

Final report

Desktop analysis of opportunities for oilenhanced forages

Project code:	B.PAS.0362
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Date published:	14 June 2023

PUBLISHED BY Meat & Livestock Australia Limited PO Box 1961 NORTH SYDNEY NSW 2059

Meat & Livestock Australia acknowledges the matching funds provided by the Australian Government to support the research and development detailed in this publication.

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Abstract

Increasing the oil content of vegetative biomass through precision genome editing could potentially increase the energy content of plants consumed by grazing livestock. The aim of this desktop analysis was to understand how the application of this technology could benefit Australian livestock industries as a precursor to further investment. First, we undertook a sectoral analysis to identify the most important pasture and forage species in Australia, and prioritise these based on suitability for future climates, potential for commercialisation and adoption, and ease of genetic transformation. Six species, representing a range of forage types, were shortlisted for further evaluation. We assessed how increasing the oil content of these forages up to 8% of dry matter could impact the productivity of grazing sheep and beef cattle and used a partial discounted cashflow analysis to understand profit-risk trade-offs. The greatest value in feeding oil-enhanced forages was to increase the liveweight gain of growing animals, especially beef cattle. At the farm level, the equivalent annual profit from oil-enhancement was highest in forage crops (*e.g.*, forage sorghum, forage oats), which are higher yielding than pastures, and can be grazed or conserved as hay/silage. At the industry scale, the greatest benefit would be from oil-enhancement of forages such as Phalaris, which are already widely adopted.

Executive summary

Background

Increasing the oil, and therefore energy content, of forage and pasture species through precision genome editing could benefit ruminant livestock industries. However, more information on the risks and benefits is required to inform future investments.

Objectives

The objectives of this project were to:

- Identify suitable forage species based on importance to industry, suitability for future climates, and ease of genetic transformation.
- Describe how changes in oil content could impact livestock production.
- Provide justification for further investment.

Methodology

This desktop analysis included three main activities:

- 1) A sectoral analysis (literature review and industry interviews) to prioritise species for further evaluation.
- 2) Modelling of animal-level impacts, to understand how increasing oil content of forages could impact livestock production.
- 3) Economic analysis to understand profit-risk trade-offs.

A business case for further investment was not completed due to changes in licensing arrangements for this technology.

Results/key findings

The sectoral analysis found that use of improved forages is concentrated in southern Australia, and the literature and interest of seed companies is also focused on species relevant to this area. This analysis also indicated that the greatest uptake of improved forages was likely to be from the sheep and dairy industries.

Increasing the oil content of biomass improved livestock productivity and reduced emissions intensity in all scenarios evaluated, with the greatest benefits observed when oil-enhanced forages were fed to growing beef cattle. There was little productivity or economic benefit in using oil-enhanced forages to support animals at maintenance (*e.g.*, dry ewes).

Of the species evaluated, oil-enhancement of forage crops (*e.g.*, forage sorghum and forage oats) provided the greatest economic benefit per ha sown because they are higher yielding than pastures and can be grazed or conserved as hay/silage. Oil-enhancement of perennial grasses, such as Phalaris, would provide some benefits in more extensive grazing systems, but are a riskier investment for farmers in below-average seasons.

At the industry scale, the greatest benefits to the red meat industry would likely arise from the application of this technology to forages which are already widely adopted and adapted to future climates.

Benefits to industry

The results from this analysis highlight best-bet options for investment into oil-enhanced forages.

Future research and recommendations

Based on a combination of the sectoral analysis and our modelling, we recommend that initial investments into oil-enhanced forages focus on high-yielding forage crops (*e.g.,* forage sorghum or forage oats) or perennial grasses, such as Phalaris, which are already widely sown.

Animal feeding trials and duty of care experiments are recommended to validate the results of our modelling and confirm the safety of feeding oil-enhanced forages to livestock.

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1. Background

Animal productivity is directly related to feed quality from pastures and forages, and higher quality pasture and forage lines will contribute to more efficient livestock production. Specifically, increasing the oil content of pastures and forages will potentially benefit livestock production systems through higher feed intake, liveweight gain and feed efficiency (kg product per kg feed consumed), and reductions in methane (CH₄) emissions.

