier due to concerns about calving difficulties, lower production and whether younger heifers are adequately grown before calving.

Key findings

This project found that well-grown Holstein heifers could be joined to calve earlier without compromising overall milk production, fertility or survival. Heifers first joined before 13 months-of-age were generally more productive and less likely to leave the herd than heifers first joined after 13 months-of-age.

Some studies included in the literature review identified lower first-lactation milk production in earlier-calving heifers, but this may be offset by fewer non-productive days before first calving and comparable longer-term reproductive and survival outcomes.

The literature review also found many traditional concerns about earlier calving could be reduced through good heifer growth and management.

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Research and results

Research approach

This project investigated the effects of age at first joining and age at first calving on dairy cow production, health, reproduction and survival. The research also examined differences between pasture-based and intensive systems, along with hormonal and metabolic influences associated with earlier breeding and calving.

This project involved:

- Analysis of international research.
- Analysis of data from six commercial herds.

Analysis of international research

This part of the project involved a literature review and statistically analysing the results from multiple international studies (meta-analysis) to identify consistent trends associated with age at first calving.

The age at first calving around the world is generally between 24 and 28 months, which is similar to Australia. In the Australian Dairy UP dataset, heifers in intensive dairy systems generally calved at about 24 months-of-age, while heifers in pasture-based systems calved later at about 26.5 months, on average.

We also examined traditional concerns about earlier calving, including joining undergrown heifers, calving difficulties and lower production. Research included in the review reported no increase in calving difficulty for well-grown heifers joined at younger ages under good management conditions.

Some studies identified lower first-lactation milk production in younger-calving heifers. However, this may be offset by fewer non-productive days before first calving, with no major differences identified in reproductive performance or survival.

Commercial herds

The study included a total of 477 Holstein heifers, managed under normal conditions on six commercial farms. Heifers were grouped according to age at first joining, with comparisons made between heifers joined before and after 13 months-of-age. The target heifers mating weight was 330kg, with most being above this target.

During heifers' pregnancy and first lactation, data on production, reproductive performance, health and survival were collected.

Additional data collected during the project included bodyweight and body condition scoring, reproductive tract assessments and blood testing. The results were used to explore hormonal and metabolic factors associated with heifer growth, pregnancy and calving.

Results

Compared with heifers joined younger than 13 months-of-age, later-joined heifers produced 814 litres less milk over a 305-day period when averaged across all heifers in the study. This reflected a higher proportion of later-joined heifers failing to conceive, aborting or leaving the herd before contributing milk production data. However, there was no difference in milk production between heifers that successfully entered lactation.

Each additional kilogram of bodyweight at the start of the study was associated with an additional 13 litres of milk over a 305-day lactation, reinforcing the importance of growing heifers to target weights before joining and maintaining performance through the lifetime.

However, younger-calving heifers had a higher risk of medical treatment and stillbirth than heifers calving after 24 months-of-age.

Heifer joining age	**Early** (<13 months)	**Late** (>13 months)
First-lactation milk yield	Similar	Similar
Herd retention	Higher	Lower
Overall milk production across study group	Higher	814 L lower
Medical treatment risk	Higher	Lower

Blood testing and biochemical analysis identified differences in several hormone and protein measures associated with age at first joining. These findings may improve understanding of the biological factors influencing fertility, growth and performance of younger-joined heifers. However, further research is required to determine how these measures could be applied in practical herd management.

There were marked differences in the vitamin and mineral concentrations of heifers within and between herds, highlighting the importance of balanced supplementation with effective feed delivery in heifers.

PhD student

Andrew Lean, Charles Sturt University

Collaborators

The P2g project was a collaboration between Dairy UP, Scibus, the University of Sydney and Charles Sturt University.

Thank you to the commercial and research herds for their participation.

This project was closely linked with the following Dairy UP projects:

- P2a Cattle Longevity
- P2d Facility Design for Cow Comfort
- P2e Calf Husbandry

Journal articles

Lean A.K.G et al. 2026 Meta-analysis of the effects of age at first calving on production outcomes, calving difficulty, and reproduction in dairy heifers, Journal of Dairy Science, Volume 109, Issue 6 Pages 5819-6654

Lean, A.K.G. et al. (in review), Age at first breeding in Holstein dairy cattle: Effects on age at first calving, production, dystocia, health and reproduction in primiparous heifers.

Lean, A.K.G. et al. (in review), Age at first breeding in Holstein dairy cattle: Associations of hormone, biochemical and protein measures.

Previous research

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More info

P2 final report

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Meta-analysis of the effects of age at first calving on production outcomes, calving difficulty, and reproduction in dairy heifers

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ABSTRACT

This study aims to determine the effects of feeding, management, and reproductive strategies used to reduce age at first calving (AFC) on production and reproductive performance through meta-analytic methods. A literature search using 3 search engines was conducted to identify prospective studies that evaluated AFC for data extraction. Inclusion criteria included specific dairy breeds, a treatment group with an AFC below 26 mo of age, and prospective enrollment of heifers. There were few prospective studies suitable for meta-analysis, with 16 studies containing 35 appropriate experiments. There was substantial heterogeneity, likely due to differences in study design and interventions used to induce an AFC difference. Outcomes were assessed using classical multilevel random effects meta-analytic models with standard mean difference, effect sizes, or risk ratios being evaluated where appropriate. Meta-regression was performed using the difference in AFC. There was a loss of production for first-lactation milk (−2.06 L/d of earlier calving), fat (−0.12 kg/d of earlier calving), and protein (−0.08 of kg/d earlier calving) yield for heifers calving at a younger age than their peers. There were no differences in second- and third-lactation milk yield, calving difficulty, heifer reproductive performance, survival to calving, and survival to the end of first lactation. Although the production in first lactation is decreased, this may be offset by fewer days to first calving without compromising the heifers' ability to calve, become pregnant for the first time, or survive to the end of first lactation. Given the lack of prospective studies, more randomized controlled experiments over multiple lactations, including health, production, and reproductive data, are needed to evaluate this topic further.

Key words: age at first calving, calving, dairy heifer, lactation, meta-analysis

INTRODUCTION

Lowering the age at first calving (**AFC**) in dairy heifers has long been viewed as a potential method to improve productivity in the dairy industry. Traditionally, calving was targeted for the spring flush in pasture-based systems, with the target AFC being 24 mo of age (Berry and Cromie, 2009). In the last 60 yr, housing of cattle and year-round calving systems have led to increased investigation into enhanced heifer feeding to increase growth rates, resulting in a decrease in the AFC (Swanson, 1961).

The objectives of decreased AFC are an earlier return on investment (Hutchison et al., 2017), improved longevity (Han et al., 2022), decreased GHG emissions (Dall-Orsoletta et al., 2019), decreased generation interval, improved reproductive performance (Lin et al., 1986), and increased lifetime milk production (Krpálková et al., 2014). Potential risks include increased dystocia (Erb et al., 1985), fatty udder syndrome (Sejrsen et al., 2000), decreased first-lactation production (Lin et al., 1986), and increased stillbirths (Ettema and Santos, 2004; Hutchison et al., 2017).

The industry is trending toward a younger AFC internationally. Hutchison et al. (2017) reported a 4-fold increase in the percentage of Holstein heifers in the USA calving at 22 mo of age compared with 1997, with a similar increase in early calving in the Jersey breed. A previous meta-analysis investigated the effects of varying prepubertal growth rates on first-lactation production (Zanton and Heinrichs, 2005). Prepubertal growth rates of 836 g/d were optimal for future milk production in the first lactation, suggesting there is a limit on how early dairy heifers can calve and perform well. Studies have variable recommendations for the youngest AFC target, from 20.5 to 22 mo (Lin et al., 1988; Curran et al., 2013; Heinrichs et al., 2017; Hutchison et al., 2017; Eastham

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The list of standard abbreviations for JDS is available at adsa.org/jds-abbreviations-26. Nonstandard abbreviations are available in the Notes.

DairyUP
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FINAL OUTPUT JOURNAL

P3
Intensification of dairy systems

DairyUP
Unlocking potential

Final Report

P3 Intensification of dairy systems

Department of Primary Industries
and Regional Development

This project was led by Sheena Carter
from NSW DPIRD

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

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Project Overview

This project was originally developed by Dairy Australia and led in NSW by DPIRD. Dairy UP enhanced its delivery for the benefit of the NSW and broader dairy industry. The project was completed in June 2023 and the final report/project summary is included here.

Item	Description
Project Title	Intensification of dairy systems
Funding Body	Dairy UP
Dairy UP Project	P3
Project Duration	2022-2023
Lead Organisation	NSW Department of Primary Industries and Regional Development
Project Lead	Sheena Carter, NSW DPIRD
Research team	Claire Waterman, Dr. John Penry, Juani Gargiulo, Karen Romano, Nikki Reichelt, Prof. Ruth Nettle,
Key Collaborators	Agriculture Victoria, Dairy Australia, NSW Department of Primary Industries and Regional Development, The University of Melbourne, The University of Sydney

DairyUP
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Project summary

P3 Intensification of dairy systems | August 2023

Transitioning towards intensification

Internal and external triggers are leading some NSW dairy farmers to consider transitioning their businesses to more intensive farm feeding systems such as total mixed rations (TMRs), and housed facilities. Farmer surveys indicate that intensification in NSW is an adaptation strategy in response to climate variability and extreme weather events.

Motivations

Some of the reasons NSW dairy farmers consider intensification include:

- As a way to adapt their farming system to better handle climate variability and extreme heat or wet conditions.
- In response to changes in water policy, milk pricing and financial pressures.
- To help achieve goals for improving animal welfare, increasing milk and feed productivity, environmental sustainability, worker-friendly environments, and land access.
- To help achieve aspirations of control and consistency.

P3 Intensification of dairy systems

This project aimed to provide information to help farmers to decide if transitioning to an intensive production/feeding system is a suitable investment for their circumstances.

This work was complemented by a similar project undertaken in northern Victoria, led by Agriculture Victoria and Dairy Australia.

The findings from both the Victorian and NSW DairyUP project underpin the Farm System Evaluator, a tool that helps assess a dairy farm's readiness to change to a new farming system involving feeding and housing infrastructure.

Dairy UP's P3 project involved two areas of work:

- P3a: Economics of intensification (7 NSW case study farms)
- P3b: Social research to explore the drivers of change (including NSW case study farms).

This document provides a summary of findings from both areas of work which were completed by June 2023.

The intensification journey

A dairy farmer's journey towards intensification is context specific. Different farms have different starting points e.g., greenfield sites or incremental transitions.

The transition involves many incremental decisions such as purchasing more land, growing of crops and/or purchase of machinery.

Intensification brings new complexities in terms of deciding to invest, build and operate a contained housing system. These decisions require individual context with the support of regionally-specific information and advice, which can be challenging to find.

Pause points occur along the way such as:

- Planning - with councils for permits, selection of housing and effluent system, environmental plans
- Securing the capital for investments
- Liaising with council
- Construction of the housed system.

Management skills set

An intensive dairy system requires different management skills to pasture-based systems and has unique operational challenges. The following diagram identifies the key management skills challenges for farmers operating housed facilities, as identified by service providers.

Economic performance of NSW intensive systems

Intensive farm systems can be as profitable as pasture-based systems in NSW. Significant capital investment is required to develop intensive farm systems with housing and effluent management and the associated equipment needed to support the intensive system.

Intensive farms that were consistently profitable:

- Were well-established businesses feeding a TMR and/or managing the transition phase very well.
- Have strategic income diversification – gross farm income had a strong livestock trading component and/or feed sales.
- Tended to have very productive and efficient cows (kg MS/kg liveweight is > 100%).
- Managed feed sources very well – largely self-sufficient with fodder or a reliable external source such as unique by-products, regular feed testing and budgeting.
- Tended to have very good overhead cost control.
- Constantly looked to identify aspects of the business they could improve.

Collaborators

Read more

- **Dairy feedpads and contained housing: national guidelines**
- **Adapting Dairy Farm Systems | Dairy Australia**
  - including access to the 'Farm System Evaluator' decision support tool.
- **Economics of Total Mixed Ration Systems in Australia, Proceedings of the Australasian Dary Science Symposium 2022, p67-70**
- **Reichelt, N., Nettle, R., & Veskoukis, S. (2023).**
  Understanding the drivers of dairy feeding system intensification in NSW, including housed systems. Research report prepared for NSW DPI Dairy team, Rural Innovation Research Group, University of Melbourne.

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Delivery organisations

Partner organisations

Project supporters

Charles Sturt University | DairyBio | Eagle Direct | Entegra | Macquarie University | smaXtec | UC Davis | University of Technology Sydney

Dairy UP (www.dairyup.com.au) is an industry driven R, D & E program led by the University of Sydney's Dairy Research Foundation (DRF) and co-delivered together with Dairy Australia, New South Wales Department of Primary Industry (both Agriculture; and Biosecurity Food and Safety; EMAI), and Scibus. The program is funded through a grant by the NSW Government (SDG scheme) with co-contributions from Australian Fresh Milk Holding Ltd., Dairy Australia, (formerly) Dairy Connect, Dairy NSW, DRF, Local Land Services (Hunter), Leppington Pastoral Co, Norco Dairy Co-Op, NSW Farmers, Scibus, and South- East Local Land Services.

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August 23

DairyUP
Unlocking potential

FINAL OUTPUT JOURNAL

P4
Maize for silage

DairyUP
Unlocking potential

Final Report

P4 – Maize for silage

THE UNIVERSITY OF SYDNEY
Dairy Research Foundation

This project was led by Rafiq Islam from the University of Sydney Dairy Research Foundation

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

Proudly funded by

Delivery organisations

Additional program supporters, collaborations or partnerships

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1. Executive Summary

Contained housing systems are becoming increasingly common in the Australian dairy industry, driven by substantial investments in large-scale operations where total mixed ration (TMR, system 5) feeding is becoming a practical reality. These systems rely heavily on maize silage as a primary dietary component to reduce feed costs and to maintain production efficiency. In this context, the P4 "Feedbase for Intensive Systems" project, funded under the Dairy UP program, was established to 1) improve the accuracy of prediction of maize silage yield through calibration and validation, 2) real-time monitoring and modelling of crops, 3) improving quality, and 4) new directions in brown midrib (BMR) maize research and development, thereby supporting informed decision making by Australian dairy farmers.

P4a — Calibration and Validation of APSIM and Remote Sensing for Maize Silage Yield and Quality: Maize silage is a key component of TMR used for dairy production in intensive system. Maize yield forecasting is important for feed planning in commercial farms which is usually implemented by modelling and remote sensing. APSIM model is used extensively to monitor yield of grain crops, and its library has 90 calibrated grain type genotypes. However, there is no silage type genotype in APSIM library. Thus, the aim of this study was to calibrate and validate data by creating maize silage library in APSIM using detailed plot and paddock level maize silage based data to simulate silage crop growth, development, and biomass partitioning across a wide range of genotype, management, and environmental conditions. Remote sensing was done by sentinel2, which was used to predict APSIM biomass. A robust APSIM parameterisation was developed for silage-specific maize genotypes, enabling accurate simulation of crop growth, phenology, biomass accumulation, and partitioning. The calibrated parameters were then used to validate maize silage rather than grain maize, grown at paddock level. Harvest index (HI) was identified to use a key proxy for silage nutritive value. The research established a framework to combine APSIM simulations with remote sensing for dynamic, in-season monitoring of maize growth, yield and quality.

P4b — Integration of APSIM with remote sensing data: APSIM is not effective to capture field variability of crop status in real time. In contrast, remote sensing provides spatial and temporal coverage of a paddock, able to detect real time canopy conditions and biomass using various indices under good weather and crop canopy conditions. However, remote sensing does not simulate physiological processes during crop growth stages such as at grain filling and other growth stage. Thus, it was hypothesised that integrating APSIM with remote sensing data may enable dynamic yield forecasts and quality predictions. Yield and quality data were collected from 11 farms in year1 (y1, 2022-23) and 6 farms in y2 (y2, 2023-24) from four regions of NSW. The aim was to integrate APSIM with satellite data to improve yield and quality predictions and enhance dairy farmers' ability to monitor and manage crop growth in real time. Both APSIM (R^2 = 0.86) and integrated APSIM-remote sensing (R^2 = 0.77-0.81) using sentinel2 NDVI through fAPAR were able to predict biomass, and HI particularly in y2 (R^2 = 0.84) across diverse environment confirming their suitability as a robust tool for silage production systems

P4c — Redefining nutritive value of maize silage through more precise evaluation: This subproject investigated the nutritive value of WCMS and addressed the long-standing observation that silages with contrasting starch contents often exhibit similar energy values. A comprehensive literature review and experimental evidence showed that increasing starch in WCMS reduces fibre digestibility by reducing rumen pH. Rapid starch fermentation lowers rumen pH, suppressing cellulolytic microbes and limiting fibre breakdown, while starch digestibility remains largely unaffected. To better represent this dynamic, a two-compartment digestion model was proposed, separating grain (starch) and stover (fibre) components rather than treating WCMS as a

homogeneous feed. This approach provides a more accurate framework for evaluating silage quality and energy supply. The findings have significant implications for ration formulation and silage management, supporting more precise feeding strategies, cost-effective and improving efficiency in intensive dairy systems.

P4d — New directions in BMR silage research: Brown midrib (BMR) maize hybrids are valued for improved fibre digestibility and animal performance but limited by lower yield and lodging. This study evaluated new short duration BMR (P07003) and conventional (CON; P0937) hybrids across three planting densities (80,000, 95,000, 110,000 plants/ha) over two years at Camden, NSW, under non-limiting nitrogen and irrigation. Biomass, grain yield, harvest index, phenology, and nutritive value were measured, and drone-based remote sensing data were taken at the flowering stage. Both hybrids achieved high yields (25-28 t DM/ha), with no lodging observed in the BMR hybrid, indicating improved resilience. The CON hybrid produced higher yields (by ~2 t DM/ha), but nutritive value was similar between the hybrids (11.8 MJ ME/kg DM). Planting density affected yield optimisation but not nutritive value- BMR performed best at 95,000 plants/ha, while CON achieved maximum yield at 110,000 plants/ha. These results indicate that modern BMR hybrid can match CON yields while maintaining quality, enabling better hybrid × density decisions in intensive dairy systems.

Together, results based on all four components, modelling and remote sensing provided the basis of real-time biomass and nutritive value proxy for quality. It also provided a new direction for BMR hybrid maize for commercial farms and proposed a two compartment model to evaluate maize silage nutritive value accurately.

2. Project Overview

Item	Description
Project Title	Feedbase for Intensive Systems
Funding Body	Dairy UP
Dairy UP Project	P4
Project Duration	2022-2026
Lead Organisation	The University of Sydney
Project Lead	Professor Sergio Garcia
Key Collaborators	University of Southern Queensland, Universidad Nacional de La Pampa, Argentina, NSW Department of Primary Industries and Regional Development, Scibus, Charles Sturt University, Pioneer Seeds, Australian Plant Phenomics Network

3. Abbreviations

ADF— Acid detergent fiber
ADFD— Acid detergent fiber digestibility
ADSA — American Dairy Science Association
APPN — Australian Plant Phenomics Network
APSIM — Agricultural Production Systems Simulator
BMR — Brown midrib
CCC— Concordance Correlation Coefficient
CON — Conventional
CSU— Charles Sturt University
DM — Dry matter
FPS — Frontiers in Plant Science
FAPAR — Fraction of absorbed photosynthetically active radiation
HI — Harvest index
JDS — Journal of Dairy Science
ME — Metabolisable energy
NDVI — Normalised difference vegetation index
NDF— Neutral detergent fiber
NDFD— Neutral detergent fiber digestibility
NSW DPIRD — NSW Department of Primary Industries and Regional Development
OMD — Organic matter digestibility
RMSE — Root Mean Square Error
RMSPE — Root Mean Square Percentage Error
TMR — Total mixed ration
USYD — The University of Sydney
WCMS— Whole crop maize silage

4. Project Background and Rationale

Australian dairy herd sizes have increased by approximately by 5% over the past decades, reflecting a shift towards more intensive systems in near future and greater reliance on TMR. This transition is evident in regions such as inland New South Wales, the Riverina, northern Victoria, and south-east Queensland. In these systems, maize silage is a key dietary component, often comprising around 50% of the total ingredients of TMR. As feed accounts for more than 50% of the total production costs, achieving high biomass yield with superior silage quality is critical for improving productivity and farm resilience. Although the Future Dairy project consistently produced maize silage yields exceeding 25 t DM/ha, silage quality did not improve with varying grain proportions.

Maize silage is highly valued by intensive farmers as it contains ~30% starch (Dairy Australia). Grain in maize silage is the primary source of starch and is one of the main factors driving energy density. Therefore, improved approaches such as Agricultural Production Systems Simulator (APSIM), remote sensing and more precise evaluation of nutritive value are needed to monitor and predict both biomass and grain yield including nutritive value of maize silage to reduce feed costs and enhance profitability in intensive dairy farming.

Therefore, the P4 “Feedbase for Intensive Systems" was undertaken to explore four areas of research: improve the accuracy of prediction of maize silage yield through calibration and real-time monitoring and modelling of crops, its validation in commercial farms, improving quality, and new directions in bmr maize research in order to support informed decision-making for both yield and nutritive value by intensively managed Australian dairy farmers.

5. Project Objectives

The overall objectives of this project were to improve the prediction of maize silage yield and nutritive value, and to develop practical tools to support farmers in optimising silage for diet formulation, thereby enhancing productivity and efficiency of intensive dairy farming. The specific objectives were to:

- **Calibrate and validate maize growth and development** using APSIM and remote sensing with a focus on biomass and grain yield, and harvest index (HI) variability relevant to silage nutritive value.
- **Predict biomass and silage nutritive value** using either by APSIM, remote sensing, or their integration to enable farm scale monitoring across NSW.
- **Redefining silage quality and determine more precise nutritive value** to explore real nutritive value of maize silage through on-farm monitoring and synthesis of published literature.
- **Evaluate next generation maize hybrids for silage** including BMR types to value to enhance both yield and nutritive quality for dairy production systems.

6. Subproject P4a — Calibration and Validation of APSIM and Remote Sensing for Maize Silage Yield and Quality

Research team: Rafiq Islam (The University of Sydney - USYD), Jonathan Ojeda (University of Southern Queensland), Diego Rotili (Universidad de Buenos Aires,) Martin Correa Luna (USYD), Juan Gargiulo (NSW Department of Primary Industries & Rural Development), Yani Garcia (USYD)

6.1 Background

The APSIM (Holzworth et al., 2014) crop model widely used to simulate crop growth, and yield responses to environmental and management conditions. Previous applications of APSIM have primarily focused on grain maize. Therefore, the APSIM cultivar library includes 90 calibrated grain type genotypes, but the model does not include any silage-specific genotypes. Silage maize differs fundamentally from grain maize in both harvest timing and biomass partitioning. Silage crops are typically harvested at ~65% grain maturity (i.e., DM% of grain at harvest; milk to dent stage), whereas grain maize is harvested closer to physiological maturity (~80% grain maturity). As a result, HI and biomass allocation patterns in silage maize are expected to differ substantially from those of grain genotypes. Therefore, parameterisation of maize silage cultivars and its validation with field data are essential to investigate model's ability to simulate silage crop growth, phenology, and biomass partitioning including nutritive value across contrasting environments to have confidence in practice.

The main goal of this study was to calibrate and validate APSIM by creating maize silage library using detailed plot and paddock level maize silage based data to simulate crop growth, development, and biomass partitioning across a wide range of genotype, management, and environmental conditions. Another approach was to investigate APSIMs ability to predict maize silage quality directly or developing an index to use as a proxy for silage quality.

6.2 Objectives

Calibrate and validate APSIM using detailed field experimental data and paddock level studies to simulate maize silage crop growth, development, and biomass partitioning.
Identify the key drivers of grain yield and HI variability and their potential to predict nutritive value of silage.
Quantify the primary sources of HI variability in silage maize across a wide range of genotype, management, and environmental conditions.

6.3 Materials & Methods

- **Data sources for calibration:** The data used for calibration were from four maize silage experiments of the Future Dairy project, the University of Sydney. These research were conducted in small plots and in commercial farms from 2009 to 2014 (Islam et al., 2011; Islam et al., 2012; Islam and Garcia, 2012, 2014). A total of five silage type maize hybrids were included for parameterisation and calibration. The experiments encompassed a wide range of management conditions, non-limiting N and water, planting densities, maturity stages, and sowing and harvesting windows.
- **Data sources for validation:** The data used for model in this study was obtained from five paddocks Silvermere Holstein and MVD Dairy of NSW (longitude 148.45-152.35; latitude 31.91-33.73). Two maize silage genotypes were grown in these farms during 2021-22 growing

seasons in paddocks (7-35 ha). Plants/ha was 7.4-8.5, N fertiliser application was 232-300 kg/ha. Crops were harvested after 112-142 days after sowing.

- **Statistical analysis:** Model was assessed by regression (R^2) and significance ($P<0.05$), Concordance Correlation Coefficient (CCC), model error was assessed by Root Mean Square Error (RMSE) and the Root Mean Square Percentage Error (RMSPE) and models' efficiency was assessed by Nash–Sutcliffe model efficiency coefficient (NSE).

6.4 Key Findings

- **Calibrated APSIM accurately predicted maize silage crop growth.** APSIM predicted total biomass, grain, cob, leaf, and stover weight (CCC=0.86-0.94) accurately (RMSPE=13%) suggesting calibration was successful (Figure 1).
- **APSIM was able to predict maize silage crop phenology:** The model accurately predicted leaf number and canopy green cover (CCC=0.79-0.97) accurately (RMSPE=23-39%).
- **APSIM also accurately predicted HI of maize silage.** APSIM accurately predicted HI of maize for silage and canopy green cover (CCC=0.78) accurately (RMSPE=12%) indicating calibrated APSIM can simulate crop growth, phenology and HI of maize silage (Figure 1).
- **APSIM validated biomass and grain yield.** APSIM validated biomass (CCC=0.75) and grain yield (CCC=0.88) but prediction error was high (RMSPE=45-57%) (Figure 2).
- **Remote sensing was able to predict APSIM simulated biomass.** However, remote sensing consistently underpredicted APSIM simulated biomass (Figure 3).

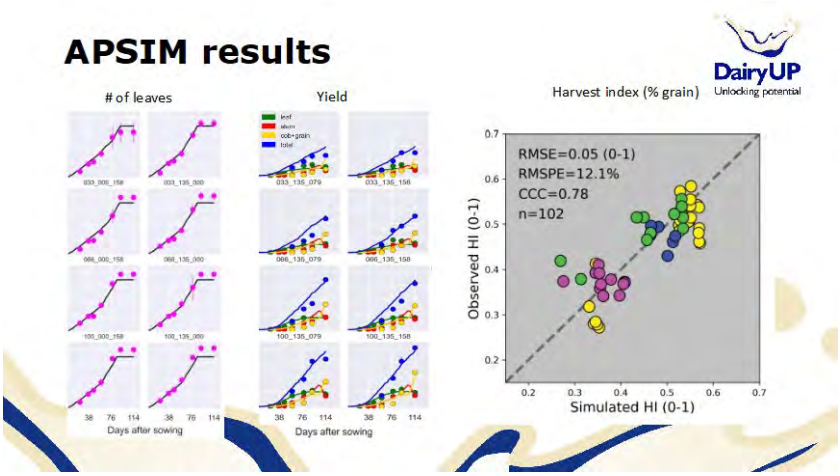


Figure 1. Parameterised and calibrated APSIM simulated biomass, leaf no. and harvest index

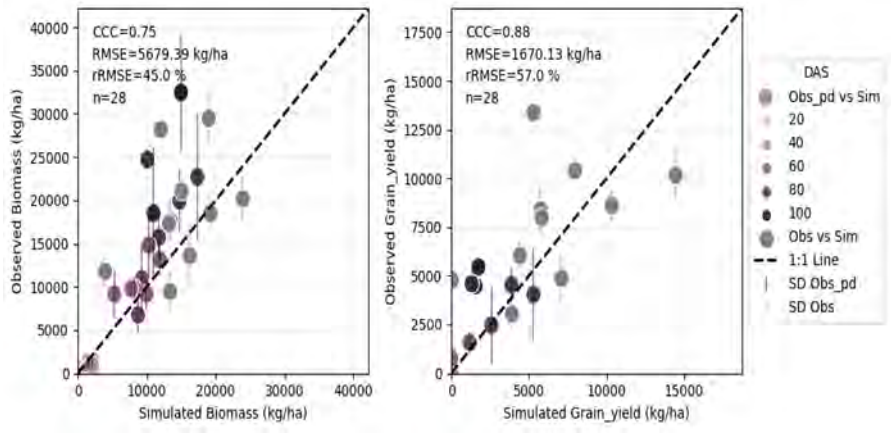


Figure 2. Observed vs. simulated biomass (left panel) and grain yield (right panel) by days after sowing (colour palette). Grey dots indicate observed data at harvest (square 5 m^2) and coloured dots indicate observed data collected from 3 plant sampling during the growing seasons. Vertical bars indicate standard deviations

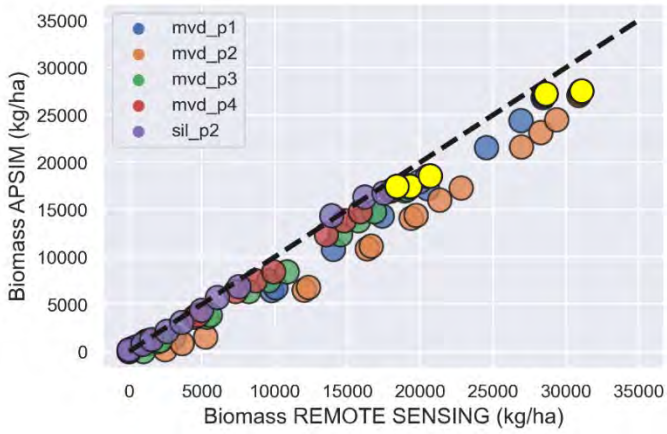


Figure 3. Simulated biomass by APSIM vs biomass estimated by remote sensing (NDVI) for each paddock across the season. Yellow circles indicate the final biomass harvested. Dashed black line represents the 1:1 line (y=x).

6.5 Applications & Impacts

The findings have direct relevance across multiple levels of the dairy farm and industry.

Improved feed planning and cost efficiency, APSIM parametrisation and calibration enables more accurate prediction of maize silage biomass and HI, which is one of the key determinants of feed quality. It also enables forecasting silage yield and quality for farmers before harvest which help farmers in decision making and improved farm-feed planning and to reduce risk.

Precision crop management for higher productivity, the model identifies key drivers of yield and HI variability across genotype, management and environment combinations. The system enables to adjust management in-season using remote sensing and supports transition towards data driven precision dairy farming system for intensive farms.

At the sustainability and environmental level, accurate prediction of biomass, grain yield, and HI improves understanding of resource use efficiency and crop performance, which supports sustainable intensification and helps meet environmental and emissions targets in dairy systems.

6.6 Key Outputs

- A robust parameterisation of APSIM was developed for silage-specific maize genotypes, enabling accurate simulation of crop growth, phenology, biomass accumulation, and partitioning. The calibrated parameters were then used to validate maize silage rather than grain maize, grown at paddock level.
- Quantified and predicted variability in HI, a key proxy for silage nutritive value. Harvest index has the potential to be used as a reliable tool to estimate nutritive value of maize silage.
- Identified key drivers of yield and HI variability e.g., genotype, N and water and a clear understanding of genotype, environment and management effects on silage productivity.

- The research established a framework to combine APSIM simulations with remote sensing for dynamic, in-season monitoring of maize growth and yield.
- Peer-reviewed publication in the *Frontiers in Plant Sciences* demonstrating HI variability in maize silage and its use as a proxy of silage nutritive value.

6.7 Future Research Opportunities

The dataset used for model calibration was based on maize silage genotypes grown at a single site (Camden) and validated at two sites (Silvermere Holstein and VMD farms) using four and two genotypes respectively, resulting in limited spatial representation. Future research should focus on validation across a wider range of commercial farms and regions, and on integrating crop models with real-time sensing and silage-specific genotype characterisation to enable precise, climate-resilient, and economically efficient intensive dairy production in Australia.

Priority areas for future research include:

- **Validation of the model across more commercial farms:** Testing the model performance across environments (genotype, N, water, plant density, large sowing and harvesting windows, heat stress) to test sensitivity and robustness of the model.
- **Integrate APSIM with remote sensing data:** Integrate APSIM with satellite platforms (e.g., Sentinel-2) through data to provide real-time decision support for farmers.
- **Development of silage-specific genotype modules:** Silage module in APSIM is not available. Further research is needed to parameterise and calibration for next generation BMR and stay-green hybrids to accurately predict yield–quality trade-offs.
- **Uncertainty and error decomposition:** High errors in some scenarios indicates structural and scale-related uncertainties. To adopt by the industry, it is necessary to improve model credibility. Thus, it is important to initiate research to identify whether the errors are related to field variability or measurement errors, or partitioning of fractions assimilation, or due to the regions or years associated with climate.

7. Subproject P4b — Integration of APSIM with Remote Sensing Data

Research team: Rafiq Islam (The University of Sydney - USYD), Jonathan Ojeda (University of Southern Queensland), Diego Rotili (Universidad de Buenos Aires), Martin Correa Luna (USYD), Juan Gargiulo (NSW Department of Primary Industries & Rural Development), Fernando Masia (USYD), Cameron Clark (Charles Sturt University), Yani Garcia (USYD)

7.1 Background

P4a demonstrated that APSIM can be used to assess interannual variability of maize silage yield and its HI, and remote sensed biomass can moderately predict APSIM simulated biomass. However, both APSIM and remote sensed biomass has limitations. While APSIM cannot easily capture field variability or reflect real time crop status, remote sensing provides spatial and temporal coverage of a paddock, able to detect real time canopy conditions and biomass using various indices under good weather conditions. However, remote sensing does not simulate physiological processes e.g., soil N status, grain filling. Therefore, it was hypothesised that integrating APSIM with remote sensing data may enable dynamic yield forecasts and quality predictions. Using field scale data from commercial maize silage systems, it was assessed whether integration improves prediction accuracy compared with standalone modelling approaches and identify key sources of uncertainty influencing model performance. The goal was to integrate APSIM with satellite data to improve yield and quality predictions and enhance dairy farmers' ability to monitor and manage crop growth in real time.

7.2 Objectives

- To evaluate APSIMs performance in predicting of biomass, grain yield and HI using data collected from commercial maize growing farms across NSW.
- Evaluate the performance of biomass and yield prediction using an integrated APSIM-remote sensing framework.
- Identify key sources of uncertainty influencing model performance.

7.3 Materials & Methods

To address these objectives, the project undertook a series of coordinated studies:

- **Data sources and crop management**: Biomass, grain yield and HI of maize data were generated from field studies conducted in commercial dairy farms growing maize for silages across NSW, covering areas from North Coast, Middle Coast, Camden and South Coast by taking 3 farms from each area and grown over two years (2022-23; y1 in 12 farms, total 145 ha, and 2023-24; y2 in 6 farms, 84 ha). Farm soil in the study areas were loam, silty loam, silty clay loam, loamy sand and silty clay. Maize hybrids sown were C29A1, hsr brutus and different hybrids of Pacific and Pioneer. Sowing dates ranged from 9 October to 20 January with 53,228-85,000 plants/ha and harvest after 99-147 days of sowing. Nitrogen fertiliser applied was 125-369 kg N/ha and irrigation water was 0-480 mm/ha. Biomass yield was recorded at 4 growth stages: V6, flowering, grain filling and at final harvest stage. At each stage, 2 random plants were harvested, dried and weight recorded. Average weight of plant was multiplied by plant density to estimate biomass at each stage. However, biomass yield at final harvest was also calculated by harvesting two 5 linear m maize rows and fresh weight was taken. Plants were chopped and dried in an oven DM biomass was calculated on DM basis. Grain yield was calculated from the proportion of grain in total biomass. Harvest index was calculated by dividing the grain yield by total biomass yield.

- **APSIM parameterisation**: Cultivar parameter sets developed for the silage hybrid '*PacificX*' by Ojeda et al. (2023) were adopted for simulations. However, *tt_emerg_to_endjuv* was changed to 260 instead of 350 and *tt_flower_to_maturity* was changed to 800 instead of 600, which provided values relatively close to the real time observations for each hybrid as the hybrids evaluated by Ojeda et al. (2023) exhibited lower HI than those grown in the current experiments.
- **Remote sensing data**: Remote sensing data were obtained from Sentinel2 satellite mission which derived NDVI, and NDVI was used to derive fAPAR.
- **Integration of APSIM and sentinel2 data**: APSIM and sentinel data were combined to investigate if integrated approach could predict biomass more accurately than APSIM. In this process, NDVI data were used to calculate potential and actual real time fAPAR. Intercepted solar radiation was calculated by multiplying fAPAR (NDVI)*intercepted radiation by APSIM. Biomass accumulation was calculated by summating the intercepted radiation (fAPAR plus RUE of APSIM). Calculations used to determine biomass using sentinel2 and APSIM were as follows:

  i) fAPAR calculation from sentinel2
  fAPAR was calculated using Zhang et al. (2015) and Gitelson et al. (2014)
  equations:
  $fAPAR_Z$ = 1.242 x EO – 0.204 (Zhang et al., 2015)
  $fAPAR_G$ = 1.35 x NDVI – 0.32 for vegetative stage (Gitelson et al., 2014)
  $fAPAR_G$ = 1.89 x NDVI – 0.7827 for reproductive stage (Gitelson et al., 2014)
  ii) potential RUE calculation from APSIM:
  intRad = fAPAR_APSIM x Rad_ APSIM
  APSIM_RUE = biomass_ APSIM/cum_intRad
  intRad, intercepted radiation; cum, cumulative; RUE, radiation use efficiency
  iii) biomass calculation from fAPAR and APSIM
  intRad_$NDVI_Z$ or intRad_$NDVI_G$ = $fAPAR_Z$ or $fAPAR_G$ x Rad_silo
  biomass_accumulated = intRad_$NDVI_Z$ or intRad_$NDVI_G$ x APSIM_RUE
- **Data analysis and model evaluation:** Observed biomass and grain including HI were evaluated against these yields obtained by 1) APSIM alone, and 2) a combination of APSIM and sentinel2 data based biomass derived using $fAPAR_Z$ or $fAPAR_G$. Model output was assessed by R^2 at $P<0.05$ including CCC, RMSE, RMSPE and NSE.

7.4 Key Findings

- **APSIM and integrated APSIM-remote sensing predicted maize biomass across years in commercial farms.** APSIM simulated biomass accurately (Figure 1; R^2 = 0.86) across regions, farms and years, which improved with outlier correction (Figure 1; R^2 = 0.88). Prediction error also improved with outlier correction (RMSPE, 32%), which improved efficiency (NSE, 0.76) and agreement (CCC, 0.86). APSIM-remote sensing also predicted biomass with similar accuracy (Figure 2; R^2 = 0.80).
- **APSIM also simulated real-time biomass yield for most farms across years.** The model generally captured the shape of growth curves well across sites. Simulated biomass closely tracked observed temporal dynamics where simulated and observed biomass were closely aligned in mostly in farms under irrigated conditions (Figure 3)
- **APSIM predicted grain yield and HI with moderate to good capacity across.** Agreement between simulated and observed grain yield was moderate (R^2 = 0.61-0.72, $p<0.001$). Model performance was better in y2 than in y1.

- **Uncertainties in model prediction.** The primary limitation lies not in biomass accumulation processes using either APSIM or integrated APSIM-remote sensing but in accurately representing partitioning dynamics under varying stress conditions which lead to measurement uncertainties in observed data. Paddock level variations also increased data deviations. In addition, phenological mismatch and discrepancies between simulated and observed data particularly at flowering and grain filling impact grain yield and HI. There is also a mismatch in observed data and sentinel2 data collection dates. These uncertainties propagate into model evaluation metrics, inflating RMSPE and potentially underestimating model performance. Improving model performance will require enhanced representation of stress interactions, phenological alignment, and better integration of high quality observational data.

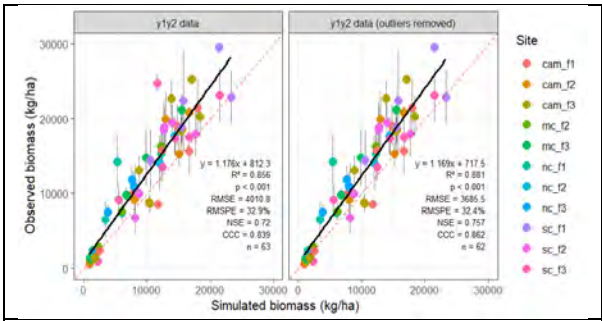


Figure 1. Simulated vs observed biomass with (left) or without outlier correction (right)

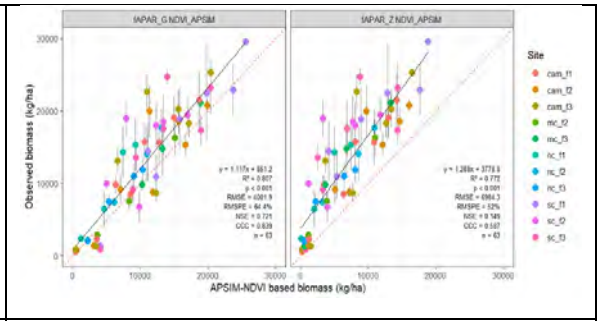


Figure 2. Integrated APSIM-NDVI derived vs observed biomass using two different fAPAR methods

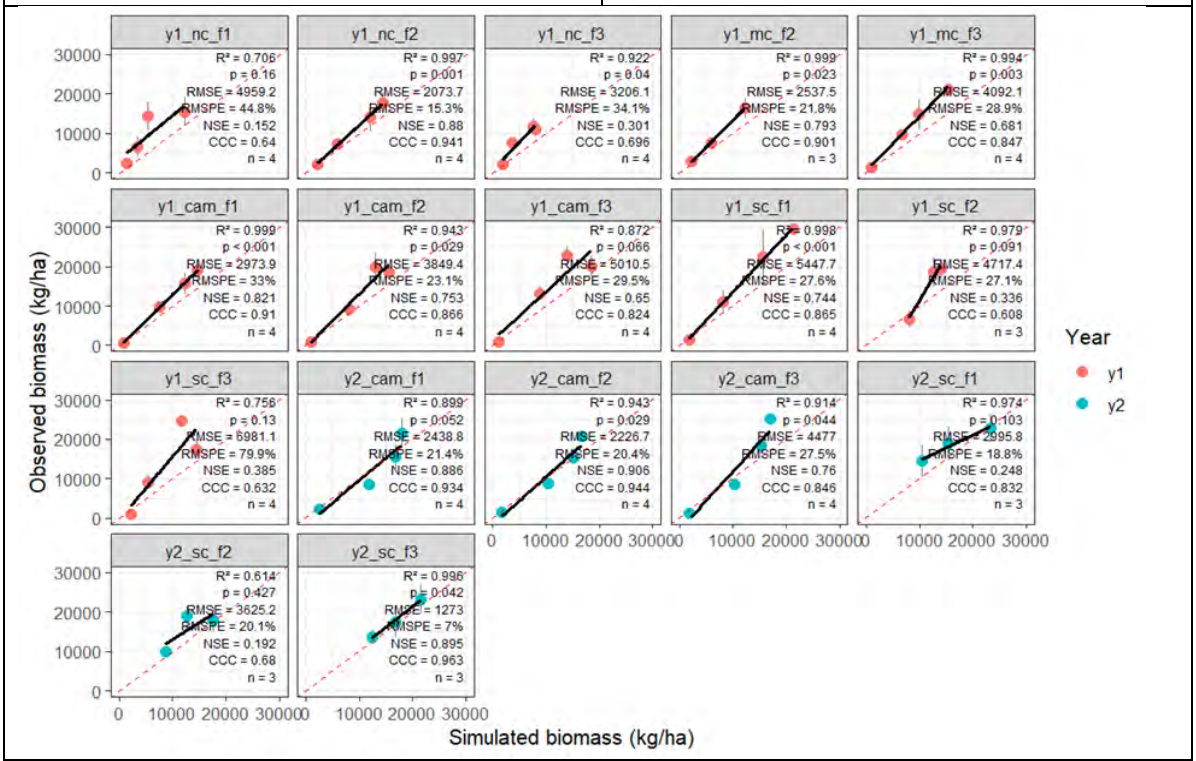


Figure 3. Simulated vs observed biomass by farms across sites, regions and year

7.5 Applications & Impacts

- **Real-time decision support for maize silage management:** APSIM alone and integration of APSIM-remote sensing enables dynamic, near real-time monitoring of crop growth and biomass accumulation, allowing farmers to adjust irrigation, nitrogen, and harvest timing more precisely. This improves management efficiency and reduces uncertainty compared to relying on either modelling or integration with satellite data.
- **Improved feed planning:** Accurate prediction of yield and HI allows better estimation of silage quantity and nutritive value, which is critical for intensive, in-housed TMR based dairy systems where maize silage constitutes a major feed component. This supports optimised diet formulation, reduced feed costs and improved farm profitability.
- **Model applicable to farm or paddock levels:** The model's strong performance across most regions, farms, and years demonstrates its ability to support decision making under diverse climatic and management conditions. By identifying key uncertainties (e.g., stress effects, phenology mismatches), the framework helps improve future modelling and supports climate resilient, data-driven intensive dairy production systems.