The CSIRO has recently developed technology allowing the accumulation of unprecedented levels of storage lipids (oils) in the vegetative plant tissues of tobacco and sorghum. This platform technology can enable engineering of energy-dense forage crops through precision genome editing (non-GM). Increasing the dietary energy content (via lipids) positively influences feed conversion efficiencies in sheep and cattle, resulting in higher meat production, and reduces the production of enteric CH₄. This approach improves the energy density of diets without increasing high-starch intake (*e.g.*, grain supplementation) or reducing fibre intake, both of which can negatively impact rumen function.

To inform future investment decisions, a better understanding of how this technology may impact livestock industries is required.

2. Objectives

- 1. Identify the most important forage species in Australia based on area sown and value/volume of seed sales information drawn from literature, industry data and interviews with at least four forage seed companies.
- 2. Assess the suitability of important forage species for future climates, based on climate projections for 2050.
- 3. Identify the suitability of forages for genetic transformation, any barriers to future work, and potential scope for increasing lipid content.
- 4. Review peer literature to determine:
 - Effect of forage oil/dietary oil content effects on neutral detergent fibre (NDF) digestibility.
 - How changes in plant structure/composition impact on nutrition and production, including how increasing oil content may contribute to changes in enteric CH₄.
- 5. Quantify the potential animal- and industry-level productivity improvements for at least four prioritised forage species.
- 6. Develop a cost-benefit analysis of the potential benefit to Australian livestock industries (sheep and beef) of targeted lipid improvement for each of the four prioritised species.
- 7. Based on suitability to changed regional climates and animal productivity benefits, provide a business case and investment plan for increasing lipid content of prioritised forages, with specific recommendations to MLA.
- 8. Identify and have documented interest from candidate seed company partners and Dairy Australia to implement the investment plan.

3. Methodology

This project comprised three key components:

1. **Sectoral analysis** – used to prioritise pasture/forage species for further analysis. This involved interviews with experts in the Australian pasture/forage industry and major seed

companies. Pasture/forages identified in the interview process were then assessed for (i) suitability for future climates, (ii) potential for commercialisation and adoption, (iii) ease of transformation. Six species, representing a range of forage types, were then shortlisted for further analysis.

- 2. Animal level analysis included a literature review of potential impacts (positive and negative) of oil-enhancement, and modelling of animal-level impacts for the six shortlisted species.
- 3. **Cost-benefit analysis** used to further inform investment decisions using outputs from the animal level analysis for the six shortlisted species.

Methods were reviewed and approved by the CSIRO Social and Interdisciplinary Science Human Research Ethics Committee (CSSHREC approval ID 103/21) and were subject to a Privacy Impact Assessment. Due to project confidentiality agreements, we are unable to provide the names of participants, or the companies/organisations/agencies they represent in this report.

CSIRO divested its interests in the oils technology during this project, and the business case and investment plan for further work were not completed. The results from this analysis are still broadly applicable to the improvement of forages for Australian grazing systems.

3.1 Sectoral analysis

The scope of this analysis was limited to non-woody forage species, including legumes, grasses and herbs that are either grazed directly by ruminant livestock, or conserved as hay or silage. This included dual-purpose crops.

3.1.1 Interviews

Fourteen individual interviews were carried out via video conference from late September to early December 2021 to identify the most important pasture and forage species in Australia. Participants included representatives of industry and major seed companies, industry consultants, practitioners, and researchers from across Australia (Figure 1) and spanned the key pasture-based livestock sectors of beef, sheep, and dairy. Despite our efforts to get even representation across the major livestock producing areas, not everyone we contacted agreed to participate in the interviews, and there is a bias towards the southern states.

Initially, major seed companies were invited to participate in interviews. Existing contacts within these companies were used to identify key individuals with extensive knowledge in national sales and research and development. Three major seed companies agreed to participate. Additional non-company contacts with a broader view of the Australian pasture/forage industry were recommended by industry participants and were also invited to participate.

Since company investment is targeted at commercially important species, researchers, industry consultants, and practioners, with extensive knowledge of the pasture/forage industry were also contacted to remove any associated bias. These participants were identified based on our existing networks, with some key individuals also recommended by other participants. This cohort of individuals represented a broad range of organisations/agencies, were nationally or internationally recognised as leaders in their field, and the majority had at least 15 years' experience (Figure 2).

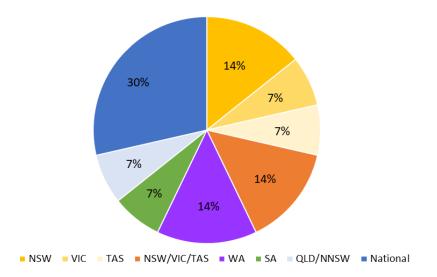


Figure 1. Summary of the location and regional expertise of interview participants

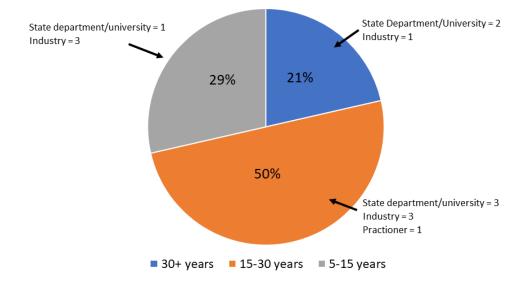


Figure 2. Summary of the number of years of experience of participants who were included in the interviews.

Interview questions were specific for each participant group and included key questions about the types of forages and pastures sown/sold within their area/company, their main use (*e.g.*, grazing by sheep, beef, dairy, or fodder production), area sown/volume of seed sold (where possible), and key traits (*e.g.*, high energy or protein, well adapted to a changing climate). Some individuals also shared summarised data, but this was very limited. See interview questions in Appendix 1.

3.1.2 Summarising interview data and literature

The most important species in Australia were separated into three groups: (1) first tier species, that were identified as a key species in at least ~50% of interviews; (2) second tier species, that were identified as a key species in ~20-50% of interviews; and (3) wildcard species that were identified once or twice or noted as a potential key species for the future. The ranking of these species was then cross-checked with data provided by interview participants, statistics from the MLA 'Analysis of Feedbase Audit (B.PAS.0297)' (Donald 2012) and the Australian Seed Federation industry review

(ASF 2022). Most of the species identified through the interview process were suited to southern production systems. At the request of MLA, we then included additional tropical and sub-tropical species widely used across northern Australia as wildcards. These species were identified through targeted discussions with scientists and extension staff from CSIRO and the Queensland Department of Agriculture and Fisheries that have extensive experience working with forages and livestock production systems in Northern Australia, and the relevant MLA program manager. Information provided was cross-checked with the Northern Australia Water Resource Assessment report (Ash et al. 2019) and a review of sown grass pastures (Peck et al. 2011).

All species were then assessed and ranked based on their suitability to future climates, potential for adoption and commercialisation, and ease of transformation. This assessment was based on expert opinion (*e.g.*, information collected from the interviews) and review of literature (*e.g.*, species descriptions from seed company websites, NSW DPI pasture factsheets). Six species were prioritised for further analysis: buffel grass, chicory, forage oats, forage sorghum, French serradella, and Phalaris.

3.2 Animal level analysis

3.2.1 Literature review

Information on the potential positive and negative animal impacts of increasing oil-content in pasture/forages was collected from the literature. This review focussed on ruminants and the potential impact of oil-enhancement on animal production (*i.e.*, liveweight gain and intake) and CH₄ emissions, including key fatty acids present in dietary lipids responsible for these changes. Due to the novelty of oil-enhancement in pasture/forages, literature was largely based on dietary lipids from supplementary sources (*e.g.*, sunflower oil, linseed oil etc.) that are currently used in ruminant production industries.

3.2.2 Animal production modelling

Animal production modelling was used to understand the productivity benefits of feeding oilenhanced forages to livestock. Ten scenarios were simulated, comprising a combination of the six species prioritised in the sectoral analysis and livestock systems where they are most often utilised (Table 1). First, we estimated the amount of grazable biomass for each forage species based on total annual DM production/ha, corrected for wastage. We then estimated potential DM intake (DMI) and productivity based on feed quality and livestock class. This information was combined to calculate economic impacts based on total grazing days and livestock productivity.