7.6 Outputs

- Validated APSIM model for maize silage across diverse environments, farms, and years confirming its suitability as a robust tool for silage production systems.
- Integrated APSIM-remote sensing using sentinel2 NDVI through fAPAR was developed, which enable spatial and temporal monitoring of crop growth and offering a foundation for real-time yield prediction tools.
- A comprehensive dataset was generated across 18 commercial farms across multiple regions over two years, capturing variability in management such as N, irrigation, plant density and environments, which is valuable for future model calibration and validation.
- This study identified key drivers and uncertainties in model performance such as biomass partitioning at grain filling stage, phenological mismatch, paddock variability and measurement uncertainties, observed and satellite data collection date mismatch including resolution and weather situation during satellite data collection.
- Presentations at major national and international scientific conferences, including the American Dairy Science Association Annual Meeting (USA, 2025) and International Precision Dairy Farming Conference (Christchurch, 2025).

7.7 Future Research Opportunities

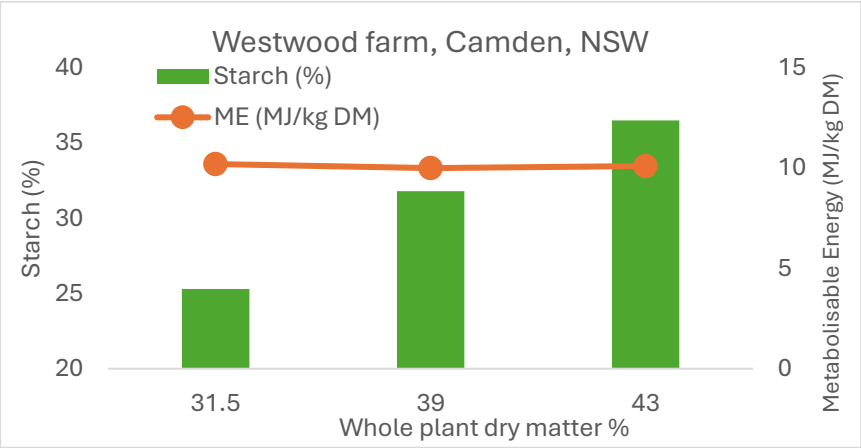
- Future studies should focus on advanced data acquisition using high-resolution remote sensing to improve real time predictions and reduce errors caused by temporal mismatch between observed and satellite data.
- Future work should also focus on stage specific biomass partitioning and stress interactions (water, N, temperature), particularly during grain filling stage to enhance prediction of grain yield and HI.
- Further works needed to develop silage specific genotype modules. Parameterisation of silage-specific maize genotypes harvested around 60-70% grain maturity to better capture variability in HI and nutritive value across environments.

8. Subproject P4c — Redefining nutritive value of maize silage

Research team: Rafiq Islam (The University of Sydney - USYD), Ian Lean (Scibus), Cameron Clark (Charles Sturt University), Martin Correa Luna (USYD), Yani Garcia (USYD)

8.1 Background

A key challenge in evaluating whole crop maize silage (WCMS) evaluation is that silages differing widely in grain proportion or starch content often exhibit remarkably similar energy values (see figure below). Future Dairy research consistently showed that WCMS containing contrasting starch (or DM) had similar energy content. This discrepancy occurs even though grain and stover ratio in silages is approximately 50:50 and grains metabolisable energy (ME) content is 14-16 MJ/kg DM but stover's ME content is 7-9 MJ/kg DM. Again, maize grain in silage is almost 100% digestible in ruminants whereas stovers digestibility is 50-60%.



We hypothesise this discrepancy stems from the fact that WCMS is defined as a forage although it is a heterogenous feed containing both forage (stover) and grain. In traditional feed analysis methods, WCMS containing both grain (starch) and stover (fibre) digested together to derive energy of maize silage. Thus, it is possible that digestion of these contrasting starch and fibre of WCMS suppresses the effect of starch and its digestibility does not reflect in the contribution of energy content of WCMS. Thus, a review of literature was taken to understand why WCMS with contrasting starch remains similar in digestibility or energy content.

This review aimed at) addressing why WCMS with contrasting starch contents consistently demonstrate similar energy values, b) identify biological mechanisms underpinning this phenomenon, and c) propose an alternative approach for evaluating WCMS that more accurately reflects its energy that defines its nutritional value. In addition, a meta-analysis on the impact of DM content at harvest on yield and nutritive value of WCMS was investigated.

8.2 Objectives

- Address why WCMS with contrasting starch contents consistently demonstrate similar digestibility and energy values
- Identify biological mechanisms underpinning this phenomenon
- Propose an alternative approach for evaluating WCMS that more accurately reflects its digestibility and energy contents in order to define its potential nutritional value
- To investigate the impact of DM content at harvest on yield and nutritive value of maize silage

8.3 Materials & Methods

- **Literature review**: Literature published from 2000-20025 on *in vivo* digestibility of WCMS using dairy cows, steers and sheep were collected.
- **Literature sorting and data analysis:** Experiments that evaluated WCMS fed at maintenance levels of feeding, with or without small amounts of concentrate supplementation were included to avoid interactions that might occur when maize silage was digested with other feeds such as concentrates, forages or TMR. Regression analysis was conducted to investigate relationships between fibre and starch content on their digestibility.
- Meta-analysis data was collected from the literature published from 2000 to 20025.

8.4 Key Findings

- **Increased starch in WCMS reduces fibre digestibility.** Contrary to our hypothesis, increased starch does not reduce its digestibility, but starch reduces fibre digestibility (Figure 1).
- **Mechanism of fibre digestibility reduction.** Greater amounts of starch of WCMS degrades rapidly in the rumen, produce volatiles and lactic acid (Nocek and Tamminga (1991). Accumulation of these acids lowers ruminal pH (Lean et al., 2013), which suppresses cellulolytic microbes and reduces fiber digestion. However, amylolytic bacteria that degrades starch are more tolerant of acidic conditions (Mackie et al., 1978). Consequently, NDFD decreases as ruminal pH decreases, whereas starch digestibility remains largely unaffected.
- **Proposed two compartment digestion models for stover and grain, and to redefine WCMS.** To explore the full energy potential of WCMS, a two-compartment digestion model proposed that separately digests the forage (stover) and grain of WCMS in contrast to the conventional approach that treats WCMS as a single, homogenous feed.
- **Meta-analysis on the impact of DM on yield and nutritive value**: The meta-analysis demonstrates that increasing DM content of maize silage enhances DM yield, harvest index, starch and energy content of maize silage.

8.5 Applications & Impacts

- **More precise evaluation and ration formulation based on WCMS:** Redefining WCMS using a two-compartment model (grain and stover) enables more precise estimation of energy supply. This allows intensive farmers to better balance starch and fibre in rations, improving diet formulation accuracy, and dairy performance.
- **Improved rumen health and feeding strategies:** Understanding that higher starch reduces fibre digestibility via rumen pH suppression, provides a mechanistic basis to design diets, that avoid suboptimal rumen conditions, thereby improving feed efficiency, milk production, and animal health.
- **Reduced feed costs and improved dairy system efficiency:** Understanding the concept that starch of high grain silage WCMS can meet a significant amount of starch in the diet, farmers can add less grain in the diet of cows, optimise diet accordingly leading to cost-effective intensive dairy production and better profitability.

8.6 Outputs

- The review demonstrated that increasing starch in WCMS does not reduce starch or overall digestibility, but instead reduces fibre digestibility, explaining why WCMS with contrasting starch contents show similar energy values.

- Review also provided biological mechanism explaining constant energy content of maize silage. WCMS starch rapidly degraded in the rumen, accumulating acids, lowers rumen pH, which ultimately reduces fibre digestibility but does affect starch digestibility. Consequently, WCMS digestibility or energy remains constant.
- Accordingly, a novel two-compartment digestion model framework to digest grain and stover separately was proposed to evaluate WCMS, providing a more realistic representation of its nutritive value than traditional single-WCMS evaluation.
- Presented orally at the ADSA at Louisville, Kentucky, USA in 2025.

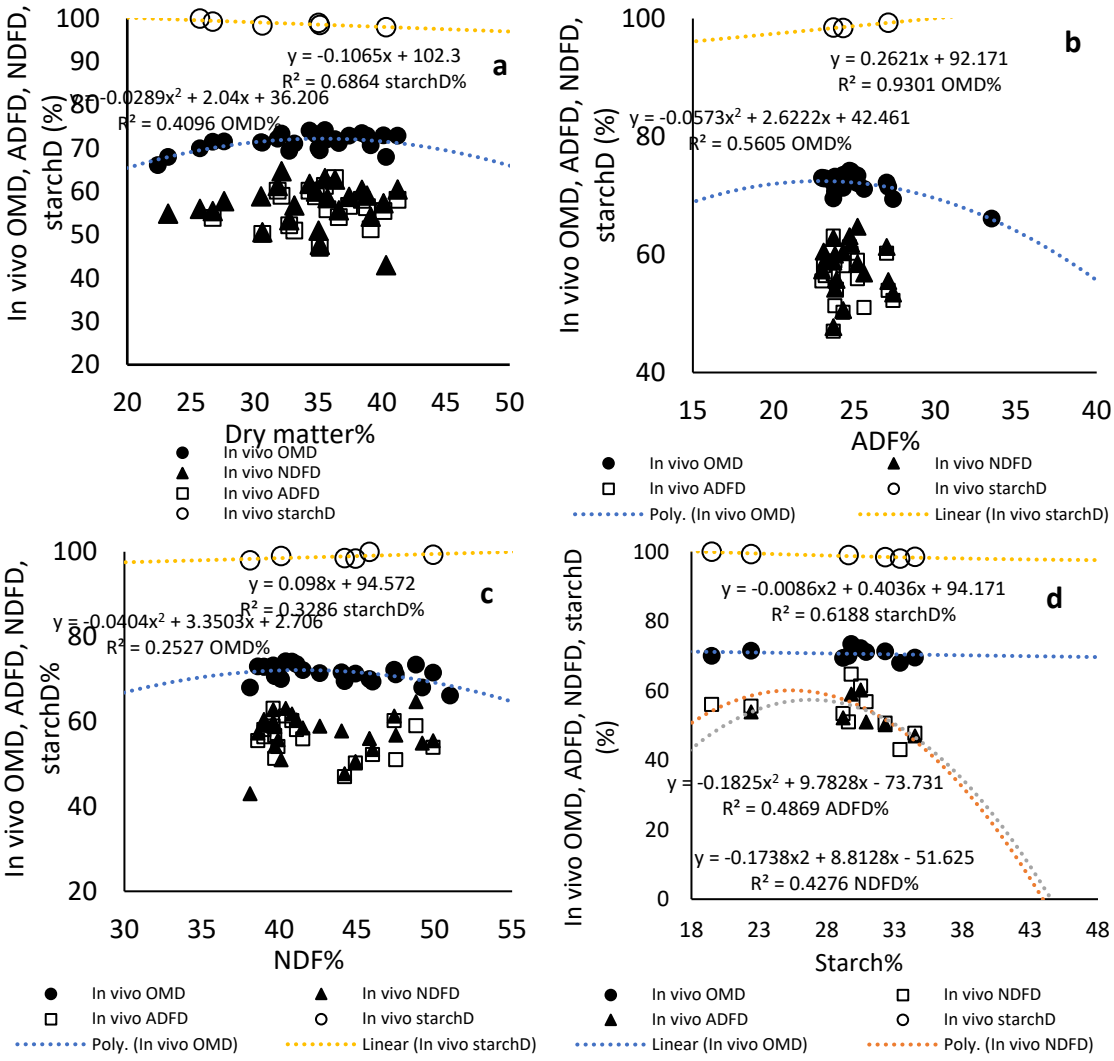


Figure 1. In vivo organic matter digestibility (OMD%), acid detergent fiber digestibility (ADFD%), neutral detergent fiber digestibility (NDFD%), and starch digestibility (starchD%) affected by dry matter, ADF, NDF and starch contents of WCMS

8.7 Future Research Opportunities

This review provided a foundation concept to re-evaluate and re-define WCMS, or other grain silages used in TMR industry for intensive dairy systems. However, the concept needs to be validated and established through further research.

Priority areas for future research include:

- **Validation of the two-compartment model:** Testing the proposed model by digesting stover and grain separately using in vitro, in situ and vivo dairy trials and compare with single-WCMS evaluation.
- **Integration with nutrition and ration formulation models:** Incorporate the two-compartment approach into feeding systems (e.g., NRC/ARC models) to investigate how much grain in the diet can be replaced by WCMS grain in the diet of cows in intensive system and its impact on cost reduction by farmers.
- **Investigating the impact of grain to stover ratio or starch of WCMS on rumen dynamics and microbiome:** Investigate how varying grain-to-stover ratios influence rumen microbiology, pH dynamics, and fibre degradation kinetics, enabling development of precision feeding strategies.

9. Subproject P4d — New Directions in BMR Silage Research

Research team: Rafiq Islam (The University of Sydney - USYD), William Salter (USYD), Anna Chlingaryan (USYD), Ben Vercoe (Pioneer Seeds), Raewyn Densley (Pioneer Seeds), Ian Lean (Scibus), Yani Garcia (USYD)

9.1 Background

During the last decades, BMR maize hybrids have gained considerable attention because of their reduced lignin content, higher fibre digestibility, increased DM intake and milk production (Tebbe and Weiss, 2018). However, it is characterised by its lodging habit, and its yield is relatively low compared to the conventional (CON) hybrids. Recently developed maize hybrids are claimed to be overcome lodging and yield gaps. Thus, in collaboration with Pioneer seeds and APPN, research was conducted to investigate trade-offs between yield and quality of maize hybrids (BMR vs CON) under different sowing density across years. Such evaluations provide the basis for recommendations of hybrid types that balance productivity, nutritive value, and crop resilience for intensive dairy farmers.

9.2 Objectives

- To evaluate the effects of maize hybrid type and planting density on yield and nutrient partitioning trade off at different stages of growth affecting nutritive value
- To investigate how hybrid type and planting density influence plant phenology, lodging and canopy characteristics
- To compare the nutritive value of BMR and CON maize hybrids, and fibre digestibility in determining silage quality
- To assess yield, phenology and nutritive value of BMR and CON maize hybrids using remote sensing

9.3 Materials & Methods

- **Crop management**: The experiment was conducted over two growing seasons (2024-25 and 2025-26) at the USYDs Lansdowne farm, Camden. A factorial design was used with two maize BMR (P07003) and CON (P0937), sown at three planting densities (80,000, 95,000 and 110,000 plants/ha) with three replicates arranged in a randomised complete block design with non-limiting N and irrigation. Data on biomass, grain yield and HI including phenology was collected at four growth stages (V6, flowering, grain filling and at final harvest). Nutritive value including NDF, ADF, starch, and ME and NDFD at 24 were determined. Data were analysed using a REML mixed model in R, with hybrid, density, and year as fixed effects and block nested within year as a random effect to evaluate main effects and interactions.
- **Remote sensing data**: Remote sensing data were obtained at flowering stage using a drone in collaboration with APPN.

9.4 Key Findings

- **Both hybrids maintained high yield under non-liming N and water.** No lodging was apparent in BMR hybrid. Both hybrids yielded 25-28 t DM/ha on average across years. Hybrid had no effect on nutritive value (11.8 MJ ME/kg DM).
- **Plant density influences yield but had no effect on nutritive value.** Plant density had no impact on yield and nutritive value. However, optimal density management was crucial in general as BMR performed best at 95,000 plants/ha (26-27 t DM/ha) in both years, but CON

hybrid performed best at 110,000 plants/ha (27-28 t DM/ha) indicating more plasticity of this hybrid (Figure 1).

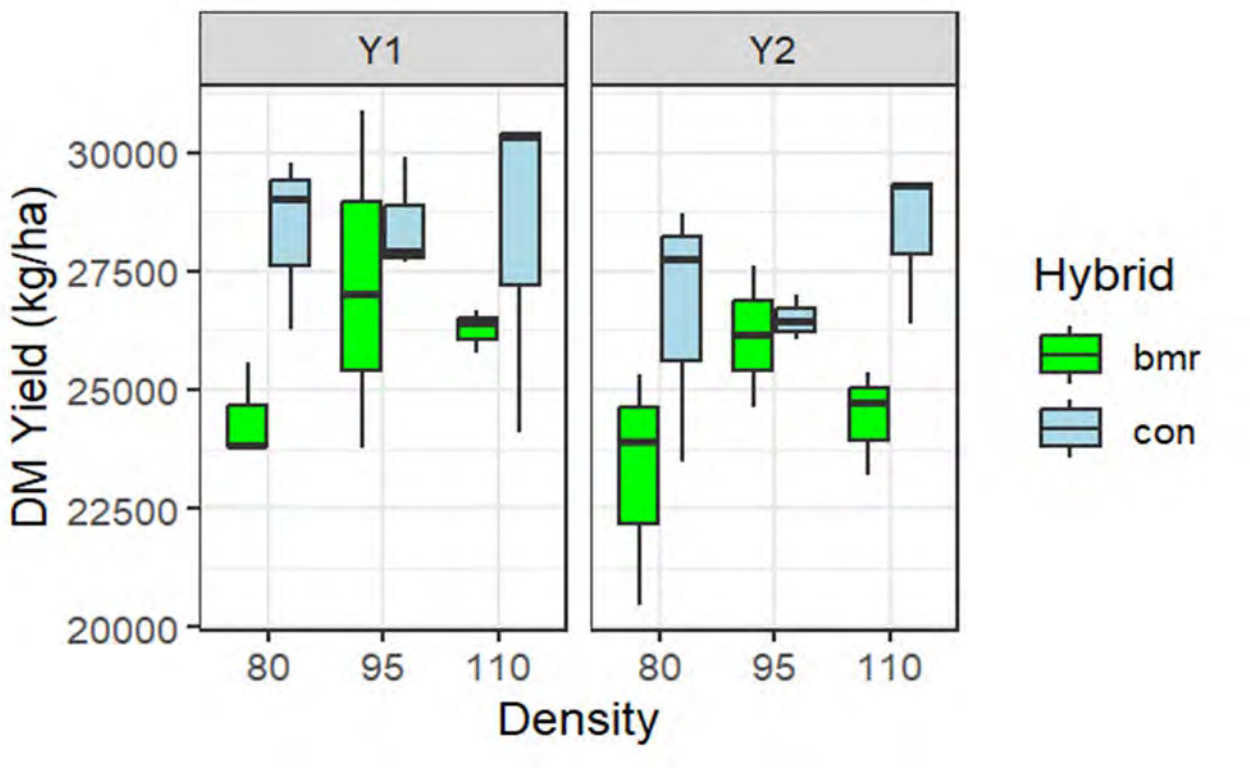


Figure 1. Yield of BMR and conventional hybrid affected by hybrid and plant density

9.5 Applications & Impacts

- **Improved hybrid selection for specific production goals:** Farmers can select hybrids based on system priorities. CON hybrid had higher yield and grain, but BMR had improved fibre digestibility. This enables more targeted decision making for TMR systems in intensive dairy farming.
- **Starch-fibre trade-off between hybrids:** Starch-fibre trade-off between CON and BMR hybrids allows farmers to better balance diets, and improving rumen function, feed efficiency and milk production.

9.6 Outputs

- The study generated robust field data on biomass, grain yield, HI, and digestibility, providing a practical benchmark for silage production in Australian dairy systems.
- This study also provided a better understanding of yield-quality trade off of CON and BMR hybrids offering Australian intensive dairy farmers a refined framework to interpret this trade-off for diet formulation and decision in feeding system based on the farm need.

9.7 Future Research Opportunities

Priority areas for future research include:

- **Multi-environment validation trial:** Hybrid effects and yield-quality relationships under real farm conditions required across climates and management system under variable input levels. System modelling also required to validate data.
- **Animal feeding trial:** Future research should investigate performance of CON and BMR hybrids in order to enable better prediction of animal performance from silage.

10. Project-wide Dissemination

One manuscript published in a high-impact international journals (FPS), which has an impact factor of 4.8 and is ranked Q1 in its field. In addition, two significant works presented orally in two significant forums. One in the American Dairy Science Association (ADSA) Annual Meeting at Kentucky in the USA, and another one present in the Precision Dairy Farming Conference in NZ, Christchurch (Table 1). These presentations collectively reached an estimated audience of over 3,700 people, spanning industry professionals, researchers and policymakers.

Table 1. Peer review publication, **c**onference presentations, abstracts and other communications

Authors	Title	Journal/Presentation Type	Conference/ Event	Location	Year	Audience
Ojeda et al	Field and in silico analysis of harvest index variability in maize silage.	Front. Plant Sci 14:1206535 (2023)				
Islam et al.	Revealing the real nutritional value for maize silage	J. Dairy Sci. Vol. 108, Suppl. 1, pp 174-175 (2025)				
Islam et al.	Integrating APSIM and remote sensing to predict maize silage biomass	Proc Precision Dairy Farming Conf, pp 171-172 (2025)				
Islam et al.	Revealing the real nutritional value for maize silage	Oral presentation	ADSA Annual Meeting, 22-25 June 2025	Louisville, USA	**2025**	1,000
Islam et al.	Integrating APSIM and remote sensing to predict maize silage biomass	Oral presentation	4th Int Precision Dairy Farming Conference , 3-5 Dec, 2025	Christ-church, NZ	**2025**	200
Garcia and Islam	Maize for silage	Dairy UP Website			**2025**	
Ojeda et al	Digitising forage options for intensive dairy systems	Dairy Up report			**2024**	

11. Conclusions and Recommendations

The P4 Feedbase for Intensive System project has delivered new insights on real-time monitoring of maize for intensive dairy farming and TMR industry and identified HI as a tool to monitor feed quality. It also investigated ways to explore its nutritive value accurately which may play a significant role to reduce feed cost and improving efficiency of farming. In addition, information help farmers in decision making and improved farm-feed planning to reduce risk in intensive dairy farms.

P4a successfully calibrated to simulate maize silage biomass using APSIM modelling across diverse environments, demonstrating strong predictive capacity for biomass yield. APSIM also provided a robust framework for analysing genotype, management an environment interaction and provides a strong foundation for further refinement of silage-specific modelling. It identified HI to use as a key determinant of feed quality.

P4b demonstrated that APSIM alone or integration of APSIM with remote sensing can be used for biomass, grain yield and HI prediction by combining mechanistic understanding with real-time observations. This hybrid approach showed strong potential for dynamic yield forecasting. However, limitations were evident in predicting grain yield and HI, particularly under variable water and nitrogen conditions, due to challenges in representing phenology, biomass partitioning, and stress responses. Integration methods require further refinement, especially in linking canopy signals (NDVI/fAPAR) with biomass partitioning during flowering and grain filling stages.

P4c demonstrated that energy content remains relatively constant despite large variation in starch concentration is driven by a biological trade-off mechanism: increased starch reduces fibre digestibility via rumen pH effects. The proposed two-compartment model (grain and stover) provides a more accurate framework for evaluating silage quality than traditional single-feed approaches, which can be used to redefine the nutritive value of WCMS.

P4d demonstrated that farmers can select hybrids based on system priorities. Conventional hybrid had higher yield and grain, but BMR had improved fibre digestibility. This enables more targeted decision making for TMR systems in intensive dairy farming.

Recommendations for future investment should prioritise:

- Research on parameterisation and calibration of silage specific hybrids including BMR enable more accurate prediction of yield and nutritive value
- Integrating APSIM with high frequency remote sensing with modelling to capture variation both at early and late growth stages particularly at flowering and grain filling stage
- Validation of the two-compartment digestibility model in dairy systems and its integration in NRC/ARC framework to improve precision feeding, productivity and sustainability in TMR based dairy systems
- Multi-environment validation trial of BMR maize and its performance in TMR and dairy industry

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Final Project Summary
P4: Feedbase – Maize for silage

May 2026

On most Australian dairy farms, the costs associated with feed represent at least half of all expenses associated with producing milk. Feed costs can be even higher for those milking cows in intensive Total Mixed Ration (TMR) systems or housed-cow operations. That's why any refinements to the feed base of a dairy farm can have a huge effect on a business' profit.

Maize silage

Maize is a key component in the feed base of many TMR or partial mix ration (PMR) systems because it is high yielding and water efficient.

Several research studies, including projects through the FutureDairy program, proved the water and nitrogen efficiency of growing maize in Australia. Work with farmers across the country demonstrated that maize silage could consistently yield 25-28 tonnes of dry matter per hectare when good management was combined with good water and nitrogen availability.

In trials where maximum amounts of irrigation water were applied, maize silage yielded 5 tonnes of dry matter per megalitre of water – five-times greater than the average response to perennial pastures in the irrigation region of Northern Victoria. With no water limitation, maize for silage also yields an average of 150kg of DM per kg of nitrogen.

Unlocking the potential of maize

Dairy UP's P4 project aimed to unlock the potential of the dairy feed base, with a focus on growing maize for intensive systems.

P4 integrated precision agriculture with real-time monitoring of plants and soils as well as advanced modelling to grow better forage crops and increase water and land-use efficiency.

This document provides the final update on P4: Feedbase – Maize for silage.

The quality challenge

The ingredients that determine profitable maize crop yields are clear but ensuring yield is converted to 'high quality' silage isn't as straight forward.

Maize silage is made from the entire plant. Each element has differing starch, fibre, and energy content.

Additionally, metabolisable energy (ME) – a traditional form of determining feed quality – isn't an accurate indication of the quality of maize silage, as the high ME of the grain (starch) can be diluted by the lower ME of the stem and leaves.

Project aim

This project aimed to provide the foundations for a future decision support tool to help dairy farmers with their maize silage production. The project investigated:

1. Predicting maize silage yield and quality using a modelling program called the Agricultural Production Systems sIMulator (APSIM).
2. Real-time monitoring of the maize crop using a combination of APISM and remote sensing.
3. Clearer indicators of maize silage energy content and nutritive value.
4. Performance of the highly digestible Brown midrib (BMR) maize variety under NSW dairying conditions.

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Key findings

The APSIM model reliably predicts maize silage yield on commercial farms, across regions, sites, years and individual farms.

Harvest index (HI) can be used as a practical on-farm proxy for silage energy content, especially in well managed conditions.

Combining the APSIM model with satellite imagery (Sentinel-2) can help track how crops are growing across a paddock, but at this stage it didn't improve yield predictions compared to APSIM on its own.

A new framework for evaluating maize silage energy was developed, showing why silages with very different starch contents often deliver similar energy.

New generation BMR maize hybrids had only 10% less yield compared to conventional hybrids but lodged less and had improved fibre digestibility and comparable energy content.

Together, these outcomes provide practical pathways for farmers to:

- Improve feed planning and budgeting.
- Reduce uncertainty in silage supply and quality.
- Make better hybrid, density, and harvest decisions
- Support profitable and resilient intensive dairy systems.

Benefits

Managed well, maize silage is an efficient use of water and nitrogen on-farm, but it's an expensive crop to plant. Tools to improve management can have a big impact on maize yield and quality and therefore the return on investment in the crop.

Understanding the potential yield and quality of the final, harvested silage will also help dairy farmers to plan in advance for their feed requirements and should decrease costs.

This work could also benefit other livestock industries that use maize silage as a feed source.

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Research and results

Yield and quality targets

A combination of a literature review and research established maize silage yield and quality targets while also identifying knowledge gaps.

Maize silage targets include:

- Dry matter yield: 25t DM/ha.
- Starch: at least 35% to dilute as much fibre as possible.
- Harvest index (ratio of grain to total dry matter): 40-50% can dilute the fibre in maize silage.

Knowledge gaps include:

- Uncertainty about how to accurately measure the dry matter percentage of maize silage and if a maize silage with 35 per cent or more dry matter ensures a lower NDF due to increase in starch content.
- How much does sowing time, plant density, water and nitrogen availability determine crop yield and quality? And how can this be monitored in real time?
- How are indicators of silage 'quality' and yield best monitored?

DairyUP
Unlocking potential

Predicting maize yield and quality

The modelling tool, Agricultural Production Systems sIMulator (APSIM) was developed and calibrated for 90 crops including maize for grain (but not maize for silage). This study tested and calibrated it to predict the yield of maize crops grown in NSW for *silage*.

The initial calibration was done with data from FutureDairy. The subsequent validation used data from maize crops on research (FutureDairy feedbase data) and six commercial dairy farms. The farm data was collected through remote sensing and real-time on-site monitoring, which involved cutting and measuring plants at four stages (V6, flowering, grain-filling, and plant maturity (harvest)).

Overall APSIM predicted yield (biomass) of maize crops with reasonable accuracy.

The APSIM calibration has room for improvement in dryland conditions, but it provided reasonable predictions of yield and harvest index for irrigated crops across regions.

Figure 1: Data combined for 6 farms

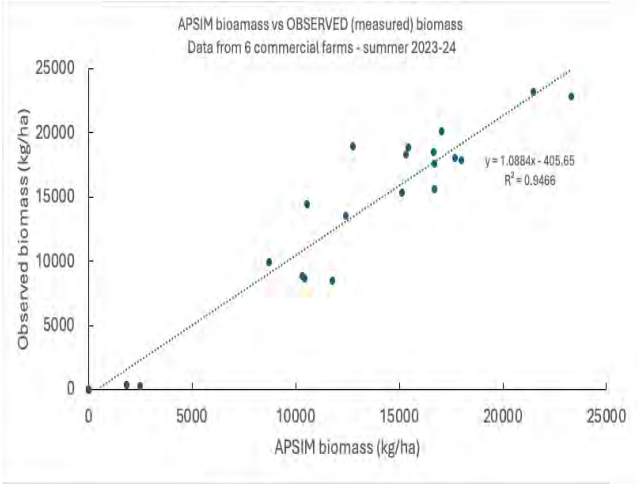


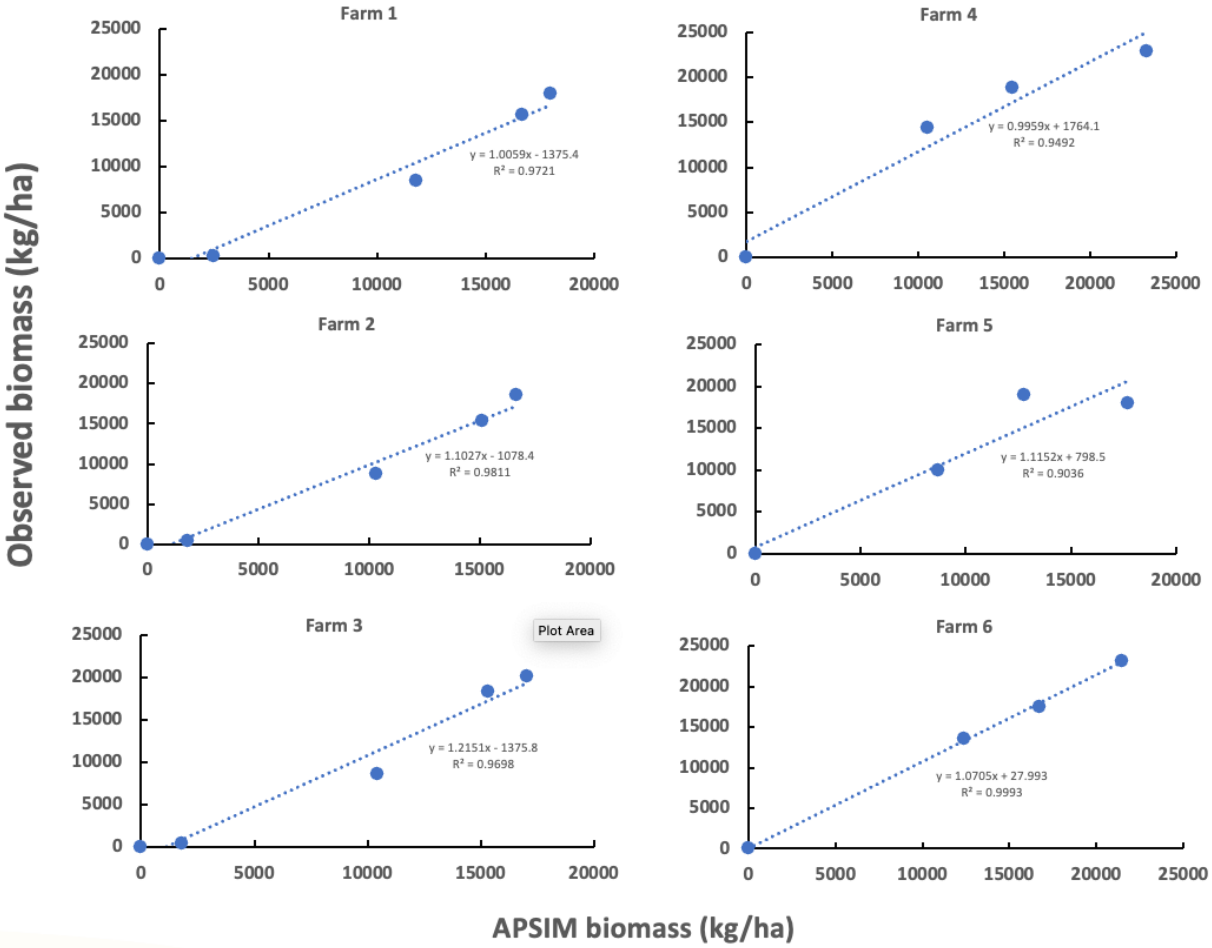
Figure 1 shows the combined data for the six farms.

Figure 2 shows the individual crop/farm data. It demonstrates good performance in all cases.

This was measured through cutting plants but variation with paddock yield was high.

Harvest index (HI) was identified as a useful proxy for silage nutritive value.

Figure 2: Individual crop/farm data



Real time monitoring of maize crop

While APSIM is provides a good overall prediction of crop growth, it doesn't pick up day-to-day differences across a paddock. Remote sensing fills that gap, using satellite and sensor data to give a full paddock view of crop condition and feed availability as it changes through the season. The Dairy UP team combined these two tools to improve real-time predictions of yield and energy for silage decisions.

On farm monitoring

Dairy UP researchers evaluated 12 different maize silages across various NSW dairy farms to determine biomass, grain yield and the quality of the crop.

On average, grain in the maize silage was 78% starch and 14 MJ/kg DM, however, there was variation between the different maize cultivars.

Neutral Detergent Fibre (NFD) percentages varied across the rest of the plant. The cob (without the grain) was 70% NDF, while the leaf and stem were at least 60% and the grain was 8%.

Metabolisable energy (ME) of the grain was 14 MJ/kg/DM, while the rest of the plant was 8 MJ/kg DM or less.

Both APSIM and the combined APSIM–remote sensing approach were able to reliably predict how much maize biomass was grown across a range of conditions.

APSIM also provided a good estimate of feed energy under well-managed conditions. This suggests these tools have the potential to be used in silage systems.

Being able to accurately predict both yield and energy helps farmers better plan how much silage they will have and its feed value — particularly important in intensive, housed systems where maize silage makes up a large part of the ration.

Maize silage nutritional quality

The Dairy UP team looked at the feed value of whole-crop maize silage and why crops with very different starch levels can still deliver similar overall energy.

The work showed that as starch increases, fibre digestion can drop. This happens because starch is broken down quickly in the rumen, lowering rumen pH and reducing the activity of the microbes that digest fibre. Starch itself is still well digested, but less fibre is used.

To better reflect how this works in the cow, the team developed a new approach that looks at grain (starch) and the rest of the plant (fibre) separately, rather than treating the silage as one uniform feed.

This gives a more accurate picture of silage quality and energy supply and improves the ability to predict feed value in real time.

This supports more precise ration planning and silage management, helping lift feeding efficiency and control costs in intensive systems.

BMR performance

In this study, the team compared a newer, short-season brown midrib (BMR) hybrid with a conventional hybrid under different planting rates at University of Sydney's "Landsdowne" farm at Camden, NSW. Crops were well managed, with adequate nitrogen and irrigation. Both yield and feed quality were measured over two seasons. Drone imagery was used to assess crop performance at flowering.

Both hybrids produced high yields (25–28 tonnes of dry matter per hectare), and importantly, the brown midrib hybrid showed no lodging, suggesting improved standability compared to older types.

The conventional hybrid produced slightly higher yields (about 2 tonnes of dry matter per hectare more) and higher starch levels. However, the brown midrib hybrid had better fibre digestibility. As a result, overall feed energy was similar between the two.

Planting rate influenced yield but not feed energy. The brown midrib hybrid performed best at moderate plant density, while the conventional hybrid achieved its highest yield at higher plant density.

Overall, these results show that newer brown midrib hybrids can close the yield gap while maintaining feed quality, giving farmers more flexibility when choosing hybrids and planting rates in intensive silage systems.

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Read more

P4 final report

Ojeda, JJ et a;. (2023) Field and in-silico analysis of harvest index variability in maize silage. Frontiers in Plant Science Volume 14

Islam et al. Revealing the real nutritional value for maize silage. J. Dairy Sci. Vol. 108, Suppl. 1, pp 174-175 (2025)

Islam et al. Integrating APSIM and remote sensing to predict maize silage biomass. Proc Precision Dairy Farming Conf, vpp 170-171 (2025)

Delivery organisations

The University of Sydney – Dairy Research Foundation
NSW Government Department of Primary Industries and Regional Development
Scibus
Dairy Australia

Partner organisations

NSW Farmers Growing the best
eastAUSmilk
Dairy NSW / Dairy Australia
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Field and in-silico analysis of harvest index variability in maize silage

Jonathan Jesus Ojeda[1,2]*, M. Rafiq Islam[3], Martin Correa-Luna[3], Juan Ignacio Gargiulo[4], Cameron Edward Fisher Clark[5], Diego Hernán Rotili[6,7] and Sergio Carlos Garcia[3]

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Maize silage is a key component of feed rations in dairy systems due to its high forage and grain yield, water use efficiency, and energy content. However, maize silage nutritive value can be compromised by in-season changes during crop development due to changes in plant partitioning between grain and other biomass fractions. The partitioning to grain (harvest index, HI) is affected by the interactions between genotype (G) × environment (E) × management (M). Thus, modelling tools could assist in accurately predicting changes during the in-season crop partitioning and composition and, from these, the HI of maize silage. Our objectives were to (i) identify the main drivers of grain yield and HI variability, (ii) calibrate the Agricultural Production Systems Simulator (APSIM) to estimate crop growth, development, and plant partitioning using detailed experimental field data, and (iii) explore the main sources of HI variance in a wide range of G × E × M combinations. Nitrogen (N) rates, sowing date, harvest date, plant density, irrigation rates, and genotype data were used from four field experiments to assess the main drivers of HI variability and to calibrate the maize crop module in APSIM. Then, the model was run for a complete range of G × E × M combinations across 50 years. Experimental data demonstrated that the main drivers of observed HI variability were genotype and water status. The model accurately simulated phenology [leaf number and canopy green cover; Concordance Correlation Coefficient (CCC)=0.79-0.97, and Root Mean Square Percentage Error (RMSPE)=13%] and crop growth (total aboveground biomass, grain + cob, leaf, and stover weight; CCC=0.86-0.94 and RMSPE=23-39%). In addition, for HI, CCC was high (0.78) with an RMSPE of 12%. The long-term scenario analysis exercise showed that genotype and N rate contributed to 44% and 36% of the HI variance. Our study demonstrated that APSIM is a suitable tool to estimate maize

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FINAL OUTPUT JOURNAL

P5
Our farm our plan

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Final Report

P5 Our farm our plan

Dairy Australia

This project was led by Greg Duncan,
Dairy Australia

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

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Project Overview

This project was originally developed and led by Dairy Australia. Dairy UP enhanced its delivery for the benefit of the NSW and broader dairy industry. The project was completed in May 2024 and the final report/project summary is included here.

Item	Description
Project Title	Our farm our plan
Funding Body	Dairy UP
Dairy UP Project	P5
Project Duration	2021-2024
Lead Organisation	Dairy Australia
Project Lead	Greg Duncan
Project Team	Neil Webster, John Steer

Final Project Summary
P5: Our Farm Our Plan

May 2026

Planning for success

In 2020 the Australian dairy industry launched an initiative called "Our Farm, Our Plan" to help dairy farmers set long term goals, improve business performance, and manage risk.

Central to this program is the development of a farm strategic plan including business and personal goals.

Within Our Farm, Our Plan is a strategic plan, known as a 'plan on a page' – a one-page document that is developed by farm business managers, with assistance from industry consultants – that provides direction by outlining goals and practical steps to get there.

It is a document that helps dairy farm businesses plan for success.

Potential benefits

Clear, long-term business and personal goals help farmers and farm businesses identify and prioritise actions, manage uncertainty, mitigate risks, and capture opportunities.

These goals – and the consultation, deep and strategic thinking required to develop them – enable business owners a unique insight into themselves and their business.

From this, they get a better understanding of their current business position, including its strengths and weaknesses, and can set a direction for the farm business.

Clear business direction is a great way to boost morale and involvement, stay on track and check progress.

An effective strategic plan also helps a business deal better with challenges or unexpected hurdles, such as changes to operating environments and climate risks.

Looking into the future

Dairy UP's P5 project, Our Farm Our Plan, was a Dairy Australia initiative, delivered in NSW through Dairy NSW, Subtropical Dairy, Murray Dairy and Dairy Australia.

The team worked with industry organisations to increase the proportion of dairy farmers in NSW with effective strategic plans to help clarify long-term business and personal goals.

Successful businesses have a strategic plan that describes goals and a plan to achieve them including practical steps.

This plan helps a business manage risk and improve its performance by encouraging reflection and a proactive business culture.

It is also a great tool for recovering from adverse events such as bushfires or droughts.

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A strategic plan is a good fit with businesses recovering from adverse events such as bushfire recovery or drought.

It also helps with investment decisions, succession planning, staff recruitment and retention as well as implementation of technology and much more.

For the dairy industry, a big part of Our Farm, Our Plan was to upskill local consultants.

This enables farmers have access to assistance quicker than flying in interstate consultants following adverse events.

Strategic approach

Our Farm, Our Plan provides a framework for identifying goals, planning, evaluation and action steps.

It equips dairy farmers with the skills and knowledge to apply strategic thinking and planning processes to achieve their agreed business and lifestyle goals.

As part of the planning process, farmers outline their business direction – including goals – assess their current position and business risks, identify, and prioritise options, understand risk management principles, records access farm performance data and develop communication skills.

NSW delivery

Dairy UP's P5 Our Farm, Our Plan ran until June 2024. One hundred and thirty farmers participated in Our Farm, Our Plan workshops and one-to-one sessions delivering more than 60 individual plans across 22 workshops.

This is a high proportion of the industry compared to other dairying states.

A breakdown of the Dairy UP roll-out included:

- Eight face-to-face workshops
- Two intensive workshops
- Three workshops focused on the next generation – those looking for a career path – through the Young Dairy Network (YDN)
- Nine national online opportunities

Ongoing delivery

Our Farm Our Plan continues to be delivered nationally by Dairy Australia. Read more.

Feedback

The workshops delivered more to dairy farmers than strategic planning skills. Feedback from dairy farmers about the workshops highlighted the benefits of meeting fellow farmers while developing a useful planning tool for their farm business.

Collaborators

Delivery of Our Farm Our Plan in NSW involves collaboration between Dairy UP, Dairy Australia. Dairy NSW, Subtropical Dairy and Murray Dairy.

More info

Project lead
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Delivery organisations

The University of Sydney – Dairy Research Foundation
NSW Government – Department of Primary Industries and Regional Development
Scibus
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Partner organisations

NSW Farmers – Growing the best
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FINAL OUTPUT JOURNAL

P6
Future systems - Dairy Da.T.A

P6a - Resilient cattle (Heat tolerance)
P6b - Resilient cattle (Health)
P6c - Future systems - Digital feeding

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Final Report

P6 – Future systems – Dairy Da.T.A

THE UNIVERSITY OF SYDNEY
Dairy Research Foundation

This project was led by Professor Cameron Clark(*)
and Dr. Anna Chlingaran from the University of Sydney Dairy Research Foundation

(*) Currently Charles Sturt University

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Delivery organisations

The University of Sydney Dairy Research Foundation | Department of Primary Industries and Regional Development | Scibus | Dairy Australia

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1. Executive Summary

The Australian dairy sector is rapidly shifting toward smarter, more resilient production systems. The P6: Future Systems – Dairy Da.T.A project funded under the Dairy UP program integrates animal science and data science to address three critical knowledge gaps affecting productivity, welfare, and resource efficiency on Australian farms. The project aims to utilise data, advanced technologies and automation to integrate information from multiple sources to enable the creation of tools that support on-farm decisions.

P6a – Resilient cattle (heat tolerance): optimising on-farm energy use and cooling systems

Heat stress (HS) imposes major economic and welfare costs on the Australian dairy industry each year, reducing milk yield, reproductive performance and overall cow comfort. Building herds with stronger heat-tolerance traits is a key strategy for reducing the impacts of HS. This subproject focused on developing new phenotypic indicators by examining how individual cows respond to heat events using continuous multi-sensor data, including core body temperature measurements and climate records. The work revealed substantial variation between animals, with rises in internal temperature occurring at around THI 67 - earlier than the 70-75 guidance commonly used on farms. Drinking behaviour also showed a clear relationship with heat load, and a model was developed to account for drinking events when interpreting temperature data. Seasonal and regional patterns were evident, with cows drinking more frequently in summer and in warmer climates. Advanced modelling approaches were applied to combine climate and production data, enabling more detailed patterns of heat response to be detected than with traditional methods alone. Through collaboration with DairyBio, these sensor-derived indicators provide a foundation for developing new heat-tolerance phenotypes that complement genomic information, and ongoing integration of phenotypic and genotypic data aims to further improve the reliability of the Heat Tolerance ABV.

P6b – Resilient cattle (health): early intervention enabled by advanced sensing

Advanced sensing technologies are reshaping herd-health management by enabling earlier detection of emerging issues than is possible through visual observation alone. Wearable and in-body systems such as CowManager and smaXtec boluses continuously capture data on activity, temperature, rumination and feeding behaviour, with algorithms identifying deviations from normal patterns that signal potential health events. Through collaboration with Moxey Farms, this subproject has successfully assembled and validated a substantial dataset linking sensor outputs with farm health and production records from DataGene, providing a strong foundation for developing early-warning indicators. Progress to date has focused on data collation, harmonising health-event definitions and securing access to external datasets. Further analytical work has been delayed due to health-related interruptions that required the PhD candidate to suspend their studies multiple times, but the project remains well positioned to advance early-intervention tools that can reduce disease impact and improve herd resilience.

P6c – Digital feeding: data-driven feeding to optimise grain allocation in pasture-based herds

Feed costs remain one of the largest expenses in Australian dairy systems, making efficient use of supplementary concentrate critical for profitability. This subproject explored how artificial intelligence can support more precise, individualised feeding decisions in pasture-based herds. Using machine learning models to predict milk yield from routinely collected cow data, and optimisation algorithms to adjust daily concentrate levels, the work demonstrated that data-driven allocation can improve feed-use efficiency without increasing total daily grain input. Across two studies, AI-based optimisation consistently increased herd-level milk yield by around 8%, highlighting the value of accounting for cow-to-cow variability rather than relying on flat-rate feeding.

These findings show strong potential for integrating machine-learning tools and optimisation algorithms into digital feeding systems to support more profitable and sustainable concentrate-use strategies in pasture-based dairying.

The P6 project delivered strong research outcomes, including two completed PhD theses and six peer-reviewed publications in high-impact international journals, with another manuscript currently under review. The work reached more than 3,810 people through about fifteen conference presentations in Australia and internationally, and further engaged industry through farmer meetings, interviews and media articles. Project results were also presented to fourth-year Bachelor of Animal and Veterinary Bioscience students at the University of Sydney as real-world examples of data-driven research applied to industry challenges, contributing to future workforce capability.

2. Project Overview

Item	Description
Project Title	Future Systems – Dairy Da.T.A
Funding Body	Dairy UP
Dairy UP Project	P6
Project Duration	2021-2026
Lead Organisation	The University of Sydney
Project Lead	Prof Cameron Clark
Key Collaborators	Charles Sturt University (CSU), Dairy commercial farms, CowManager, smaXtec, DataGene, Scibus, DairyBio

3. Abbreviations

AI – Artificial Intelligence
AIiA – Artificial Intelligence in Agriculture
BOM — Bureau of Meteorology
COMPAG — Computers and Electronics in Agriculture
CSU — Charles Sturt University
DIM — Days in Milk
HAIM — Hybrid AI-based Model
HS — Heat Stress
LLM — Linear Mixed Model
ML — Machine Learning
MY — Milk Yield
RRM — Random Regression Model
RRT — Reticulorumen Temperature
SAT — Smart Agricultural Technology
THI — Temperature–Humidity Index
USyd — The University of Sydney

4. Project Background and Rationale

The Australian dairy industry faces increasing pressure to improve productivity, sustainability, and resilience. Rising input costs, climate variability, tightening environmental regulation, and competitive global markets are pushing producers and processors to find new ways to extract more value from existing resources. Dairy UP was established to accelerate applied research that directly addresses some of these pressures.

Within this context, the P6 "Future Systems – Dairy Da.T.A" program was funded to explore sensor-enabled, large-scale datasets and develop advanced data-driven approaches to enhance the selection of heat-tolerant cattle and strengthen early-intervention tools for animal health, as well as maximise herd-level milk production through optimised grain allocation in pasture-based herds. These focus areas were selected because they address core constraints on herd resilience and productivity in Australian dairy systems, and because each represents a clear opportunity to translate emerging sensor technologies and large-scale data into practical, on-farm decision tools.

5. Project Objectives

The project aimed to deliver practical, data-driven solutions to help dairy farmers improve herd resilience, animal health and feed efficiency under increasingly variable climatic conditions. The specific objectives were to:

- **Improve understanding of HS** by clarifying key terms and concepts so farmers, advisors and researchers can communicate consistently and make better decisions about heat-abatement strategies.
- **Enhance the use of sensor data on farm** by developing reliable methods to interpret reticulorumen temperature and drinking-behaviour data, improving the accuracy of heat-stress monitoring tools.
- **Identify cows that cope better with heat** by analysing large-scale datasets to understand individual differences in heat response and determine practical indicators that can support future breeding and management decisions.
- **Evaluate simple, on-farm indicators of heat tolerance** such as milk-yield changes, drinking patterns and temperature responses, to help farmers identify animals that are more vulnerable during hot conditions.
- **Develop advanced AI-enabled approaches to support breeding for heat tolerance** by integrating machine learning with statistical modelling to improve the accuracy of identifying heat-tolerant cows without compromising milk production.
- **Maximise herd-level milk production through optimised feed allocation** by developing machine-learning and optimisation tools that allocate grain more efficiently across the herd, improving milk yield without increasing feed costs.
- **Build integrated datasets that support future digital tools** by linking climate, production, behaviour and physiological data to enable next-generation decision-support systems.
- **Share practical insights with industry** through publications, presentations, farmer meetings and teaching activities to support adoption and build capability across the sector.

6. Subproject P6a – Resilient cattle (heat tolerance)

Research team: Cameron Clark (CSU), Yani Garcia (The University of Sydney - USyd), Anna Chlingaryan (USyd), Peter Thompson (USyd), Alice Shirley (USyd)

6.1 Background

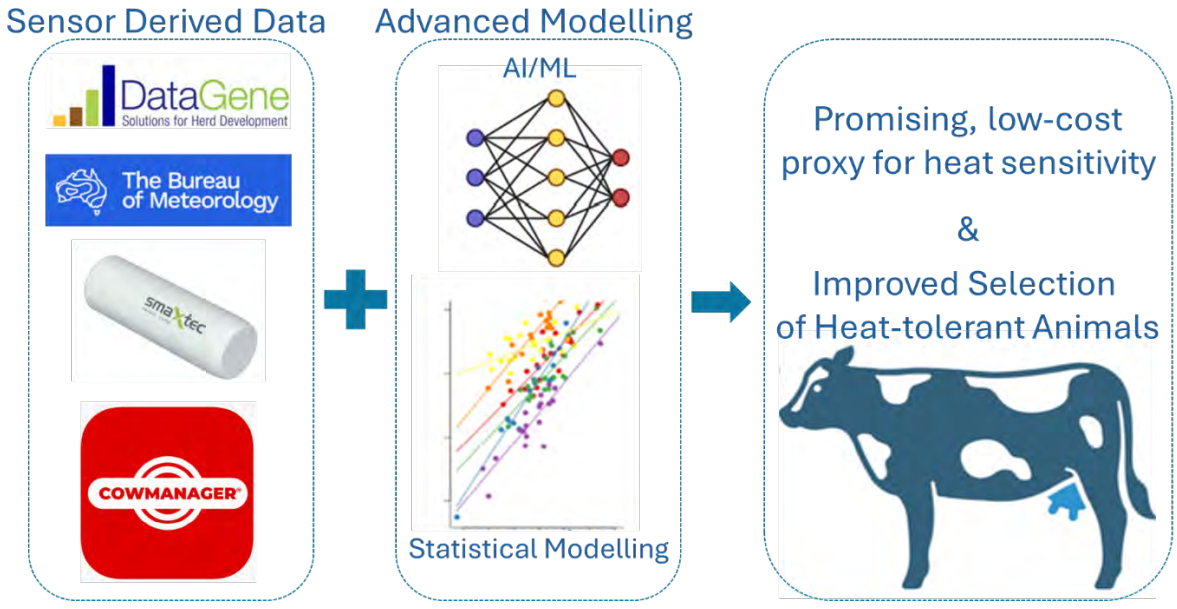
Dairy farming in Australia spans diverse production environments that are increasingly exposed to climate variability and rising HS risk. Heat stress reduces feed intake, milk yield, fertility and cow comfort, and can compromise animal welfare. Cattle respond to heat load through a range of behavioural and physiological mechanisms aimed at restoring thermal balance, but these responses vary widely among individuals. Understanding this diversity is essential for developing climate-smart dairy systems.

Recent advances in sensor technologies, particularly reticulorumen temperature (RRT) boluses, have created new opportunities to monitor physiological responses to heat in real time. However, interpreting these data is challenging. Drinking events cause rapid, pronounced drops in reticulorumen temperature, masking true core-temperature patterns and limiting the reliability of HS detection tools. Without robust methods to distinguish behavioural effects from physiological responses, sensor data cannot be confidently used for on-farm decision-making.

At the same time, the dairy industry lacks consistent terminology for describing HS responses. Terms such as tolerance, resistance, resilience and susceptibility are used inconsistently across research and advisory contexts, creating confusion and limiting the translation of scientific findings into practice.

There is also limited understanding of the extent to which cows differ in their sensitivity to heat load. Some animals show sharp increases in core temperature or declines in milk yield at relatively mild THI levels, while others maintain stable performance. Identifying this diversity is critical for both management and breeding.

Finally, while genetic selection for heat tolerance is recognised as a long-term strategy, current phenotyping approaches rely on established statistical models. Artificial intelligence (AI) and Machine Learning (ML) methods offer new opportunities to improve the accuracy and efficiency of heat-tolerance phenotyping (Fig. 1).



6.2 Objectives

1. Standardise HS terminology to improve communication and decision-making across the dairy sector.
2. Improve interpretation of sensor data by reliably separating drinking events from true core-temperature responses.
3. Identify variation in cow heat-stress responses using large-scale climate, temperature and production datasets.
4. Evaluate practical indicators of heat tolerance such as milk yield, drinking behaviour and temperature changes.
5. Develop AI-enabled heat-tolerance phenotypes to support future breeding and management decisions.

6.3 Materials & Methods

- Data sources:
    - For objectives 2, 3 and 4, the analysis used RRT data collected from 1,429 dairy cows across three pasture-based farms in Victoria, obtained via smaXtec boluses, together with matched climate data. Reticulorumen temperature was recorded every 10 minutes, producing approximately 75 million records. To develop and validate the drinking-event detection model, an additional 28 heifers from the USyd farm were monitored, contributing around 1,000 video-validated drinking events.
    - For objective 5, model development used ~3 million herd-test records (provided by DataGene) from 201,836 cows across 97 NSW herds (2003–2022). These records included daily milk yield, animal age, parity and DIM. The dataset was matched with 20 years of daily THI values (≈7,300 records) from the BOM to support AI-enabled heat-tolerance phenotyping.
- Statistical analysis: A range of statistical models were applied depending on the research aim.
    - Sensor-data processing: Time-series decomposition and threshold-based algorithms were developed to detect drinking events from characteristic temperature-drop patterns. Multiple detection settings were evaluated to ensure reliable separation of drinking events from true core-temperature responses.
    - Heat-response modelling: Mixed models were used to quantify associations between THI and RRT deviation, estimate individual sensitivity to heat load, and assess the influence of baseline THI conditions.
    - Phenotypic-indicator evaluation: Relationships among RRT, MY and drinking frequency were assessed using mixed models and slope-based analyses to identify potential proxy traits for heat tolerance.
    - AI-based phenotyping: A hybrid AI-statistical model was developed by integrating ML algorithms with LLMs to estimate heat-tolerance phenotype based on MY decline above THI threshold.

o

6.4 Key Findings

- **Heat-stress terminology requires standardisation.** A critical review revealed major inconsistencies in the use of *tolerance*, *resistance*, *resilience* and *susceptibility*.
- **Drinking events can be accurately detected from sensor data.** A transparent, high-accuracy model (F-score = 0.99) was developed to identify drinking events (Fig. 1), improving interpretation of RRT and the accuracy of heat-stress monitoring tools.
- **Seasonal variation in drinking behaviour reflects underlying physiological diversity.** Drinking frequency, temperature-drop magnitude and recovery time differed between summer and winter.
- **Cows vary widely in their physiological response to heat.** Reticulorumen temperature increased with THI, but the magnitude of response differed substantially among individuals. Temperature deviations began at THI ≈ 65, suggesting current industry thresholds may underestimate heat load.
- **Drinking frequency influences cooling response.** Cows that drank more frequently showed a smaller temperature drop per drink, indicating reduced cooling efficiency at higher drinking rates (Fig. 2). Individual-animal differences in this relationship were substantial, highlighting variation in how cows regulate core temperature through drinking behaviour.
- **Multiple traits show heat-related variation.** Milk yield, drinking frequency and RRT all responded to thermal deviations, with strong individual differences. Inter-trait correlations confirmed that these measures are interconnected in how cows react to heat (Fig. 3). Drinking frequency emerged as a promising, low-cost proxy for heat sensitivity, offering farmers an early means to identify and support more vulnerable animals.
- **AI improves heat-tolerance phenotyping.** The hybrid AI-based model (HAIM) reduced residual error compared with RRM, improving milk-yield prediction accuracy by 3.56% (Table 1). The RRM and HAIM overlapped in identifying 84% of heat tolerant animals, but the HAIM replaced about 8% of the cows that weren't identified with the statistical method, while accounting for a greater proportion of the total observed variability (i.e. increasing accuracy).

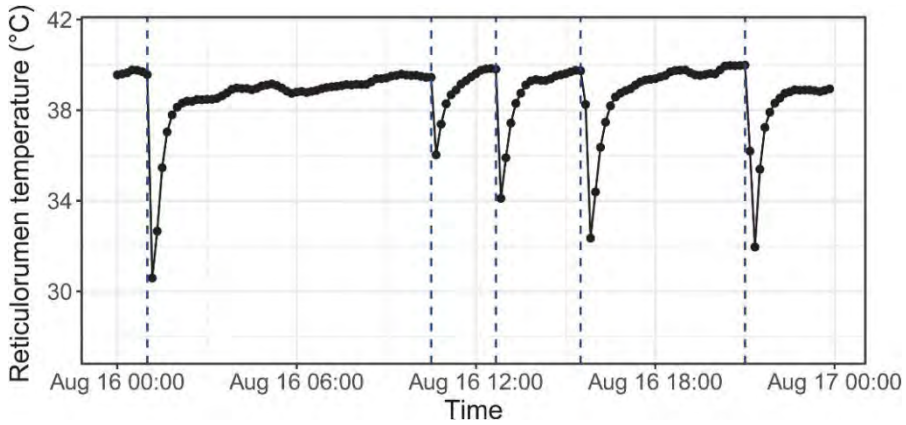


Figure 1 *Synchronisation of raw sensor data (black dots indicate 10-minute interval reticulorumen temperature readings) and manual video annotations (dashed blue lines indicate observed drinking events).*

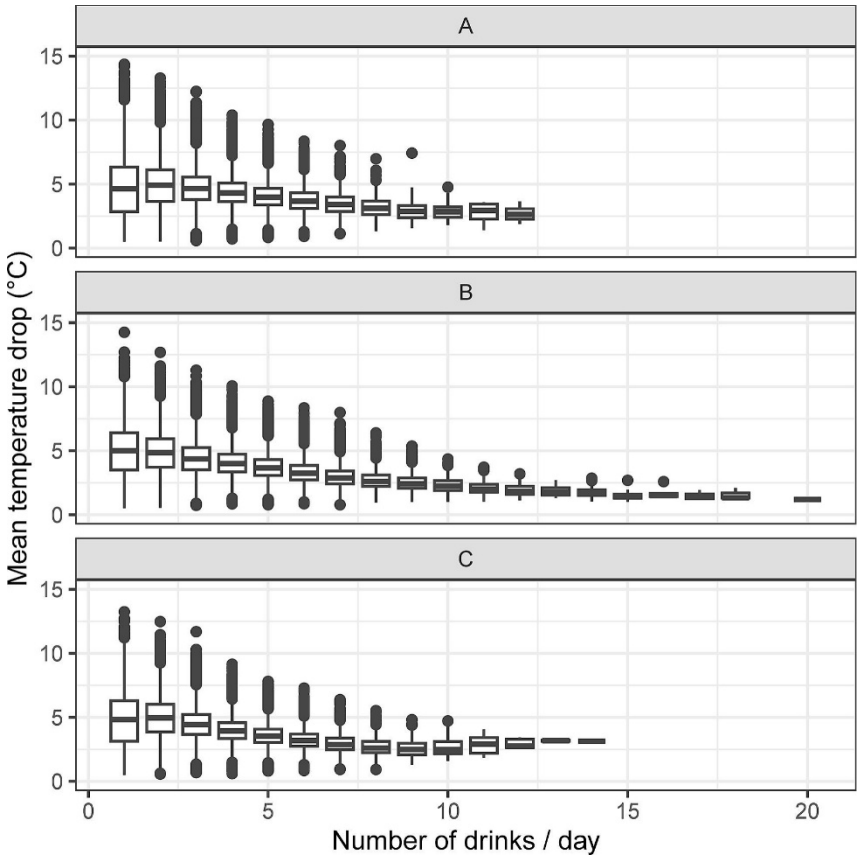


Figure 2 *Association between number of drinks per day and average drop in reticulorumen temperature (°C)*

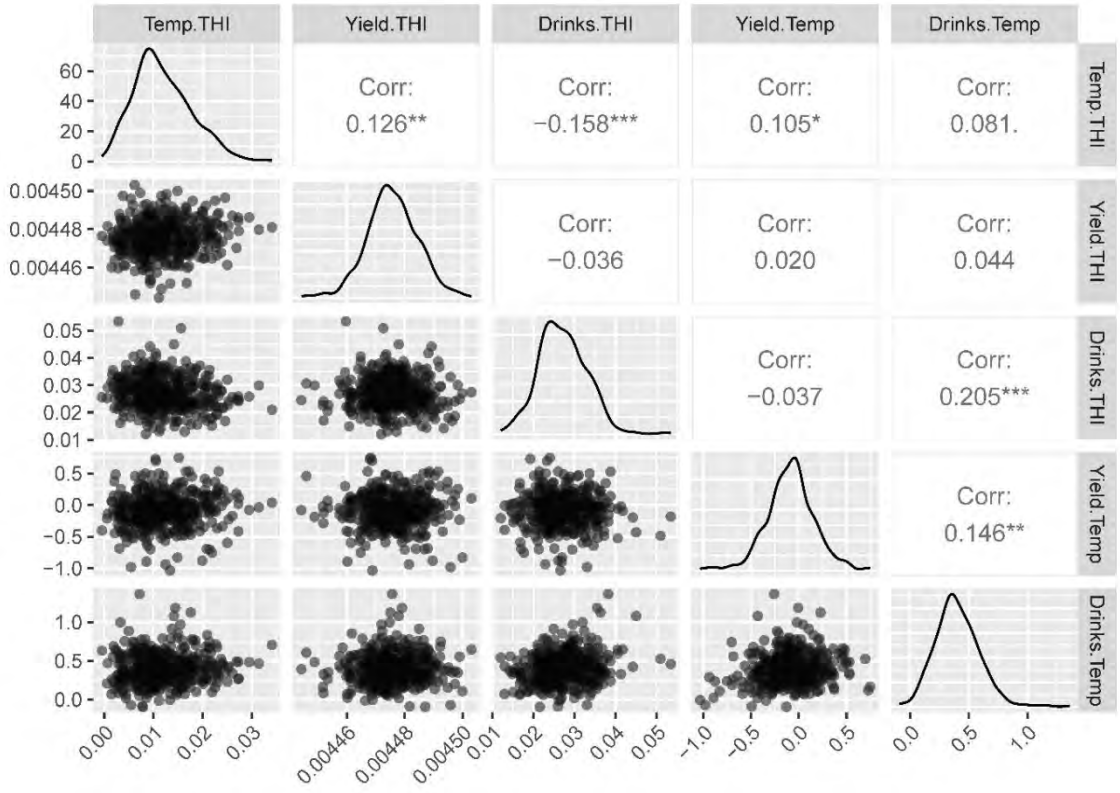


Figure 3 *Generalised pairs plot revealing the association between slopes of Tempdev per change in THImax_dev (Temp.THI), Yielddev per change in THImax_dev (Yield.THI), ndrinkdev per change in THImax_dev (Drinks.THI), Yielddev per change in Tempdev (Yield.Temp), and ndrinkdev per change in Tempdev (Drinks.Temp) for each individual.*

Table 1 Model performance comparison - key findings

	HAIM vs RRM
Predictive ability	
Statistical performance metric: SD of residuals (L/cow/test date)	3.25 for HAIM vs 3.37 for RRM, representing 3.56 % reduction of milk yield estimation error
Estimated random slopes / intercepts	High association between the random slopes (R^2 = 0.93) and intercepts (R^2 = 0.98) showing general agreement between the models with HAIM outperforming RRM (as evidenced by the HAIM's lower SD of residuals)
Practical relevance	
Heat-tolerant cattle classification	• Heat-tolerant: HAIM re-classified ~8 % of cattle in the sense of heat tolerance • High-producing and heat-tolerant: 18.1 % (of the total cow population) by HAIM and 18.24 % by RRM • Agreement RRM and HAIM: ~84 % out of the identified high-producing and heat-tolerant cattle
Importance of both traits: random slopes and intercepts	Negative association between random slopes and random intercepts estimated by both models

6.5 Applications & Impacts

The findings have direct relevance for improving heat-stress monitoring, management and long-term selection of heat-tolerant cattle.

- **Improved heat-stress monitoring**: Reliable detection of drinking events supports more accurate interpretation of temperature patterns by distinguishing true heat-stress responses from cold-water effects. This capability can already inform more targeted heat-abatement decisions within existing smaXtec systems. Although P6a incorporated climate data for research analysis, fully automated early-warning platforms that integrate climate, behaviour and predictive modelling will require further development before commercial release.
- **Better identification of vulnerable cows:** Understanding individual variation in heat responses can already help farmers recognise which cows are more vulnerable during hot conditions and prioritise shade, cooling and feeding strategies accordingly. While this provides immediate, practical value, developing automated vulnerability scores or decision-support tools that classify cows by heat-risk level will require further validation across farms and seasons before wider industry adoption.

- **Foundations for heat-tolerance breeding:** AI-enabled phenotyping and validated proxy traits provide a pathway for incorporating heat tolerance into breeding programs without compromising production.
- **Enhanced welfare and climate resilience:** By revealing meaningful diversity in physiological responses to heat, P6a provides insights that farmers can already use to support more welfare-focused management during hot conditions. These findings also contribute to longer-term progress toward climate-resilient dairy systems, where individual animal responses are incorporated into breeding, management and decision-support frameworks as they mature.
- **Improved decision-support tools:** Integrated datasets linking climate, physiology and production create a foundation for next-generation digital tools that support real-time management decisions.

6.6 Key Outputs

- A high-quality dataset integrating climate records, sensor measurements, drinking behaviour and milk production assembled across multiple farms. The structure enables seamless addition of new herds, supporting continuous expansion and long-term system learning.
- A high-accuracy method was developed and validated to identify drinking events from RRT data collected via smaXtec sensors, improving interpretation of physiological responses to heat.
- A methodology was developed that integrates AI with current statistical approaches, improving the selection of heat-tolerant cattle.
- Multiple phenotypic indicators were identified that respond consistently to thermal load, providing complementary insights into individual heat-tolerance profiles.
- The project contributed to ongoing DairyBio efforts to extend the Heat Tolerance ABV by incorporating sensor-based phenotypic indicators, supporting identification of animals that maintain production under hot and humid conditions.
- Five peer-reviewed publications in high-impact Q1 journals such as *Computers and Electronics in Agriculture, Smart Agricultural Technology, Scientific Reports*, and *Animal*
- Conference presentations, industry reports, and media interviews and articles disseminated findings to researchers, advisors and producers.

6.7 Future Research Opportunities

Building on the advances made in sensor interpretation, physiological characterisation and AI-enabled phenotyping, several opportunities remain to further strengthen heat-tolerance research and accelerate translation into breeding and management tools. These opportunities focus on deepening biological understanding, improving prediction accuracy and ensuring practical on-farm application.

- **Genomic marker discovery:** Identify genetic variants associated with heat-tolerance phenotypes derived from sensor and AI models.
- **Refinement of THI thresholds:** Develop region- and breed-specific thresholds based on physiological data rather than historical assumptions.
- **Integration into commercial decision-support tools:** Embed drinking-event detection and heat-response modelling into farm-management platforms.
- **Validation of proxy traits:** Confirm the utility of drinking frequency and MY slopes as practical indicators of heat sensitivity.
- **Development of amelioration strategies:** Use sensor-derived insights to design targeted cooling, feeding and management interventions.
- **Long-term selection strategies:** Evaluate the economic and production impacts of selecting for heat tolerance across different herd structures and environments.

7. Subproject P6b – Resilient cattle (health)

7.1 Background

Improving the resilience and health of dairy cattle requires earlier identification of emerging disorders than is possible through visual observation alone. Subproject P6b focuses on establishing the foundations for early-intervention health sensing by integrating advanced sensor technologies with detailed farm health and production records.

Health disorders both in the periparturient period and throughout lactation are an important factor impacting animal welfare and production in both the immediate and long term and are rarely observed in isolation within the dairy industry. The incidence of health disorders, including metabolic and reproductive illness, predominantly occur within the first month post calving. Production illnesses, including mastitis and lameness, generally occur throughout lactation and after drying off. Understanding the interconnected relationship between health disorders in conjunction with relevant risk factors is an essential aspect of developing prediction of potential illness with high sensitivity and specificity. This in turn enables earlier detection of disease and more timely interventions put in place to reduce both the incidence and severity of impact of the health disorders.

7.2 Objectives

- **Establish a strategic data partnership** with Moxey Farms to secure access to their large, year-round calving herd and associated health, production and management records, enabling high-resolution analysis of disease patterns in a commercial setting.
- **Integrate multi-source sensor and farm data** by linking outputs from wearable and in-body sensing systems with harmonised health-event and production datasets to create a reliable, research-ready database for modelling early-intervention indicators.
- **Quantify health-event patterns** by characterising the incidence, timing and production impacts of major metabolic, reproductive and production-related disorders across the periparturient period and throughout lactation, using harmonised herd-management and production records.
- **Develop predictive risk models** that identify animals at elevated likelihood of developing key health disorders, and map the relationships between these conditions, enabling earlier detection and more targeted intervention strategies.

7.3 Materials & Methods

- **A retrospective analysis using herd-level production and management records from a large year-round calving dairy herd.** Data were collected for 3,154 primiparous and multiparous cows over a six-month period (1 August 2022 – 31 January 2023). Health events were categorised into metabolic disorders (milk fever, displaced abomasum, ketosis, gut illness), reproductive disorders (retained foetal membranes, metritis), mastitis, and other production-related conditions such as respiratory disease, lameness and foot-rot. These events were harmonised into consistent categories to support comparative analysis. A descriptive epidemiological assessment was conducted to quantify event frequency and timing relative to calving, followed by a Kruskal–Wallis test to evaluate differences in temporal patterns between disorder groups.

- **Extension to a larger dataset:** Incorporate cows from multiple farms over an 18-month period (1 August 2022 – 31 January 2024), enabling assessment of cross-herd variation, seasonal influences and broader disease patterns. The expanded dataset also includes disorders occurring in late pregnancy to provide a more complete representation of the periparturient interval. Planned analyses include descriptive comparisons of incidence across herds and evaluation of differences in post-calving timing between disorder categories, forming the basis for subsequent predictive-model development.

7.4 Key Findings

- **Robust data partnership established** with Moxey Farms, enabling access to their large-scale herd health, production and management records for comprehensive analysis of disease patterns under commercial conditions.
- **Validated multi-source dataset** created by linking CowManager sensor outputs with harmonised farm health events and DataGene production records, providing a reliable, research-ready foundation for developing early-warning indicators.
- **Early postpartum identified as highest-risk period:** Analysis of 3,154 cows recorded 142 post-calving health events, with 40% (57/142) occurring within the first seven days after calving (Fig. 5). Event timing differed significantly between disorder categories ($P = 4.5 \times 10^{-7}$), confirming that metabolic, reproductive and production-related illnesses follow distinct temporal patterns and warrant further investigation using the larger multi-farm dataset

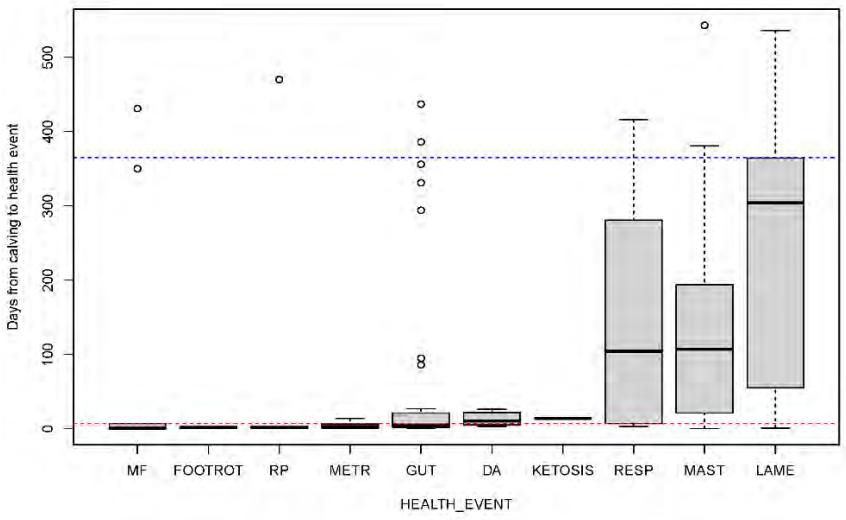


Figure 4 *Number of health events by category relative to number of days post-calving. The red dotted line indicates 7 days post calving, and the blue dotted line indicates 365 days post-calving. MF represents milk fever (hypocalcaemia), FOOTROT represents foot-rot, RP represents retained placenta/foetal membranes, METR represents metritis, GUT represents gut illness, DA represents displaced abomasum, KETOSIS represents clinical ketosis, RESP represents respiratory illness, MAST represents clinical mastitis, and LAME represents lameness requiring treatment.*

The subproject generated a peer-reviewed abstract presented at the ADSS 2024. Subproject activities were delayed due to health-related suspension of the PhD candidature.

8. Subproject P6c — Digital feeding

Research team: Yani Garcia (USyd), Anna Chlingaryan (USyd), Cameron Clark (CSU), Blessing Azubuike (USyd), Dr Martin Correa-Luna (USyd)

8.1 Background

Feed costs account for 40–60% of milk production expenses in Australian dairy systems, making efficient use of supplementary concentrate a major driver of farm profitability. Despite this, most pasture-based herds still rely on flat-rate feeding, where all cows receive similar concentrate levels regardless of their individual production potential, stage of lactation, or response to heat and pasture variability. This approach overlooks substantial cow-to-cow differences in feed efficiency and milk yield response.

Advances in ML and optimisation algorithms now offer the opportunity to move beyond flat-rate feeding toward individualised, data-driven concentrate allocation. By predicting each cow's expected milk yield from routinely collected data, and then optimising grain distribution across the herd, digital feeding systems can increase total milk production without increasing total daily concentrate use.

Subproject P6c evaluated this opportunity through two complementary studies. Both demonstrated that integrating ML prediction models with optimisation algorithms can increase herd-level milk yield by approximately 8%, highlighting the value of accounting for individual variability in pasture-based systems. These results provide a strong foundation for future digital feeding tools that support more profitable and sustainable concentrate use strategies.

8.2 Objectives

- Identify opportunities to improve concentrate use efficiency in pasture-based dairy herds.
- Develop data-driven methods to adjust grain allocation based on individual cow needs.
- Demonstrate how digital feeding strategies can increase herd-level milk production without increasing feed costs.
- Evaluate different digital optimisation approaches to determine which are most effective and practical for on-farm use.
- Provide a foundation for future digital feeding tools that support more profitable and sustainable concentrate use.

8.3 Materials & Methods

- **Data sources:** As the project included two complementary studies, it analysed large datasets from two commercial pasture-based dairy herds. The first pre-processed dataset contained 456,078 daily records from 1053 cows, including MY, concentrate intake, DIM, parity, breed and stage of lactation, matched with local climate information (temperature, humidity and vapour pressure). The second dataset included 32,504 records from 130 Holstein-Friesians for model development, with an additional 7,371 records from 81 cows used to test digital feeding strategies.
- **Data analysis to understand herd feeding patterns:** Milk yield, concentrate intake and animal characteristics were analysed to identify how cows differ in their response to grain. This included grouping cows with similar production levels and highlighting where flat-rate feeding may under- or over-supply concentrate.

- **Machine Learning for MY prediction:** Routine farm data (MY, DIM, parity, concentrate intake and weather) were used to build and evaluate sixteen ML models that estimate how much milk each cow is likely to produce under different feeding levels. These prediction models provided the foundation for testing alternative feeding strategies.
- **Optimisation tools for testing digital feeding strategies:** Optimisation tools, such as evolutionary algorithms and Dirichlet-Rescale algorithm with Monte Carlo simulation, were used to explore how concentrate could be redistributed across the herd without increasing total grain use. Feeding limits reflected normal farm practice, including minimum and maximum concentrate levels and limits on daily changes.
- **Comparing feeding approaches:** The performance of digital feeding strategies was compared with the farm's existing rule-based or flat-rate feeding system. This allowed the project to quantify potential improvements in milk production and feed efficiency under real-world conditions.

8.4 Key Findings

- **Digital feeding predicted an increase in herd-level milk production of 6.6–8.8% without increasing grain use.** The best-performing optimisation method (NSGA-II) consistently delivered around 8.6% more milk, demonstrating clear potential for improving feed efficiency on pasture-based farms (Fig. 7).
- **Cows differ widely in feed efficiency.** Milk produced per kg of concentrate ranged from 0.17 to 15.8 L/kg, showing that some cows convert grain into milk far more effectively than others. This variation highlights the limitations of flat-rate feeding and the opportunity to target grain to cows that respond best.
- **Machine learning-based milk yield prediction models performed reliably using routine farm data:** Machine learning models built from MY, DIM, parity, concentrate intake and weather explained around 61% of the variation in daily MY on *unseen* data. Days in milk and concentrate intake were the strongest predictors, confirming that accurate predictions can be made without liveweight data, which is often unavailable in pasture-based systems.
- **Individualised feeding outperformed the farm's existing feeding rules:** Digital feeding strategies shifted grain from low-responding cows to high-responding cows while respecting practical limits (e.g., minimum/maximum concentrate levels and daily change limits). This resulted in higher total milk production without extra feed costs.
- **Environmental conditions had only a small direct effect on milk yield:** Weather variables such as temperature and humidity added only minor improvements to prediction accuracy. Cow-level factors remained the main drivers of milk response, meaning farms can adopt digital feeding without needing additional sensors or climate monitoring equipment.
- **Digital feeding systems are practical for commercial pasture-based herds:** The approach worked with existing in-parlour feeding systems and used data already collected on most farms. Optimisation respected real-world feeding constraints and ran consistently across multiple days and seasons, demonstrating strong potential for on-farm adoption.

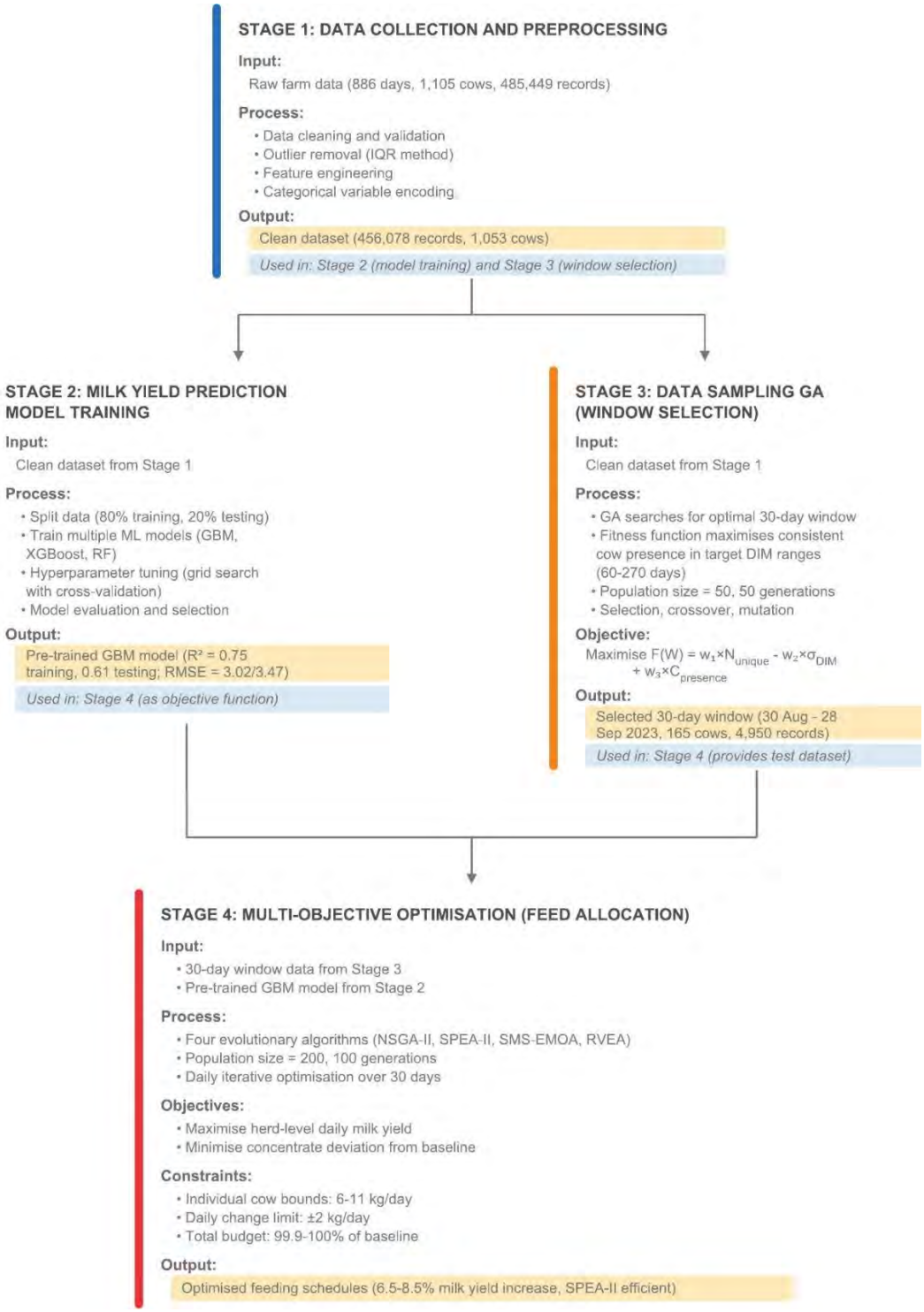


Figure 5 *Methodological framework showing the four-stage workflow. Stage 1: Data preprocessing; Stage 2: Machine learning model training (GBM selected as best model); Stage 3: Data Sampling GA for optimal 30-day window selection (165 cows); Stage 4: Multi-objective optimisation using evolutionary algorithms integrated with the pre-trained model. Arrows indicate data flow between stages, demonstrating how the Data Sampling GA (Stage 3) serves as a data selection tool for testing the optimisation algorithms (Stage 4).*

8.5 Applications & Impacts

- **Farm-level benefits:** Data-driven concentrate allocation provides a potential practical way for farmers to increase milk production by 6.6–8.8% without increasing total grain use. By shifting concentrate toward cows that respond best, farms can improve feed conversion efficiency, reduce waste, and lift overall herd performance. This approach supports more profitable and resilient feeding decisions in pasture-based systems.
- **Environmental and sustainability impacts:** Improving concentrate use efficiency reduces the amount of grain that is fed without contributing to milk output, lowering the environmental footprint of production. More efficient feeding supports reduced emissions intensity per litre of milk and aligns with sustainability expectations from processors, retailers and consumers.
- **Operational and management impacts:** The digital feeding approach works with existing in-parlour feeding systems and relies on data already collected on most farms. This means farmers can adopt precision feeding without major infrastructure changes. The system respects practical feeding limits, making it suitable for day-to-day use in commercial herds.
- **Technical and innovation impacts:** The successful development of ML models capable of predicting milk yield from routine farm data (without requiring liveweight) removes a major barrier to adoption in pasture-based systems. The integration of optimisation tools demonstrates that individualised feeding can be automated and scaled, providing a foundation for future digital decision-support tools.
- **Industry-wide opportunities:** The project provides clear evidence base for moving beyond flat-rate feeding. As digital feeding becomes more common, processors and advisory services can support farmers to improve feed efficiency, reduce production variability, and enhance the profitability of grain use across the sector.

8.6 Outputs

- **High-quality datasets linking production, feeding and climate data:** Compilation and preprocessing of two large commercial herd datasets, including over 456,000 cow-day records in Study 1 and 32,504 records in Study 2, providing robust evidence base for understanding concentrate use efficiency in pasture-based systems. The data-processing framework developed in this project is directly transferable to other commercial farms, meaning additional herds can be incorporated into the database to expand model accuracy, support benchmarking, and enable broader industry adoption.
- **Characterisation of cow-level variability in concentrate response:** Identification and quantification of substantial differences in feed efficiency between cows, providing a foundation for targeted concentrate allocation and future precision-feeding tools.
- **Predictive and optimisation tools for precision feeding:** Development and evaluation of sixteen MY prediction models using routine farm data, together with implementation of optimisation approaches demonstrating proof-of-concept for individualised concentrate allocation in pasture-based herds.
- **Peer-reviewed scientific contributions:** Publication of two peer-reviewed papers in high-impact journals (*Artificial Intelligence in Agriculture* (AIiA) and *Smart Agricultural Technology* (SAT)), along with conference presentations detailing the modelling framework, optimisation methods and implications for precision feeding in pasture-based dairy systems.

8.7 Future Research Opportunities

This subproject established proof-of-concept for a data-driven precision-feeding tool using commercial farm datasets, creating a strong foundation for further scientific development and industry adoption. Its key limitation is the absence of individual health indicators, forage intake and high-resolution pasture nutritional value data, which constrains the precision of cow-level response modelling. Building on this foundation, several opportunities exist to enhance the scientific robustness, practical utility and industry uptake of the approach.

- **Expansion of datasets and refinement of response modelling:** Incorporating additional commercial herds from diverse regions and production systems and extending the modelling framework to capture dynamic changes in feed efficiency across lactation, seasons and management conditions. Addressing current data gaps (individual health, forage intake and pasture nutritive value) would substantially enhance model accuracy and biological interpretability.
- **Testing the effectiveness of digital feeding on commercial farms:** this would require a well-designed experiment to address factors such as system type, region, pastures and concentrate types, etc.
- **Future industry engagement and knowledge transfer:** Further collaboration with farmers, advisors and technology providers to demonstrate the tool in real-time commercial settings, support adoption pathways, and facilitate integration into existing farm decision-support systems.
- **Economic modelling of return on investment:** Quantifying the financial benefits of precision-feeding strategies under varying milk prices, feed costs and pasture conditions to demonstrate the economic value proposition for commercial dairy farms.
- **Development of farmer-friendly decision-support interfaces:** Designing intuitive, user-centred interfaces that present model outputs in clear, actionable formats, enabling farmers to apply precision-feeding recommendations in daily management.

9. Project-wide Disseminations

The project has produced two PhD theses and six peer-reviewed publications in high-impact international journals, including *Artificial Intelligence in Agriculture* (Impact Factor (IF): 12.4), *Computers and Electronics in Agriculture* (IF: 8.9), *Smart Agricultural Technology* (IF: 5.7), *Animal* (IF: 4.2), and *Scientific Reports* (IF: 3.9), accumulating 12 citations to date (Table 2). These are leading Q1 journals in digital agriculture, sensor technologies and animal science, reflecting the scientific quality and international reach of the project's outputs.

Findings were also disseminated through multiple international and national conference presentations and industry-facing events, ensuring engagement across research, advisory and producer communities. This included presentations at the American Dairy Science Association (ADSA) Annual Meeting in the USA, as well as range of Australian and New Zealand forms, collectively reaching an estimated audience of more than 3,810 people (Table 3). A list of these outputs is provided in Tables 3 and 4 (Annexes).