Scenario	Pasture/forage species	Annual/Biennial/Perennial	Livestock type/class
1	Buffel grass	Perennial	Beef steers
2	Chicory-clover/grass mix (50:50) ¹	Biennial	Prime lambs
3	Forage oats	Annual	Prime lambs
4			Beef steers
5	Forage sorghum	Annual	Beef steers
6	French serradella	Annual	Prime lambs
7			Dry ewes
8	Phalaris	Perennial	Prime lambs
9			Dry ewes
10			Beef steers

Table 1. The 10 scenarios included in the animal production modelling and economic analysis.

¹Chicory is most often utilised as a mixed forage and so was simulated as a 50:50 mix.

3.2.2.1 Pasture/forage biomass

Total biomass production was calculated using average growth rates for each month, summed to give total annual biomass production (t DM/ha.year, Table 2). This approach accounts for different growth rates of each species throughout the year, and months where no growth occurs. Regionally specific estimates of annual biomass production of Phalaris, forage oats and forage sorghum were calculated using the MLA Feed Demand Calculator (Meat & Livestock Australia Limited 2022) under standard-year seasonal conditions with growing wastage and carryover wastage of 20% each. The region selected for the annual biomass production estimates of these pasture/forage species best represented the region (or agroclimatic conditions) in which they are most typically grown. French serradella and buffel grass were not available in the Feed Demand Calculator, so average monthly growth rates were obtained from the literature (Bowen and Chudleigh 2018, Thomas et al. 2021). For the chicory-grass/clover mix, average annual biomass production was estimated using a combination of monthly growth rates of chicory from Hayes et al. (2006), and perennial ryegrass/white clover that was available in the Feed Demand Calculator (Table 2).

	NDF (%DM)	CP (%DM)	Ash (%DM)	Lipid (% DM)	Total biomass (t DM/ha.yr)	Grazeable biomass (t DM/ha.yr)
Buffel grass	63.7	9.7	10.0	3.0	5.1	3.06
Chicory-grass/clover mix ¹	33.4	23.0	10.4	3.2	9.6	5.76
Forage oats	47.9	17.0	10.7	2.5	11.9	7.14
Forage sorghum	55.0	16.8	9.0	3.9	14.3	8.58
French serradella	33.0	22.0	10.0	2.3	4.8	2.88
Phalaris	38.5	20.0	9.7	3.4	7.0	4.20

Table 2. Baseline nutritive value and biomass of modelled pastures
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¹50% chicory mix. Nutritive value for chicory-grass/clover mix accounts for proportion of species in mix. Total fat lipid content was only adjusted for the chicory.

Since not all biomass produced is consumed by livestock, yearly biomass production was adjusted to account for wastage of both fresh and carryover biomass, with a biomass utilisation rate of 60% (Table 2).

(Equation 1) Grazable biomass (t DM/ha.yr) = Total yearly biomass (t DM/ha.yr) × 0.6

3.2.2.2 Livestock productivity

The potential impact of increasing lipid content on average ME content of forages, DMI, daily liveweight gain (LWG), and CH₄ production of grazing ruminants was investigated using the Small Ruminant Nutrition System (SRNS) and Large Ruminant Nutrition System (LRNS) (Tedeschi and Fox 2020). These software platforms were selected due to the ability to set specific animal details and modify individual dietary components, including total fat lipid content. However, the SRNS model was unable to predict changes in wool yields, and this was not captured in our analysis.

The SRNS and LRNS models calculate the ME content of forages based on NDF adjusted for crude protein, crude protein (CP), digestibility of CP, indigestible acid detergent insoluble CP, ash, lipid content and lignin. Pasture species were selected from feed libraries available within the SRNS and LRNS software, and nutritive value parameters were adjusted based on average values detailed in the literature for each species (Table 2; Hayes et al. 2010, Lang 2001, Niderkorn et al. 2019, NSW DPI 2022, Pereira-Crespo et al. 2012, Uebergang 2016, Watson et al. 2000). For each pasture species, we simulated the baseline total lipid level (unique to each species) as well as the impact of increasing lipid content to 5%, 6%, 7%, and 8% DM. A maximum lipid content of 8% DM was selected as levels >10% DM are known to adversely impact upon animal production and health.