In addition, the P6 outcomes were also presented to fourth-year Bachelor of Animal and Veterinary Bioscience students at the University of Sydney as real-world examples of data-driven research applied to industry challenges, contributing to future workforce capability.

***Table 2**. Peer-reviewed publications in international journals.*

Author	Title	Journal	Year Published	Citations (to May 26)
Azubuike et al. (2026)	Leveraging artificial intelligence and evolutionary algorithms for optimising cow supplementation and milk production	AliA	2026	-
Shirley et al. (2026)	Probing the diversity in dairy cattle reticulorumen temperature for adaptation selection	SR	2026	-
Chlingaryan et al. (2025)	An AI-based hybrid model for dairy cattle heat tolerance phenotype	SAT	2025	1
Azubuike et al. (2025)	A data-driven approach for optimising supplement allocation to individual lactating dairy cows in pasture-based systems	SAT	2025	1
Shirley et al. (2025)	The diversity in dairy cattle reticulorumen temperature: Identifying water intake events	COMPAG	2025	6
Shirley et al. (2024)	Review: Ruminant heat-stress terminology	Animal	2024	4

10. Conclusions and Recommendations

The P6 Future Systems – Dairy DaTA project has delivered important progress across two interconnected areas of dairy research, generating evidence-based findings with clear, pathways to industry application.

P6a demonstrated that advances in sensor interpretation, physiological characterisation and AI-enabled phenotyping improve the reliability of heat-stress monitoring and support the development of more effective management and breeding approaches. Successful identification of drinking events from raw sensor data enabled large-scale analyses showing substantial individual variation in heat responses, with drinking frequency emerging as a practical, low-cost indicator of heat sensitivity. The AI-based phenotyping model further improved the accuracy of identifying heat-tolerant animals compared with traditional statistical approaches. The outcomes of P6a provide a strong foundation for data-driven accurate heat-stress detection, better identification of vulnerable cows, and future breeding strategies aimed at improving herd resilience under increasing climate variability. A key next step is to extend this work through ongoing collaboration with DairyBio to integrate sensor-derived and AI-enabled phenotypes into genomic selection pipelines.

P6c demonstrated that digital precision feeding can substantially improve concentrate use efficiency in pasture-based dairy herds. By replacing flat-rate feeding with ML-based milk yield predictions and optimisation algorithms, the project showed that herd-level milk production can increase by 6.6–8.8% without raising total concentrate use. The work highlighted large cow-to-cow variation in feed efficiency and confirmed that routine farm data are sufficient to support reliable prediction models. While the approach proved technically feasible and compatible with existing in-parlour feeding systems, further refinement of prediction accuracy and on-farm real-time validation will be required before commercial deployment.

Across the two subprojects, the project has produced several peer-reviewed publications, engaged audiences across Australia and internationally, and built productive industry partnerships, laying a solid foundation for the next phase of research.

Recommendations for future investment should prioritise:

- Development of a national herd-level heat-tolerance phenotype tailored to pasture-based systems, in collaboration with DairyBio, to enable genomic selection that reflects real-world grazing environments.
- Validation of sensor-derived and behavioural indicators across diverse production environments to ensure they are robust enough for incorporation into national heat-tolerance selection and herd-management frameworks.
- Large-scale on-farm trials to evaluate the real-time performance of digital precision feeding systems and quantify their economic benefits under commercial pasture-based conditions.

11. Annexes

Table 3. *Conference presentations, abstracts and other meetings*

uthors	Title	Presentation Type	Conference /Event	Location	Year	Audience
Shirley, A.K.	Revealing and exploiting the diversity in dairy cattle reticulorumen temperature data for heat stress amelioration	Final PhD Presentation (oral)	SOLES HDR Showcase (USYD)	Sydney	**2025**	40
Chlingaryan A.	Artificial intelligence and heat stress	Oral presentation	DRF Symposium	Wollongong	**2025**	150
Shirley A.K.	Association between three phenotypic indicators of heat stress in dairy cattle	Oral presentation	PDF	Christchurch, New Zealand	**2025**	300
Chlingaryan A.	AI for cattle tracking and behaviour identification from video footage	Oral presentation	PDF	Christchurch, New Zealand	**2025**	300
Chlingaryan A.	AI for dairy cattle heat tolerance phenotype	Oral presentation	ADSA, Annual Meeting	Kentucky, USA	**2025**	1000
Azubuike et al	Leveraging artificial intelligence and evolutionary algorithms for optimising cow supplementation and milk production	Abstract/Oral presentation	ADSS	Christchurch, New Zealand	**2024**	150
Shirley A.K.	The diversity in dairy cattle reticulorumen temperature: Correlation with drinking event activity.	Oral presentation	ADSS	Christchurch, New Zealand	**2024**	150
Pearce M.F.	Dairy disease associations: predicting and preventing illness incidence and impact	Oral presentation	ADSS	Christchurch, New Zealand	**2024**	150
Azubuike B.N.	A Data-Driven Approach for Optimising Supplement Allocation to Individual Lactating Dairy Cows in Pasture-Based Systems	Poster presentation (Intermediate PhD)	SOLES HDR Showcase (USYD)	Sydney	**2024**	40
Shirley et al.	The diversity in dairy cattle reticulorumen temperature: Identifying water intake events	Abstract/Oral presentation	ECPLF	Bologna, Italy	**2024**	200

Chlingaryan et al.	An AI-based phenotype for dairy cattle heat tolerance	Abstract/Oral presentation	ECPLF	Bologna, Italy	**2024**	200
Shirley et al.	The diversity in dairy cattle reticulorumen temperature: Exploring the influence of temperature humidity index.	Abstract/Oral presentation	EAAP Annual Meeting	Florence, Italy	**2024**	300
Shirley et al.	The diversity in dairy cattle reticulorumen temperature: Identifying variability in drinking behaviour	Abstract/Oral presentation	WAAFL	Florence, Italy	**2024**	200
Shirley A.K.	Heat stress – findings and considerations for design.	Oral presentation	Raising the Roof	Sydney	**2024**	50
Shirley A.K.	Hot Cows, Cool Solutions	Oral presentation	Thesis competition (USYD)	Sydney	**2023**	60
Cameron C.E.F.	P6 – Resilient Cattle	Farmer meeting presentation	Dairy UP Roadshow	Bega and Cobargo	**2023**	30
Shirley A.K.	Development of a threshold model to isolate and investigate water intake in dairy cattle fitted with a reticuloruminal sensor.	Oral presentation	DRF Symposium	Camden	**2023**	150
Shirley et al.	An exploratory analysis of rumen temperature for three dairy herds in Australia: a potential path for GHG emission reduction	Abstract/Oral presentation	International Symposium on the Nutrition of Herbivores	Florianopolis, Brazil	**2023**	200
Shirley A.K.	Revealing and exploiting the diversity in dairy cattle core body temperature data for heat amelioration	Oral presentation (Introductory PhD)	SOLES HDR Showcase (USYD)	Sydney	**2023**	40
Cameron C.E.F.	Better milk and dairy beef markets	Oral presentation	DRF Symposium	Sydney	**2022**	100

***Table 4**. Technical reports, pamphlets, published material and other media engagements*

Authors	Title	Place Published	Year
Cameron C.E.F. & Chlingaryan A.	P6a Resilient cattle (heat tolerance)	Dairy UP Website	2026
Cameron C.E.F.	P6: Future Systems – Dairy DaTA	Dairy UP Website	2023

Final Project Summary

P6a Resilient Cattle (Heat Tolerance)

May 2026

P6a Resilient Cattle (heat tolerance)

Managing dairy cattle in hot, humid conditions is an increasing issue for the Australian dairy industry, with climate change and the trend towards intensification. Improving heat tolerance in dairy cattle has benefits for both the business (maintaining production and reproductive performance in hot months) and animal welfare (improved cow comfort in hot conditions).

Under the supervision of Dr Anna Chlingaryan, Dairy UP PhD student, Alice Shirley, explored the diversity in dairy cattle responses to heat events. By collaborating with DairyBio and DataGene, the outcomes of this work will contribute to developing an improved tool for breeding dairy cattle with improved heat tolerance.

Heat Tolerance ABV

Published by DataGene, the Heat Tolerance Australian Breeding Value allows farmers to identify and breed animals with greater ability to tolerate hot, humid conditions with less impact on milk production.

The current Heat Tolerance ABV is derived from genomics only – the DNA testing of animals. It has a relatively low reliability which has contributed to slow uptake by farmers.

Data, Advanced Technology and Automation (DATA)

Dairy UP's P6 project explored ways to use existing farm, climate and industry data to develop ways to monitor cows and systems. Reports and tools based on this data could be used by farmers to make better decisions, for example about heat management, health and feeding.

The Australian dairy industry collects large amounts of farm and production data. Until now this has been stored in numerous, separate databases.

This project aimed to utilise data, advanced technologies and automation to integrate information from multiple sources to enable the creation of tools that support on-farm decisions.

P6 was a suite of three projects that combine animal science and data science. Each project was undertaken by a PhD student.

- P6a: Resilient Cattle (heat tolerance).
- P6b: Resilient Cattle (health): early intervention for improved animal health, enabled by advanced sensing.
- P6c: Digital Feeding – data-driven feeding to optimise grain allocation in pasture-based herds.

This document provides a final overview of Project P6a Resilient Cattle (heat tolerance).

This Dairy UP project aimed to develop phenotypes for heat tolerance to add to the model for the Heat Tolerance ABV. Phenotypes are indicators of animal performance that can be measured in commercial animals. Combining genomics with traditional and new measures of heat tolerance should improve the reliability (accuracy) of the Heat Tolerance ABV.

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Collecting phenotypes with sensor technology (Dairy UP)

The Dairy UP team analysed data from innovative sensor technologies (core body temperature) through advanced statistical methods.

Data automatically downloaded via API were used to build animal-level datasets to improve our understanding about which animals are more susceptible to heat.

Data was collected from cows on three Australian pasture-based dairy farms fitted with rumen sensors (reticuloruminal boluses) to monitor core body temperature, every 10 minutes, 24 hours a day. A total of 1429 animals were involved in this research, plus 28 heifers from the University of Sydney farm in NSW.

In addition, climate data from the past 20 years was obtained from the Bureau of Meteorology to match the observations from the cows.

A water threshold model was developed to account for water intake, isolating the impact of drinking events on core body temperature.

Drinking behaviour

Analysis of the sensor data confirmed significant variation between individual animals' reticulorumen temperature (core body temperature) and their drinking behaviour over time. This work also demonstrated the core body temperature of cattle increased at lower Temperature and Humidity Index (THI) levels than previously thought.

For example, the rumen sensors were indicating a rise in core body temperature at 67 THI. Until now, it has been recommended that herd managers prepare for a heat stress event at 70 THI, with negative effects on production expected at 75 THI.

The research also confirmed that cows drink more often during summer and less in winter. As expected, cows in herds in warmer regions also returned to the trough to drink more times, on average, in a day compared to those in cooler climates.

Further analysis found that drinking behaviour can be used as a reliable indicator of heat stress.

Drinking was a consistent behavioural response to cope with internal and external heat. Those animals that drank more often during rising THI displayed a decline in reticuloruminal temperature. They were also able to maintain production under higher internal temperature. These findings open up new opportunities to integrate additional phenotypic indicators into the genomic-only model currently used to calculate the Heat Tolerance ABV, thereby improving its reliability.

Enhancing phenotype calculation (Dairy UP)

The Dairy UP team developed a hybrid artificial intelligence (AI)-based model (HAIM) to improve the assessment of heat tolerance in dairy cattle.

They used machine learning techniques and 20 years of historical climate and dairy cattle production data to enhance the heat tolerance phenotype calculation.

Traditional methods for determining heat tolerance in dairy cattle involve statistical models that use the rate of decline in milk yield as Temperature-Humidity Index rises above 60. Dairy UP's HAIM combines the predictive capabilities of machine learning algorithms with these established statistical models, allowing for the detection of intricate relationships within the extensive data.

The HAIM revealed patterns that might remain hidden when using traditional models alone, enhancing the understanding of heat tolerance in dairy cattle and the identification of more heat-tolerant animals.

The traditional statistical method and HAIM model overlapped in identifying 84% of heat tolerant animals, but the HAIM replaced about 8% of the cows that weren't identified with the statistical method, while accounting for a greater proportion of the total observed variability (i.e. increasing accuracy).

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Combining phenotypes and genotypes (DairyBio)

This collaborative project was undertaken by DairyBio and La Trobe University PhD student, Laura Jensen under the supervision of Professor Jennie Pryce.

This project combined the genetic and performance data from animals with a variety of approaches including the sensor technologies from the Dairy UP project.

This information may allow the model for the Heat Tolerance ABV to be extended to incorporate sensor phenotypes which could help to identify animals that better tolerate hot and humid conditions with less impact on their milk production.

Sensors are one example of new research methods that could improve how we select for heat tolerance, capturing its full complexity.

Success in this area will come from collaboration among animal scientists, combining genomics with traditional and new measures of heat tolerance.

Collaboration

Dairy UP
DairyBio *(Dairy Australia, Gardiner Foundation & Victorian Government)*
DataGene
Charles Sturt University
SmaXtec
University of Sydney

Journal articles

Chlingaryan et al. (2025) An AI-based hybrid model for dairy cattle heat tolerance phenotype. Smart Agriculture Today Volume 12, December 2025, 101455

Shirley et al. (2026) Probing the diversity in dairy cattle reticulorumen temperature for adaptation selection. *Nature Scientific Reports* (2026).

Shirley A. K. et al (2024) Review: Ruminant heat-stress terminology. Animal Volume 18, Issue 9.

Shirley A.K. et al (2025), The diversity in dairy cattle reticulorumen temperature: Identifying water intake events, Computers and Electronics in Agriculture, Volume 235, 2025, 110357

Pryce Jennie E et al (2022) Impact of hot weather on animal performance and genetic strategies to minimise the effect. Animal Production Science 62, 726-735

More info

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P6 final report

Delivery organisations

The University of Sydney – Dairy Research Foundation | NSW Department of Primary Industries and Regional Development | Scibus | Dairy Australia

Partner organisations

NSW Farmers Growing the best | eastAUSmilk | Dairy NSW | Dairy Australia | AFMH | Leppington Pastoral Company Pty Ltd | NSW Local Land Services | Norco

Additional program supporters, collaborations or partnerships

Charles Sturt University | DairyBio | DataGene | Eagle Direct | Entegra
Macquarie University | NSW EPA | smaXtec | UC Davis | University of Technology Sydney

Project Update
P6b: Resilient Cattle (Health)

May 2026

New opportunities from sensor data

The number of dairy cattle managed in confinement systems in Australia is increasing, with the trend towards intensification.

Cattle housed indoor can be fitted with sensors (in and on the animal) to monitor behaviour in near-real time.

These systems can continuously record information such as activity, rumination, feeding behaviour and temperature.

This project explored opportunities to combine sensor data with farm health and production records to better understand patterns of disease and support future development of early-intervention tools.

Research approach

The Dairy UP team established a data partnership with Moxey Farms, a large commercial dairy herd in NSW. The Dairy UP team was given access to production and management records from the farm.

The project established a linked dataset combining CowManager sensor data and DataGene records, creating a pipeline to bring together farm health, production and management information that had previously been

Data, Advanced Technology and Automation (DATA)

Dairy UP's P6 project explored ways to use existing farm, climate and industry data to develop ways to monitor cows and systems. Reports and tools based on this data could be used by farmers to make better decisions, for example about heat management, health and feeding.

The Australian dairy industry collects large amounts of farm and production data. Until now this has been stored in numerous, separate databases.

This project aimed to utilise data, advanced technologies and automation to integrate information from multiple sources to enable the creation of tools that support on-farm decisions.

P6 was a suite of three projects that combined animal science and data science. Each project was undertaken by a PhD student.

- P6a: Resilient Cattle (heat tolerance).
- P6b: Resilient Cattle (health): early intervention for improved animal health, enabled by advanced sensing.
- P6c: Digital Feeding – data-driven feeding to optimise grain allocation in pasture-based herds.

This document provides a final update on P6b Resilient Cattle (Health).

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stored in separate systems.

Integrating these previously separate data sources created a research-ready dataset and a proof-of-concept that could support future research and decision support tools.

Results

Analysis of records from more than 3100 cows confirmed that the period immediately after calving was the highest-risk time for health events, with 40% of recorded cases occurring within the first seven days after calving.

The study also found that different health disorders followed different timing patterns after calving.

Subproject activities were delayed due to health-related interruptions to the PhD candidature. However, the project established a valuable research dataset that provides a foundation for future work developing early-intervention tools for dairy cattle health.

PhD student

Maddi Pearce, University of Sydney

Collaborators

Dairy UP participating commercial farms

Read more

Pearce M.F. — *Dairy disease associations: predicting and preventing illness incidence and impact* — Abstract from oral presentation — ADSS 2024.

More info

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P6 final report

Delivery organisations

The University of Sydney – Dairy Research Foundation
NSW Government – Department of Primary Industries and Regional Development
Scibus
Dairy Australia

Partner organisations

NSW Farmers – Growing the best
eastAUSmilk – Advocacy · Unity · Service
Dairy NSW
Dairy Australia
AFMH
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An AI-based hybrid model for dairy cattle heat tolerance phenotype

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ARTICLE INFO

Keywords:
Cattle heat stress
Individual cattle level
Extreme weather events
Machine learning
Milk production

ABSTRACT

Extreme weather events pose substantial risks to dairy cattle production and welfare, with heat stress being a major disruptor. Current approaches for heat tolerance phenotype selection in dairy cattle primarily utilise statistical models. We propose a novel hybrid AI-based method (HAIM) to enhance the selection of heat-tolerant cattle. It integrates AI with a statistical genetics model that links heat tolerance to decline in milk yield as the Temperature-Humidity Index (THI) rises above 60. This decline is modelled at both herd and individual levels, with the individual decline per unit increase in THI represented by the random slope and the intercept providing the baseline.

For this modelling, 20 years of climate data were combined with New South Wales Holstein dairy cow production data, covering the same period and resulting in 3 million records. The HAIM model demonstrated a smaller standard deviation of residuals (3.25 L/cow/test day) compared to the statistical model (3.37 L/cow/test day). A high correlation ($R^2 = 0.93$) between the random slopes estimated by both models indicated similar heat tolerance estimates, with the AI-based model showing lower standard deviation of residuals. A negative association between estimated random intercepts and slopes highlighted that high-producing animals are more vulnerable to heat stress. These findings emphasise the importance of considering both traits (random slopes and intercepts) to improve heat tolerance selection without compromising overall production levels. The proposed model combines the strengths of statistical modelling with the capabilities of AI, resulting in a more accurate and efficient means of identifying heat-tolerant cattle.

1. Introduction

The global demand for dairy production is surging due to population growth [7]. Nevertheless, climate change and extreme weather events pose substantial risks to dairy cattle production and welfare with heat stress being a major disruptor [27]. Heat stress, caused by high temperatures and humidity, can significantly reduce milk yield, fertility, and overall well-being of cattle [3,26,29,35]. Such changes in climate are particularly pertinent for the Australian dairy industry, as the country encounters some of the world's most elevated temperatures and climate variability, while simultaneously being a significant exporter of dairy products [48].

Although the adoption of efficient management practices can substantially reduce the effects of heat stress, advancements in genomics offer further solutions. Techniques such as genomic selection and gene editing now allow breeders to pinpoint animals with superior genetics at a younger age [50]. Genomic selection is a breeding technique that has been increasingly adopted in the dairy cattle industry [23,51]. This has enabled the creation of new breeding values that utilise phenotypes from specific reference populations of genotyped cows. Egger-Danner et al. [20] have examined prospective candidate traits for new breeding values, focusing on expected health and fitness traits. In this regard, heat tolerance in dairy cattle could be improved using genomic selection, with the development and application of genomic breeding values for heat tolerance which have been predicted to reduce the decline in milk production and reduce increases in core body temperature [21]. Recently, Australian researchers have made significant strides in this area, demonstrating the first attempt to incorporate heat tolerance (HT) into selection indices for dairy cattle [32]. Furthermore, they introduced a new innovative trait: HT genomic breeding values [31], which has been incorporated into the national selection indices of Australia [36].

The development of the HT breading values, as presented by Nguyen et al. [33]; Nguyen et al. [31], is based on prior research that employed temperature-humidity indices (THI) as an indicator of heat load and

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The diversity in dairy cattle reticulorumen temperature: Identifying water intake events

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ARTICLE INFO

Keywords:
Heat stress
Drinking behaviour
Sensors
Threshold algorithm
Precision agriculture

ABSTRACT

Climate change and associated weather variability across the Australian landscape has lent themselves to an increased incidence of cattle heat stress. Water consumption can have a sizeable, sustained impact on reticulorumen temperature readings, thereby impacting our interpretation of an individual's underlying physiological response to changing environmental conditions. To distinguish drinking events, we developed a drinking event detection model based on observed drinking events (video recording) from 28 dairy heifers, alongside sensor-derived reticulorumen temperature (smaXtec Animal Care GmbH) profiles. The optimised model identified drinking events with high accuracy (F-score = 0.99), as predicted when the average reticulorumen temperature declined by at least 0.5°C per 10-minutes, over a 10-, 20-, or 30-minute period. To account for differences in rapidity of decline, smaller reductions of 0.25°C per 10 min were considered valid indicators of a drinking event, provided the 0.5°C per 10-minute threshold was also met in a consecutive observation period. The temporal variability in drinking behaviour for 1,429 lactating dairy cattle across three dairy farms was then determined. Daily drinking events were greater in summer (mean 4.1) than winter (mean 3.3), while the change in reticulorumen temperature with each drinking event was smaller in summer (mean 3.7°C) than winter (mean 4.9°C). Drinking-recovery duration averaged 97.8 min/event. By revealing temporal differences in drinking behaviour for pasture-based dairy cattle, this work provides the basis for an improved understanding of core body temperature diversity.

1. Introduction

Dairy farming in Australia is primarily pasture-based and occurs across a wide range of environments (Cheruiyot et al., 2019). As such, Australian dairy cattle are increasingly exposed to climate variability and with more frequent, intense heat events the associated risk of cattle heat stress (HS) also increases (Blunden and Boyer, 2022; Cowley et al., 2015). Climate change is of particular importance for high-yielding dairy cows as they already experience an elevated internal heat load compared to lower producing individuals (Pryce et al., 2022), and as a result, the impact of heat accumulation is exacerbated for these cows (Bernabucci et al., 2015; West, 2003). As a response to escalating global temperatures, there is an increasing need to understand the role of climate factors and their impact on the prediction and assessment of HS for dairy cattle (Yan et al., 2020). Several thermal indices are used by livestock industries (Wang et al., 2018a) including the temperature humidity index (Mader et al., 2006), heat load index (Gaughan et al., 2008), comprehensive climate index (Mader et al., 2010), index of thermal stress for cows (Da Silva et al., 2015), dairy heat load index (Lees et al., 2018), and equivalent temperature index for cattle (Wang et al., 2018b), among others. However, the success of a thermal index is intimately linked with its ability to predict animal responses to heat (Hahn et al., 2009). As most thermal indices predictions are at a herd-level, methods to determine individual animal responses to HS are required.

An animal's primary response to a challenging thermal environment is the alteration of its physiology and/or behaviour (Ji et al., 2020; Polsky and von Keyserlingk, 2017; West, 2003). Cattle response to HS can appear in a variety of forms, however each response aims to reduce metabolic heat production and enhance heat dissipation into the environment (Islam et al., 2021; West, 2003). Due to advances in precision livestock farming, the use of objective technology is enabling

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Review: Ruminant heat-stress terminology

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ABSTRACT

With increasing climate variability, there is a rise in the exposure to, and incidence of, ruminant heat stress (**HS**), increasing the requirement for focused research. As such, precise terminology is crucial to maintain effective communication and knowledge advancement. Despite this, several key terms are currently defined inconsistently, leading to confusion and misinterpretation. This paper examines the historical and contemporary use of the terms 'resistance', 'tolerance', 'resilience', and 'susceptibility' across various disciplines, revealing significant ambiguities that hinder both research and practice. Through this comprehensive review, we propose new definitions for each term as they are used relating to HS, with a focus on ruminant production. Proposed definitions align with current scientific understanding, providing a robust framework for future research and application. As further research is conducted, we hope these definitions can be improved through the inclusion of quantitative measures which align with these classifications. This present review provides definition clarity for common heat abatement terminology, enabling consistency and from this, progress in the field to ameliorate HS for ruminants.

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Implications

The exposure of livestock to climate variability and the associated risks of heat stress is increasing, warranting the development of heat abatement techniques. Whilst various mitigation strategies have been identified, poor clarity and consistency across heat tolerance terminology remain. This review provides definition clarity for common heat abatement terminology, enabling consistency and from this, progress in the field to ameliorate heat stress.

Introduction

Ruminant production in Australia occurs in a wide range of environments, with most livestock exposed to climate variability and the associated risk of heat stress (**HS**) (Cowley et al., 2015; Cheruiyot et al., 2019). Alongside these climate changes has been production intensification, compounding the impact of HS worldwide (Renaudeau et al., 2012; Polsky and von Keyserlingk, 2017; Islam et al., 2021). High-producing animals are particularly vulnerable to HS due to their greater metabolic rates and energy requirements as compared to lower−producing contemporaries (Ravagnolo and Misztal, 2000; Collier et al., 2019). With global projections requiring milk production to double in the next five decades (Britt et al., 2018) and the World Health Organization recognising a notable rise in the demand for animal-derived protein in all regions (Henchion et al., 2021), investigation of heat abatement techniques is vital to minimise and ameliorate heat stress and with this, maintain or enhance animal welfare standards and production outputs.

To ameliorate HS in ruminants, there are three management strategies: genetic development of resistant breeds, nutritional management, and physical modification of the environment (Beede and Collier, 1986; Johnson, 2018). Irrespective of strategy, the identification of an individual animal's ability to cope under HS conditions is essential for effective implementation. Despite this, there is poor clarity across heat tolerance terminology, leading to ambiguity. Ambiguity with regard to HS terminology is problematic due to resulting confusion and miscommunication among researchers, disciplines, and industries. For instance, the terms 'resistance', 'tolerance', 'resilience', and 'susceptibility' are used interchangeably despite their distinct meanings.

Resistance is typically viewed as an innate ability that enables an organism to defend itself from disease agents (Best et al., 2008; Schneider and Ayres, 2008), though variability in the classification of this term remains. Tolerance is primarily associated with a degree of endurance, defined by the slope of a reaction norm (West-Eberhard, 2008), but has other interpretations. Definitions of resilience primarily account for an encapsulation of change and ability to recover at speed (Gunderson and Holling, 2002;

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Final Project Summary

P6c: Future Systems – Digital Feeding

May 2026

Feed costs are one of the biggest expenses on Australian dairy farms, so improving the efficiency of supplementary feeding has a direct impact on farm profitability. Accurate individual grain allocation offers the opportunity for many pasture-based herds to improve milk response to supplementation.

This project investigated how existing on-farm data and climate data could be used to support more precise, individualised grain allocation decisions for pasture-based herds with automatic feeders. It built on earlier work by the University of Sydney that identified the potential to increase milk production by 10% through improved concentrate allocation to individual cows.

Digital feeding systems

The project explored how artificial intelligence (AI), machine learning and optimisation algorithms could be used to predict milk yield from routinely collected farm data and adjust grain allocation accordingly.

The work focused on developing a "digital feeding" approach that could operate within practical farm constraints and existing in-parlour feeding systems. The models tested whether overall milk production could be improved through more advanced individualised feeding based on each cow's predicted response to different feeding levels.

Data, Advanced Technology and Automation (DATA)

Dairy UP's P6 project explored ways to use existing farm, climate and industry data to develop ways to monitor cows and systems. Reports and tools based on this data could be used by farmers to make better decisions, for example about heat management, health and feeding.

The Australian dairy industry collects large amounts of farm and production data. Until now this has been stored in numerous, separate databases.

This project aimed to utilise data, advanced technologies and automation to integrate information from multiple sources to enable the creation of tools that support on-farm decisions.

P6 was a suite of three projects that combined animal science and data science. Each project was undertaken by a PhD student.

- P6a: Resilient Cattle (heat tolerance).
- P6b: Resilient Cattle (health): early intervention for improved animal health, enabled by advanced sensing.
- P6c: Digital Feeding – data-driven feeding to optimise grain allocation in pasture-based herds.

This document provides a final update on P6c Digital Feeding.

Key findings

Our modelling predicted that herd-level milk production could increase by 6.6–8.8% when grain allocation was optimised using AI-predicted individual cow response to feeding.

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Cows differed widely in how efficiently they converted grain into milk. One kg of grain has energy for up to about 2.4 L of milk. However, due to variations in the intake and composition of other feeds, as well as differences in feed conversion efficiency, the 'apparent' total conversion efficiency varies widely. In our studies, some cows produced only 0.17 L/kg, while others produced up to 15.8 L/kg.

This highlights the importance of accounting for cow-to-cow variability when making feeding decisions.

Machine learning models reliably predicted milk yield using routinely collected farm data, without requiring liveweight measurements or additional climate-monitoring equipment.

Implications for farmers

The findings suggest there are opportunities to improve feed efficiency by directing more grain towards cows that respond most effectively.

Artificial intelligence has the potential to enhance existing individualised feeding systems by predicting how individual cows are likely to respond to different feeding levels.

More info

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P6 final report

Research and results

Research approach

The project used existing commercial farm data to test whether artificial intelligence and optimisation tools could improve grain allocation decisions in pasture-based dairy herds.

Researchers analysed large datasets from two commercial pasture-based dairy farms, including milk yield, concentrate intake, days in milk, lactation number, breed and weather conditions.

Study 1 used 32,504 records from 130 cows to develop machine learning models for predicting milk yield. Digital feeding strategies were tested using 7,371 of these records from 81 cows.

Study 2 used 456,078 daily records from 1,053 cows to investigate variation in cow response to grain feeding and evaluate digital feeding optimisation strategies using the developed machine learning models.

Optimisation algorithms were then used to test whether redistributing the same total amount of grain across the herd could improve overall milk production while remaining within practical farm feeding limits, such as minimum and maximum concentrate levels and restrictions on day-to-day changes in feeding.

Results

The project demonstrated that redistributing grain according to predicted individual cow response could improve feed-use efficiency and herd-level milk production without increasing overall grain use.

The research highlighted major differences between cows in how efficiently they converted grain into milk. Without considering other diet components, milk produced per kilogram of concentrate ranged from 0.17 to 15.8 litres, highlighting the importance of accounting for cow-to-cow variability when making feeding decisions.

Modelling predicted herd-level milk production could increase by 6.6–8.8% when grain allocation was based on AI-predicted individual cow response to feeding. The best-performing optimisation approach consistently predicted increases of around 8.6%.

The optimisation models redistributed grain from lower-responding cows to cows predicted to respond more efficiently to concentrate feeding, while remaining within practical farm feeding limits.

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Machine learning models reliably predicted milk yield using routinely collected farm data, including milk yield, concentrate intake, days in milk, lactation number and weather conditions. Days in milk and concentrate intake were identified as the strongest predictors of milk production. The models explained about 61% of the variation in daily milk yield when tested on separate farm records not used to develop the models.

Environmental variables such as temperature and humidity added only small improvements to prediction accuracy compared with cow-level production and feeding data. This suggests digital feeding systems may be able to be implemented using data already collected on many commercial farms, without requiring additional climate-monitoring equipment or liveweight measurements.

Further research

The project was a modelling and simulation study using commercial herd datasets. The next step would be to validate the performance of digital feeding systems under real-time farm conditions.

PhD Student

Blessing Azubuike, University of Sydney

Collaborators

Participating dairy farms: Ryan Apps, Bega NSW; University of Sydney Corstorphine farm.

Journal articles

Azubuike et al. (2026) Leveraging artificial intelligence and evolutionary algorithms for optimising cow supplementation and milk production. Artificial Intelligence in Agriculture,Volume 16, Issue 2, Pages 926-939.

Azubuike et al. (2025) A data-driven approach for optimising supplement allocation to individual lactating dairy cows in pasture-based systems, Smart Agricultural Technology, Volume 12,101669

Delivery organisations

The University of Sydney – Dairy Research Foundation
NSW Government Department of Primary Industries and Regional Development
Scibus
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Research Paper

Leveraging artificial intelligence and evolutionary algorithms for optimising cow supplementation and milk production

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ABSTRACT

Efficient allocation of grain-based concentrate is essential for maximising milk yield and improving profitability in dairy farming. This study optimised concentrate allocation for dairy cows by integrating machine learning with evolutionary algorithms to enhance milk yield. Data from 1105 cows were analysed to model milk yield using predictor variables including breed, days in milk, stage of lactation, lactation number, daily concentrate allocation and climatic factors. For optimisation, records for 165 cows within a 30-day window were identified using a Genetic Algorithm. Three machine learning models (Gradient Boosting Machines, Extreme Gradient Boosting and Random Forest) were evaluated, with GBM showing superior predictive accuracy ($R^2 = 0.75$ training, 0.61 testing; RMSE = 3.02 L/cow·day^{-1} training, 3.47 L/cow·day^{-1} testing). The pre-trained GBM was integrated into four evolutionary algorithms (NSGA-II, SPEA-II, SMS-EMOA and RVEA) to simultaneously maximise herd-level daily milk yield and minimise deviations in daily concentrate allocation. Concentrate levels per cow were constrained between 6 and 11 kg per day, with adaptive bounds allowing no more than a 2 kg change per cow per day from the previous allocation. Without increasing herd-level supplementation allowance, statistical analysis based on 10 independent runs per algorithm revealed NSGA-II achieved the highest mean milk yield increase of 8.64 ± 0.10%, significantly outperforming SPEA-II (7.94 ± 0.13%), RVEA (7.41 ± 0.10%), and SMS-EMOA (6.63 ± 0.24%). One-way ANOVA confirmed significant differences among all algorithms ($F = 303.24$, $p < 0.001$), with NSGA-II also demonstrating superior computational efficiency (74.9 ± 0.7 s per day). These findings demonstrate the potential of combining machine learning and evolutionary algorithms to optimise feed use efficiency, highlighting promising practical applications of individualised feeding approaches to enhance milk production in dairy systems.

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1. Introduction

Efficient feed allocation is pivotal in dairy farming, directly influencing profitability and milk production. In pasture-based dairy systems, grain-based concentrates, rich in energy, are commonly used to enhance milk yield (MY) and supplement pasture intake; however, they significantly contribute to operational costs, often comprising up to 60% of total production expenses (Craig et al., 2022; Uyeh et al., 2018). Traditional feeding practices in these systems typically employ a flat-rate approach, distributing concentrates uniformly across the herd without accounting for individual cow variations such as breed, lactation stage, days in milk (DIM), and historical production performance. This homogenised method can lead to feed inefficiencies and inconsistent MY, ultimately affecting the economic viability of the farm (Dela Rue and Eastwood, 2017; Hills et al., 2015).

Optimising feed allocation remains a complex challenge despite advancements in agricultural technology. Current strategies often fail to utilise the extensive data generated on modern farms (Cockburn, 2020) or adapt to the dynamic nutritional requirements of individual cows (Dela Rue and Eastwood, 2017; Hills et al., 2015). This results in suboptimal feeding regimes that do not fully exploit the potential of each cow for milk production, leading to underutilisation of resources and limited productivity gains. Innovative approaches are needed to process complex datasets, informing feeding decisions and tailoring concentrate allocation to maximise MY while maintaining cost-effectiveness (Brady et al., 2022; Dale et al., 2016; Henriksen et al., 2019).

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P7
Dairy businesses for future climates

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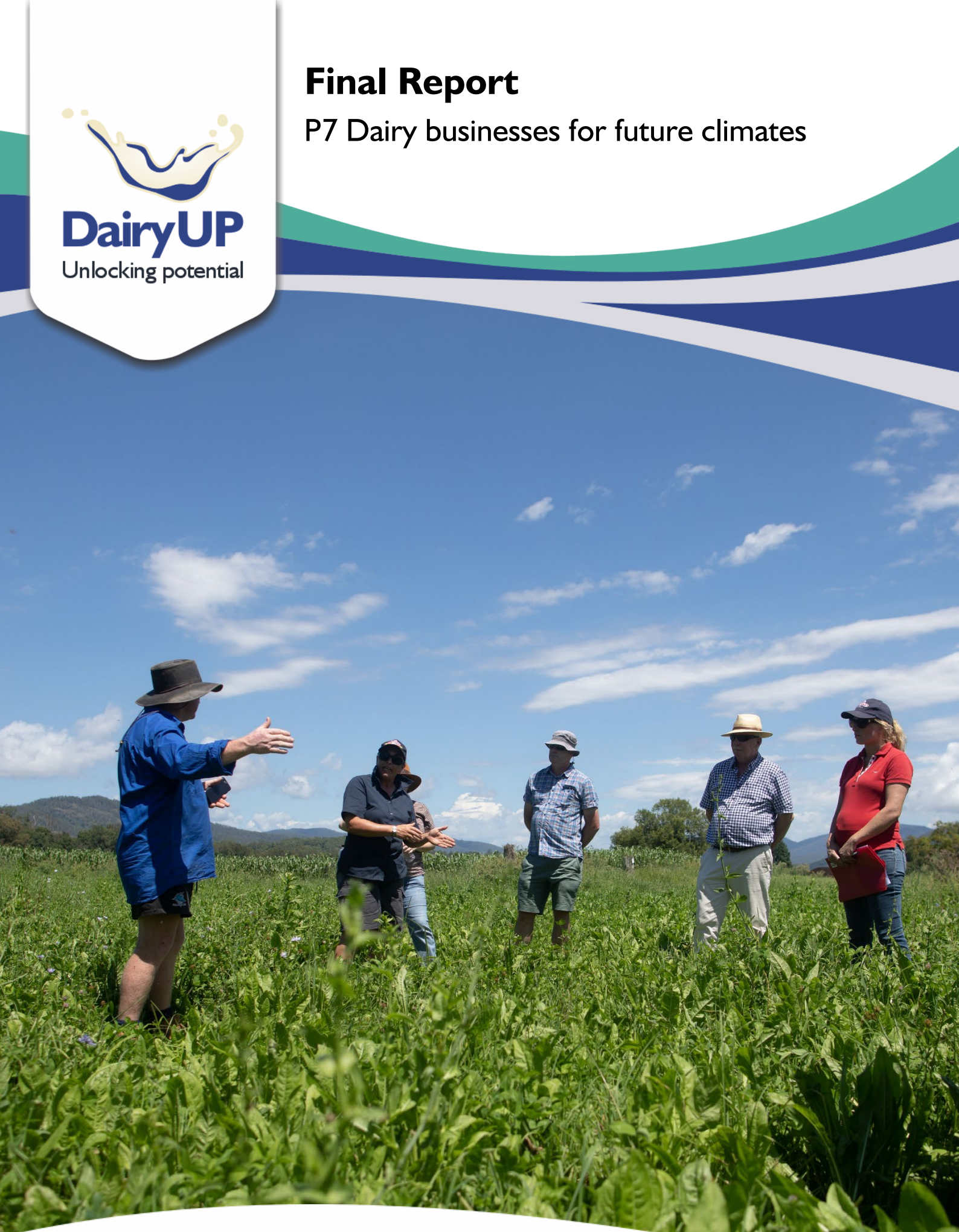
P7 Dairy businesses for future climates



Department of Primary Industries
and Regional Development

This project was led by Zita Ritchie,
formerly of NSW DPIRD

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI (Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

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Project Overview

This project was originally developed by Dairy Australia and led in NSW by DPIRD. Dairy UP enhanced its delivery for the benefit of the NSW and broader dairy industry. The project was completed in June 2022 and the final report/project summary is included here.

Item	Description
Project Title	Dairy businesses for future climates
Funding Body	Dairy UP
Dairy UP Project	P7
Project Duration	2021-2022
Lead Organisation	NSW Department of Primary Industries and Regional Development
Project Lead	Zita Ritchie, NSW DPIRD
Research team	Ashley Black, Brendan Cullen, Dan Armstrong, Paul van Wel
Key Collaborators	Dairy Australia, Dairy NSW, NSW Department of Primary Industries and Regional Development, University of Melbourne

DAIRY BUSINESSES FOR FUTURE CLIMATES

MID-NORTH COAST NEW SOUTH WALES

KEY FINDINGS

Options to manage climate impacts in 2040 were analysed for a case study farm on mid-north coast of NSW.

The feedbase (mix of temperate and tropical pastures and crops), was quite resilient to the changed climate.

Heat stress on cows will be a key issue in the 2040 climate for this farming region.

Milk production was predicted to decline by 3-4% by 2040.

If no changes are made to the case study farm, annual operating profit is predicted to decrease by about 20% under 2040 climate.

None of the development options analysed were clearly superior to the others. All have positive and negative features.

Profitability of all options analysed were negatively affected by the 2040 climate change scenarios.

The Adapt option (shade and feedpad) enables better heat stress management in 2040 with moderate capital investment.

Dairy farm managers will need to continue to adapt their farm systems to manage risks.

Aim of the case study research

To explore how dairy farm systems in the mid-north coast region of New South Wales (near Kempsey) might perform under predicted climate changes (out to 2040) and how they could adapt to a changing climate.

A dairy farm near Kempsey was selected as a case study farm, supported by a reference group of farmers and service providers. Three development options for the base farm (case study farm) were modelled in a 2040 climate by an economist and biophysical modellers.

Changes in 2040

- Temperatures will increase by 1.0°C (from 1990-2009 base period).
- Heat stress is predicted to increase in 2040, and milk production to decline by 3-4% for the base farm.
- Predicted to be little change in total annual rainfall but with a 5% decline in winter, 10% increase in autumn and little change to summer rainfall.
- Fewer, larger rainfall events are expected, with longer dry spells in between.
- Pasture feedbase is quite resilient to the predicted climate changes in 2040 (and more so than in southern Australia where climate change is predicted to lead to a contraction of the pasture growing season, lower pasture production and grazed intake).
- Monthly average pasture growth rates were predicted to increase in autumn and winter but decline slightly in late spring.

Base farm:
• 230 milking cows • Year-round calving (except summer months) • Grain feeding 2 t/cow per year • Production of 535 kg milk solids/cow per year • Feedbase mix of temperate and tropical pastures and crops.

Three development options explored:

Option 1: Feedlot	Option 2: Adapt	Option 3: Simplify
• 300 milking cows with an increase in milk solids/cow • Freestall barn, no grazing • Invest in a barn, effluent system and machinery • Year-round calving • 3 t concentrate/cow per year.	• 230 milking cows (same as base farm) • Shade infrastructure and concrete feedpad • Cover yard at the dairy • Increase cow intake, production per cow and less wastage • Keep feedbase and operating costs similar.	• Decrease to 180 milking cows • No capital investment in infrastructure • Split calving pattern to 66% in autumn and 33% in spring • 1 t concentrate/cow per year • Feedbase changed to more perennial pasture to fit calving pattern.

What differences did we find between the options?

The predicted profitability of the base dairy farm business and all three development options was lower than current under the 2040 climate change scenarios modelled.

None of the development options analysed were clearly superior to the others. All options have positive and negative features and the profitability of all are sensitive to milk price. The Adapt option appears to enable better heat stress management in the predicted 2040 climate without such substantial capital investment as the Feedlot option. The most suitable option will depend on resources available to the business and the long-term plans and goals of the people involved.

In the Base farm and Simplify option, where no additional infrastructure was provided, milk production was predicted to decline by 3-4%. But for the Adapt and Feedlot options, with shade infrastructure, the reductions in milk production were predicted to be 1% and 1.5% respectively, with the high-producing feedlot-cows being more susceptible to heat stress.

Feedlot option

- Higher variability in profit/risk than the other options and predicted profit would not justify the additional risk if the average milk price was $8.00/kg milk solids or less.
- Higher exposure to supplementary feed prices means this option is impacted by dry periods (with high imported feed prices).
- A high and stable milk price makes this option a more attractive investment.
- Obtaining planning approval for a feedlot is likely to be difficult in this location.
- Higher initial equity is important for this option.

Adapt option

- The Adapt option was least impacted by 2040 climate due to better infrastructure to manage heat stress with lower capital investment than the Feedlot option.
- Reasonable strategy to mitigate heat stress with moderate capital investment.
- Little difference between the profitability of the Base Farm and the Adapt option in the Historic climate.

Simplify option

- While the % return is comparable to the Base Farm, the annual operating profit is smaller which would impact on ability to service debt and grow the business.
- Other risks are reduced.
- Performs relatively well in dry periods with high supplementary feed prices due to lower exposure to the supplementary feed market.

This case study provides some indicative data for the region, but care needs to be taken about making regional generalisations from individual case study farms. For this region, most of the impact stems from increased heat stress impacting milk production. Other regions around Australia, where case studies have been conducted, are more affected by reduced pasture/crop production.

Successful implementation of all options will be heavily dependent on excellent management skills.

In addition to 2040 climate impacts, 'one-off' events such as a large flood or bushfire can be very costly to farm businesses but are difficult to represent in the modelling and should be considered where possible in future planning decisions.

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FINAL OUTPUT JOURNAL

P8 Farmer capability

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Final Report

P8 Farmer Capability

Dairy Australia

This project was led by Paul van Wel,
Dairy Australia

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

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This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

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Project Overview

This project was originally developed and led by Dairy Australia. Dairy UP enhanced its delivery for the benefit of the NSW and broader dairy industry. The project was completed in June 2023 and the final report/project summary is included here.

Item	Description
Project Title	Farmer capability
Funding Body	Dairy UP
Dairy UP Project	P8
Project Duration	2021-2023
Lead Organisation	Dairy NSW
Project Lead	Paul van Wel, Dairy NSW
Key Collaborators	Dairy Australia, The University of Sydney

Final Project Summary
P8: Farmer Capability

May 2026

Australian dairy businesses face ongoing challenges in sourcing and keeping people with the skills needed to work on farms.

The issues existed before Covid but escalated during and after it. Dairy Australia research found that 58% of dairy workers who resign move to a role outside the dairy industry. It identified issues around both attracting and retaining people working in the industry.

This project aimed to enhance retention by building the skills and capabilities of the current and next generation of dairy farmers, both employers and employees.

It delivered a series of one-on-one consultations and developed a suite of resources.

One-on-one consultations

A one-on-one consultation package was developed and tested for upskilling farmers in HR and people management. It involved on-farm visits by a specialist HR consultant to provide guidance to improve HR management and skills, particularly in compliance and safety.

Farmer capability

Dairy UP's P8 set out to help NSW dairy farm businesses and their workforce develop their capability in compliance and management.

Completed in June 2023, this was a collaborative project with Dairy Australia, with NSW activities delivered by Dairy NSW. Dairy Australia is continuing to expand its resources and delivery activities in this area, as part of its People in Dairy portfolio.

This document provides a summary of the work delivered for NSW under the Dairy UP banner.

Seventeen NSW farmers participated in one-on-one consultations through the project. Upon completion, they reported increased reported confidence to manage HR issues (rising from 6.1 to 8.2 out of 10).

Resources

The project developed/updated a suite of training resources for farmers focussing on human resources (HR), safety and developing people capability.

These resources have been pilot tested, refined and incorporated into Dairy Australia's online training platform (Enlight) - which is available to all Australian dairy farmers.

The project created the content for 'Building your farm system' which covers chemical handling, quad bikes, introduction to farm safety and confined spaces.

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The 'Managing People' course was completed and uploaded to Enlight following two online pilots. Farmer feedback is that it helps them develop valuable skills such as consideration of different learning styles thus creating more efficient and engaged workplaces contributing to increased retention.

The Employment Basics course was finalised following a lengthy review including survey of past participants and two pilots. The course directly supports compliance with IR legislation thus encouraging employee attraction and retention.

Next steps

DA has since made significant investment through its Workforce attraction project that funded delivery of these programs in NSW.

Collaborators

Dairy UP, Dairy Australia

Read more

Dairy Australia's Farmer Capability Guide

Dairy Australia Employment Basics training package

Dairy Australia Managing People training package

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The University of Sydney Dairy Research Foundation
NSW Government Department of Primary Industries and Regional Development
Scibus
Dairy Australia

Partner organisations

NSW Farmers Growing the best
eastAUSmilk
Dairy NSW
Dairy Australia
AFMH
Leppington Pastoral Company Pty Ltd
NSW Government Local Land Services
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FINAL OUTPUT JOURNAL

P9
Designer milk

P9a - Producing milk with less lactose
P9b - Milk as an indicator of heat load
P9c - Adding value to dairy waste

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Final Report

P9 Designer Milk

NSW GOVERNMENT
Department of Primary Industries and Regional Development

This project was led by Juan Gargiulo from the Department of Primary Industries and Regional Development

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1. Executive Summary

The Australian dairy industry is under growing pressure to produce more efficiently, manage climate risk, and reduce waste. The P9 "Designer Milk" project was funded under the Dairy UP program to address three specific gaps across the supply chain, each with direct relevance to Australian producers and processors.

P9a — Less lactose, less water, more milk solids: Milk is approximately 87% water, and it is the sugar lactose that draws most of that water into the udder. Cows that synthesise less lactose while maintaining equivalent milk solids output (fat + protein), can effectively produce more valuable milk per litre — reducing transport, processing costs, and resource use. Despite this potential, no large-scale Australian dataset existed to determine whether such cows exist in meaningful numbers or whether the trait could be selected for. Analysis of over 30,000 NSW cows identified a group of 80 individuals that consistently produced equivalent milk solids output with around 15% less lactose and water volume. Heritability estimates suggest the trait has a meaningful genetic component, indicating that selection is feasible. The potential scale of impact is substantial: a 1% reduction in lactose across the national herd could reduce milk transport volumes by around 80 million litres annually — saving an estimated 2,700 truck trips and 20,000 litres of water per farm per year. Though the underlying molecular mechanisms remain to be confirmed, the work establishes an evidence-based foundation for a genetic selection program.

P9b — Milk as an indicator of heat load: Heat stress (HS) costs the Australian dairy industry millions of dollars each year in lost production, poorer reproduction, and compromised animal welfare. Yet most monitoring approaches rely on environmental measures that do not directly reflect what is happening inside the animal. This subproject evaluated heat shock protein 70 (HSP70), a stress-response protein naturally present in milk, as a non-invasive indicator of cellular HS measurable during routine herd testing. Reliable, low-cost measurement methods were validated, and sample handling requirements were established. A key finding is that milk HSP70 responds to HS with a delay of approximately 33–44 hours, meaning it reflects prior or cumulative cellular stress rather than current conditions. During sustained heatwave conditions it showed strong diagnostic performance, though its discriminatory power was more limited outside extreme heat events and may be influenced by health or physiological status. Further validation across herds and seasons is needed, but the work positions milk HSP70 as a promising complementary tool within a broader HS monitoring framework.

P9c — Adding value to dairy food waste (DFW): More than 1,300,000 tonnes or ~16% of milk produced in Australia is lost across the supply chain as food waste, representing a significant economic and environmental cost to the industry. While the potential to convert DFW into value-added products through microbial fermentation has been demonstrated internationally, the waste streams most amenable to this approach in the Australian context, the biological pathways best suited to them, and the most commercially relevant bioproducts had not been systematically evaluated. This subproject addressed those gaps by mapping Australian DFW streams and identifying manufacturing by-products — particularly cheese whey and whey permeate — as strong candidates for scalable bioproduction, with microbial biomass and derived products representing the most immediate valorisation opportunities. As a proof of concept, a food-grade yeast strain was successfully engineered to grow on lactose as its sole carbon source and produce pro-vitamin A, demonstrating that the biological pathway is viable. Significant technical and economic development is required before commercial application; however, the work establishes a clear foundation and a defined set of next steps toward scalable bioproduction from Australian DFW.

Collectively, the Designer Milk project generated 6 peer-reviewed publications in high-impact international journals, one PhD thesis and one Honours thesis. The project reached audiences of over 2,700 people across more than ten conference presentations in Australia and internationally, and engaged the broader industry through podcasts, media articles and farmer meetings. The three subprojects offer evidence-based, interconnected pathways for improving dairy productivity, animal welfare, and environmental sustainability — and together provide a sound basis for continued investment in applied dairy research in NSW.

2. Project Overview

Item	Description
Project Title	Designer Milk
Funding Body	Dairy UP
Dairy UP Project	P9
Project Duration	2022-2026
Lead Organisation	NSW Department of Primary Industries and Regional Development (NSW DPIRD)
Project Lead	Dr. Juan Gargiulo
Key Collaborators	The University of Sydney (USYD); Macquarie University (MCU); University of California, Davis (UC Davis)

3. Abbreviations

ADSA — American Dairy Science Association
AGF — Australian Genome Foundry
CV — Coefficient of Variation
DFW — Dairy Food Waste
DIM — Days in Milk
ELISA — Enzyme-Linked Immunosorbent Assay
HS — Heat Stress
HSP70 — Heat Shock Protein 70
JDS — Journal of Dairy Science
JDS Comms — Journal of Dairy Science Communications
LALBA — Alpha-lactalbumin
LP — Lactose Percentage
LY — Lactose Yield
MCU — Macquarie University
MSY — Milk Solids Yield (kg fat + kg protein)
MSY:LY — Milk Solids Yield to Lactose Yield Ratio
NSW DPIRD — NSW Department of Primary Industries and Regional Development
SCC — Somatic Cell Count
THI — Temperature–Humidity Index
UC Davis — University of California, Davis
USYD — The University of Sydney

4. Project Background and Rationale

The Australian dairy industry faces increasing pressure to improve productivity, sustainability, and resilience. Rising input costs, climate variability, tightening environmental regulation, and competitive global markets are pushing producers and processors to find new ways to extract more value from existing resources.

Dairy UP was established to accelerate applied research that directly addresses some of these pressures. Within this context, the P9 "Designer Milk" was funded to explore three interconnected opportunity areas: improving the efficiency of milk composition, enhancing the tools available for animal health monitoring, and recovering value from DFW streams. These areas were selected because they span the full supply chain, from cow genetics and on-farm management through to processing and waste, and because each represented a gap between existing research knowledge and practical industry application in the Australian context. Key strategic alignments of each subproject with the priorities of Dairy UP, Dairy Australia, and NSW DPIRD are summarised below.

Subproject	Dairy UP Priorities	Dairy Australia Strategic Plan 2030	DPIRD RD&E Strategy 2025–2030
P9a Less Lactose, Less Water, More Milk Solids	Productivity & profitability — unlock the true potential of NSW dairy systems; developing value-add dairy products	Priority 2.2 — Building herd productivity: genetic gain; nutrition efficiency; higher milk income	Future-ready production — driving innovation in established industries; strengthening supply chains; circular economy
P9b Milk as an Indicator of Heat Load	De-risking — new tools to better prepare dairy businesses for future challenges (climate)	Priority 2.3 — Adapting to a changing operating environment: tools to adapt to a changing climate; data-driven decision making	Climate resilient primary production - Resilience and improved management to extreme weather
P9c Adding Value to Dairy Food Waste	Developing new markets — greater economic contribution through value-adding; improved long-term food security of NSW	Priority 3.1 — Advancing industry sustainability: coordinated action on emerging issues; industry sustainability framework	Future-ready production — driving innovation in established industries; circular economy and sustainability in primary industries

Dairy UP Priorities; DA Strategic Plan 2030; NSW DPIRD RD&E Strategy 2025-2030

5. Project Objectives

The project aimed to generate applied, industry-relevant knowledge to improve the efficiency, sustainability, and resilience of the Australian dairy supply chain. The specific objectives were to:

- **Improve milk production efficiency** by quantifying variation in lactose synthesis and identifying cows capable of producing equivalent milk solids with reduced lactose and water volume (i.e., concentrated milk).
- **Develop novel indicators of animal stress and welfare** by evaluating milk-based biomarkers, particularly heat shock protein 70 (HSP70), as non-invasive tools for monitoring heat stress (HS) in dairy cows.
- **Identify and develop pathways for dairy food waste (DFW) valorisation** by characterising Australian waste streams and assessing their suitability for conversion into value-added products through microbial fermentation.
- **Generate integrated datasets and analytical frameworks** linking production, genetics, environment, and physiology to support future research and industry application.
- **Translate research findings into practical insights** for producers, processors, researchers and industry stakeholders through targeted dissemination and engagement activities.

6. Subproject P9a — Less Lactose, Less Water, More Milk Solids

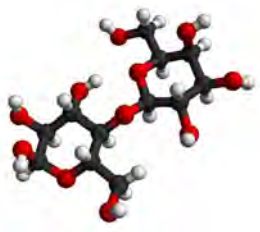
Juan Gargiulo (Research Scientist, NSW DPIRD); Russ Hovey (Prof. Animal Science, UC Davis); Yani Garcia (Prof. of Dairy Science and Dairy UP Program Leader, USYD); Rezaul Rakib (PhD Student, USYD).

6.1 Background

Milk lactose is the primary carbohydrate in bovine milk and plays a central role in regulating milk volume by generating an osmotic gradient that draws water into the mammary gland (Figure 1). Given that milk is approximately 87% water, even small changes in lactose synthesis can substantially influence milk volume. Across breeds, higher concentrations of milk components are typically associated with a proportional reduction in milk volume, often resulting in lower total daily fat and protein yields. However, some cows may exhibit reduced lactose synthesis while maintaining total kilograms of fat and protein which could improve dairy efficiency. Such animals would likely have lower energy requirements, reduced water transport and processing costs, and enhanced resource use efficiency across the supply chain.

Previous research demonstrated that the administration of exogenous glucocorticoid (dexamethasone) can reduce lactose synthesis and water volume by approximately 45% without affecting fat and protein output (Shamay et al., 2000). This response has been linked to the downregulation of α-lactalbumin (LALBA), a key enzyme in lactose synthesis (Sadovnikova et al., 2022). These findings suggest that reduced-lactose milk production has a biological basis, can vary among individuals, and may be heritable.

Despite this evidence, large-scale data characterising natural variation in lactose production across Australian dairy herds remain limited. The factors regulating lactose synthesis under commercial conditions have not been systematically evaluated in the Australian context, and no validated phenotypic or genetic framework currently exists to identify and select animals with improved lactose efficiency.



Lactose is the main milk sugar and a key determinant of milk volume. Lactose concentration is relatively stable

Some species produce milk with lower lactose and higher fat and protein content

Glucocorticoids reduced lactose synthesis and milk volume (~45%), associated with changes in gene expression of key mammary gland proteins

Figure 1. Milk lactose is the primary determinant of milk volume and is relatively conserved across cows; however, substantial variation exists across species and cattle breeds, and lactose synthesis can be experimentally manipulated, resulting in marked changes in milk volume, and concentration of milk solids.

6.2 Objectives

- Quantify variation in lactose yield (**LY**) and lactose percentage (**LP**) in NSW dairy herds.
- Characterise associations between lactose traits and key variables, including milk yield, milk solids yield (**MSY,** kg fat + protein), cow characteristics, and environmental factors.
- Identify cows with high MSY relative to lactose output and characterise the genetic and phenotypic attributes.

6.3 Materials & Methods

- **Data sources:** The analysis used almost 400,000 herd test records from 33,200 cows across 85 NSW herds (2008–2022), including milk yield, milk composition (lactose, fat, protein), reproductive performance, and breed. Supplementary datasets included sire genetic values and local meteorological data**.** Records were collated, cleaned, and merged with genetic and climate datasets.
- **Statistical analysis:** Linear mixed models were used to assess the contributions of parity, stage of lactation, year, season, climate indices (THI), and sire genetics to variation in LY, LP, and MSY. Phenotypic correlations between lactose traits and other production variables were also estimated.
- **Identification of high-efficiency cows:** Cows were ranked by their milk solids yield to lactose yield ratio (MSY:LY), a metric aiming to capture the efficiency of milk solids production relative to lactose output. An enhanced methodology was subsequently developed, incorporating lactation curve modelling and adjustment for parity, days in milk, and mastitis events, to enable more precise and equitable comparisons across animals.

6.4 Key Findings

- **Substantial phenotypic variation exists across the population.** LY, LP, and MSY varied considerably across cows and herds, confirming that meaningful phenotypic diversity exists within the commercial population, a necessary precondition for selection.
- **Breed influences lactose production traits:** On average, Holsteins produced the highest LY and MSY, but had the lowest MSY:LY ratio (~1.50) (Figure 2 a-d). In contrast, Jerseys had the lowest LY and MSY, yet achieved the highest MSY:LY ratio (~1.75), indicating that, they produce milk solids more efficiently relative to lactose and water output. Crossbred animals showed intermediate values for most traits. LP was broadly similar across breeds (~4.95 - 4.98%).
- **Stage of lactation and parity are significant determinants.** LY and LP peaked in early lactation and declined progressively thereafter. LY increased with lactation number while LP decreased (Figure 2e-h).
- **Lactose is negatively associated with heat load and somatic cell count (SCC).** A negative phenotypic correlation was observed between LY and the temperature-humidity index (THI, $r = -0.20$), and between LP and SCC ($r = -0.30$), consistent with known effects of HS and mammary health on lactose synthesis.
- **Lactose traits have a meaningful genetic component.** Heritability estimates for LY and LP ranged from 0.24 to 0.33, indicating that a substantial proportion of phenotypic variation is attributable to genetic effects. This is a critical finding, as it supports the feasibility of genetic selection for improved lactose efficiency.

- **A cohort of high-efficiency cows was identified.** Eighty cows were identified that consistently produced milk with a high MSY:LY ratio, delivering equivalent MSY output to the broader population with approximately 15% less lactose and water volume (Table 1). These animals were descendants of 13 sires spanning Jersey, crossbred lines and Holsteins, suggesting that the relevant genetic variation is already present within the commercial population.
- An enhanced methodology for identifying high-efficiency cows was subsequently developed and validated on a Dairy UP dataset, identifying a cohort of 90 cows. Blood and milk samples have been collected from some of these animals. DNA and milk LALBA concentrations are being analysed to identify genetic variants associated with the high-efficiency phenotypes.

***Table 1**. Predicted means for cows with high milk solids yield to lactose yield ratio (MSY:LY) and the rest of the population for the main variables analysed.*

Variable	High MSY:LY cows (n = 80)	Rest of population (n = 33,200)	Difference (%)
Milk Yield (kg/cow/d)	20.36*	23.97	-15
Lactose Yield (kg/cow/d)	0.98*	1.19	-18
Fat Yield (kg/cow/d)	1.01*	0.96	+5
Protein Yield (kg/cow/d)	0.75*	0.8	-6
Milk Solids Yield (kg/cow/d)	1.76	1.76	0
Lactose (%)	4.81*	4.96	-3
Fat (%)	5.39*	4.14	+30
Protein (%)	3.89*	3.38	+15
Milk Solids (%)	9.29*	7.53	+23
MSY:LY	1.94*	1.53	+27
Somatic cell count (SCC, '000)	114*	91	+25

**groups are statistically different ($p < 0.05$)*

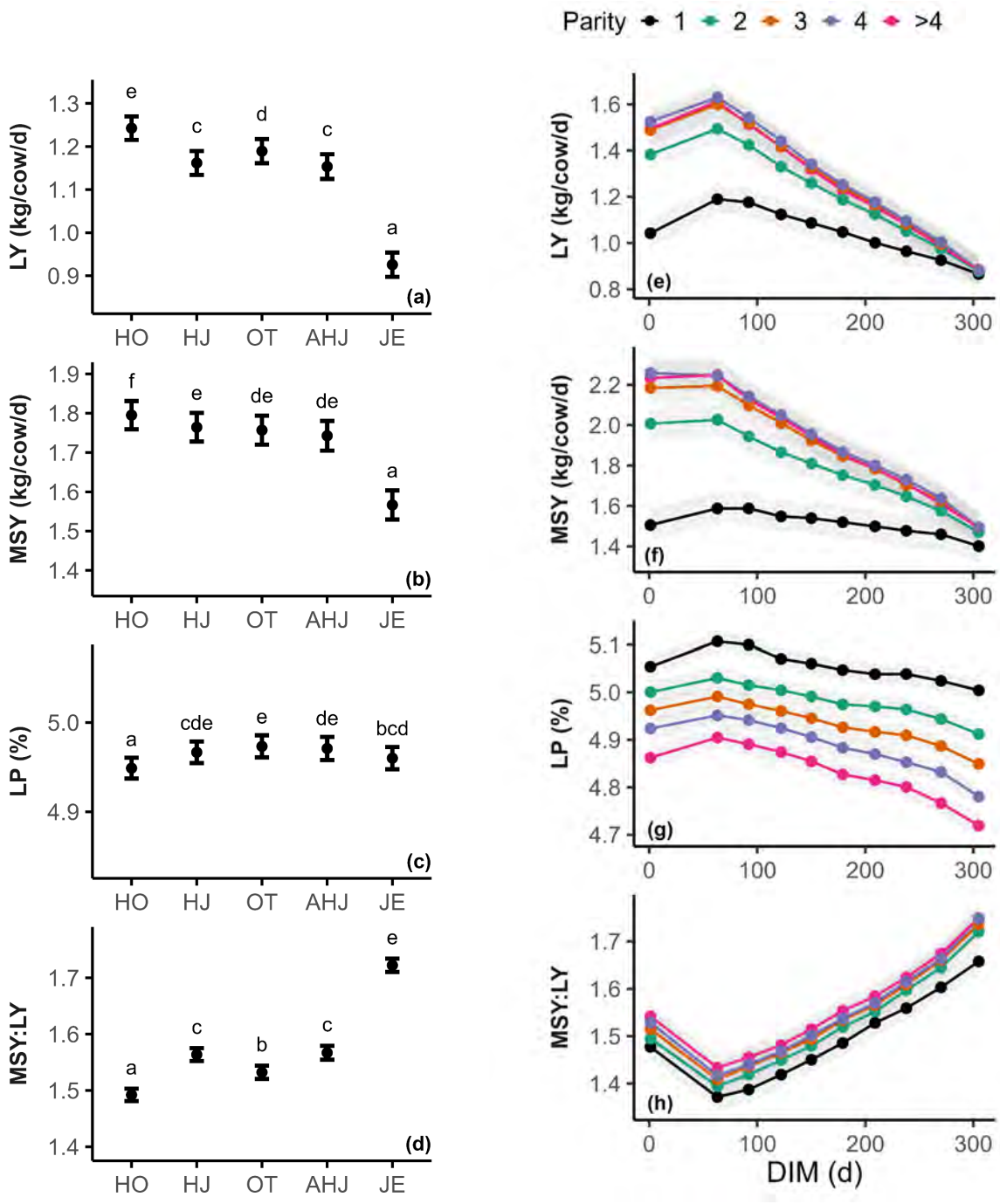


Figure 2. *Effect of breed* ***(a-d)*** *and parity and DIM* ***(e-h)*** *on key variables. HO = Holstein; HJ = Holstein × Jersey; OT = other; AHJ = Ayrshire × Holstein × Jersey; JE = Jersey. Means with different letters are significantly different (P < 0.05).*

6.5 Applications & Impacts

The findings have direct relevance across multiple levels of the dairy supply chain:

At the cow level, reducing lactose synthesis can lower the metabolic energy demand associated with milk production. For example, a 1% reduction in lactose output in a 7,000-litre cow would reduce glucose requirements by ~3.5 kg per lactation — representing a meaningful improvement in energy balance, particularly during periods of peak demand.

At the farm level, lower milk volume per cow may reduce milking frequency requirements in high-producing or automated milking systems, with associated benefits for labour and equipment utilisation.

At the processor level, a higher milk solids concentration reduces the quantity of liquid requiring transport and processing. A 1% reduction in lactose output across the national herd is estimated to reduce annual milk transport volumes by approximately 80 million litres, equivalent to approximately 2,700 truck trips or 800,000 kilometres travelled per year.

At the environmental level, reduced lactose production lowers both the energy and water requirements associated with milk production and processing. For example, a 1% reduction in lactose is estimated to save approximately 20,000 litres of water per farm annually, with considerable cumulative benefit at national scale.

6.6 Key Outputs

- A high-quality, integrated dataset linking lactose production traits with genetics, environment, and production variables across 85 NSW commercial herds, the first such dataset compiled for the Australian dairy industry.
- Development and validation of an enhanced phenotypic methodology for identifying high-efficiency cows based on modelled lactation curves, adjusted for parity, stage of lactation, and health status.
- Peer-reviewed publication in the *Journal of Dairy Science (JDS)* characterising lactose variability and its determinants in Australian dairy herds.
- Conference presentations and industry reports disseminating findings to researchers, producer and processor audiences.

6.7 Future Research Opportunities

The identification of a cohort of cows consistently exhibiting high MSY:LY ratios, combined with moderate heritability estimates for lactose traits and previous experimental research, provides an empirical foundation for a genetic selection program. The leading hypothesis is that these animals may carry variation in regulatory regions governing LALBA expression, resulting in reduced lactose synthesis without affecting fat or protein secretion. Confirmation of this mechanism would represent a significant advance in understanding mammary gland function and could unlock a practical selection pathway for the Australian industry.

Priority areas for future research include:

- **Genetic marker discovery:** Genome-wide association analysis of high-efficiency cows to identify variants associated with elevated MSY:LY, as a basis for genomic selection tools.
- **Molecular characterisation:** Targeted investigation of LALBA and related lactation genes in sampled animals to confirm the proposed biological mechanism and define the molecular basis of the phenotype.

- **Early-life predictors:** Identification of markers measurable early in life that are predictive of long-term lactose efficiency, enabling earlier and more cost-effective selection decisions.
- **Economic and systems modelling:** Quantitative assessment of the farm- and industry-level impacts of selective breeding for this trait across different herd structures, breeds, and payment systems.

7. Subproject P9b — Milk as an Indicator of Heat Load

Juan Gargiulo (Research Scientist, NSW DPIRD); Rezaul Rakib (PhD student, USYD); Yani Garcia (Prof. of Dairy Science and Dairy UP Program Leader, USYD), Valeria Messina (Technical Specialist, USYD); Jade Nguyen (Honours student, USYD); Nicolas Lyons (Head of Science, DairyNZ); Indunil Pathirana (Prof. in Animal Science, University of Ruhuna).

7.1 Background

HS is a major constraint in Australian pasture-based dairy systems, reducing milk yield, reproduction, and animal welfare. Traditional HS indicators, such as the THI, rectal temperature or reticulorumen temperature, provide useful environmental and immediate physiological information but offer limited insight into cellular-level stress responses.

Heat shock protein 70 (HSP70), a conserved molecular chaperone, is rapidly upregulated during cellular stress and has recently been detected in milk, offering a potential non-invasive biomarker that can be incorporated into routine herd testing (Figure 3). Subproject P9b evaluated HSP70 as a practical indicator of cellular stress by validating analytical methods, defining sample-handling requirements, characterising seasonal variation, and quantifying lag times relative to physiological HS measures.

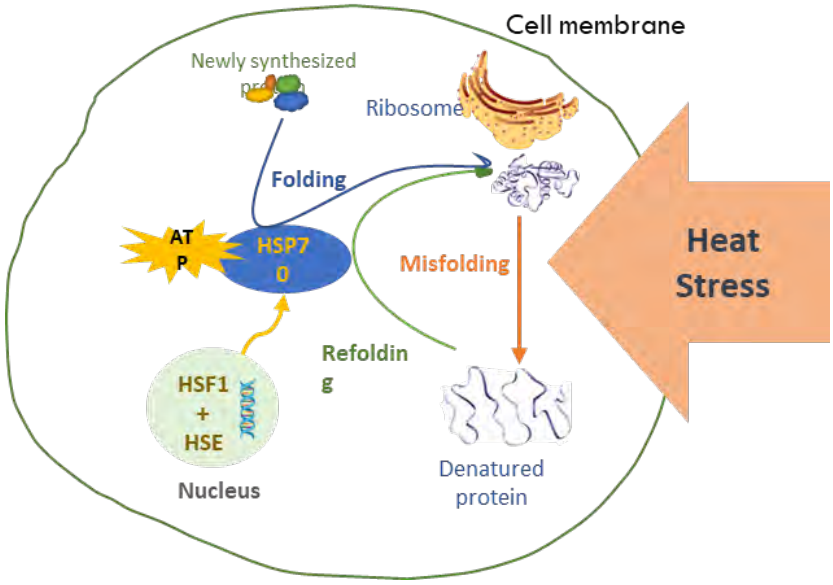


Figure 3. HPS70 is induced in response to heat stress (HS) to help stabilise and protect proteins, and has been previously measured in blood and saliva, and recently in detected in milk [adapted from Rakib et al. (2024)]

7.2 Objectives

- Review current knowledge of HS detection in dairy cows, focusing on the potential and limitations of HSP70 as a molecular biomarker.
- Validate reliable and cost-effective laboratory methods for detecting HSP70 in milk, blood, and saliva.
- Establish milk sample handling and storage protocols to ensure accurate HSP70 detection.
- Characterise between-cow and seasonal variation in HSP70.
- Compare milk HSP70 with environmental and physiological HS indicators.
- Quantify time lags between heat load and milk HSP70 response.
- Identify on-farm and industry applications for HSP70-based HS monitoring.

7.3 Materials & Methods

To address these objectives, the project undertook a series of coordinated studies:

- **Analytical validation**: A newly developed in-house competitive ELISA was benchmarked against a commercial ELISA across milk, blood, and saliva. Agreement, precision, working range, and cost were evaluated.
- **Sample handling and storage**: Milk samples were processed either skimmed before storage or skimmed after storage and stored under controlled conditions (room temperature, refrigeration, freezing, refrigeration + preservative). HSP70 stability was assessed across multiple time points.
- **Seasonal biological characterisation**: HSP70 concentrations were measured in milk, blood, and saliva across contrasting seasons and linked to THI, milk yield, and milk composition. Mixed-effects models were used to quantify environmental and physiological influences.
- **Comparative evaluation with physiological indicators**: Simultaneous measurements of THI, rectal temperature, infrared thermography, and reticulorumen temperature were collected to compare the sensitivity and responsiveness of milk HSP70 with established HS indicators. Time-lagged correlations (0–48 h) were calculated.
- **Heatwave trial:** A five-day heatwave event was monitored intensively to validate temporal behaviour, evaluate threshold values, and assess diagnostic performance using ROC analysis.

7.4 Key Findings

- **Milk HSP70 can be measured reliably using a validated, low-cost method.** Strong agreement was demonstrated between the in-house competitive ELISA and commercial ELISA across all matrices, confirming that accurate and affordable detection of HSP70 in dairy samples is feasible for routine application.
- **Sample handling conditions materially affect HSP70 detection accuracy.** Skimming milk before storage and holding samples refrigerated or frozen for up to three days preserved HSP70 integrity. Storage at room temperature or with bronopol preservative reduced accuracy, with direct implications for how samples should be managed in commercial herd-testing workflows.
- **HSP70 concentrations vary substantially between cows and across seasons.** Levels were highest in summer and lowest in winter, consistent with seasonal variation in heat load. Milk HSP70 exhibited greater coefficient of variation than blood or saliva (CV 54% vs. 34% and 32%, respectively), indicating a higher degree of biological variability in the milk fraction that must be accounted for in interpretation.
- **Milk HSP70 reflects prior heat exposure rather than current conditions.** Peak milk HSP70 concentrations occurred approximately 33–44 hours after peak THI (Figure 4) and 48–50 hours after peak reticulorumen temperature, confirming that the biomarker reflects cumulative cellular stress over the preceding one to two days rather than providing a real-time indicator of heat load. This temporal lag has direct implications for how and when the biomarker can be usefully applied in practice.

- **Diagnostic performance was strong during sustained heat events but more limited under typical seasonal conditions.** During a five-day heatwave, a milk HSP70 threshold of approximately 550 ng/mL demonstrated good diagnostic performance. Under non-heatwave seasonal conditions, discriminatory power was more limited, consistent with the view that milk HSP70 functions most reliably as an indicator when cellular stress is both substantial and persistent. HSP70 levels may also be influenced by other biological factors not assessed in this study, including stage of lactation, lactation number, production level, and health status, which should be taken into account when interpreting results outside of extreme heat conditions.

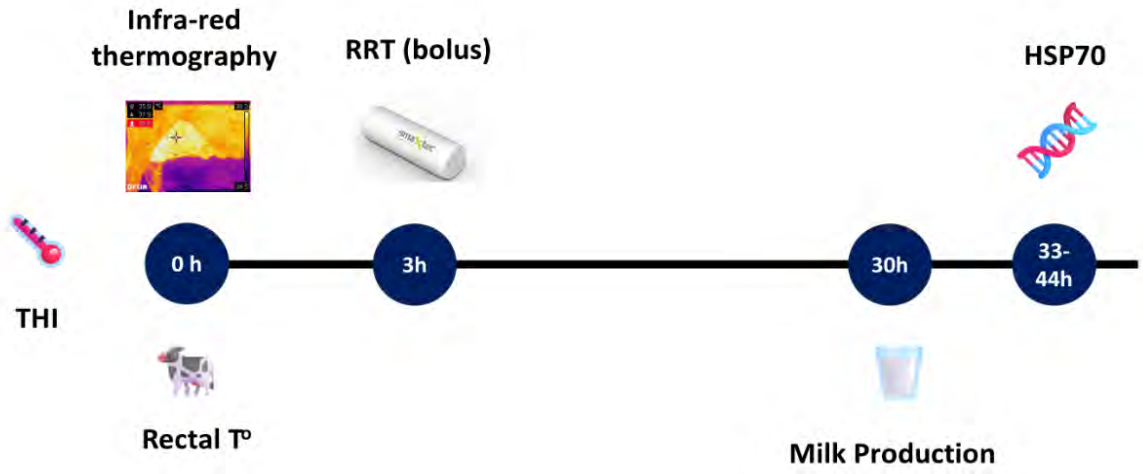


Figure 4. Comparison of heat stress (HS)detection methods and their lagged relationship with temperature-humidity index (THI)

7.5 Applications & Impacts

- **At the cow and farm level,** milk HSP70 provides a specific, quantitative indicator of cellular HS that can be collected during routine milking without additional animal handling or discomfort. Its delayed response profile, reflecting cumulative stress over the preceding 33–44 hours, makes it best suited to retrospective assessment of heat exposure rather than real-time intervention decisions. Its specificity to cellular stress, rather than environmental temperature alone, represents an advance over existing THI-based approaches, though its interpretation requires consideration of other factors known to influence HSP70 expression (e.g. stage of lactation or health status). The validated threshold of approximately 550 ng/mL provides an objective, repeatable basis for comparing HS severity across cows and seasons in research and monitoring contexts, pending further field validation.
- **At the herd management level,** milk HSP70 measurement has potential for integration into herd testing, enabling surveillance of cumulative cellular stress at the herd level without additional labour requirements. It could also serve as an objective tool for evaluating the effectiveness of HS mitigation strategies — such as cooling systems, shade provision, or adjusted milking schedules — providing evidence for both on-farm management decisions and industry-level welfare reporting.
- **At the processor and industry level,** consistent and quantifiable HS monitoring can support processors in managing milk supply quality and variability and provides data to underpin sustainability and animal welfare reporting requirements increasingly expected by domestic and international markets.

7.6 Outputs

- Validation of a low-cost, in-house competitive ELISA for HSP70 detection in milk, blood, and saliva, benchmarked against a commercial assay — providing a practical analytical tool for adoption in research and industry settings.
- Establishment of evidence-based sample handling and storage protocols required for accurate milk HSP70 detection in commercial workflows.
- Characterisation of the temporal lag between heat load and milk HSP70 response, establishing the biomarker's behaviour relative to established physiological indicators across seasonal and acute heat conditions.
- Four peer-reviewed publications in the JDS and JDS Comms (Table 2), with two additional manuscripts under revision
- PhD thesis (Rezaul Rakib, USYD) documenting the full body of research conducted under this subproject.
- Honours thesis (Jade Nguyen, USYD) on "Assessing Alternatives to Rectal Temperature for Heat Stress Detection in Dairy Cattle"
- Presentations at major national and international scientific conferences, including the American Dairy Science Association Annual Meeting (USA, 2024 and 2025), the Australasian Dairy Science Symposium (New Zealand, 2024), and the Dairy Research Foundation Symposium (Australia, 2023 and 2025).

7.7 Future Research Opportunities

- Future studies should extend beyond HS alone to evaluate milk HSP70 as an integrative marker of overall physiological stress and health status in dairy cows, including potential associations with disease, ageing, mastitis, and immunological activation.
- Significant variations in HSP70 responses were observed among cows under similar environmental conditions, suggesting that animals may differ in how their cells respond to heat load. Further research is needed to determine whether these individual variations are consistent across lactations and environmental conditions, and to better understand the biological processes underlying them.

8. Subproject P9c — Adding Value to Dairy Food Waste

Juan Gargiulo (Research Scientist, NSW DPIRD); Briardo Llorente (Associate Prof. and Chief Scientist AGF, MCU); Yani Garcia (Prof. of Dairy Science and Dairy UP Program Leader, USYD); Ian Paulsen (Prof. and Director ARC Centre of Excellence in Synthetic Biology and AGF, MCU); Belinda Fabian (Business Manager AGF, MCU), Artur Wlodarczyk (Synthetic Biologist, MCU); Hugh Goold (Research Scientist, NSW DPIRD); Ken Garner (Manager Dairy, NSW DPIRD).

8.1 Background

Dairy food waste (DFW) represents a substantial and underutilised resource across the Australian supply chain, totalling 1,363,500 tonnes or ~16% of national milk production (Dairy Australia, 2024). Manufacturing is the dominant source, generating lactose-rich by-products, and process waste as inherent outputs of milk processing (Figure 5). Manufacturers spend approximately $967 million per year on waste management, with a further $786 million lost through finished products discarded across retail, food service, and households. Beyond these direct costs, DFW generates significant environmental burdens, including greenhouse gas emissions, water use, and waste disposal.

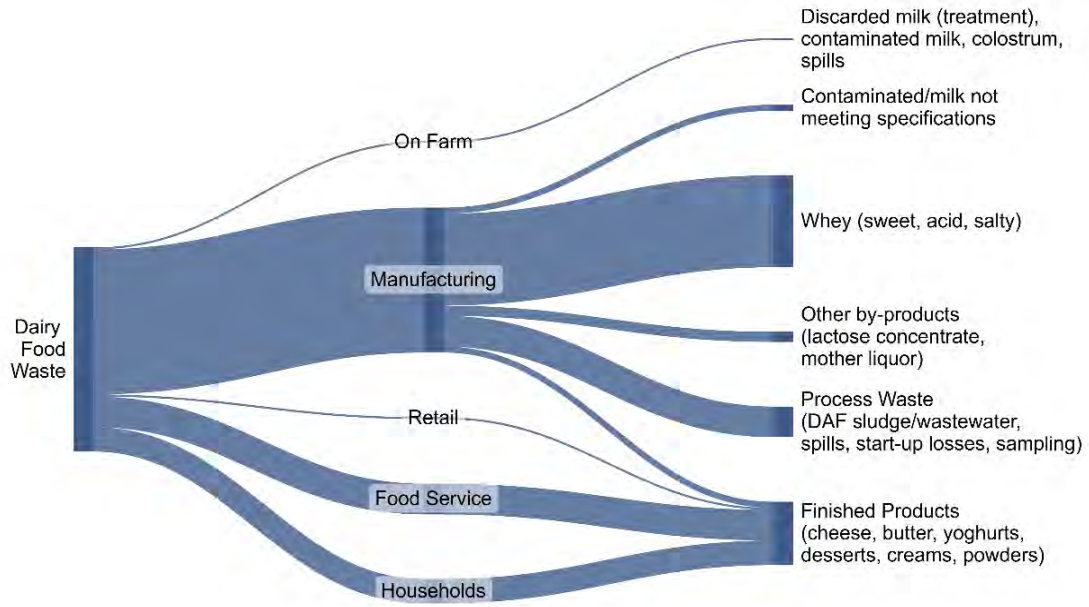


Figure 5. Dairy food waste across the supply chain [adapted from Dairy Australia (2023)].

Valorisation of DFW through microbial fermentation offers a pathway to recover value by converting these streams into useful products, including animal feed ingredients and other high value bioproducts, while reducing disposal costs and environmental impacts. This approach aligns with circular economy principles and has been demonstrated at laboratory scale internationally.

Despite this potential, the DFW streams most amenable to microbial fermentation in the Australian context had not been systematically evaluated. In addition, key biological and engineering constraints—particularly the inability of standard microbial strains to efficiently utilise lactose, the principal carbohydrate in DFW—had not been addressed in a food-grade production context. Limited proof-of-concept existed for producing value-added compounds from Australian DFW using engineered food-grade microorganisms. This subproject was designed to address these gaps.

8.2 Objectives

- Identify DFW streams in Australia that are most suitable for microbial fermentation
- Identify value-added compounds that can be produced from DFW
- Develop a food-grade yeast strain capable of growing on DFW and producing target compounds as a proof of concept.

8.3 Materials & Methods

- **Literature review and stakeholder consultation:** A comprehensive review was conducted to identify DFW sources, compositional variability, and their potential as substrates for microbial bioproduction. Industry stakeholders were consulted to prioritise feasible DFW sources and value-added products.
- **Biological and engineering strategies:** Approaches to overcome key constraints, particularly lactose utilisation and substrate variability, were evaluated.
- **Yeast engineering:** Synthetic biology and adaptive laboratory evolution were applied to engineer food-grade *Saccharomyces cerevisiae* capable of growing on lactose as the sole carbon source and producing pro-vitamin A as a proof of concept.

8.4 Key Findings

- **Microbial fermentation represents a viable and strategically relevant pathway for DFW valorisation.** A systematic review of the Australian DFW landscape confirmed that the sector produces waste streams well-suited to bioproduction in terms of volume, composition, and consistency, supporting the case for investment in this approach as a practical circular economy strategy.
- **Cheese whey and derived by products were identified as the most suitable streams for microbial valorisation.** These streams are produced in concentrated and relatively consistent volumes at manufacturing facilities, making them more amenable to scalable bioproduction than household or food service waste, which is compositionally variable and logistically dispersed. On-farm DFW is only ~1%, though a significant proportion comprises milk with antibiotics fed to calves, representing a potential risk and future opportunity for valorisation.
- **Microbial biomass and derived products represent the most immediate valorisation opportunity.** Analysis of potential bioproducts identified microbial biomass and its derivatives as the most scalable near-term opportunity, primarily for animal nutrition applications. Other compounds, including amino acids, organic acids, and enzymes, were identified as potential targets for higher-value applications requiring further technical development.
- **The inability of standard yeast strains to metabolise lactose is the principal technical constraint.** Conventional *S. cerevisiae* strains cannot efficiently utilise lactose. Alternative species, such as *Kluyveromyces marxianus*, offer partial solutions but present performance and regulatory limitations. Engineered S. cerevisiae strains can overcome this constraint but require further optimisation for industrial-scale efficiency**.**
- **A food-grade yeast strain capable of growing on lactose and producing pro-vitamin A was successfully engineered.** Using a combination of synthetic biology and adaptive laboratory evolution, the research team at MCU engineered a food-grade S. cerevisiae strain capable of growing on lactose as its sole carbon source and producing pro-vitamin A (Figure 6). The strain also demonstrated growth on 50% milk supplemented with lactose, confirming feasibility for DFW valorisation. While currently at laboratory scale, this establishes a functional platform for further development.

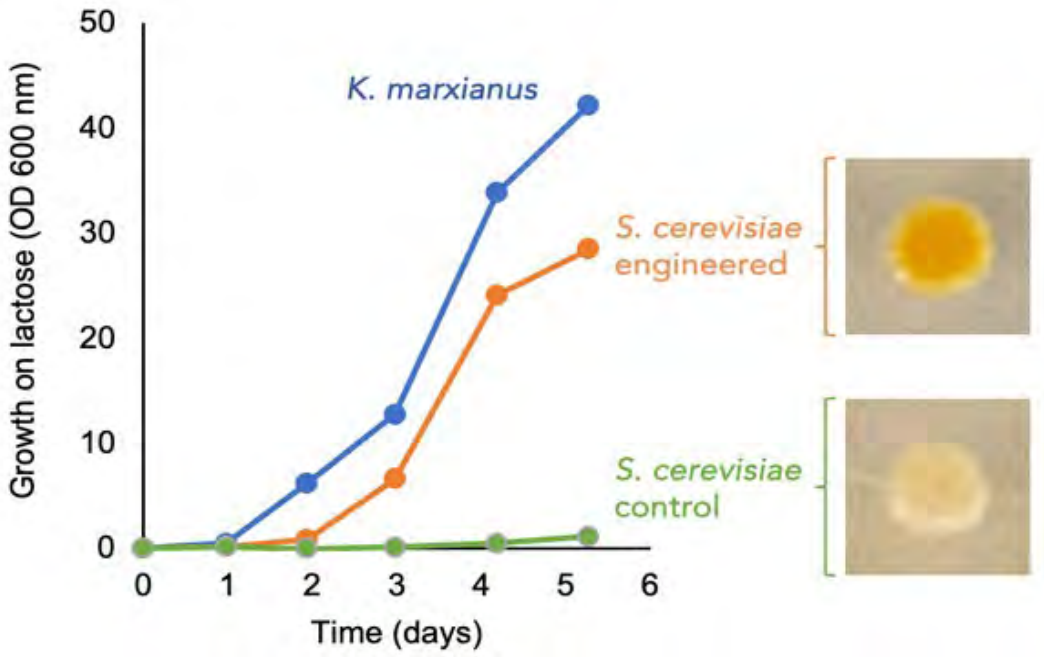


***Figure 6**. Engineered S. cerevisiae (orange) can grow on lactose and produces provitamin A, which gives the cells an orange colour. Kluyveromyces marxianus (blue), a yeast capable of growing on lactose is shown for comparison. Control, non-engineered S. cerevisiae (green) cannot grow on lactose and cells are white.*

8.5 Applications & Impacts

- **At the manufacturer level**, microbial valorisation of concentrated DFW streams — particularly cheese whey and whey permeate — provides a practical pathway to generate value-added products, reduce waste disposal costs, and create new revenue streams. For processors currently spending significant resources on waste management, even partial valorisation of these streams could materially improve the economics of waste handling.
- **At the environmental level**, converting DFW into useful products reduces the volume requiring disposal, lowering associated greenhouse gas emissions, water use, and environmental compliance costs. This approach directly supports the circular economy principles that are increasingly expected by regulators, retailers, and consumers in domestic and international dairy markets.
- **At the technical level,** the successful engineering of a food-grade yeast strain capable of lactose utilisation and pro-vitamin A production demonstrates that a key biological constraint can be overcome. This establishes a platform that can be extended to produce a broader range of target compounds.