Model parameters for beef steers, prime lambs and dry ewes are summarised in Table 3. The production goal of prime lamb and steer scenarios was to support continual growth to attain a target market or feedlot weight. The production goal of the dry ewe scenario was to maintain mature weight. Animal management and environmental parameters were kept constant for each livestock species/class, including those in the buffel grass simulations since the area of use for this species spans across a broad range of environments of varying climatic conditions, and is not only isolated to northern Australia (Friedel et al. 2006). The LRNS and SRNS models were used to predict potential grazed DMI of each pasture/forage species and subsequent livestock production (both on a per animal/day basis), with feed intake limited by animal size and feed quality, rather than availability. The models predict DMI using equations that account for the energy required to support the growth of a specified species/livestock class based on their age, current liveweight, and mature and/or target liveweight, as well as accounting for the nutritive value of the feed.

Livestock	Livestock breed	Mature LW	Starting age	Starting LW	Target LW	Price
class		(kg)	(months)	(kg)	(kg)	(\$/kg LW)
Beef steers	Angus	500	6	240	300	5.40
Prime lambs	Merino x Border Leicester	55	4	28	50	3.38
Dry ewes	Medium Merino	50	24	50	NA	2.48

Table 3. Details of beef steer, prime lamb, and dry ewe scenarios

Livestock prices were obtained from the Meat and Livestock Australia website (<u>https://www.mla.com.au/prices-markets/</u>). Sheep prices in the report were based on a carcase weight basis and a 45% dressing percentage was applied to convert prices to a LW basis.

3.2.2.3 Productivity comparisons

Grazable biomass production and modelled DMI and LWG were used to calculate grazing day value to allow comparisons between the different scenarios. Total grazing days was calculated using grazable biomass and predicted DMI outputted by the model (Equation 2).

(Equation 2) Total grazing days (days/ha.yr) = Grazable biomass (kg DM/ha.yr) / predicted DMI (kg/head.day)

To calculate total grazing day value, a meat profit was calculated using average daily LWG outputted by the model and liveweight prices for the different livestock classes (Table 2). Total grazing day value was then calculated using Equation (3).

(Equation 3) Total grazing day value (\$/ha.yr) = Total grazing days × meat profit (\$/kg LWG.head.day)

Daily CH₄ emissions were calculated using DMI of sheep (Howden and White 1994) (Equation 4) and cattle (Charmley et al. 2016) (Equation 5) using the same approach applied in the Australian National Greenhouse Gas Inventory (Commonwealth of Australia 2021).

(Equation 4) CH_4 emissions from sheep $(g/d) = (DMI (g/d) \times 0.0188 + 0.00158) \times 1000$

(Equation 5) CH_4 emissions from cattle (g/d) = 20.7 (± 0.28) × DMI (kg/d)

Methane efficiency was calculated based on calculated CH₄ emissions estimates and average daily LWG outputted by the model (Equation 6).

(Equation 6) CH_4 efficiency (g $CH_4/kg LWG$) = CH_4 emissions (g/d)/average daily LWG (kg/head.d)

3.3 Cost-benefit analysis

3.3.1 Scenarios

A partial discounted cash flow analysis was carried out on the 10 scenarios used in the animal production modelling (Table 1). Results from the animal production modelling identified linear improvements in average daily LWG, CH₄ efficiency, and total grazing day value for all pasture/forage species and livestock classes with increasing fat lipid content to a maximum of 8%. Since fat lipid content of 8% was always optimal, only these were included in the cost-benefit analysis.

3.3.2 Partial discounted cash flow analysis

A partial discounted cash flow analysis was used on a per hectare basis to compare the alternative scenarios over a 10-year period. A partial discounted cash flow analysis only includes resources that will be changed, *i.e.*, it does not consider the resources in the business that are left unchanged. The long-term profitability of each scenario is summarised as the net present value of future cash flows, or the sum of future net cash flows discounted to their present value. Net present value (\$) is defined as:

(Equation 7)

Net present value =
$$\sum_{t=0}^{t=10} \frac{I-0}{(1+r)^t}$$

where the sum of annual cash inflows (*I*) over a period of 10 years, is calculated as the revenue from total grazing day value (\$/ha.year), and the sum of annual cash outflows (*O*) includes annual cost of seeding the forage crop (\$/ha per reseeding year) and the cost of annual CH₄ emissions – calculated as CH₄ efficiency converted to CO₂ equivalents using the global warming potential (GWP) of 34 for CH₄ relative to CO₂ (IPCC 2022) and multiplied by an assumed price for CO₂ of \$25/tonne. A real discount rate (r) of 4.0% is used in the analysis to determine the present value of future cash flows, based on rates of return experienced for general investments in agriculture in recent years. The relative benefit of oil-enhanced scenarios is discussed in terms of percent change relative to baseline.

3.3.3 Parameterisation

The input parameters for the cost-benefit analysis included pasture/forage biomass production, total grazing day value, and CH₄ efficiency outputs from the 10 scenarios in the animal production modelling. For all pasture/forage species, biomass production was kept fixed every year, except buffel grass, which is grown in the northern livestock systems that rarely invest in improved pastures. Hence a decay rate of 55% over 10 years was included in the biomass calculation (Peck et al. 2011).

The cost of establishment for each scenario required data on sowing rate (kg/ha), seed cost (\$/kg), fertiliser cost (AUD\$/ha; applied at sowing), and cost of the sowing/fertiliser operation based on \$19/ha with a medium tractor of 245 hP (incl. diesel and lubricants for tractor, labour at \$27.60/h) (Brook Anderson, CSIRO, pers. comm., 2022). All scenarios were assumed to incur an extra 10% seed cost premium for the oil-enhancement technology, with an expected annual decrease of 0.5% p.a. (Srinivas Belide, CSIRO, pers. comm., 2022). Sowing costs were incurred only in the reseeding years throughout the analysis. The exception was buffel grass, which was assumed to be sown in the first year and then renovated (via blade ploughing or deep ripping) every 10 years to remove woody weeds and reactivate soil nitrogen that can limit biomass production otherwise. Resowing of buffel grass is an uncommon practice for buffel grass (Kylie Hopkins, QDAF, pers. comm., 2022); hence, only machinery costs were included every 10 years (\$19/ha). Input data for the partial discount cash flow analysis is shown in Table 4.

Pasture/forage	Years to re-seed	Sowing rate (kg/ha)	Seed price ² (\$/kg)	Fertiliser/ inoculant cost (\$/ha)	Total cost of establishment (\$/ha.yr) ³
Buffel grass	10 ¹	3	15.0	0	64
Chicory-grass/clover mix	2	6	25.5	50	222
Forage oats	1	50	1.92	50	165
Forage sorghum	1	8	10.9	50	156
French serradella	4	6	10.0	60	139
Phalaris	10	3	20.0	0	79

Table 4. Assumed input parameters for the six pasture/forage species as used in the partial discounted cash flow analysis.

¹Buffel grass is very rarely resown and is rather renovated every 10 years (assumed a ploughing/ripping operation cost of \$19/ha with a medium tractor of 245 hP); ²As sold – treated and coated; ³Calculated as the sum of seed cost [(seed rate x seed price) + cost of fertiliser/inoculum + cost of sowing/fertilising operation].

3.3.4 Sensitivity analysis

Sensitivity analyses were carried out to account for changes in several input parameters for the partial discount cash flow analysis (Table 5). A higher seed premium of oil-enhanced technology of 25% was tested from the default 10% assumption. The cost of seed varies widely, depending on variations in seed price, that is driven by species cultivar, seasonal seed supply, and reseller profit margins as well as sowing rate (varies between production environments). For example, while forage oats is rarely sown at more than 50 kg/ha in lower production environments where it is more typically used for grazing only (*e.g.*, sub-tropic, semi-humid environments), it can be sown at 100 kg/ha in higher rainfall, temperate environments in the southern Australia, where it may be used for both grazing and hay production. Hence, we conducted a sensitivity on the seed cost (+50%). Given potential changes in the market price of beef and lamb, a +20% change in livestock meat price was also applied.

Table 5. Changes made to parameters to test for sensitivity of the cost-benefit analysis for the six	
pasture/forage species.	