The proof-of-concept demonstration of growth on lactose substrates and production of a nutritionally relevant compound provides the evidence base required to justify further investment in scale-up and techno-economic evaluation.

8.6 Outputs

- Systematic identification and characterisation of DFW streams suitable for microbial fermentation, with whey and derived by-products identified as primary candidates — providing an evidence base for industry investment decisions.
- Literature review characterising DFW valorisation opportunities and the biological pathways

available for their realisation.

- Engineering and characterisation of a food-grade S. cerevisiae strain capable of lactose utilisation and pro-vitamin A production, demonstrating proof-of-concept for value-added bioproduction from DFW.
- Established relationships with dairy manufacturers and industry stakeholders, facilitating access to dairy by-products and providing practical insight into supply chain constraints and potential commercial pathways.
- Compilation of historical datasets on milk composition, by-products, and waste streams that informed experimental design and provide a foundation for future techno-economic modelling.

8.7 Future Research Opportunities

This subproject established proof-of-concept for the valorisation of DFW through engineered yeast fermentation, confirmed the suitability of key Australian waste streams, and demonstrated that a major technical constraint—lactose utilisation by food-grade yeast—can be overcome. The work remains at laboratory scale, and translation to commercial application will require coordinated efforts in optimisation, scale-up, and economic validation.

Priority areas for future research include:

- **Techno-economic analysis:** Evaluation of production targets, input costs, and revenue potential for priority bioproducts (e.g. microbial biomass, pro-vitamin A) to define conditions for commercial viability.
- **Enzyme engineering for improved lactose assimilation:** Development of β-galactosidase variants with enhanced activity and stability under dairy waste conditions.
- **Strain optimisation:** Improvement of growth rates, product titres, and tolerance to substrate variability under industrial conditions.
- **Platform strain development:** Engineering strains for production of additional value-added compounds using the lactose-utilising platform developed in this project.
- **Pilot-scale validation:** Scale-up of promising strain–substrate systems in collaboration with industry partners to generate process and economic data required for commercial assessment.

9. Project-wide Dissemination

The project has produced one PhD thesis, one Honours thesis, and six peer-reviewed publications in high-impact international journals (JDS and JDS Comms), accumulating 34 citations to date (Table 2). The JDS, the leading journal in dairy science, has an impact factor of 4.4 and is ranked Q1 in its field, reflecting the quality and reach of the outlets targeted by this project.

Findings were also presented at multiple conferences and scientific events, including three major international forums, most notably the American Dairy Science Association (ADSA) Annual Meeting in the USA, as well as Australian and New Zealand venues (Table 3). These presentations collectively reached an estimated audience of over 2,700 people, spanning industry professionals, researchers and policymakers.

Beyond peer-reviewed and conference outputs, the project actively engaged internally and externally with industry and farming communities through a range of channels, including farmer meetings, webinars, podcasts, newsletters, and online articles. A list of these outputs is provided in Tables 3 and 4 (Annexes).

***Table 2**. Peer-reviewed publications in international journals.*

Author	Title	Journal	Year Published	Citations (to May 26)
Gargiulo (2026)	Invited Review: Turning Dairy Food Waste into Valuable Products via Yeast Bioproduction (In Press)	JDS	2026	-
Rakib et al. (2026a)	Detecting heat shock protein 70 in milk, blood, and saliva of dairy cows exposed to different seasonal conditions	JDS	2026	-
Rakib et al. (2026b)	Comparison between in-house competitive and commercial ELISA for the detection of heat shock protein 70 in milk, blood, and saliva of dairy cows	JDS	2026	-
Rakib et al. (2025)	Skimming and storage factors affect the detection of heat shock protein 70 in raw bovine milk	JDS Comms	2025	2
Gargiulo et al. (2025)	Sources of variation underlying the production of lactose by dairy cows	JDS	2025	7
Rakib et al. (2024)	Graduate Student Literature Review: Potential use of HSP70 as an indicator of heat stress in dairy cows—A review	JDS	2024	25

10. Conclusions and Recommendations

The P9 - Designer Milk project has delivered meaningful progress across three interconnected areas of dairy research, generating evidence-based findings with clear, still early-stage, pathways to industry application.

P9a demonstrated that substantial variation in lactose production exists across NSW dairy herds. The project identified a subset of cows capable of maintaining equivalent milk solids output with significantly lower lactose and water volume, with potential implications for transport, processing costs, and resource use efficiency. Heritability estimates indicate a genetic component to this variation, supporting selective breeding as a plausible long-term pathway for improving milk efficiency. However, the underlying genetic mechanisms remain unconfirmed, and further molecular research is required before practical selection tools can be developed.

P9b validated milk HSP70 as a measurable, non-invasive biomarker and established sample-handling requirements for reliable detection. However, its relationship with HS was not consistently strong, with concentrations influenced by additional biological and environmental factors, and a delayed response limiting its use for real-time monitoring. These findings highlight both the potential and current limitations of HSP70 as an indicator, indicating that further work is required to improve its specificity before integration into routine herd testing. Results suggest that HSP70 is better positioned as a complementary component within a broader, multifactorial biomarker framework for assessing HS and animal resilience.

P9c identified key DFW streams and prioritised bioproducts for valorisation through microbial fermentation. A proof-of-concept was established developing an engineered yeast strain capable of growing on lactose and producing pro-vitamin A, demonstrating the technical feasibility of this approach. The work remains at laboratory scale, and significant technical optimisation and economic evaluation are required before commercial application.

Across all three subprojects, the project has produced several peer-reviewed publications, engaged audiences across Australia and internationally, and built productive industry partnerships, laying a solid foundation for the next phase of research.

Recommendations for future investment should prioritise:

- Molecular studies to identify genetic markers associated with high-solids, low-lactose cows in P9a
- Field validation of HSP70 monitoring in commercial herd-testing workflows in P9b
- Techno-economic analysis and scale-up optimisation for dairy waste valorisation in P9c

11. Annexes

Table 3. Conference presentations, abstracts and other meetings

Authors	Title	Presentation Type	Conference /Event	Location	Year	Audience
Rakib, R.	Investigations of heat shock protein 70 for non-invasive detection of heat stress in dairy cows	Final PhD Presentation (oral)	SOLES HDR Showcase (USYD)	Sydney	**2025**	40
Rakib, R.	Milk speaks when cows are heat-stressed	Oral presentation	Thesis competition (USYD)	Sydney	**2025**	60
Rakib, et al.	Milk speaks when cows are heat-stressed	Abstract/oral presentation	DRF Symposium	Wollongong	**2025**	150
Gargiulo, J.	Unlocking the Potential of Milk	Oral presentation	DIAA Dairy Conference	Parramatta	**2025**	40
Gargiulo et al.	Factors influencing lactose production in dairy cows	Refereed abstract/poster presentation	ADSA Annual Meeting	Louisville, USA	**2025**	1,000
Rakib et al.	Potential of HSP70 as a non-invasive biomarker for detecting heat stress in dairy cows	Refereed abstract/poster presentation	ADSA Annual Meeting	Louisville, USA	**2025**	
Rakib et al.	Influence of management and storage factors on detection of HSP70 in raw bovine milk	Refereed abstract/oral presentation	ADSA Annual Meeting	Florida, USA	**2024**	1,000
Rakib et al.	Influence of management and storage factors on detection of HSP70 in raw bovine milk	Poster presentation (Intermediate PhD)	SOLES HDR Showcase (USYD)	Sydney	**2024**	100
Rakib et al.	Detection of HSP70 in milk, blood, and saliva of dairy cows exposed to different climatic conditions	Refereed abstract/Oral presentation	ADSS	Christchurch, NZ	**2024**	200
Rakib et al.	Can heat shock protein 70 be monitored in dairy cows under heat stress?	Oral presentation (Introductory PhD)	SOLES HDR Showcase (USYD)	Sydney	**2023**	40
Rakib et al.	Can heat stress be detected in the milk of dairy cows?	Oral presentation	DRF Symposium	Camden	**2023**	100
Gargiulo, J.	P9 – Designer Milk	Farmer meeting presentation	Dairy UP Roadshow	Bega and Cobargo	**2023**	30

Table 4. Technical reports, pamphlets, published material and other media engagements

Authors	Title	Place Published	Year
Gargiulo, J.	Producing milk with less water?	DPIRD Dairy Newsletter	2025
DairyNews Australia	Less water, more milk solids	DairyNews Australia	2025
Gargiulo, J.	Producing milk with less lactose - why?	The Business of Dairy Podcast	2025
Gargiulo, J.	HSP70 as an indicator of heat load	DPIRD Dairy Newsletter	2025
DairyNews Australia	Research aims to add value to dairy waste	DairyNews Australia	2024
Monks, L.; Gargiulo, J.; Hovey, R. & Garcia, S.	Producing milk with less water?	Dairy UP Website	2024
Monks, L.; Gargiulo, J.; & Llorente, B.	Adding value to milk waste	Dairy UP Website	2024
Gargiulo, J. & Llorente, B.	Revalorisation of dairy food waste	Dairy UP Webinar	2024
Monks, L.; Gargiulo, J.; Rakib R. & Garcia, S.	Milk as an indicator of heat load	Dairy UP Website	2023
Garcia, S. & Gargiulo, J.	Dairy UP - fast tracking the NSW dairy industry through R, D & E	The Business of Dairy Podcast	2023
Gargiulo, J.	New opportunities for milk?	Dairy UP Webinar	2023
Gargiulo, J.	More milk income from less milk volume? Exploring lactose variability in NSW herds to concentrate milk solids	Dairy UP Webinar	2023

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Rakib, M. R. H., V. Messina, J. I. Gargiulo, N. A. Lyons, I. N. Pathirana, P. C. Thomson, and S. C. Garcia. 2025. Skimming and storage factors affect the detection of heat shock protein 70 in raw bovine milk. JDS Communications 6(4):578-582.https://10.3168/jdsc.2025-0759

Rakib, M. R. H., V. Messina, J. I. Gargiulo, N. A. Lyons, I. N. Pathirana, P. C. Thomson, and S. C. Garcia. 2026a. Detecting heat shock protein 70 in milk, blood, and saliva of dairy cows exposed to different seasonal conditions. Journal of Dairy Science 109(4):4647-4658.https://10.3168/jds.2025-27609

Rakib, M. R. H., V. Messina, J. I. Gargiulo, I. N. Pathirana, N. A. Lyons, and S. C. Garcia. 2026b. Comparison between in-house competitive and commercial enzyme-linked immunosorbent assays for the detection of heat shock protein 70 in milk, blood, and saliva of dairy cows. Journal of Dairy Science 109(4):4596-4603.https://10.3168/jds.2025-27811

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Final Project Summary
P9a Producing milk with less lactose

May 2026

Climate change is increasing pressure to produce food with less water. As milk is 87% water, producing milk with less water is an opportunity for the dairy industry to improve its carbon footprint, reduce energy costs and improve water use efficiency. This could create more value and profit along the supply chain.

This project aimed to find ways to produce cows' milk with high concentrations of solids.

Lactose was the focus of this work as it plays a key role in determining milk's water content.

The project investigated the theory that those cows that produce less lactose would produce milk with less water and higher concentrations of solids (fat and protein), reducing transport, processing costs, and resource use.

Potential benefits

Lactose has broad roles in the physiology of the dairy cow, which means the potential to influence lactose production has a wide range of potential benefits.

Unlocking the potential of cows

Milk underpins the dairy industry. Getting more from this precious commodity has the potential to increase its value throughout the supply chain.

The Dairy UP team investigated novel ways to get more value from milk.

There were three elements to this research:

- P9a: Producing milk with less lactose
- P9b: Milk as an indicator of heat load
- P9c: Adding value to dairy waste

This document provides an overview of P9

Animal performance, health and welfare
It takes a lot of energy for a cow to produce lactose so reducing the production of lactose could improve the energy balance of the cow, especially during the transition period. Improved energy balance could also have a role in reproduction and fertility.

Lactose is also associated with some animal health traits. For example, milk lactose content could be used to monitor or detect mastitis and ketosis in dairy cows.

Farm labour
If reducing lactose production means cows produce milk containing less water, it may be possible to reduce the frequency of milking. This could provide a labour-saving opportunity.

Processing and transport
A reduction in milk volume could also lead to more efficient transport – carting less water or volume overall – and gains in the processing sector.

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Key findings

Out of a dataset of more than 30,000 cows, researchers identified 80 individuals that consistently produced milk with about 15% less water and lactose content or lactose yield while maintaining similar fat and protein levels.

Heritability estimates suggest the trait has a meaningful genetic component, indicating that breeding selection is feasible.

The potential scale of impact is substantial: a 1% reduction in lactose across the national herd could reduce milk transport volumes by about 80 million litres annually — saving an estimated 2,700 truck trips and 20,000 litres of water per farm per year.

Although the underlying molecular mechanisms have not been confirmed, the work established an evidence-based foundation for a genetic selection program.

The findings also highlight the potential to develop management strategies to influence lactose production and enhance the milk production efficiency of cows and potentially reduce their environmental impact.

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P9 final report

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Research and results

This project built on earlier work in California led by Dairy UP collaborator Prof Russ Hovey that demonstrated it is possible to reduce the lactose in milk production without negatively affecting the total milk solids output. To identify potential management interventions to reduce lactose production, Dairy UP researchers investigated the factors that influence lactose secretion in the cow, for example milk composition, genetics and environmental conditions.

There were two elements of this project. The first project investigated the impact of genetics and other management or environmental influences. It involved creating and analysing a large-scale dataset from Dairy UP monitor farms, NSW herd test results and DataGene's Central Data Repository.

The second was research to better understand lactose synthesis in the dairy cow and how it is regulated. This work was being undertaken at the University of California, Davis, USA.

What influences milk lactose levels?
To understand the factors affecting lactose production the team examined datasets of herd records, genetic data for dairy sires and meteorological records. Spanning 14 years (2008-2022) the dataset included 393,772 herd records from 33,280 cows in 85 herds, representing 5% of the NSW herd records.

The following summarises some of the high-level findings about the factors affecting lactose production in Australian dairy cows.

Parity (number of calvings)
The lactose percentage was higher for heifers throughout their lactation but their total lactose yield wasn't (because heifers' total milk yield per lactation is less than mature cows).

Stage of lactation
Stage of lactation had a strong impact on lactose output, peaking in early lactation and decreasing as the lactation progressed (similar to the milk yield curve).

Seasonal conditions
Hot, humid weather negatively affected yield of milk, lactose and milk solids but it did not affect lactose percentage or other milk components. A decline in milk yield, lactose yield and protein

yield coincided with severe drought conditions from 2016.

Breed

Holsteins had the highest lactose yields. Jerseys had the lowest lactose yields and produced more milk solids per unit of lactose. This finding wasn't surprising given that Jerseys are well recognised for producing milk with higher concentrations of solids for a given volume of milk compared with other dairy breeds like Holsteins.

Genetic link

Building on these findings, DairyUP researchers determined that lactose traits have a meaningful genetic component.

Heritability estimates for lactose yield and lactose percentage ranged from 0.24 to 0.33. This suggests that genetic selection for improved lactose efficiency is feasible.

A cohort of 80 high-efficiency cows was identified. These cows consistently produced milk with a high ratio of milk solids yield to lactose yield, delivering equivalent milk solids output to the broader population with approximately 15% less lactose and water volume.

These animals were descendants of 13 sires spanning Jersey, Holstein, and crossbred lines, suggesting that the relevant genetic variation is already present within the commercial population.

All the female descendants of these sires produced a similar milk yield, but with more fat, and showed a trend towards lower protein production. There was no difference in daughter fertility.

An enhanced methodology for identifying high-efficiency cows was subsequently developed and validated on a Dairy UP dataset, identifying a cohort of 90 cows. Blood and milk samples have been collected from some of these animals. DNA and milk LALBA concentrations are being analysed to identify genetic variants associated with the high-efficiency phenotypes.

Lactose synthesis

A 'proof of concept' study showed it is possible to change the proportion of lactose in milk without affecting fat and protein production.

The study involved Holsteins in a total mixed ration farming system that were on their second calf and at peak lactation.

It demonstrated that a single treatment with the drug dexamethasone temporarily reduced the amount of lactose in the cow's udder. Fat and protein production increased in response to the treatment as milk volume decreased.

Blood and milk samples were collected from some of these animals. DNA and milk alpha-lactalbumin concentrations were analysed to identify genetic variants associated with the high-efficiency animals.

The team uncovered a possible explanation for this finding. Advanced genetic testing pointed to a regulatory molecule involved in lactose synthesis (alpha-lactalbumin) that was suppressed by the dexamethasone treatment.

This finding offers new insights for researchers to better understand the factors regulating milk yield (volume) relative to the fat and protein content which could lead to interventions.

Priorities for future research

Priorities for future research could include:

- Genetic marker discovery: associated with elevated ratio of milk solid yield to lactose yield.
- Molecular characterisation: to confirm the proposed biological mechanism.
- Early-life predictors of long-term lactose efficiency.
- Economic and systems modelling of on-farm and industry impact of selecting for lower lactose production.

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Unlocking potential

Collaborators

This project was a collaboration between Dairy UP researchers based at the University of California Davis (USA), DPI NSW and the University of Sydney. A large proportion of the herd records were provided by DataGene.

Read more

J.I. Gargiulo, et. al., Sources of variation underlying the production of lactose by dairy cows, Journal of Dairy Science, Volume 108, Issue 4, 2025, Pages 4403-4421.

Sadovnikova A, et. al. Transcriptomic changes underlying glucocorticoid-induced suppression of milk production by dairy cows *Frontiers in Genetics* Volume 13 - 2022

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Delivery organisations

The University of Sydney Dairy Research Foundation | NSW Department of Primary Industries | Scibus | Dairy Australia

Partner organisations

NSW Farmers Growing the best | eastAUSmilk | Dairy NSW | Dairy Australia | AFMH | Leppington Pastoral Company Pty Ltd | NSW Local Land Services | Norco

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Sources of variation underlying the production of lactose by dairy cows

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ABSTRACT

This study explored variability in the production of lactose by dairy cows, and the factors underlying it, using herd testing data from New South Wales, Australia. The dataset spanned 14 years (2008–2022) and comprised 393,772 records from 33,280 cows across 85 herds, alongside meteorological and dairy sire genetic data. Variables included milk yield, composition, and quality; reproductive data; breed information; and environmental factors. We found significant variation in lactose yield (LY), lactose percentage (LP), and milk solids yield (MSY) across breed, parity, and stage of lactation. Holsteins had the highest LY and MSY and the lowest LP, whereas Jerseys produced more MSY per unit of LY (MSY:LY ratio). We observed a negative correlation between LY and milk composition variables (fat and protein percentages) and environmental factors such as the temperature-humidity index. Heritability estimates indicated a moderate genetic influence on LY, LP, and MSY:LY (0.24–0.33). The identification of cows producing milk with a consistently lower LP or higher MSY:LY ratio highlights the potential for selective breeding against lactose output to increase the production of milk components and suppress milk volume. These findings highlight the potential to modulate lactose synthesis that could enhance the milk production efficiency of dairy cows, and potentially reduce their environmental impact.
Key words: lactose, variability, dairy industry, productivity

INTRODUCTION

Lactose is the main carbohydrate in cow milk and makes a substantial contribution to its energy content (Tyrrell and Reid, 1965). The biosynthesis of lactose occurs in mammary epithelial cells, which utilize ~65%–70% of the available glucose present in the bloodstream of cows during peak lactation (Baumgard et al., 2017). This intracellular synthesis of lactose establishes an osmotic gradient, drawing water into the epithelium, thereby increasing milk volume (Sadovnikova et al., 2021). Lactose output is positively correlated with the output of milk fat and protein, where the concentration of these components in milk can vary due to a range of different genetic and environmental factors (Osorio et al., 2016; Costa et al., 2019b).

Lactose is also a versatile ingredient that functions as a sweetener, bulking agent, source of energy, and flavor enhancer in various food products (Huppertz and Gazi, 2016). In the pharmaceutical industry, lactose is used as an excipient in drug delivery systems, and it also plays a pivotal role in the production of lactulose (Gänzle et al., 2008). In countries such as New Zealand, lactose is imported to standardize the level of milk components in products such as whole milk powder (Sneddon et al., 2013). However, it is noteworthy that lactose is often excluded from milk payment systems for dairy farmers. In fact, many companies apply volume charge deductions, which inadvertently penalize for lactose production, and only a few incentivize dairy farmers to increase lactose production by factoring it into the payment system (Costa et al., 2019a).

Reducing lactose synthesis and consequent milk volume, without affecting the secretion of other milk components, could result in economic, environmental, and social benefits for the dairy industry. For example, processors would benefit from reduced transport and storage costs (Garrick and Lopez-Villalobos, 2000), increased yields of dairy products, lower water and energy usage, and reduced waste (Haile-Mariam and Pryce, 2017). Additionally, producing the same amount of saleable milk solids (i.e., fat and protein) with a lower lactose output would provide comparable income to farmers while potentially reducing their cows' energy demands, lowering their risk for metabolic diseases, and improving their reproductive performance (Collard et al., 2000). A similar amount of milk solids could also be harvested each day from fewer milkings, or with flexible milking times, leading to positive impacts on

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The list of standard abbreviations for JDS is available at adsa.org/jds-abbreviations-25. Nonstandard abbreviations are available in the Notes.

Final Project Summary

P9b: Milk as an Indicator of Heat Load

May 2026

Dr Rezaul Rakib completed his PhD on a project to investigate the potential to use milk samples as an indicator of heat load.

Milk samples can be collected during routine milking without additional animal handling or discomfort. This offers a non-invasive and convenient alternative to testing though blood and saliva. There's also less room for error or subjectiveness compared to visually assessing a herd for heat load.

This project investigated the potential to use the level of the 'heat shock protein' HSP70 in milk as an indicator of heat load in dairy cattle. HSP70 is a stress-response protein that is naturally present in milk.

Key findings

The research found that milk HSP70 levels reflect heat load with a lag of approximately 33–50 hours, meaning it reflects prior or cumulative cellular stress rather than current conditions.

During sustained heatwave conditions it showed strong diagnostic performance, though its discriminatory power was more limited outside extreme heat events and may be influenced by health or physiological status.

Further validation across herds and seasons is needed, but the work positions milk HSP70 as a promising complementary tool within a broader framework for monitoring heat load. Potential applications include monitoring heat load at the individual cow level, at the herd management level and at the processor level. Additionally, it has the potential for remote monitoring and could be a service offered by milk processors.

Getting more from milk

Milk underpins the dairy industry. Getting more from the precious commodity has to potential to increase its value throughout the entire supply chain.

The Dairy UP team investigated novel ways to get more value from milk.

There were three elements to this research:

- P9a: Producing Milk with Less Lactose
- P9b: Milk as an Indicator of Heat Load
- P9c: Adding Value to Dairy Waste

This document provides an overview of P9b.

More info

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Research and results

The project developed and validated reliable, low-cost measurement methods and sample handling protocols. It also characterised seasonal variation and quantified lag times relative to physiological heat stress measures.

Analytical validation
An "in-house" ELISA test was adapted to detect HSP70 in milk, blood and saliva samples. The method was initially developed by Dr Indunil Pathirana in a collaboration with the Dairy Research Foundation.

ELISA is a laboratory testing technique that detects certain proteins, antibodies, hormones and much more. This ELISA test is cost effective and detects a broader range of HSP70 than commercial kits.

The research found that the ELISA test is reliable way to measure milk HSP70. Strong agreement was demonstrated between the in-house competitive ELISA and commercial ELISA across all sample types (saliva, blood and milk). This confirmed that the ELISA test is a feasible, accurate and affordable option for routine monitoring of heat load from milk samples.

Sample handling and storage
Milk samples were processed, either skimmed before storage or skimmed after storage and stored under controlled conditions (room temperature, refrigeration, freezing, refrigeration + preservative). HSP70 stability was assessed across multiple time points.

The research found that sample handling conditions materially affect HSP70 detection accuracy. Skimming milk before storage and holding samples refrigerated or frozen for up to three days preserved HSP70 integrity.

Storage at room temperature or with bronopol preservative reduced accuracy, with direct implications for how samples should be managed in commercial herd-testing workflows.

Seasonal variation
HSP70 concentrations were measured in milk, blood, and saliva across contrasting seasons and linked to temperature humidity index (THI), milk yield, and milk composition. Statistical models were used to quantify environmental and physiological influences.

The research found that HSP70 concentrations vary substantially between cows and across seasons. Levels were highest in summer and lowest in winter, consistent with seasonal variation in heat load.

Milk HSP70 showed greater variation between samples than blood or saliva, indicating that results from milk are more variable and need to be interpreted with this in mind.

Test sensitivity
Simultaneous measurements of temperature humidity index (THI), rectal temperature, infrared thermography (IRT), and reticulo-rumen temperature were collected to compare the sensitivity and responsiveness of milk HSP70 with established heat stress indicators. Time-lagged correlations (0–48 h) were calculated.

The research found that milk HSP70 reflects prior heat exposure rather than current conditions. Peak milk HSP70 concentrations occurred approximately 33–44 hours after peak temperature humidity index and 48–50 hours after peak reticulo-rumen temperature. This confirmed that HSP70 levels reflect cumulative cellular stress over the preceding one to two days rather than providing a real-time indicator of heat load. This time lag has direct implications for how and when the HSP70 levels can be usefully applied in practice.

Heatwave trial
A five-day heatwave was closely monitored to understand how HSP70 levels changed over time, identify useful thresholds, and assess how accurately it can detect when cows are under heat stress.

The results show that diagnostic performance is strong during sustained heat events but more limited under typical seasonal conditions.

During the heatwave, a milk HSP70 threshold of approximately 550 ng/mL demonstrated good

diagnostic performance.

Under non-heatwave seasonal conditions, discriminatory power was more limited, consistent with the view that milk HSP70 functions most reliably as an indicator when cellular stress is both substantial and persistent.

HSP70 levels may also be influenced by other biological factors not considered in this study, including stage of lactation, lactation number, production level, and health status, which should be considered when interpreting results outside of extreme heat conditions.

Applications

These findings pave the way for future applications for monitoring heat load at the cow, herd and processor/industry level.

Cow/farm level
At the cow and farm level, milk HSP70 provides a quantitative indicator of heat load that can be collected during routine milking. It best suited to retrospective assessment of cumulative heat load rather than real-time intervention decisions. Its specificity to cellular stress, rather than environmental temperature alone, represents an advance over existing THI-based approaches, though its interpretation requires consideration of other factors known to influence HSP70 expression (e.g. stage of lactation or health status). The validated threshold of approximately 550 ng/mL provides an objective, repeatable basis for comparing heat stress severity across cows and seasons in research and monitoring contexts, pending further field validation.

Herd management
Milk HSP70 measurement has potential for integration into herd testing, enabling surveillance of cumulative heat load at the herd level without additional labour requirements. It could also serve as an objective tool for evaluating the effectiveness of heat stress mitigation strategies — such as cooling systems, shade provision, or adjusted milking schedules — providing evidence for both on-farm management decisions and industry-level welfare reporting.

Processor/industry application
Being able to consistently monitor heat load could help processors manage milk supply quality and variability. It could also be used to provide data to underpin sustainability and animal welfare reporting requirements which are increasingly expected by domestic and international markets.

Collaborators

The P9b project was a collaboration between Dairy UP researchers at the University of Sydney and NSW DPIRD and the University of Ruhuna, Sri Lanka.

This project involved a PhD study undertaken by Rezaul Rakib and an Honours thesis prepared by Jade Nguyen.

Related work

Other DairyUP projects investigated heat load from different angles:

- P2d: Heat stress
- P6: Cow response to climate extremes.

Read more

P9 final report

M.R.H. Rakib et. al. Skimming and storage factors affect the detection of heat shock protein 70 in raw bovine milk, JDS Communications, Volume 6, Issue 4, 2025, Pages 578-582.

M.R.H. Rakib et. al. Comparison between in-house competitive and commercial enzyme-linked immunosorbent assays for the detection of heat shock protein 70 in milk, blood, and saliva of dairy cows, Journal of Dairy Science, Volume 109, Issue 4, 2026, Pages 4596-4603

M.R.H. Rakib et. al. Detecting heat shock protein 70 in milk, blood, and saliva of dairy cows exposed to different seasonal conditions, Journal of Dairy Science, Volume 109, Issue 4, 2026, Pages 4647-4658

M.R.H. Rakib, et. al. Graduate Student Literature Review: Potential use of HSP70 as an indicator of heat stress in dairy cows—A review*, Journal of Dairy Science, Volume 107, Issue 12, 2024, Pages 11597-11610

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Delivery organisations

The University of Sydney – Dairy Research Foundation

NSW Government – Department of Primary Industries and Regional Development

Scibus

Dairy Australia

Partner organisations

NSW Farmers – Growing the best

eastAUSmilk – Advocacy · Unity · Service

Dairy NSW | Dairy Australia

AFMH

Leppington Pastoral Company Pty Ltd

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Detecting heat shock protein 70 in milk, blood, and saliva of dairy cows exposed to different seasonal conditions

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ABSTRACT

Heat stress (HS) negatively affects the health, reproduction, and milk production of dairy cows, indicating a major challenge to animal welfare and farm profitability. Although environmental and physiological indicators are commonly used to monitor HS, cellular biomarkers such as heat shock protein 70 (HSP70) offer a promising approach for detecting HS at the molecular level. This research aimed to evaluate and compare HSP70 levels in the milk, blood, and saliva of lactating dairy cows across seasons, assess their environmental responsiveness using the temperature-humidity index (THI) as the standard environmental indicator of HS under temperate Australian conditions, and investigate the potential of milk HSP70 as a noninvasive biomarker of HS at the cellular level. Twenty multiparous Holstein Friesian cows were monitored across 4 seasons (summer, autumn, winter, and spring) over 1 yr, and HSP70 concentrations were measured using a competitive ELISA system. Weather data, including THI, as well as milk yield and composition traits, were also recorded. Results showed significant seasonal variation in HSP70 levels, with the highest concentrations observed in summer across all sample types. Blood exhibited the highest (416.45 ng/mL) HSP70 concentrations overall, followed by saliva (304.89 ng/mL) and milk (279.02 ng/mL). Milk HSP70 showed the highest variability (CV = 53.76%), indicating that additional factors beyond HS may influence its expression, whereas blood and saliva appeared to be more consistent. The associations between HSP70 concentrations in biological fluids were initially evaluated using Pearson correlation analysis. To account for repeated measurements in cows and identify key predictors, linear mixed-effects models with cow identification as a random effect were then fitted. Moderate positive correlations were found between the concentration of milk HSP70 and both blood (r = 0.58) and saliva HSP70 (r = 0.53), indicating that milk HSP70 may reflect systemic stress responses in dairy cows. Multivariable linear mixed models identified average THI as a significant predictor of HSP70 concentrations across fluids. In the milk HSP70 model, DIM was also a significant predictor, and milk yield was not. Overall, this study provides preliminary evidence that milk HSP70 may have potential as a noninvasive indicator of HS in dairy cows. These findings also contribute to a better understanding of HS physiology and may inform future approaches for herd-level monitoring and management under climate change.
Key words: biomarkers, heat stress, HSP70, noninvasive

INTRODUCTION

Heat stress (**HS**) poses a major challenge in dairy farming, reducing milk production, impairing reproduction, and compromising animal welfare (West, 2003; Sammad et al., 2020). Effective monitoring and mitigation of HS are crucial for maintaining dairy cow productivity and farm sustainability. A range of methods, including environmental indicators and physiological, behavioral, and molecular biomarkers, have been developed to detect and monitor HS in dairy cows (Rakib et al., 2024). Among these, visual observation remains the simplest and most widely used method on farms, including signs such as increased respiration rate, panting, drooling, and reduced activity (Shu et al., 2021). Declines in milk yield are also used as indirect evidence of HS; however, they reflect

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Comparison between in-house competitive and commercial enzyme-linked immunosorbent assays for the detection of heat shock protein 70 in milk, blood, and saliva of dairy cows

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ABSTRACT

Heat shock protein 70 (HSP70) has emerged as a potential physiological indicator of heat stress (HS), recognized for its substantial upregulation in response to increased temperatures and cellular stress. The accurate measurement of HSP70 is crucial for its application in research and on-farm monitoring. Commercial ELISA kits are widely available and exhibit high sensitivity; however, their limited working ranges require the dilution of high-concentration samples, which can potentially introduce variability. A recently developed in-house competitive ELISA provides a cost-effective alternative, previously validated for bovine HSP70 detection; however, its performance across various biological matrices has not been systematically evaluated against commercially available systems. A secondary objective was to determine whether HSP70 levels measured in less-invasive matrices such as milk and saliva, correspond to those obtained from blood. This study compared an in-house competitive ELISA to a commercial sandwich ELISA kit for detecting HSP70 in milk, blood, and saliva samples of dairy cows. Samples were analyzed with optimized methods, and assay performance was evaluated using descriptive statistics, Pearson correlations, and linear regression models. As expected for assays using different formats and calibration systems, the in-house ELISA indicated elevated absolute concentrations across all matrices; however, these differences were systematic and did not affect the strong agreement between methods. Blood samples had the lowest CV in both assays (2.76% in-house; 5.89% commercial), indicating greater repeatability than milk (5.69% and 10.24%) and saliva (7.5% and 9.95%). The consistently lower CV for the in-house method across all fluids indicates that it is more precise and can lead to higher statistical power than commercial assays for a given sample size. Strong positive correlations were observed between assays across all matrices (milk r = 0.91; blood r = 0.88; saliva r = 0.87). Regression analysis validated these relationships, with R^2 values of 0.777, 0.833, and 0.752 for milk, blood, and saliva, respectively. The findings indicate that the in-house ELISA closely aligns with the performance of the commercial kit. Despite the lower detectable minimum concentration of the commercial assay, the in-house assay provides a more economical alternative for evaluating physiologically high and moderate HSP70 levels; however, the commercial kit remains widely available for purchase. Among biological matrices, plasma exhibited the highest cross-assay correlation, whereas milk and saliva showed consistent relative trends, indicating their potential as noninvasive alternatives for HS monitoring in dairy cows.

Key words: biomarkers, heat stress, heat shock protein 70, noninvasive

INTRODUCTION

Heat stress (**HS**) is a major constraint to dairy production worldwide, negatively affecting the welfare, health, and productivity of dairy cows (Wankar et al., 2021; Thornton et al., 2022). Among the physiological indicators of HS, heat shock protein 70 (**HSP70**) has gained substantial attention as a potential biomarker due to its upregulation in response to elevated temperatures and cellular stress (Hyder et al., 2017). Heat shock protein 70 plays a key role in maintaining protein homeostasis by promoting proper protein folding, stabilizing newly synthesized proteins, and preventing the aggregation of

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Short Communication
Physiology

Skimming and storage factors affect the detection of heat shock protein 70 in raw bovine milk

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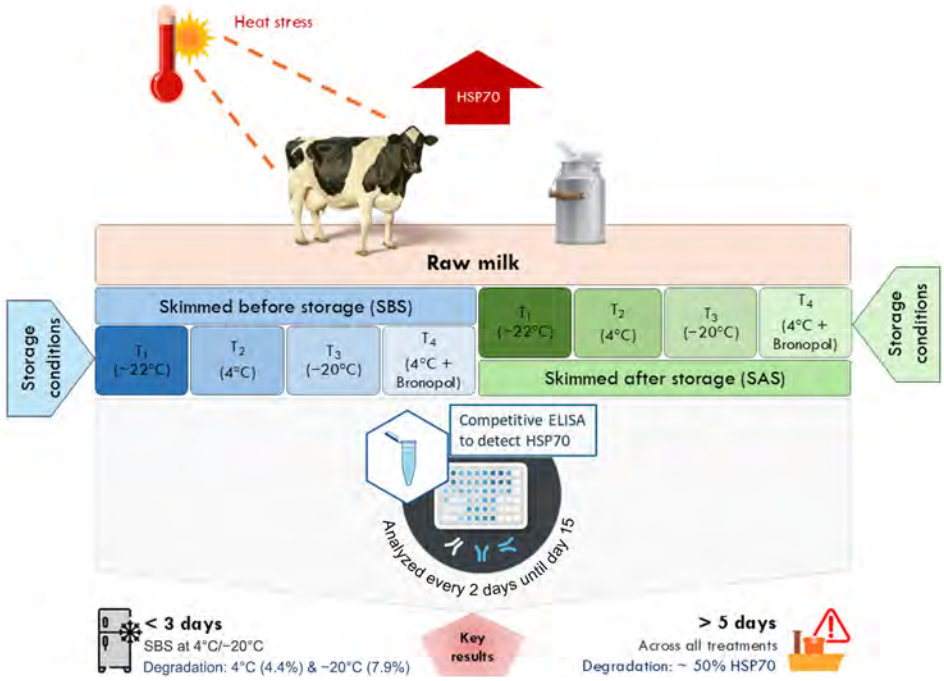
Graphical Abstract



Summary

This study evaluated the impact of milk sample handling on the detection of heat shock protein 70 (HSP70) in bovine milk by using a competitive enzyme-linked immunosorbent assay (ELISA) system. Samples were processed as skimmed before storage (SBS) or skimmed after storage (SAS) and stored under various conditions: room temperature (~22°C; T_1), refrigeration (4°C; T_2), freezing (−20°C; T_3), and refrigeration with preservative (4°C + bronopol; T_4). Samples were analyzed every 2 days over 15 days to assess HSP70 stability. Results showed that SBS samples stored at 4°C or −20°C for up to 3 days preserved HSP70 levels with minimal degradation (4.4% and 7.9%, respectively), whereas SAS samples showed significantly higher degradation (44.2%–53.9%). Room temperature storage led to the greatest degradation, and bronopol use did not consistently maintain HSP70 levels. These findings highlight the importance of optimizing milk storage to preserve HSP70, a potential biomarker for cellular and heat stress in dairy cows.

Highlights

- Skimming milk before storage reduces HSP70 degradation compared with after storage.
- Skim milk stored at 4°C or −20°C preserves HSP70 levels effectively for up to 3 days.
- Storage beyond 5 days causes ~50% HSP70 degradation across all treatments.

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Short Communication
Physiology

Skimming and storage factors affect the detection of heat shock protein 70 in raw bovine milk

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Abstract: Heat shock proteins (HSP) play a vital role in maintaining cellular function under elevated temperatures. Among these, heat shock protein 70 (HSP70) has recently been detected in bovine milk using competitive ELISA, highlighting its potential as a noninvasive marker of cellular stress and a tool for detecting heat stress (HS) in dairy cows. However, optimal management and storage conditions of milk samples for detection of HSP70 are yet to be determined. This study aimed to evaluate how different management and storage practices affect detection of HSP70 in milk samples. Samples were skimmed by centrifugation either before storage (SBS) or after storage (SAS), and then stored under different temperatures: (a) room temperature (T_1, ~22°C), (b) refrigerated (T_2, 4°C), (c) frozen (T_3, −20°C), and (d) refrigerated at 4°C with a preservative (bronopol; T_4). Samples were analyzed every 2 d until d 15 (d 1, d 3, …, d 15) using a competitive ELISA system (sensitivity range 31.25–2,000 ng/mL) to detect HSP70 levels. Result showed that SBS samples had the smallest reduction in HSP70 levels on d 3 when stored at T_2 or T_3, with decreases of 4.4% and 7.9%, respectively. In contrast, SAS samples exhibited greater reductions (44.2% at T_2 and 53.9% at T_3). Samples stored at room temperature (T_1) showed the greatest degradation, especially in SAS samples, whereas the use of a preservative (T_4) did not consistently maintain HSP70 levels in either SBS or SAS samples. The HSP70 detection showed lower variability during early storage (d 1–3) across all treatments, with SBS samples demonstrating greater stability than SAS; however, variability significantly increased by d 9 in both sample types. An apparent increase in HSP70 levels was observed from d 11 to 15 in both SBS and SAS, suggesting the influence of protein stability and interactions affecting ELISA reactivity, where further research is required. Overall, samples stored as skim milk until d 3 at 4°C or −20°C preserved HSP70 levels more effectively than whole milk, with room temperature storage being the least favorable. This pioneering study explored how sample storage and management affect HSP70 detection, providing practical guidelines for farmers and processors to preserve milk sample integrity for assessment of HSP70 as indicator of cellular stress and, potentially, HS.

Heat stress (**HS**) in lactating dairy cows not only reduces feed intake, milk yield, and reproductive performance, but also affects milk quality and composition, which has direct implications for dairy processing and product consistency (Becker et al., 2020; Rakib et al., 2020). Managing HS has become more complex due to the increasing number of high-producing animals, which are metabolically more active (Polsky and Von Keyserlingk, 2017), and the ongoing impact of climate change, with more frequent and severe heat events (IPCC, 2021).

Several methods for detecting HS in dairy cows have been explored, ranging from physiological and behavioral measurements to advanced applications of remote sensing and machine learning technologies (Rakib et al., 2024). Identifying practical biomarkers for HS is crucial for improving herd management and minimizing production losses. Among potential biomarkers, HSP70 is of particular interest due to its critical role in cellular stress response in dairy cattle and other livestock species, including pigs and poultry (Gaughan et al., 2013). HSP70 helps protect cells under stress by facilitating proper protein folding and preventing the aggregation of denatured proteins. By binding to misfolded proteins, it helps maintain protein homeostasis and limits the accumulation of cellular damage during HS (Mosser and Morimoto, 2004; Mayer and Bukau, 2005). Additionally, HSP70 is involved in signaling and regulatory pathways that enhance the cellular adaptive response. Its expression is upregulated through a complex network involving heat shock factors (**HSF**), element-binding proteins, and co-chaperones (Voellmy, 2004). Given its role in maintaining cellular balance and its release both within and outside cells in response to stress, HSP70 shows strong potential as an indicator of tissue-level stress (Rakib et al., 2024). However, HSP70 is not specific to HS alone; its expression can also be induced by other physiological stressors, including inflammation and disease conditions such as mastitis (Hyder et al., 2017). Understanding these influences is essential for interpreting HSP70 levels accurately and for exploring its broader diagnostic potential. Nevertheless, HSP70 potentially remains as a valuable supplementary tool for identifying heat stress or resilience in individual animals, supporting genetic selection, and informing herd management strategies (Rakib et al., 2024).

Although increases in HSP70 levels in blood and saliva have been associated with HS in cattle (Haque et al., 2012; Lamy et al.,

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Graduate Student Literature Review: Potential use of heat shock protein 70 as an indicator of heat stress in dairy cows—A review*

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ABSTRACT

Heat stress (HS) poses significant challenges to the dairy industry, resulting in reduced milk production, impaired reproductive performance, and compromised animal welfare. Therefore, understanding the molecular mechanisms underlying cellular responses to HS is crucial for developing effective strategies to mitigate its adverse effects. Heat shock protein 70 (HSP70) has emerged as a potential player involved in cellular thermotolerance in dairy cows. This review provides a comprehensive overview of the role of HSP70 as a molecular chaperone in cellular thermotolerance in dairy cows under HS. HSP70 facilitates proper protein folding and prevents the aggregation of denatured proteins. By binding to misfolded proteins, it helps maintain protein homeostasis and prevents the accumulation of damaged proteins during HS. Additionally, HSP70 interacts with various regulatory proteins and signaling pathways, contributing to the cellular adaptive response to HS. The upregulation of HSP70 expression in response to HS is regulated by a complex network involving heat shock factors (HSF), heat shock element-binding proteins, and HSF co-chaperones. Therefore, HSP70 holds the potential to be a useful indicator of tissue stress due to its role in maintaining cellular balance and because it is released both inside and outside cells in response to stress. Traditional methods of measuring HSP70 in blood samples are labor intensive, and because the process is potentially stressful for the animals, this may subsequently affect the results. Therefore, measuring HSP expression in cow milk has shown promise as an easy, noninvasive, and accurate way to detect HS in dairy cows. Monitoring HSP70 levels in milk can be applied as a supplementary approach to identify HS or HS resistance of individual cows, select suitable animals, and guide targeted management strategies. However, despite the potential advantages of using HSP70 as a biomarker for monitoring HS on dairy cows, challenges remain in standardizing measurement protocols, establishing species-specific reference ranges, addressing interindividual variations, and determining the specificity of changes in HSP70 due to HS. Future research should focus on developing noninvasive techniques for HSP70 detection, with consideration of climatic conditions and unraveling the molecular interactions and regulatory networks involving HSP70.
Key words: animal welfare, biomarker, ELISA, heat shock protein, milk

INTRODUCTION

Heat stress (**HS**) in dairy cows refers to an environment that raises the body temperature of cows due to exposure to high temperatures and humidity levels beyond their ability to dissipate heat effectively (Dunshea et al., 2013; Hyder et al., 2017). This can lead to reduced feed intake, lower milk production, and compromised reproductive performance in cows (Bernabucci et al., 2014; Polsky and Von Keyserlingk, 2017; Becker et al., 2020; Rakib et al., 2020). To cope with HS, cows attempt to regulate their body temperature through panting and sweating, leading to increased water consumption and dehydration (Islam et al., 2021). A combination of observed behavioral changes and physiological indicators are used to diagnose HS in cows, including monitoring increased respiration rates, rectal temperature, panting, drooling, and reduced activity (Tresoldi et al., 2018).