Parameter	Change (%)
Biomass (t DM/ha.yr; standard year seasonal conditions)	+40 (good year seasonal conditions) -55 (poor year seasonal conditions
Seed premium of oil-enhanced technology (\$/ha)	+25
Seed cost (\$/kg)	+50
Livestock meat price (\$/kg)	+20
Combined seed premium (\$/ha) + seed cost (\$/kg)	+25 and +50, respectively
Combined seed premium (\$/ha) + livestock meat price (\$/kg)	+25 and +20, respectively

3.4 Adoption of oil-enhanced forages

We investigated the use of CSIRO's ADOPT tool (Kuehne et al. 2017; https://adopt.csiro.au) to explore the potential adoption of oil-enhanced pastures/forages across Australian livestock industries. The tool uses a series of 22 questions (Appendix 2) to estimate the peak adoption level

and time to peak adoption based on the relative advantage of the innovation, population-specific influences on the ability to learn about the innovation, the learnability characteristics of the population, and the relative advantage for the population. The tool was piloted with a small number of experts. However, we found that their answers were highly context-dependant, with experts providing different answers for different species, and for different farming systems or regions. Despite their expertise, the researchers and practitioners interviewed also expressed low confidence in their responses to many questions. Kuehne et al. (2017) note that ADOPT makes predictions based on a stable environment, with factors such as the cost of oil-enhanced forages, livestock market prices, legislation relating to gene editing, and variable climates all changing rapidly and likely to influence the acceptability and uptake of this technology. These likely contributed to the high variation and low confidence in the answers received, giving us low confidence in the results predicted by ADOPT. Based on this, the decision was made not to pursue this analysis.

4. Results

4.1 Forages of importance to Australian livestock industries

Grazing is largest agricultural land use in Australia, covering 341 million ha in 2016-17 (ABS 2018). Of this, 36 million ha (10%) are sown to improved pastures, with the rest being natural or unimproved pastures and grazing lands (Table 6). We used the area and number of agricultural businesses with improved pasture as an indication of where the oils transformation technology would have the highest potential uptake. As a proportion of pasture area, improved pastures are concentrated in Victoria and Tasmania, with 75% and 67% of grazing land in these states sown to improved pasture. The highest proportion of businesses with improved pastures are found in Tasmania (94%), Victoria (90%), Western Australia (85%), South Australia (75%) and NSW/ACT (73%). While Queensland had the largest area of improved pastures, this represents only a small proportion (12%) of total grazing area.

	Grazing area (million ha)	Area improved pastures (million ha)	Proportion of grazing area with improved pasture (%)	Proportion of businesses with improved pasture (%)
Tasmania	1.0	0.7	67.3	94
Victoria	5.6	4.2	75.1	90
Western Australia	69.8	3.9	5.5	85
South Australia	42.5	2.9	6.9	75
NSW/ACT	42.6	8.7	20.4	73
Queensland	129.4	15.1	11.7	59
Northern Territory	49.9	0.1	0.2	22
Total National	340.8	35.6	10.4	77

Table 6. Area of land mainly used for grazing, area of improved pastures and proportion of businesses with improved pastures (ABS 2018). States are ranked based on the proportion of businesses with improved pastures.

Recent data on area of specific pasture species sown is not publicly available, and raw data collected during industry interviews is unable to be included in this report due to privacy reasons. However, a recent assessment on the value of the Australian pasture seed industry (ASF 2022) estimated that of the 2.4 million ha of pastures and forage crops sown in 2020-21, 33% was temperate perennial pastures (*e.g.*, perennial ryegrass, lucerne), 25% was summer forage crops (*e.g.*, forage brassicas and

sorghum), 21% was annual pastures (*e.g.*, annual ryegrass, sub clover), 13% was winter forage crops (*e.g.*, forage oats, triticale, barley) and 8% was sub-tropical perennial pastures (*e.g.*, Rhodes grass, buffel grass). While these data do not reflect inter-year variation in the types of pastures sown, they show the importance of temperate perennial pastures, summer forage crops and annual pastures to Australian livestock industries. The Australian Seed Federation also reported that improved pastures provided most value to sheep and dairy industries, contributing 21%, 14% and 12% of farm gate value to wool, milk, and sheep meat production, respectively, but only 6% to beef production. Combined with the information gleaned from national statistics summarised above, this suggests that the greatest uptake of improved pastures is likely to be in the southern states.