The ability of an animal to maintain homeostasis in response to thermal stress is known as thermotolerance, and it is essential for survival under these conditions. Heat shock proteins (**HSP**) are a class of molecular chaperones (i.e., assistants or helpers) that play a crucial role in maintaining cellular homeostasis and promoting thermotolerance in cells exposed to high temperatures (Mayer and

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Final Project Summary

P9c: Adding value to dairy waste

May 2026

What a waste

In Australia, about 16% (equivalent to 1,300,000 tonnes/year) of total on-farm milk production is discarded as waste. Most of this wastage occurs post farm gate, for example at processing, distribution, food service and consumer stages of the supply chain.

The effect of this wastage is not just economic; it also has an impact on the environment and food security.

Although some efficiency could be gained by reducing total waste, it is also possible to turn what would be wasted into a valuable product. For example, dairy waste can be recycled and repurposed using existing methods such as reverse osmosis, drying, hydrolysis, ultrafiltration, and electrodialysis.

Fermentation

The Dairy UP team investigated fermentation opportunities to convert dairy wastes into value-added products with an existing commercial market. Fermentation uses microbes such as yeasts to convert dairy wastes into valuable products.

Fermentation can be cost-effective, can generate new income streams for farmers and processors as well as reduce waste.

Fermentation can produce a range of compounds, including stockfeed supplements.

Getting more from milk

Milk underpins the dairy industry. Getting more from this precious commodity has the potential to increase its value throughout the supply chain.

The Dairy UP team investigated novel ways to get more value from milk.

There were three elements to this research:

- P9a: Producing milk with less lactose
- P9b: Milk as an indicator of heat load
- P9c: Adding value to dairy waste

This document provides an overview of P9c.

Key findings

Fermentation is feasible

This work confirmed that microbial fermentation of dairy food waste streams at the manufacturer level, could be a practical pathway to generate value-added products, reduce waste disposal costs, and create new revenue streams. This is particularly the case for cheese whey and whey -derived byproducts. For processors currently spending significant resources on waste management, even partial use of these streams could significantly improve the economics of waste handling.

From an environmental perspective, converting dairy food wastes into useful products reduces the volume requiring disposal and lowering associated greenhouse gas emissions, water use, and environmental compliance costs. This approach directly supports the circular economy principles that are increasingly expected by regulators, retailers and consumers in Australian and overseas markets.

Overcoming a technical constraint

In the laboratory, the team identified a technical constraint: standard yeast strains are unable to

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metabolise lactose. To address this, they were able to engineer a food-grade yeast strain capable of growing on lactose and producing provitamin A. The strain also demonstrated growth on 50% milk medium supplemented with lactose, confirming feasibility of fermenting dairy food waste. While currently at laboratory scale, this established a functional platform for further development.

Application
Significant technical and economic development is required before commercial application; however, the work establishes a clear foundation and a defined set of next steps toward scalable bioproduction from Australian dairy food waste.

More info

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Research and results

The project aimed to:

- **Identify opportunities** by reviewing and mapping dairy food waste streams in Australia and overseas and consulting with industry stakeholders to determine those with the greatest opportunity for reduction.
- **Investigate microbial fermentation** options for adding value to dairy food waste.
- **Pilot/proof-of-concept studies** to develop yeast strains capable of producing valuable compounds from dairy waste.

Fermentation opportunities

The review confirmed that dairy food waste streams are well suited to bioproduction in terms of volume, composition and consistency. This supports the case for investment in this approach, as a practical circular economy strategy. The two biggest opportunities for using fermentation to add value to dairy food waste identified were in the manufacturing sector and on farm.

On farm
Although dairy food waste on farm represents only about 1% of the total, a significant proportion comprises milk with antibiotics fed to calves. This represents a potential risk and opportunity for further investigation. For example, waste milk could be used to produce microbial protein (also called single-cell protein) as an animal feed or ration additive to improve digestive efficiency. It may also be possible to use yeast to degrade antibiotics present in waste milk and then feed the antibiotic-free milk or by-product to calves, thus preventing the development of antibiotic resistance.

Manufacturing
Manufacturing waste makes up 70% of all dairy food waste in the supply chain (Dairy Australia, 2023).

Cheese whey and derived by-products were identified as the most suitable streams for microbial fermentation. They are produced in concentrated and relatively consistent volumes at manufacturing facilities, making them more amenable to scalable bioproduction than household or food service waste, which is compositionally variable and logistically dispersed.

Yeasts

The Dairy UP team focussed on fermentation using brewer's or baker's yeast (*Saccharomyces cerevisiae)* which is safe for humans and animals. Yeast is already widely used for fermentation, so processes and equipment are readily available. There are also existing paths to market for the end products.

Lactose is a key component of dairy wastes, so the focus was on developing yeast that can grow on lactose. The team identified opportunities to use yeast fermentation of lactose in dairy waste to produce:

- Functional milks that are high in specific compounds that could be used as dietary supplements in the cow's diet.

DairyUP
Unlocking potential

- Enzymes and binders that reduce human and animal health risks such as mycotoxins in stockfeed.
- Enzymes and organic acids that improve stock feed quality.
- Supplements that optimise cow nutrition (e.g. proteins, amino acids and probiotics).
- Production of ethanol as a biofuel.

Determining viability

Researchers at Macquarie University's Genome Foundry used state-of-the-art synthetic biology technology to develop a yeast capable of efficiently fermenting lactose to produce valuable compounds.

As a proof-of-concept, yeast strains were engineered to produce provitamin A from lactose. Provitamin A is a precursor of vitamin A, has antioxidant properties and is widely used as a dietary supplement for humans and stockfeed.

The research identified a key technical constraint in that standard yeast strains (e.g. *S. cerevisiae*) are unable to metabolise lactose. Alternative species, such as *Kluyveromyces marxianus*, offer partial solutions but present performance and regulatory limitations. Engineered *S. cerevisiae* strains can overcome this constraint but require further optimisation for industrial-scale efficiency.

A food-grade yeast strain capable of growing on lactose and producing provitamin A was successfully engineered. The strain also demonstrated growth on 50% milk supplemented with lactose, confirming feasibility of fermenting dairy food waste. While currently at laboratory scale, this established a functional platform for further development.

The proof-of-concept demonstration of growth on lactose substrates and production of a nutritionally relevant compound provides the evidence base required to justify further investment in scale-up and techno-economic evaluation.

Read more

www.dairyup.com.au

P9 final report

Dairy Australia 2023; Dairy sector food waste action plan.

J.I. Gargiulo, et. al. INVITED REVIEW: Turning Dairy Food Waste into Valuable Products via Yeast Bioproduction, Journal of Dairy Science, 2026

Collaborators

The P9c project was a collaboration between Dairy UP, NSW DPIRD, Macquarie University's Australian Genome Foundry and the University of Sydney.

Delivery organisations

The University of Sydney Dairy Research Foundation | NSW Department of Primary Industries and Regional Development | Scibus | Dairy Australia

Partner organisations

NSW Growing the best | eastAUSmilk | Dairy Australia | AFMH | Leppington Pastoral Company Pty Ltd | NSW Local Land Services | Norco

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Invited review: Turning dairy food waste into valuable products via yeast bioproduction

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ABSTRACT

Dairy food waste (DFW) is a major global issue, with estimates indicating that almost 20% of production is wasted in some regions. Such losses result in economic and environmental challenges but also create opportunities to recover valuable nutrients, reduce costs, and support a more circular dairy industry. This review explores the sources and composition of DFW and highlights strategies to transform it into valuable bioproducts through yeast fermentation, focusing on lactose-fermenting species as near-term platforms and *Saccharomyces cerevisiae* as the longer-term alternative. Key DFW streams, particularly cheese whey and its derivatives, offer nutrient-rich substrates for bioproduction. Yeast fermentation can convert these into value-added products, such as microbial biomass, AA, vitamins, minerals, fatty acids, and enzymes with direct applications in dairy cattle nutrition. Despite this potential, challenges persist, including the inability of industrially preferred yeasts to natively metabolize lactose, the variability in DFW composition, microbial contamination, and economic, logistical, and regulatory barriers. Emerging approaches such as strain engineering, adaptive evolution, and biofoundry-based synthetic biology offer promising solutions. By integrating bioproduction into the dairy cycle, the industry has the potential to reduce DFW, lower greenhouse gas emissions, and create new revenue streams.

Key words: cheese whey, lactose fermentation, single cell protein, synthetic biology

INTRODUCTION

Approximately one-third of the food produced globally is wasted, resulting in substantial economic losses as well as significant environmental and social impacts (FAO, 2019). The dairy industry is no exception, with estimates indicating that ~20% of production is wasted in some regions (FAO, 2011; Dairy Australia, 2024). Dairy food waste (**DFW**) includes milk, milk-derived products, and byproducts that are lost, spilled, discarded, or otherwise not consumed as intended at any stage of the supply chain (Campbell and Feldpausch, 2022). The majority of DFW arises from milk processing byproducts, waste generated during manufacturing operations, and discarded finished products from food service establishments and household consumption.

While traditional waste management approaches can mitigate some of the environmental impacts, they often fail to address the root causes of waste generation, frequently resulting in the loss of nutrients. Waste prevention is therefore recognized as the most effective strategy within the food waste hierarchy (Papargyropoulou et al., 2014). When prevention is not feasible, repurposing, recycling, or adding value to waste streams represents a more sustainable alternative. However, these approaches can be costly and energy-intensive and present challenges for small and mid-sized processors, which frequently lack the capital, scale, or technology required for efficient valorization (Peters, 2005).

Biological valorization strategies have gained increasing attention as pathways to recover value from DFW while supporting circular production systems. In particular, microbial fermentation offers an opportunity to transform waste streams into high-value products, including microbial biomass, enzymes, organic acids, biopolymers, and bioenergy (Sar et al., 2022). Among microbial platforms, yeast stands out due to its robust-

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The list of standard abbreviations for JDS is available at adsa.org/jds-abbreviations-26. Nonstandard abbreviations are available in the Notes.

DairyUP
Unlocking potential

FINAL OUTPUT JOURNAL

P10
Sustainable pathways for surplus calves

P10a - Sustainable pathways for surplus calves (NSW Coast calf-pathways)
P10b - Calf pathways economics

DairyUP
Unlocking potential

Final Report

P10a Sustainable pathways for surplus calves (NSW Coast calf-pathways)

Dairy Australia

This project was originally led by Sarah Bolton, formerly of Dairy Australia

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
- De-risking the industry and
- Developing new markets.

A key part of Dairy UP was a coordinated network of partner farms across New South Wales (and beyond) providing valuable insights into real world application of new practices, including the challenges and benefits of new innovative technologies.

Dairy UP made a big contribution to dairy research and development rejuvenation, (attracting new researchers, PhD students and Industry investment).

Dairy UP was funded through the Australian and NSW Government's Bushfire Industry Package – Sector Development Grant (BIP-SDG) program, with cash co-contributions from Dairy Australia, The University of Sydney's Dairy Research Foundation, Local Land Services, Norco, Leppington Pastoral Co Ltd and Dairy NSW; and in kind contributions by all the above organisations plus NSW DPI Biosecurity and Food Safety; Agriculture), Scibus, Australia Fresh Milk Holding Ltd, Dairy Connect and NSW Farmers.

This project was delivered jointly by University of Sydney's Dairy Research Foundation, Scibus, Department of Primary Industries and Regional development.

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Delivery organisations

The University of Sydney Dairy Research Foundation | Department of Primary Industries and Regional Development | Scibus | Dairy Australia

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Project Overview

This project was originally developed and led by Dairy Australia. Dairy UP enhanced its delivery for the benefit of the NSW and broader dairy industry. The project was completed in June 2023 and the final report/project summary is included here.

Item	Description
Project Title	Sustainable pathways for surplus calves (NSW Coast calf-pathways)
Funding Body	Dairy UP
Dairy UP Project	P10a
Project Duration	2021-2023
Lead Organisation	Dairy Australia
Project Lead	Sarah Bolton
Research team	Pip Band, Howard Parry-Husbands. Morgan Owen, Ellie Holes
Key Collaborators	Band Consulting, Dairy Australia, Pollinate

Link to Final Report from Band Consulting – Dairy Australia – Pollinate:

[Read more]

METAMORPHOSIS

Final Report: NSW South Coast Calf-pathways

Pip Band, Howard Parry-Husbands, Morgan Owen, Ellie Holmes

October 2022

Pollinate BAND CONSULTING DairyUP Unlocking potential Dairy Australia

Executive Summary

The South Coast Surplus Calf Project ran from October 2020 to October 2022. This timeline was extended by almost 12 months due to a combination of factors including COVID and repeated flooding on the South Coast.

This report provides an overview of the project approach, results and learnings. The report then provides a high-level summary of each phase of the project.

The project successfully:

- Identified multiple pathways for surplus calves on the South Coast
- Formed relationships resulting in project development
- Developed a roadmap for how to effectively engage dairy farmers in co-creation with multi-stakeholders
- Identified insights from consumers and supply chain that enabled a reframe and shift in mindset for both farmers and consumers
- Proved that it's possible to build trust across different stakeholder groups using a co-design approach

Key farmer insights

- Economics dominates
- See a power imbalance with processors / retailers having all the power
- Sharing consumer perspective with farmers broke down the 'us' vs. 'them' mentality, motivating them to listen and align with consumers
- Not in the right headspace to feel motivated
- Solutions felt to have high input of time and capital, when they already feel constrained by time
- A lack of 'proof' further fuels hesitancy
- Sense they're risking more than what they will gain
- BUT They want to be involved in finding a viable solution!

Key consumer insights

- Generally unaware, but not surprised
- Don't see farmers as the bad guys
- View farmers as victims of the food system
- Consumer guilt, not animal welfare is the real issue…
- Details can be accessed through the above link.

Key supply chain insights

- Universal acceptance of significant risk
- Farmers need to understand what the market wants
- Gap is closing to address the issue
- Circular economy opportunity to create value out of a current 'waste product'
- Prejudice to dairy changing at processor/ retailer but challenges remain

Ultimately, farmers need "sideways support" – build capability nor dependency.

Most farmers lack the capacity to easily invest in change. Their time is short, every day is busy, money is tight, innovation exposed knowledge and skills gaps.

The rest of the supply chain needs to invest/build capacity.

There is no guarantee of support, reward or return. So, why would farmers take the risk?

Even if they had 'spare' capacity, there is no certainty which makes it impossible to calculate the risk.
Farmers needed support/certainty to invest in becoming better farmers.

In conclusion

Farmers needed "sideways support" to kick start solutions/s and help make farmers even better at what they do.

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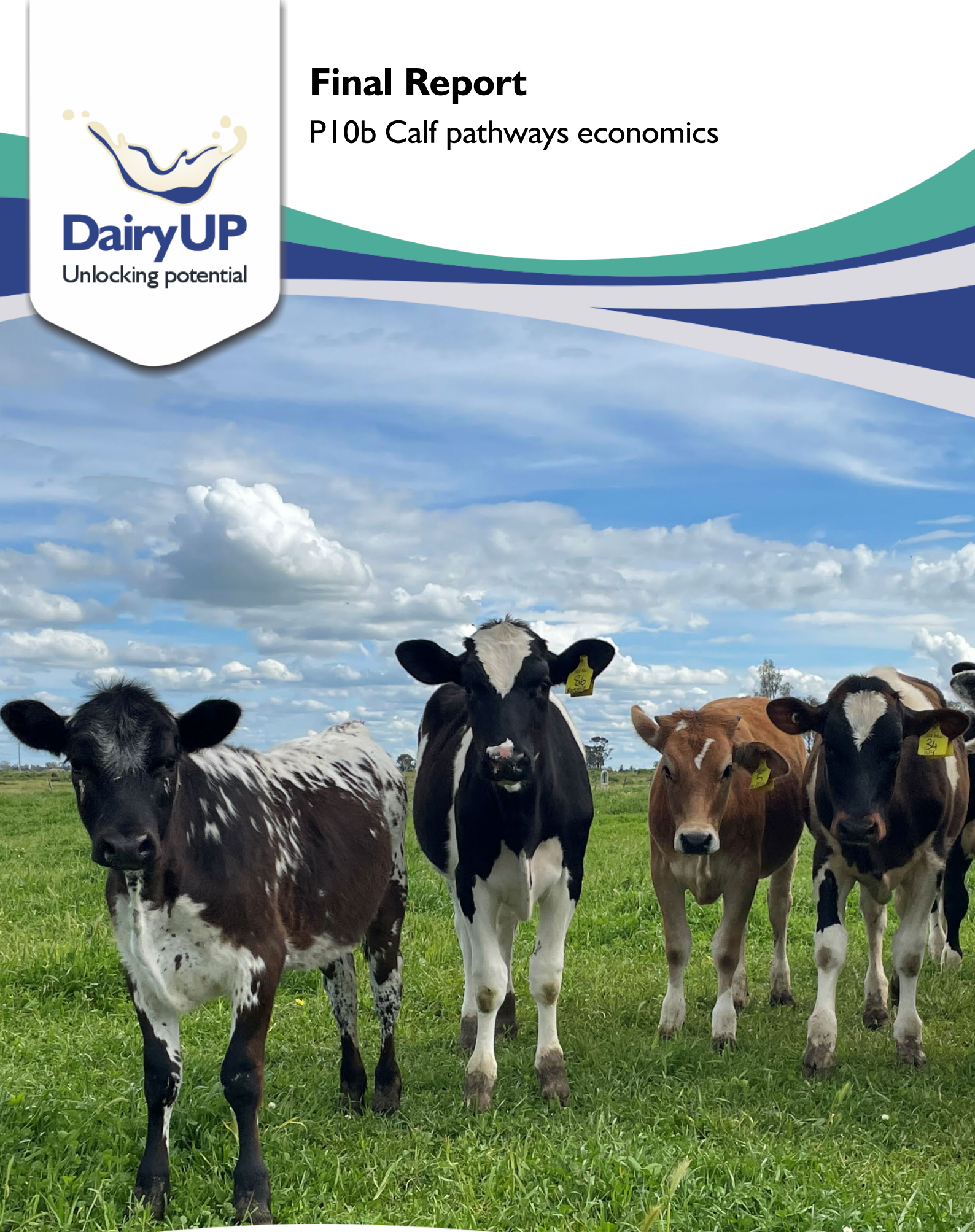
Final Report

P10b Calf pathways economics



RMCG

This project was led by Dan Armstrong,
RM Consulting Group

Dairy UP (Phase 1) was a $16 million, five-year industry driven project with a portfolio of 10 research, development and adoption projects collectively aiming to realise three primary objectives:

- Increase Productivity and Profitability by unlocking the potential of milk, the cow and water,
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Project Overview

This project was originally developed by Dairy Australia and led by RM Consulting Group. Dairy UP enhanced its delivery for the benefit of the NSW and broader dairy industry. The project was completed in June 2023 and the final report/project summary is included here.

Item	Description
Project Title	Calf pathways economics
Funding Body	Dairy UP
Dairy UP Project	P10b
Project Duration	2021-2023
Lead Organisation	RM Consulting Group
Project Lead	Dan Armstrong
Research team	Tristan Wardley, Lauren Jones, Sheena Carter and Blake Cheer
Key Collaborators	Dairy Australia, NSW Department of Primary Industries and Regional Development, RM Consulting Group

RMCG

AUGUST 2023

Calf Pathways Phase 2: Economic Impact Assessment

Summary Report

Dairy Australia

135 Mollison Street, Bendigo Victoria 3550

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Victoria — Tasmania — NSW

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Executive Summary

This project aimed to gain a comprehensive understanding of the costs, benefits, and risks associated with raising surplus dairy calves for the beef market and investigate the economic viability of alternative 'dairy-beef' calf pathways. Through collaboration with a Farmer Reference Group (FRG) and industry, we gained valuable insights into the factors influencing on-farm decisions, and identified limitations in terms of understanding the economic risks associated with rearing surplus calves. We found some of the underlying reasons were due to the anticipated effort required to manage an additional beef enterprise, a lack of available feed and land, beef market volatility and a primary focus on milk production. To address these concerns and delve deeper into the issue, we conducted a detailed economic analysis of two case study farms that examined the additional infrastructure and management requirements associated with raising dairy calves for the beef market. Our analysis yielded the following key findings:

- On average, raising surplus calves for beef resulted in roughly a break-even result. However, the range in net benefit varied from approximately $1000/head in good years to -$600/head in poor years.
- The profitability of the enterprise was primarily influenced by fluctuating beef market prices, which emerged as the largest source of variation in the extra profit/loss generated by raising surplus calves.
- Access to premium markets, such as feedlots, showed potential for increasing profitability. However, it did not significantly reduce the worst-case scenarios associated with market volatility.
- Some dairy farms may require more significant investment in calf rearing facilities if they are to rear all of their surplus calves and will find it challenging to achieve an attractive return on investment unless they can obtain a more stable beef price.
- Growing dairy calves to 450+ kilograms showed potential for higher profitability. However, this approach also exposed the business to downside risks, including increased feed costs, limited land availability, climate variations, and beef market volatility.
- It is important to consider that an increased focus on managing a beef enterprise can divert attention away from the core goal of profitable milk production, which remains the primary business for dairy farmers.

In light of these findings, it is crucial to explore alternative strategies that can enhance the economic sustainability of surplus calves. The active involvement of specialised calf rearers holds potential in bridging the gap between dairy farmers and feedlot operators/meat processors and to help manage economic risks. The contribution that milk processers can make to help manage risk also warrants further investigation.

For this particular region (Taree/Kempsey), some follow up support to foster a regional working group/producer demonstration site may be very beneficial.

A decision support calculator has been developed (Appendix 1) and presented to the FRG and could potentially be tested, refined and circulated more widely.

For this project we adopted a participatory approach with the FRG. This method allows the project team to adapt the project design to meet the needs of participants as the project evolves. This approach proved its value when the FRG identified a key barrier to raising surplus calves was a lack of confidence in the potential demand and value of their product in Australian beef markets. To address this concern, we (RMCG) facilitated discussions between MLA, feedlot representatives and the group, these constructive discussions shed light on the market opportunities associated with dairy beef and outlined actionable steps that dairy farmers can take to improve their products attractiveness to meat processors and feedlots. Although this aspect was not initially in the project scope, it played a pivotal role in bolstering the confidence of dairy farmers within the group, that legitimate pathways for surplus dairy calves into beef markets do indeed exist.

This project has highlighted the need for a better understanding of the costs, benefits and risks associated with raising surplus calves for beef. By understanding the economic implications and collaborating with industry stakeholders (especially within the beef industry), there is potential to develop surplus calf management practices that are both socially and economically sustainable. However, these solutions will be regionally and farm business specific and significant attention needs to be devoted to reduce the proportion of years when there is likely to be a negative return.

ACKNOWLEDGEMENT OF COUNTRY

We acknowledge the Traditional Owners of the Country that we work on throughout Australia and recognise their continuing connection to land, waters and culture. We pay our respects to their Elders past and present, and we acknowledge emerging leaders. Moreover, we express gratitude for the knowledge and insight that Traditional Owners and other Aboriginal and Torres Strait Islander people contribute to our shared work in Australia.

We pay respects to all Aboriginal and Torres Strait Islander communities. We recognise that Australia was founded on the genocide and dispossession of First Nations people and acknowledge that sovereignty was not ceded in this country. We embrace the spirit of reconciliation, working towards self-determination, equity of outcomes, and an equal voice for Australia's First People.

1 Introduction

1.1 BACK GROUND

The dairy industry is a vital component of the agriculture sector and holds an important place in the structure of rural communities throughout Australia. Social license has become an important consideration for the dairy industry as farmers are beginning to face security issues regarding the treatment of bobby calves as part of the dairying process. This problem has led to a growing demand for alternative calf pathways where farmers can diversify their income streams while adhering to societal expectations. One such opportunity is raising dairy calves for the beef market. Traditionally, dairy farmers direct their non-replacement calves either into beef production or into early life slaughter. The pathway undertaken has been highly motivated by economic factors with market perceptions of “dairy-beef” and beef prices being major contributors.

1.2 PURPOSE

The primary objective of this project is to gain a comprehensive understanding of the costs, benefits, and risks associated with raising dairy calves for the beef market. Through this research, we aim to enhance the knowledge and understanding of dairy farmers regarding economically sustainable practices that can effectively extend the life of non-replacement calves on their farms negating potential social licence issues. We aim to aid farmers in making informed decisions that contributes to a future where surplus calves are consistently raised for beef, using practices that can withstand seasonal and commodity price volatility, as well as navigate regionally specific challenges.

1.3 APPROACH

The delivery of this program involved identifying two case study farms and developing a Farmer Reference Group (FRG) in the mid-north coast of NSW. Two workshops were held with the FRG to provide a participatory learning experience and assist with uptake of changes relating to surplus calf management.

By utilising data from the two case study farms, one with year-round calving and one more batch calving, implications for capital infrastructure, feedbase, labour and general trade-offs and interactions with dairy business were identified and used to create a partial budget.

To add value to this project we have also undertaken risk modelling to show the impact of normal variation in our main input prices and production outcomes on the investment outcome. This will allow an estimate, for example, of the relative break-even point of the investment compared to the beef price, expressed as the number of years in ten it will be profitable.

1.4 REPORT

This report will be useful for dairy farmers who are considering diversifying their income streams while also managing expectations around social license. The report will also be relevant to agricultural extension services, policymakers and academics interested in the economics of dairy calf rearing for the beef market. The results of this research project will provide insight into the economic viability of this alternative revenue stream and help farmers make informed decisions on the allocation of their resources, while also considering the societal expectations of their practices.

2 Scenarios

2.1 CASE STUDY FARMS

Two case study farms were used with two broadly different scenarios where we looked at keeping calves to different weights. The farms were located in the central/north coast area of NSW and both operated profitable dairy businesses.

The first case study farm had existing infrastructure and only required a small upgrade to comfortably manage keeping calves for longer.

The second farm required capital investment in an additional calving shed. This farm used a split calving autumn/spring system (not calving in summer and few in winter).

Table 2-1: Farming system description of case study farms

FARM	NO. MILKING COWS	CALVING PATTERN	GRAIN FED (T/COW/YEAR)	FEEDBASE	ADDITIONAL INFRASTRUCTURE REQUIREMENT
Bale	320	Year – round, batch calving	2.1	Milking area (92 ha) Support area (135ha). Kikuyu/annual ryegrass pastures.	$30,000 extension to calf shed
Grey	230	Split – Autumn to Spring and not many in winter	2.0	Milking area (75 ha): 25% perennial pasture (prairie grass, cocksfoot, lucerne and chicory mix) 25% ryegrass/sorghu m 50% ryegrass/ kikuyu Runoff area (33 ha) 55% ryegrass/maize (silage) 45% ryegrass / kikuyu	$75,000 calf shed

3 Approach and Key Assumptions

3.1 OPERATIONAL COSTS

The operational costs associated with raising dairy calves for beef plays a significant role in determining the feasibility of alternative calf pathways. The costs involved include feed, labour, animal health, and other inputs required for the care and management of animals.

Developing robust cost assumptions was a crucial component of the research project, Barber et al. (2020) was a key resource in this regard. By closely examining the operational costs, we can assess the feasibility and potential profitability of this alternative revenue stream. These cost profiles were developed in conjunction with the PRG as well as industry experts and published literature. The following tables present our assumptions and estimates for each of the costs that were impacted by the management changes and are based on our case study farms.

Table 3-1: Operational costs

ASSUMPTION	DESCRIPTION	VALUE
Animal health	$50 Weaning, $50 Weaning to sale	$100.00
Labour costs	Industry average at approximately $34 per hour, 6 hours per day to raise calves and manage beef	$34.00
Electricity and fuel	Extra costs annual	$1,000.00
Forgone income	Price of calf 1-2 weeks old	$50.00
Transport	Cost per head agistment and sales	$8.50
Sales and marketing	5% of total sales	5%

Table 3-2:Production assumptions

ASSUMPTION	DESCRIPTION	VALUE
Weaning days	10 weeks 7 days a week	$70.00
Weaning to 5 months	10 weeks 7 days a week	$70.00
Supplementary feed days fed (Months/days) 5- 10 months old	6 months	$182.50
Average consumption of milk per day per calf (litres)	This may vary from birth to end of weaning average 5.5	$5.50
Average consumption of calf pellets per day (kg)	As above may vary, average per day	$0.50
Average consumption of grain 5-10 Months (kg/day)	As above, average per day	$2.00
Weight at end of weaning (kg)		$100.00

Table 3-3: Feed costs

FOOD SOURCE	DESCRIPTION	ANNUAL COST
Weaning stage		
Milk formula ($/day/calf)	Calf rearing using milk powder – milk powder costed at 47 cpl ($4.00/kg DM) fed at 5.5 litres/calf per day for 10 weeks until weaning.	$231.00
Oaten hay ($/kg DM)	Birth to weaning (KG DM/day) 0.54 - Weaning to 150kg (10 weeks) 0.9	$17.50
Pellets	0.5 kg/Day to weaning	$15.75
Grain cost per week	2kg/day 5-10 months	$127.75
Total weaning cost	Includes: Milk, pellets, hay	$264.25

3.2 ECONOMIC ANALYSIS

Partial budgets were developed according to Malcolm et al. (2005) comprising a 15-year discounted cash flow analysis considering the capital and operational costs and benefits of keeping surplus calves longer to be sold as a mature animal into the beef mark. The key measures used in comparing the profitability and performance of the alternative development options were:

Annual Net Benefit, estimated once the options had been implemented and were fully operational. This comprised of the additional income minus the additional operating costs.

Net Present Value: the sum of future cashflows (positive or negative) over the life of an investment that are discounted to represent their present value.

3.3 @ RISK

The excel add in @Risk (ver. 8.2; Palisade Corporation, Newfield, NY, USA), was used to incorporate risk analysis into the economic model through Monte Carlo simulation. This powerful technique allows us to explore how various factors and uncertainties can influence the project's outcomes. Instead of relying on a single estimate, Monte Carlo simulation considers a range of possible values for key variables (such as beef price or grain price) and generates a distribution of possible results. By analysing this distribution, we gain insights into the likelihood of different outcomes and identify the factors that have the greatest impact on the project's success. This approach improves the information available and allows managers to make better- informed decisions and help understand the uncertainties involved in decisions. The results reported in this analysis are based on 50,000 iterations of 15-year periods.

The program was used to develop a number of probability distributions that are shown below in Table 3-4.

Table 3-4: Probability distribution data sources

DISTRIBUTION	DATA SOURCE
Milk prices	FRG and Dairy Businesses for Future Climates (DBFC) project
Grain prices	FRG and DBFC project
Beef prices	MLA (2023), Prices and Markets. https://www.mla.com.au/prices-markets/
Hay prices	FRG and DBFC project

DISTRIBUTION	DATA SOURCE
Pasture costs	Case study farms DairyBase data

3.4 PRICE DISTRIBUTIONS

The price distributions have been created by capturing information from a range of data sources, as referenced above, and combined with the local knowledge gained through the PRG.

Included in the cost distribution are both the manufacturing and feeder markets across different liveweights. The reason for including the variances is to provide a range of options when selling and to be able to plan a suitable program.

Table 3-5: Distribution of prices used for costs and prices

DESCRIPTION	LOWER 5TH PERCENTILE	UPPER 95 TH PERCENTILE	AVERAGE
Beef price $/kg liveweight manufacturing	$2.21	$4.15	$3.07
Beef price $/kg liveweight feeder	$2.34	$5.48	$3.51
Grazed pasture cost $/kg dry matter (Grey)	$0.15	$0.20	$0.17
Grazed pasture cost $/kg dry matter (Bales)	$0.09	$0.15	$0.11
Conserved pasture cost $/kg dry matter (Grey)	$0.27	$0.33	$0.30
Conserved pasture cost $/kg dry matter (Bales)	$0.19	$0.25	$0.21
Grain price $/tonne	$290.00	$555.00	$390.00
Hay price $/tonne	$270.00	$450.00	$320.00
Milk replacement price $/litre	$0.60	$0.95	$0.75

4 Results and Discussion

4.1 COSTS TO REAR TO VARIOUS AGES

The results from the cost analysis of the Bale farm shows expected costs during different stages of growth below in Table 4-1. If a week-old calf is held until it reaches 600kg, it could be expected that the cumulative cost per animal would be $1,263. During the weaning period, the cost per kilogram of weight gain is significantly higher than the cost per kilogram in subsequent stages of growth because of the high cost of milk per kilogram gained.

Table 4-1: Costs to rear to various ages – Bale Farm

STAGE OF GROWTH	COST PER ANIMAL	CUMULATIVE COST PER ANIMAL	COST PER KG WEIGHT GAINED	FEED INTAKE
Weaning	$338.00	$338.00	$6.76	5.5 Litres milk/Day
100–150kg	$103.00	$441.00	$2.06	5Kg/DM/Day
150–300kg	$319.00	$760.00	$2.13	7Kg/DM/Day
300–450kg	$240.00	$1,000.00	$1.60	11Kg/DM/Day
450–600kg	$262.00	$1,263.00	$1.75	11Kg/DM/Day

At the Bale farm the gathered costs for each kilogram gained are displayed in Figure 4-1 and Table 4-2 below. The range of costs per kilogram during weaning were well above the mean beef price of $3.07 and ranged from $8.08/kg down to $5.65/kg.

After weaning the costs per kg of weight gained drops to below the mean beef price. Calves were moved on to grain and hay until the point where they reached 150kg, at which point pasture was added. Once they reached 300kg they were taken off grain and fed mainly on pasture with some supplementary hay. When the animals reached 300kg and were mainly fed pasture, the costs were low with a maximum of just $2.07 and minimum of $1.37 and there was less variability in the range of cost prices. During the later stages of growth, the cost per kg of weight gain improves, leading to more efficient weight gain outcomes. However, the area of land required to rear the surplus animals increases if they are kept until the later stages.

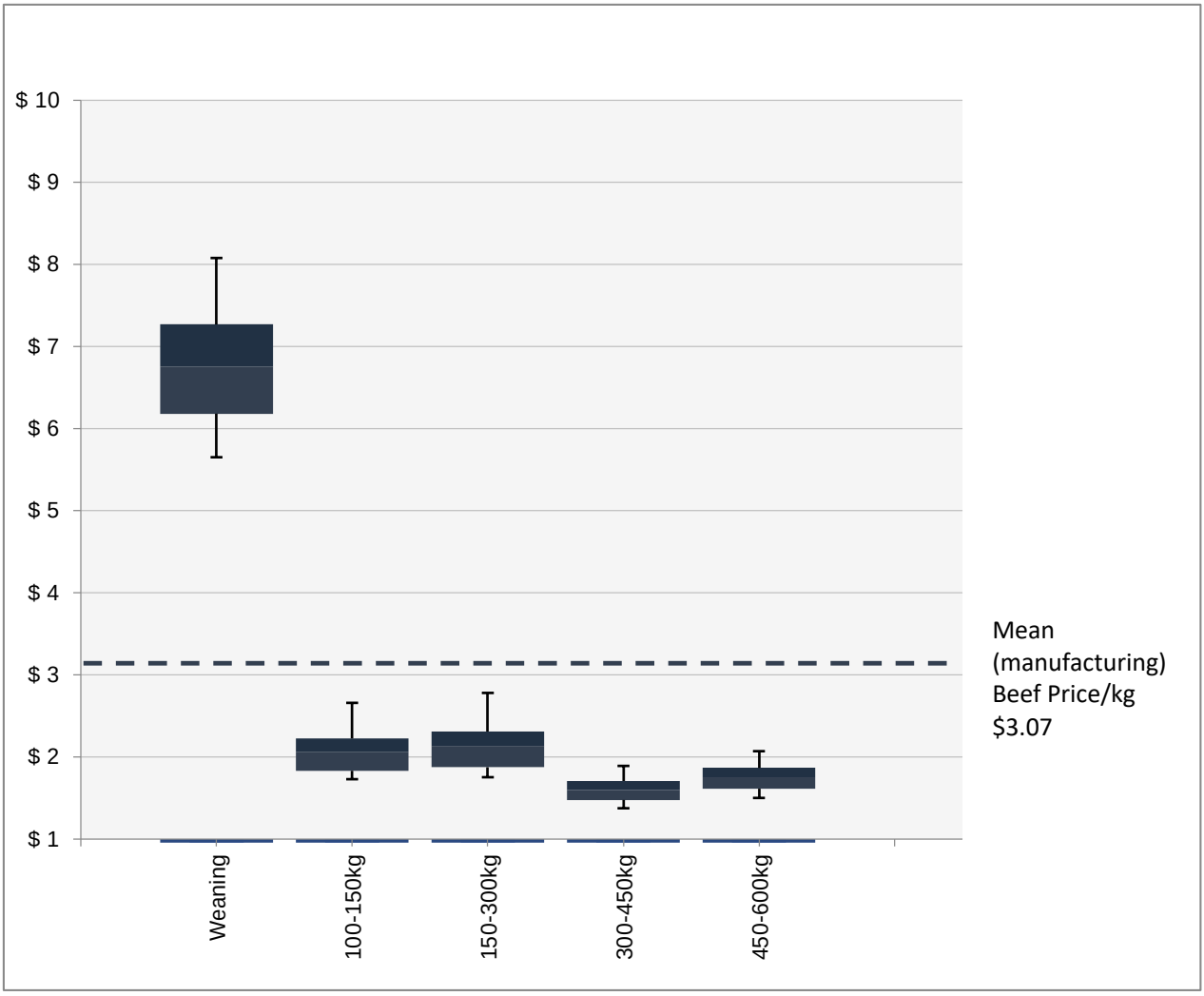

Figure 4-1: Costs per kilogram of weight gained during each stage of growth including the mean beef price per kg of $3.07 – Bale Farm

Table 4-2: Cost ranges per kilogram of weight gained during each stage of growth – Bale Farm

STAGE OF GROWTH	MAXIMUM PRICE PER KG	UPPER 75TH PERCENTILE PRICE PER KG	MEDIAN PRICE PER KG	LOWER 25TH PERCENTILE PRICE PER KG	MINIMUM PRICE PER KG
Weaning	$8.08	$7.27	$6.76	$6.18	$5.65
100–150kg	$2.66	$2.22	$2.06	$1.83	$1.73
150–300kg	$2.78	$2.31	$2.13	$1.87	$1.75
300–450kg	$1.89	$1.71	$1.60	$1.48	$1.37
450–600kg	$2.07	$1.87	$1.75	$1.61	$1.50

The Grey farm costs were analysed below in Table 4-3. In comparison to the Bale farm, the cumulative cost of an animal held until it reached 600kg was $1,474. The Grey farm had an additional $211 per head in costs.

The costs up to 150kg are identical to the Bale farm. However, the cost per animal and cost per kg weight gained is higher than the Bale farm following that growth stage.

Due to the Grey farm being further north than the Bales, it is more difficult to grow ryegrass and more challenging to maintain the quality of kikuyu pasture. Hence, they have moved to more cropping, which has some additional costs.

Table 4-3: Grey Farm Cost to rear to various ages

STAGE OF GROWTH	COST PER ANIMAL	CUMULATIVE COST PER ANIMAL	COST PER KG WEIGHT GAINED	DAILY INTAKE
Weaning	$338.00	$338.00	$6.76	5.5 Litres milk/Day
100–150kg	$103.00	$441.00	$2.06	5Kg/DM/Day
150–300kg	$335.00	$776.00	$2.23	7Kg/DM/Day
300–450kg	$333.00	$1,109.00	$2.22	10Kg/DM/Day
450–600kg	$365.00	$1,474.00	$2.43	11Kg/DM/Day

The Grey farm had a similar pattern to the Bale farm for costs per kilogram of weight gained as shown in Figure 4-2 and Table 4-3 below. The weaning prices were identical and well above the mean beef price, however following weaning the costs were below the mean beef price. It appears that during the growth stages between 100- 300kg they have lower minimum costs of $1.73 and $1.86 per kg with a higher variability in costs of close to $1. During the 300–450kg stage, minimum costs are $1.99 per head and variability is just $0.52 between the minimum and maximum cost. The minimum cost during the 450–600kg stage is $2.18 per head with a variability of $0.57 to the maximum cost.

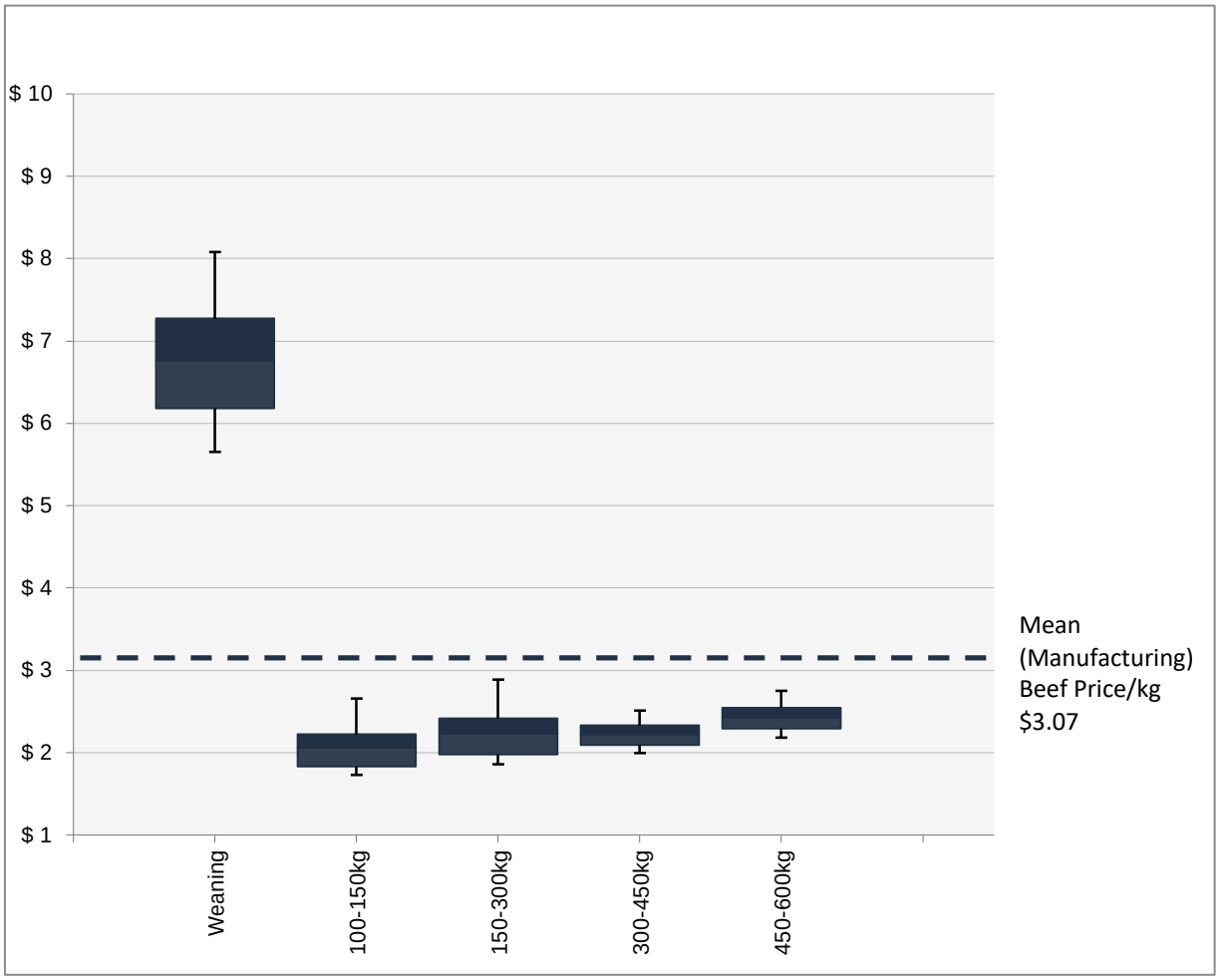

Figure 4-2: Costs per kilogram of weight gained during each stage of growth including the mean beef price per kg of $3.07 – Grey Farm

Table 4-4: Cost ranges per kilogram of weight gained during each stage of growth– Grey Farm

STAGE OF GROWTH	MAXIMUM PRICE PER KG	UPPER 75TH PERCENTILE PRICE PER KG	MEDIAN PRICE PER KG	LOWER 25TH PERCENTILE PRICE PER KG	MINIMUM PRICE PER KG
Weaning	$8.08	$7.27	$6.76	$6.18	$5.65
100–150kg	$2.66	$2.22	$2.06	$1.83	$1.73
150–300kg	$2.89	$2.41	$2.23	$1.98	$1.86
300–450kg	$2.51	$2.33	$2.22	$2.10	$1.99
450–600kg	$2.75	$2.55	$2.43	$2.29	$2.18

4.2 ANNUAL NET RETURN FROM RAISING SURPLUS CALVES FOR BEEF

The annual net return range for each farm has been calculated to work out the additional profit by market for manufacturing steers and feeder steers and the range of returns that can be expected. It is important to note that these outcomes are a result of all farm inputs. Therefore, the range of outcomes is influenced by input prices as well as beef prices. In years where beef prices are high and feed costs are low higher profits will be achieved. The counterfactual is true for high input costs and low beef market prices.

The Bale farm annual average net return is displayed below in Figure 4-3, and the profit range in Table 4-5. In the manufacturing market, the Bales could expect a profit if the beef price is at or above the median price. The maximum profit they could expect in the manufacturing market is $173k for a 600kg animal or $106k for a 450kg animal.

In the feeder market, annual net return is at the highest for a 450kg animal, reaching a maximum profit of $228k and losses are not as high. A 300kg animal sold in the feeder market reaches a maximum profit of $115k and is slightly below break-even if the beef price is at the median range.

A profit would not be achieved, and in fact, a loss would be made, if the beef price in both the manufacturing and feeder market is at the lower 25th percentile or at the minimum price.

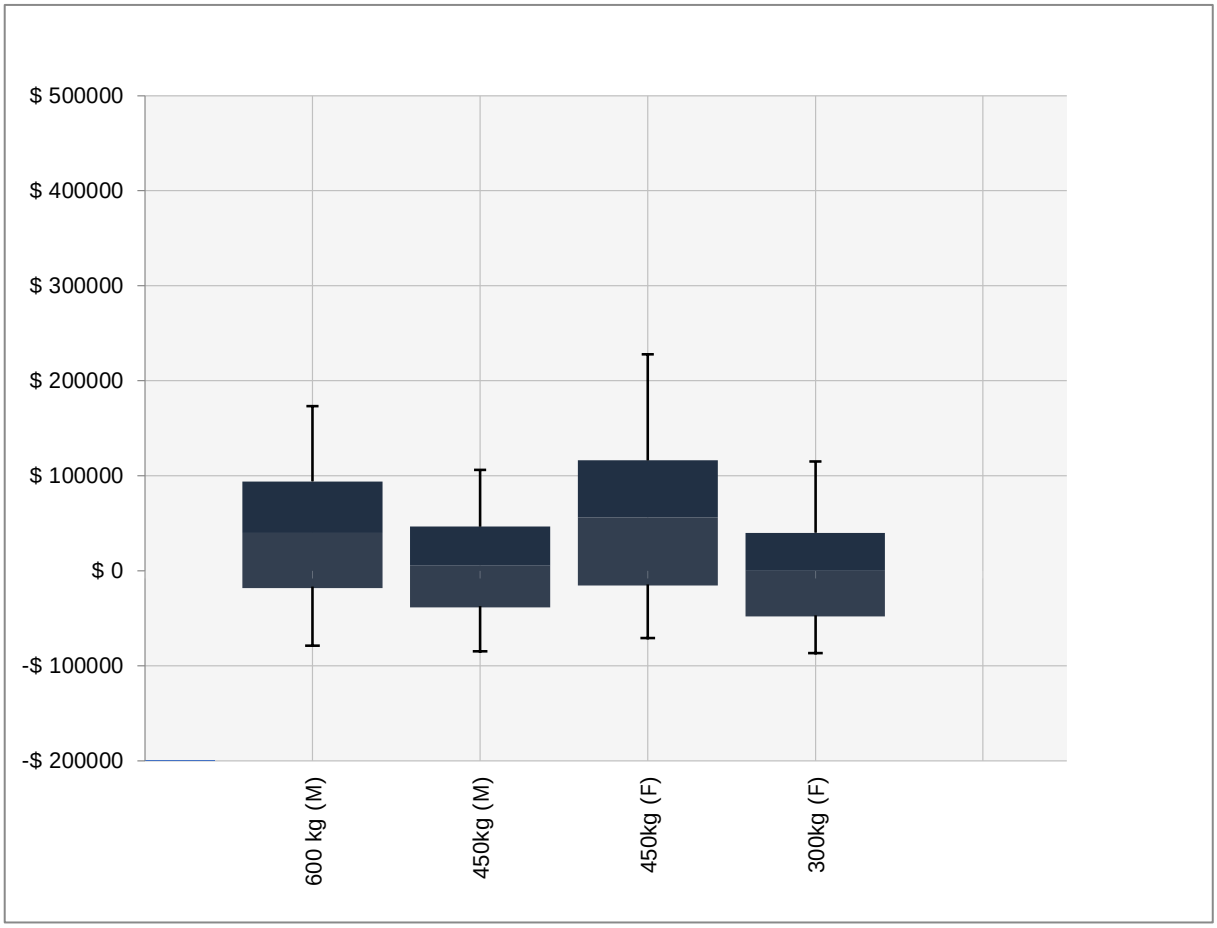

Figure 4-3: Annual average net return – Bale Farm

Table 4-5: Annual profit range – Bale Farm

BEEF MARKET	MAXIMUM PROFIT	UPPER 75TH PERCENTILE PROFIT	MEDIAN PROFIT	LOWER 25TH PERCENTILE PROFIT	MINIMUM PROFIT
600 kg Manufacturing	$173,429.00	$94,306.00	$39,945	-$17,974.00	-$78,733.00
450 kg Manufacturing	$106,270.00	$46,445.00	$5,458	-$38,351.00	-$84,549.00
450 kg Feeder	$227,998.00	$116,134.00	$56,177	-$15,537.00	-$70,745.00
300 kg Feeder	$115,160.00	$40,206.00	-$85.00	-$48,044.00	-$86,645.00

In comparison, the second farm is displayed below in Figure 4-4 and Table 4-6 In the manufacturing market, the beef price needs to be at the upper 75th percentile or at a maximum price for the farm to gain a positive annual net return. For a 600kg manufacturing steer the farm could receive a maximum profit of $85.5k and for the 450kg manufacturing steer the farm could expect a maximum $54k profit.

In the model the feeder market provides a maximum $143k profit for a 450kg steer or $77k for a 300kg steer. While a 450kg steer could still provide a profit if the beef price was at a median, the 350kg feeder steer and both the sizes in the manufacturing market would result in a profit loss if the beef price was at or below the median price. It is important to take into consideration the impacts of a 'good' year (where input costs are low and beef prices are high) Vs a 'bad' year (where inputs costs are high and beef prices are low).

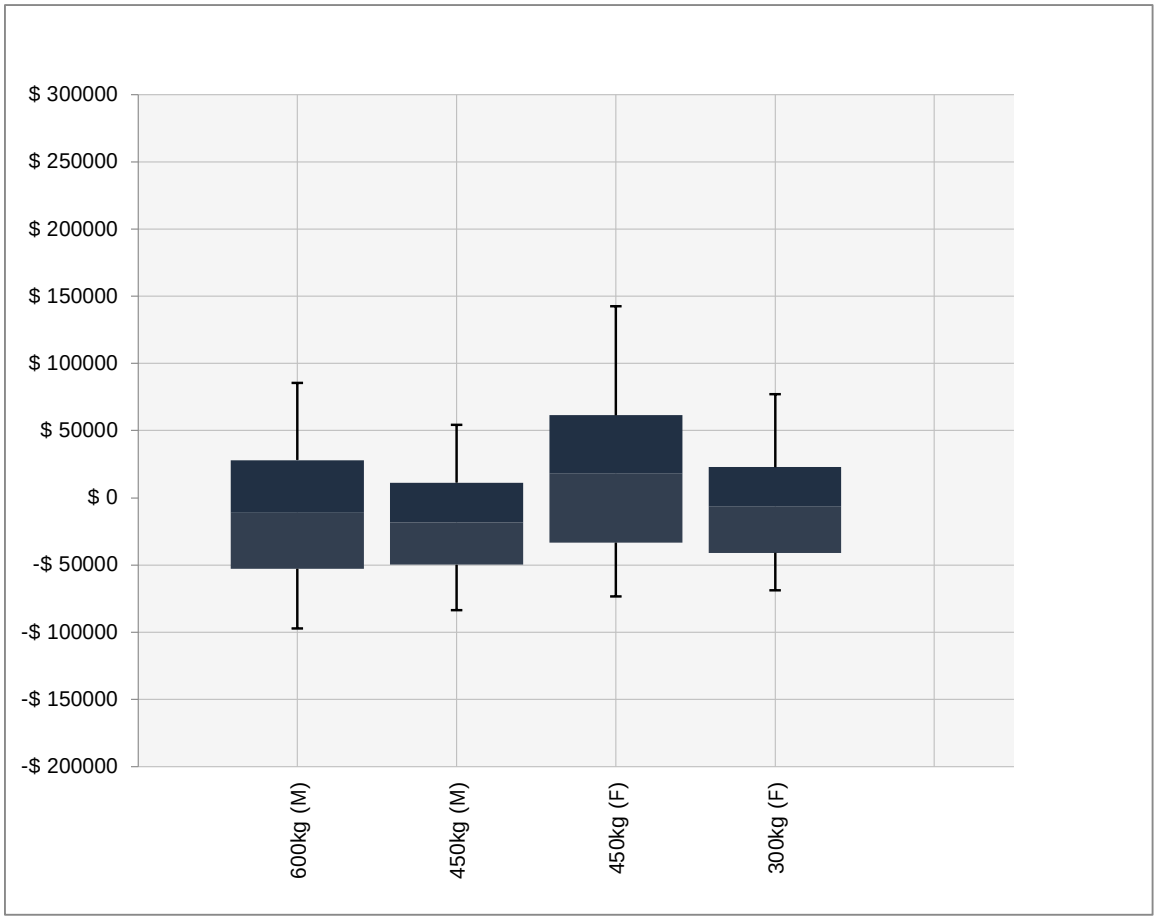

Figure 4-4: Annual average net return – Grey Farm

Table 4-6: Annual profit range – Grey Farm

BEEF MARKET	MAXIMUM PROFIT	UPPER 75TH PERCENTILE PROFIT	MEDIAN PROFIT	LOWER 25TH PERCENTILE PROFIT	MINIMUM PROFIT
600 kg Manufacturing	$85,513.00	$28,147.00	-$11,020.00	-$52,723.00	-$97,256.00
450 kg Manufacturing	$54,242.00	$11,333.00	-$18,408.00	-$49,862.00	-$83,500.00
450 kg Feeder	$142,581.00	$61,361.00	$18,119.00	-$33,422.00	-$73,415.00
300 kg Feeder	$77,118.00	$22,804.00	-$6,411.00	-$40,822.00	-$68,800.00

The Bale farm annual average net return per head of cattle is displayed below in Figure 4-5 and in Table 4-7. It shows the annual average net return but is further broken down to return per head of cattle.

In the manufacturing market, the Bales could expect a return if the beef price is at or above the median price. The maximum return per head they could expect for a 600kg animal is $796 or $487 for a 450kg animal.

In the feeder market, the maximum return per head they could expect for a 450kg animal is $1,046 and $528 per head for a 300kg animal.

A return per head would not be achieved, and in fact a loss would be made, if the beef price in both the manufacturing and feeder market is at the lower 25th percentile or at the minimum price, and the price per head for 300kg Feeder would be at break-even if the beef price was at the median price.

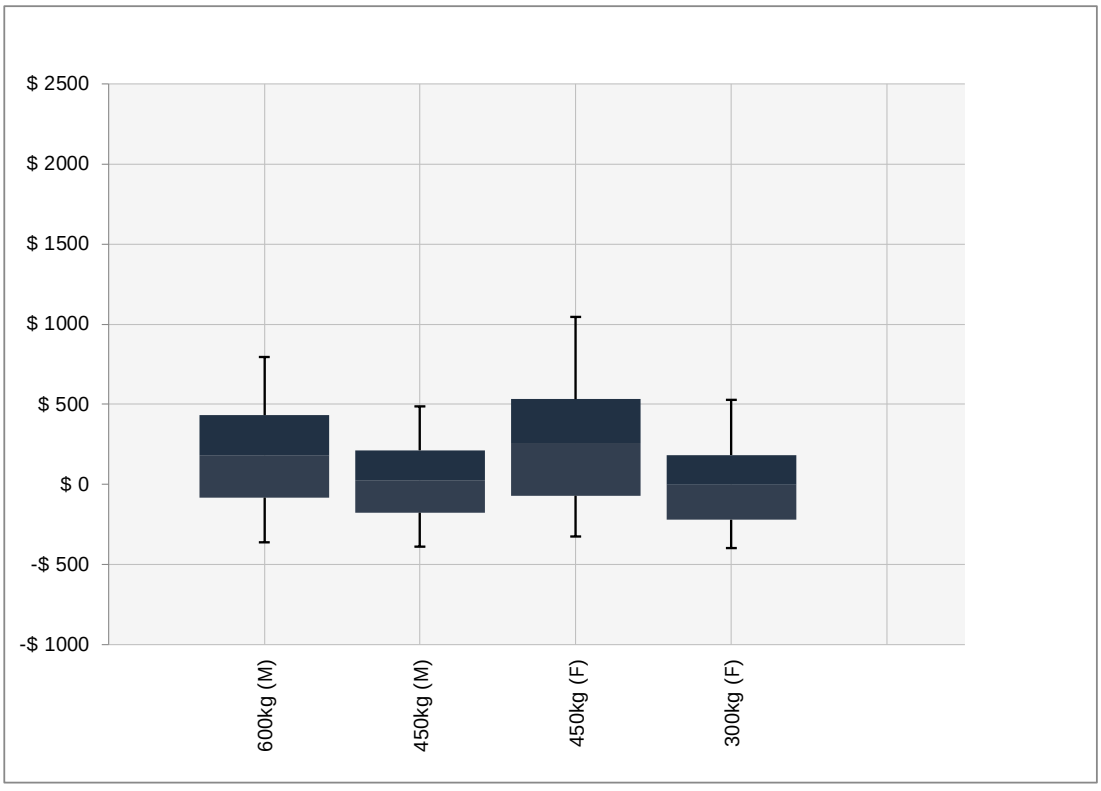

Figure 4-5: Profit per head – Bale Farm

Table 4-7: Profit per head range – Bale Farm

BEEF MARKET	MAXIMUM PROFIT	UPPER 75TH PERCENTILE PROFIT	MEDIAN PROFIT	LOWER 25TH PERCENTILE PROFIT	MINIMUM PROFIT
600 kg Manufacturing	$796.00	$433.00	$183.00	-$82.00	-$361.00
450 kg Manufacturing	$487.00	$213.00	$25.00	-$176.00	-$388.00
450 kg Feeder	$1,046.00	$533.00	$258.00	-$71.00	-$325.00
300 kg Feeder	$528.00	$184.00	-$0.00	-$220.00	-$397.00

The net return per head for the second farm is displayed below in Figure 4-6 and Table 4-8. For a 600kg manufacturing steer the farm could receive a maximum return per head of $545 and $345 for the 450kg manufacturing steer.

The feeder market could provide a maximum $908 net return per head for a 450kg steer or $491 for a 300kg steer. While a 450kg steer could still provide a return if the beef price was at a median, the 350kg feeder steer and both the sizes in the manufacturing market would result in a negative net return per head if the beef price was at or below the median price.

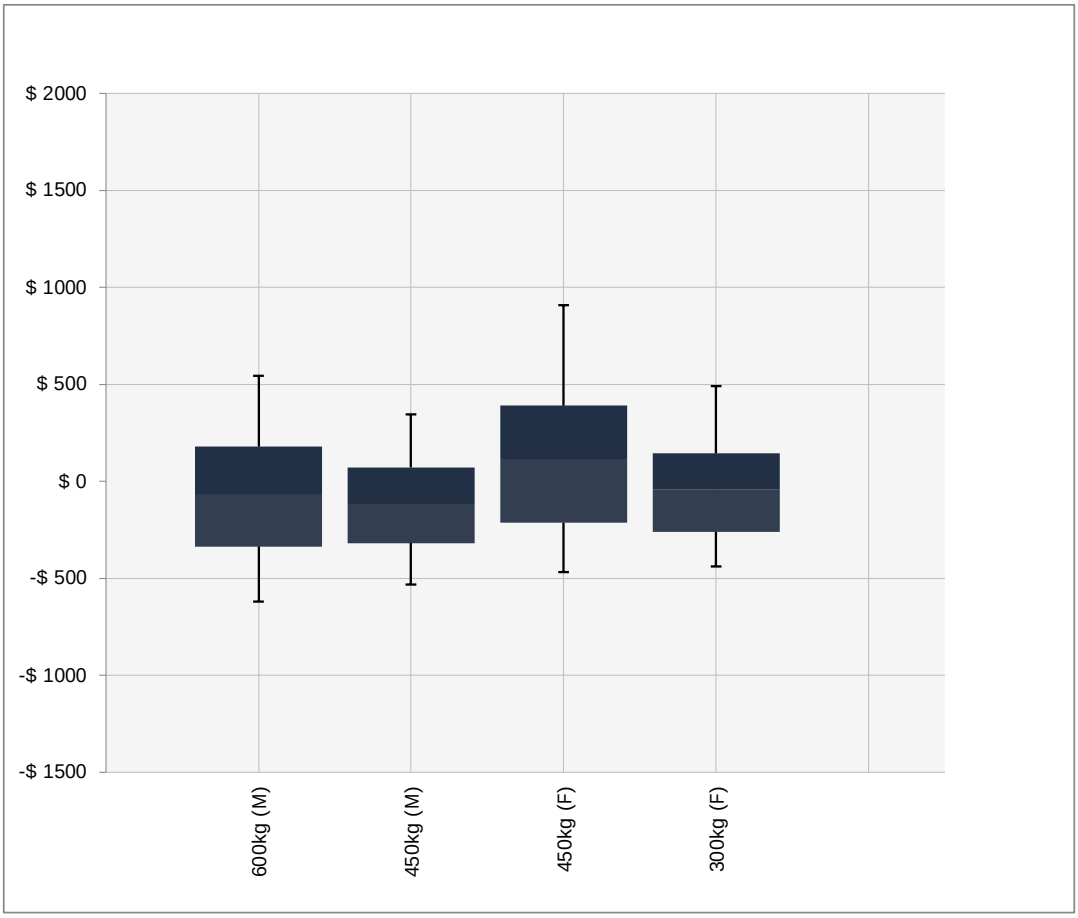

Figure 4-6: Profit per head – Grey Farm

Table 4-8: Profit per head range – Grey Farm

BEEF MARKET	MAXIMUM PROFIT	UPPER 75TH PERCENTILE PROFIT	MEDIAN PROFIT	LOWER 25TH PERCENTILE PROFIT	MINIMUM PROFIT
600 kg Manufacturing	$545.00	$179.00	-$70.00	-$336.00	-$619.00
450 kg Manufacturing	$345.00	$72.00	-$117.00	-$318.00	-$532.00
450 kg Feeder	$908.00	$391.00	$115.00	-$213.00	-$468.00
300 kg Feeder	$491.00	$145.00	-$41.00	-$260.00	-$438.00

4.3 RETURN ON INVESTING IN EXTRA IN FRASTRUCTURE

A 15-year discounted cashflow analysis was developed to show the economic outcome of the investments and incorporates all capital and operational changes to the business as benefits and costs that are associated with each scenario. The two case study farms had slightly different requirements for extra investment in calf rearing facilities if they are to rear all their surplus calves to weaning.

The results are displayed in boxplots, shown in Figure 4-7 and Figure 4-8 below. The box plots in these figures give the range of results associated with each tested scenario.

The Bale farm had more consistent positive results with the NPV for three out of the four scenarios on average being positive. The most consistent returns were from the heavier weight classes for both pricing scenarios with 600kg manufacturing steers averaging a NPV of $285K with a range of -$645K to $1.3M and 450kg Feeder steers averaging a NPV of $411K with a range of -$581K to $1.75M. The Bale farm had a capital investment of $35K and the payback period ranged from one to fifteen (plus) years (shown in Table 4-9).

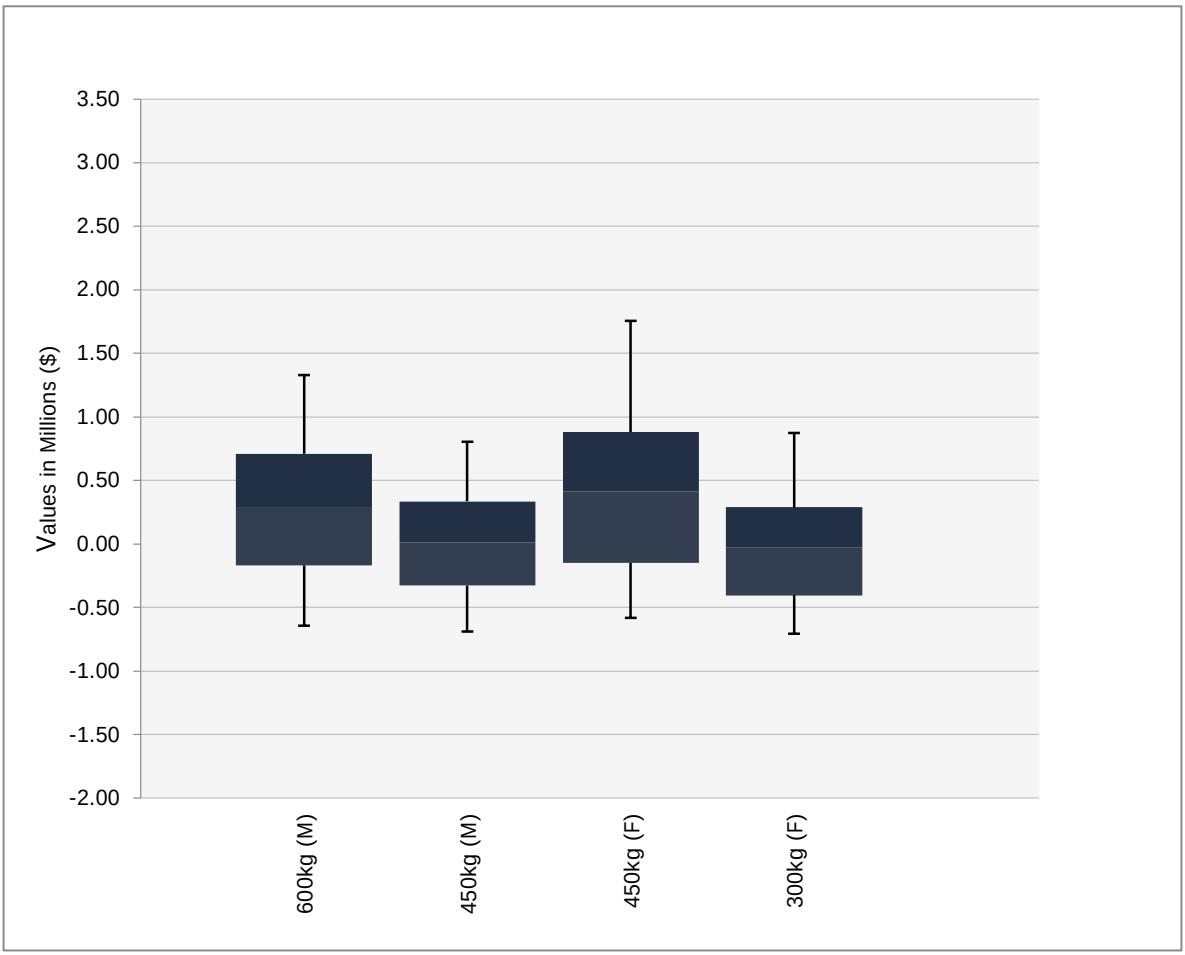

Figure 4-7: NPV - Bale farm

Table 4-9: Payback period - Bale farm

DESCRIPTION	600KG MANUFACTURING	450KG MANUFACTURING	450KG FEEDER	300KG FEEDER
Payback period	1	7	1	15+

The Grey farm had less consistent results. This was mainly due to a higher infrastructure requirement $75K, as well as consistently higher pasture costs. Their geographic position and the need for a larger more consistent cropping program contributed to these results. The NPV for three out of the four scenarios on average was negative, results are shown in Figure 4-8 below. As was the case with the Bale farm, better results were achieved from the heavier weight classes with the 450kg Feeder steers returning an average NPV of $18K. The Greys farm had a capital investment of $75K and the payback period ranged from two to fifteen (plus) years. Results are shown in Table 4-10.

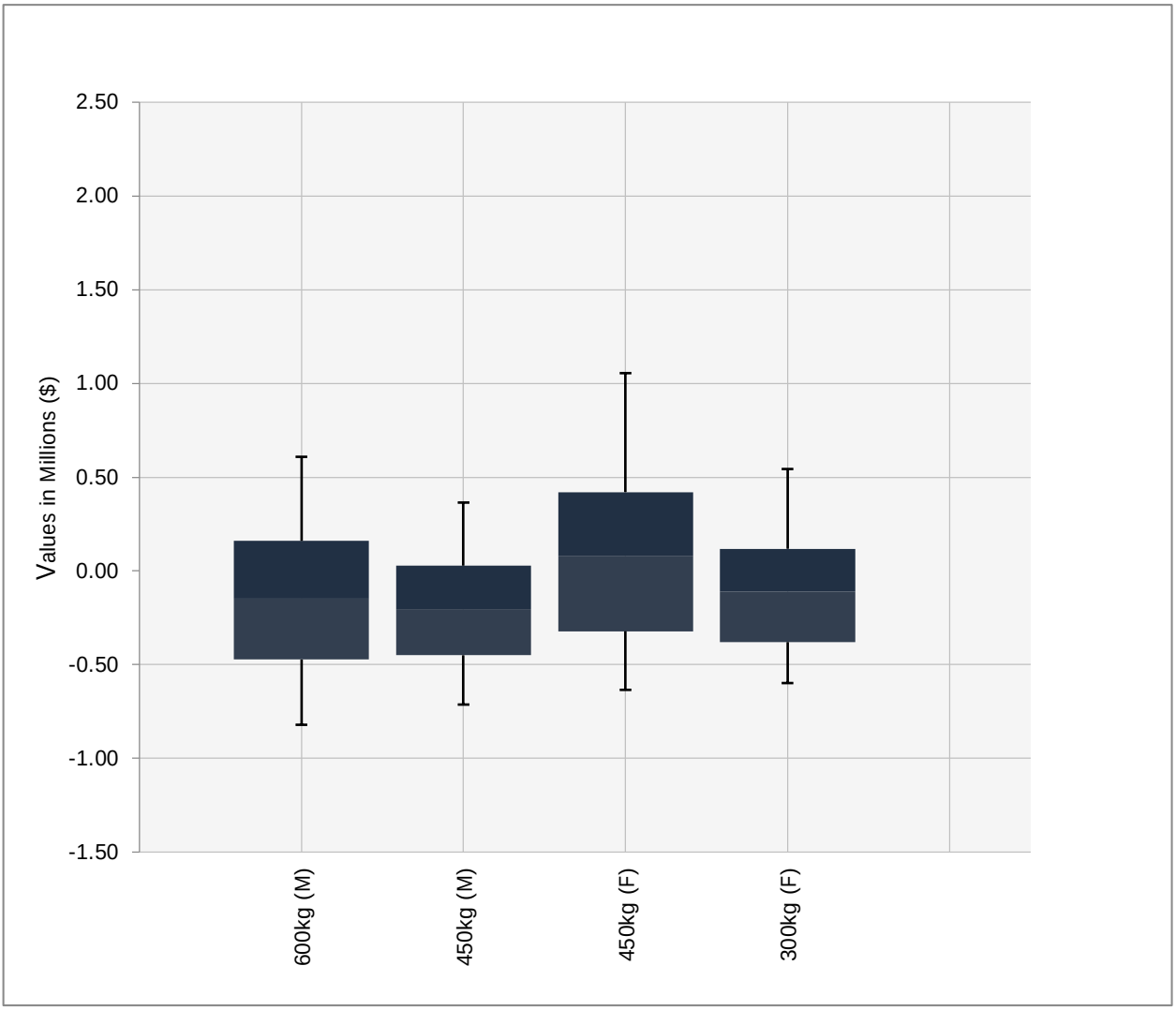

Figure 4-8: Net Present Value - Grey Farm

Table 4-10: Payback period - Grey farm

DESCRIPTION	600KG MANUFACTURING	450KG MANUFACTURING	450KG FEEDER	300KG FEEDER
Payback period	15	15+	2	15+

Some dairy farms may require more significant investment in calf rearing facilities if they are to rear all of their surplus calves and will find it challenging to achieve an attractive return on investment unless they can obtain a more stable beef price. It is important to acknowledge the infrastructure challenges posed by a tight single calving pattern compared to year-round or split calving patterns observed in our case study farms. Further consideration should be given to the required infrastructure and capital investment to support the change on a farm with a tightly scheduled single calving pattern, particularly in regions with colder winters. as our case study farms may not be representative of these scenarios.

5 Conclusions/Future Directions

Currently, in the Australian dairy industry, non-replacement calves are often culled early which causes societal upset due to the perceived poor treatment of the animals. Although it is undesirable, there are many factors that influence the farmers decision to cull early. Through this project, and in collaboration with the FRG, we found some of the underlying reasons were due to the anticipated effort required to manage an additional beef enterprise, a lack of available feed and land, a primary focus on milk production, and market volatility, particularly around beef prices. To delve into this problem further, we developed a detailed economic analysis of two case study farms evaluating the additional infrastructure and management requirements for alternative 'dairy-beef' calf pathways. Our key findings indicate:

- On average, raising surplus calves for beef resulted in roughly a break-even result. However, the range in net benefit varied from approximately $1000/head in good years to -$600/head in poor years.
- The profitability of the enterprise was primarily influenced by fluctuating beef market prices, which emerged as the largest source of variation in the extra profit/loss generated by raising surplus calves. If the beef market price is below the median, it is unlikely that a profit could be made for any size dairy-beef animal.
- As farmers are currently predicted to have a loss in profit or break even from a dairy-beef enterprise about 50% of the time, the expectation that a large number of dairy farms will pursue a 'dairy-beef' enterprise is unrealistic, unless more stable and profitable markets can be developed.
- Access to premium markets, such as feedlots, showed potential for increasing profitability. However, it did not significantly reduce the worst-case scenarios associated with market volatility.
- Some dairy farms may require more significant investment in calf rearing facilities if they are to rear all of their surplus calves and will find it challenging to achieve an attractive return on investment unless they can obtain a more stable beef price.
- Growing dairy calves to 450+ kilograms showed the potential for higher profitability. However, this approach also exposed the business to downside risks, including increased feed costs, limited land availability, climate variations, and beef market volatility.
- It is important to consider that an increased focus on managing the beef enterprise can divert attention away from the core goal of profitable milk production, which remains the primary business for dairy farmers.

The value of the participatory approach adopted in this project was demonstrated when the FRG identified a key barrier being their lack of confidence that there would be demand for their product if they were to raise surplus calves. We were able to organise MLA and feedlot representatives to talk to the group about what could be achieved with dairy beef and what dairy farmers could do to ensure that they have a product that is attractive to meat processors or feedlots. This aspect was not part of the original project brief but made an important contribution to increased confidence of dairy farmers in this region that there could be demand if they raised surplus dairy calves for beef.

This project has highlighted the need for a better understanding of the costs, benefits and risks incorporated with raising surplus calves for beef. Additionally, there is a strong need for dairy farmers, processors, beef markets, and industry bodies to come into partnership and make significant changes to processes and adjust market ideals. By understanding the economic implications and collaborating with industry stakeholders (especially within the beef industry), there is potential to develop surplus calf management practices that are both socially and economically sustainable. However, these solutions will be regionally, and farm business specific and significant attention needs to be devoted to reducing the proportion of years when there is likely to be a negative return.

5.1 FUTURE DIRECTIONS

In light of these findings, it is crucial to explore alternative strategies that can enhance the economic sustainability of surplus calves. Common themes within the FRG were a lack of land and labour coupled with limited access to premium beef markets. In order to bridge the gap (between dairy farmers and feedlot operators/meat processors) in the study region, the development of specialised calf rearers that raise the calves on leased land holds potential. Specialised calf rearers can provide the necessary effort, expertise and infrastructure to effectively raise and market surplus calves, alleviating the burden on dairy farmers while creating new sustainable opportunities for the beef market. This currently appears to be the most practical pathway to reduce the week old slaughter of surplus calves.

For this particular region (Taree/Kempsey) some follow-up support to foster a regional working group/producer demonstration site may be very beneficial.

While exporting dairy heifers is not an option in this region, further analysis of rearing surplus heifers for the export market could provide valuable insights for other dairying regions. Assessing the economic viability and market demand for these markets can provide additional revenue streams and profitable diversification options for dairy farmers in other regions.

It is important to acknowledge the infrastructure challenges posed by a tight single calving pattern compared to year-round or split calving patterns observed in our case study farms. Further consideration should be given to the required infrastructure and resource allocation necessary to support a tightly scheduled calving pattern particularly in regions with colder winters as these case studies may not be representative of these scenarios.

The contribution that milk processers can make to help manage risk also warrants further investigation. By offering premium prices for milk and/or providing incentives, milk processors could help mitigate the financial risks associated with rearing surplus calves. This may encourage more dairy farms to actively participate in raising surplus calves, which would foster a more sustainable industry in terms of social licence.

A decision support calculator has been developed (Appendix 1) and will be presented to the FRG and could potentially be tested, refined and circulated more widely.

In conclusion, the economic analysis emphasises the significance of market conditions, farm management efforts, and the goals of dairy farmers when exploring alternative pathways for surplus calves. Continued collaboration within the regional farmer working group, coupled with the active involvement of milk processors, meat processors and specialised calf rearers, holds potential for the development of viable surplus calf pathways. We see merit in a continued effort working towards sustainable and profitable calf solutions that will enable the dairy industry to continue to thrive while meeting the evolving demands of both market and societal expectations.

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Appendix 1: Decision Support Calculator

Scenario assumptions	Value
Number of cattle	150
Current age	
Current value	
Target weight (kg/animal)	600
Target weight gain (kg/day)	1

Profit/loss	Value
Per head	$524
Total	$78,559

Summary	Value
Sale price per animal	$1,842.00
Total feed costs	$984.14
Weaning costs	$334.13
Age at sale (years)	1.6
Age at sale (months)	19
Days to target weight	563
Total DM fed (tonnes) (after weani	4.6

Price assumption	Default	User	Value
Beef price ($/kg lw)	$3.07	*$3.07*	$3.07
Grain price ($/tonne)	$390		$390
Milk replacement price ($/lt)	$0.75		$0.75
Calf pellets ($/tonne)	$450		$450
Pasture costs ($/kg DM)	$0.17		$0.17
Hay price ($/tonne)	$320		$320
Labour cost ($/hour)	$34		$34
Additional costs (fuel, electricity, et	$5		$5
Transport (per head)	$8.50		$8.50

Feed Intake	Default	User	Value
Weaning (10 weeks)	5.5	*5.50*	5.5
100-150 kg	5		5
150-300 kg	7		7
300-450 kg	10		10
450-600 kg	11		11

Appendix 2: Draft Factsheet Example

PROJECT BACKGROUND

The Economic Impact Assessment for Calf Pathways project aimed to gain a clear understanding of the costs, benefits, and risks associated with raising surplus dairy calves for the beef market and investigate whether it would be economically practical to hold onto surplus dairy calves and pursue alternative pathways.

The delivery of this program involved identifying two case study farms and developing a Farmer Reference Group (FRG) in the mid-north coast of NSW.

Quote from the FRG “I’m interested in if it’s economically positive to raise surplus calves because I’m interested in making this a sideline business”

The need for the project arose due to social licence priorities and the importance for dairy farmers to meet societal expectations around the treatment of bobby calves.

Quote from FRG “It’s not just about the numbers but also what the consumer wants”.

The aim is to aid farmers in making informed decisions that contributes to a future where surplus calves are consistently raised for beef, using practices that can withstand seasonal and commodity price unpredictability, as well as navigate regionally specific challenges.

Farm 1 The Bales - runs a year-round calving pattern with 320 milking cows. The Bales had existing infrastructure and only required a small extension to their calf shed to comfortably manage keeping calves for longer.

Farm 2 The Greys - runs a split Autumn to Spring calving pattern with not many calves in winter and 230 milking cows. They required capital investment for an additional calving shed.

The factsheet content is attached.

Appendix 3: Beef Prices

Table A3-1: Market beef prices (low, average, high) used in sensitivity analysis[1]

				MARKET PRICE (C/KG LWT)			VARIANCE TO AVERAGE OF CATEGORIES		
Category	**Lwt Kg**	**Muscle score**	**Fat score**	**low**	**avg**	**high**	**low**	**avg**	**high**
Grown Steer	400-500	A-C	3-4	225	309	418	-2	-10	-38
Grown Steer	500-600	A-C	3-4	232	317	367	6	-2	-89
Grown Steer	600--750	A-C	3-4	240	338	437	14	19	-19
Grown Steer	750+	A-C	3-4	230	325	425	4	7	-31
Manufacturing Steer	540+	A-C	3-4	221	307	415	-6	-11	-41
Yearling Steer	330-400	A-C	3-4	244	376	558	18	57	102
Yearling Steer	400+	A-C	3-4	236	331	479	10	13	23
Vealer Steer	200-280	A-C		168	262	544	-58	-57	88
Vealer Steer	280-330	A-C		211	281	453	-15	-37	-3
Vealer Steer	330+	A-C		256	340	465	29	22	9
Average of categories				226	319	456			

[1] MLA, Australian Saleyard Cattle Transactions Armidale, https://www.mla.com.au/prices-markets/statistics/cattle-transactions/, viewed 5 May

Figure A3-2: Distribution of Manufacturing Steers 540+ kg, Armidale (1 July 2017 – 5 May 2023)

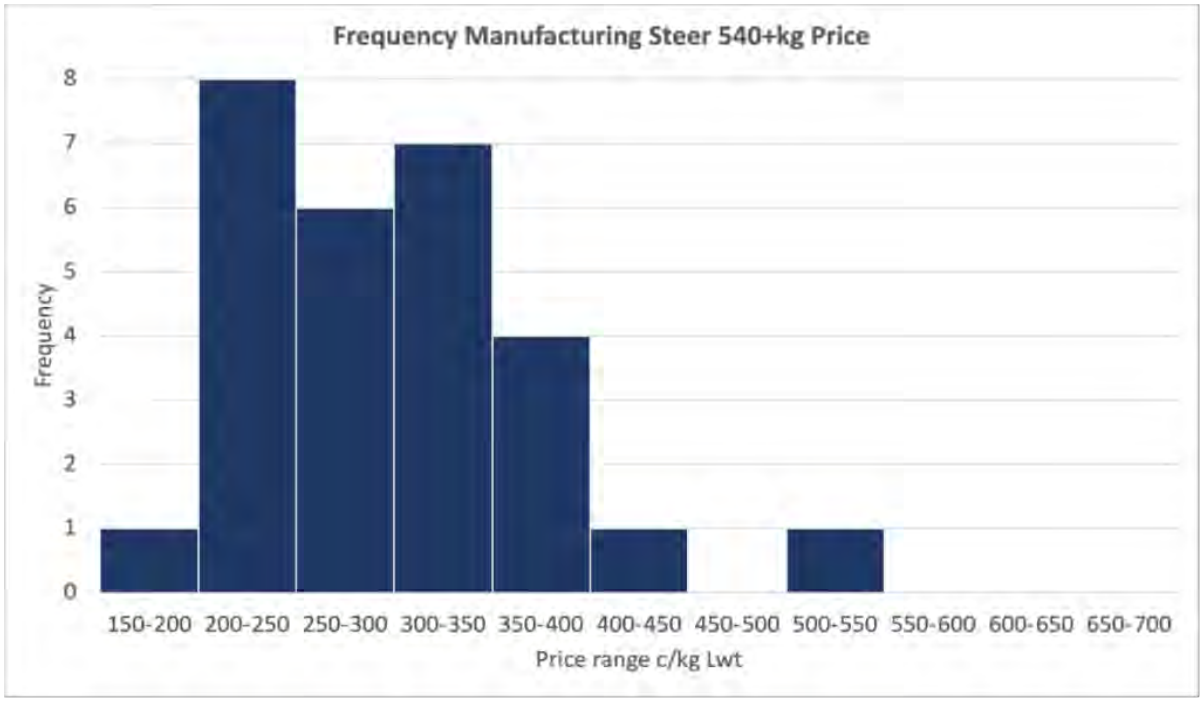

Figure A3-3: Frequency Histogram of Manufacturing Steers 540+ kg Prices, Armidale (1 July 2017 – 5 May 2023)

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Document review and authorisation

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1.0	Final	25/08/2023	D. Armstrong T. Wardley L. Jones	D. Poole	L. McKenzie E. Kelly	D. Armstrong	Dairy Australia

Final Project Summary

P10b: Economics and Risks of Raising Non-Replacement Calves in a Dairy Business

May 2026

This project aimed to analyse the economics and risks of raising non-replacement calves in a dairy business, with the view to selling them as dairy beef.

The market for dairy beef varies across regions, depending on the scale and infrastructure of the local beef supply chain including backgrounders, feedlots and processors.

This means the economics and risks of raising dairy beef tend to be regionally specific making it difficult to offer blanket recommendations.

The benefits and risks are also influenced by the existing dairy farming system and infrastructure for calf rearing. A year round calving system is likely to require less investment in modifying calf rearing systems than a farm operating with a batch calving system. For example, herds with tight batch calving systems (such as in Victoria and Tasmania) may require up to three times the existing calf shed facilities to rear non-replacement calves.

Case study approach

A case study approach was chosen to provide relevant insights for farmers in the project group. Two case study farms were chosen from the Kempsey/Taree area where some dairy businesses had indicated they were keen to participate.

Farm 1: Year round calving system
Farm 2: Batch calving system.

P10 Sustainable pathways for surplus calves

Dairy UP's P10 aimed to develop sustainable options for managing surplus calves born on NSW dairy farms such as male (bobby) calves or females that are not destined for the milking herd. It complemented other Dairy Australia work to address the issue of surplus calves.

Designed to be regionally relevant and applicable to local communities, the Dairy UP project involved two phases:

- Consultation along the entire supply chain and collaborative design of alternative management pathways (Nowra, NSW community)
- An analysis of the economics and risks of raising non-replacement calves in a dairy business.

This document provides an update on the analysis of the economics and risks of raising non-replacement calves in a dairy business.

While P10 is now complete, work on this topic continues through a follow-on project Growing Beef from Dairy which funded jointly by Dairy Australia and MLA.

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For each farm, the study compared two scenarios:

- 'Base scenario': all cows joined to conventional dairy semen or dairy bulls with non-replacement calves sold at 3 weeks old.
- Change scenario: Sexed semen used to breed dairy replacements with the rest of the herd joined to beef semen/bulls and beef calves raised to various ages.

Market fluctuations

Market fluctuations during the study period highlighted the risks associated with variations in demand and price for dairy beef. When the project concept was being developed in 2022, demand for dairy beef was strong and project participants were confident they could make a profit from dairy beef. This confidence was challenged over during 2023 when demand (and price paid) for dairy beef weakened significantly.

To get an indication of risks, the analysis looked at best and worst case years and an average.

Key findings

Profitability (net return per head)
Beef market prices were the biggest contributor to variation in net return per head. The feedlot market (and beef market in general) is highly volatile and this has an impact on the prices paid for dairy beef calves.

Net returns varied from $900/head in good years to a loss of $600 in bad years, with about break even on average.

Net returns are also influenced by how long animals are reared before they are sold (from weaning through to 300-600kg).

Risk
Growing out animals to above 450kg can be more profitable but exposes the business to additional downside risk such as land limitations, higher feed costs, seasonal variation (drought, flood) and beef market fluctuations.

Accessing premium markets could reduce some of this risk, but this option varies across regions.

Payback period
The payback period for investment in infrastructure was shorter (1-7 years) for Farm 1 (year round calving) because needed only minor upgrades to accommodate rearing dairy beef calves.

The payback period for Farm 2 (batch calving) could extend to beyond 15 years as it needed more investment in calf rearing infrastructure.

Other considerations
The decision to rear non-replacement calves involves significant non-financial considerations such as animal welfare, ethics and social licence. For some, breaking even on average may be a reasonable balance for these benefits.

Other things to consider include:

- Effort involved in rearing non-replacement calves and the potential distraction from the core business of producing milk.
- Goals of individual farm business owners (specialist dairy farmer versus interest in diversifying)
- Role and availability of a specialist calf rearer.

Next steps

A decision support calculator developed during the project could be tested more widely.

The study authors recommended further analysis of the following situations:

- raising more surplus heifers for export (recognising this is not an option for every Australian dairying region).

- Tight single calving herds where infrastructure investment is greater.

Given dairy beef is an evolving industry, the report recommended updating results as new knowledge and options become available.

DairyUP
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Collaborators

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More info

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Delivery organisations

The University of Sydney – Dairy Research Foundation
NSW Government – Department of Primary Industries and Regional Development
Scibus
Dairy Australia

Partner organisations

NSW Farmers – Growing the best
eastAUSmilk – Advocacy · Unity · Service
Dairy NSW | Dairy Australia
AFMH
Leppington Pastoral Company Pty Ltd
NSW Government – Local Land Services
Norco – Quality since 1895

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