The best estimate of the area of pasture species sown in southern Australia is the report of Donald et al. (2012). In this report, the authors estimated the area of various pasture legumes and grasses across NSW, South Australia, Tasmania, Victoria, and southwest Western Australia in 2011 (Table 7). While this report is now a decade old, and some of these species were relatively new to Australian livestock industries at the time the research was conducted, this data highlights the huge importance of sown crops, subterranean clover, annual ryegrass, lucerne, Phalaris and perennial ryegrass.

Species	Common name	Area (thousand ha)
Various	Sown crops	6,020
Trifolium subterraneum	Subterranean clover	3,556
Lolium rigidum	Annual ryegrass	2,782
Medicago sativa	Lucerne	2,277
Phalaris aquatica	Phalaris	1,965
Lolium perenne	Perennial ryegrass	1,591
Dactylis glomerata	Cocksfoot	874
Trifolium repens	White clover	603
Medicago polymorpha	Burr medic	579
Pennisetum clandestinum	Kikuyu	530
Festuca arundinacea	Fescue	453
Medicago littoralis	Strand medic	377
Medicago truncatula	Barrel medic	377
Ornithopus sp.	Serradella	149
Trifolium resupinatum	Persian clover	108
Setaria sp.	Seteria	88
Trifolium michelianum	Balansa clover	73
Trifolium fragiferum	Strawberry clover	41
Trifolium pratense	Red clover	20
Biserrula pelecinus	Biserrula	16
Trifolium pilulare	Ball clover	16
Puccinellia ciliata	Puccinellia	8

Table 7. Estimated area of improved pasture species across southern Australia in 2011 (Donald et
al. 2012). Species are ranked from greatest to least area sown.

Information gleaned from the industry interviews in this project provided a similar story (Table 8). Species identified were primarily suited to temperate, Mediterranean, and sub-humid climates, reflecting the current market focus and greater use of improved forages in southern regions. Species identified as Tier 1 species (most often ranked species) from the interview process; ryegrasses, subterranean clover, lucerne, and Phalaris, were also reported by Donald et al. (2012) as species with greatest area sown. Wild card species were not mentioned in Donald et al. (2012) but were highlighted in the industry interviews and by MLA program managers as species that may be of increasing importance to Australian livestock industries in the future.

Table 8. Forages of importance to Australian livestock industries. Tier 1 species were prioritised by >50% of interviewees and Tier 2 species by at least 20% of interviewees. Wildcard species were noted as potentially important species for northern Australia or future livestock industries. Species within each Tier are listed in alphabetical order.

Grouping	Species	Common name	Plant type	Climate	Use
Tier 1	<i>Lolium rigidum</i> Gaud.	Annual ryegrass	Annual grass	Temperate	Dairy, beef
	<i>Lolium multiflorum</i> Lam.		Annual grass	Temperate	Dairy, beef
	Medicago sativa L.	ryegrass Lucerne	Perennial legume	Mediterranean; Temperate, sub- humid	Sheep, some hay/silage
	Lolium perenne L.	Perennial ryegrass	Perennial grass	Temperate	Dairy (80-90%), high rainfall beef/prime lambs (10-20%)
	Phalaris aquatica L.	Phalaris	Perennial grass	Widespread. Temperate; temperate, sub- humid	Sheep and beef (dryland)
	Trifolium subterraneum L.	Subterranean clover	Annual legume	Mediterranean; Temperate, sub- humid	Sheep and beef (dryland)
Tier 2	Dactylis glomerata L.	Cocksfoot	Perennial grass	Temperate; temperate, sub- humid	Sheep and beef (dryland)/dairy (newer types)
	Triticum aestivum L.	Dual-purpose wheat	Annual grass	Temperate; Temperate, sub- humid	Sheep
	Festuca arundinacea L.	Fescue	Perennial grass	Temperate; temperate, sub- humid	Sheep and beef (dryland) - beginning to be used in dairy
	Avena sativa L.	Forage oats	Annual grass	Widespread	Sheep and beef (dryland), export hay markets
	Brassica napus var. biennis L.	Forage rape	Annual/biennial brassica	Temperate; Temperate, sub- humid	Sheep (~70%) and dairy (~30%)
	<i>Ornithopus sativus</i> Brot.	French serradella	Annual legume	Mediterranean	Sheep
	Trifolium repens L.	White clover	Perennial legume	Temperate	Dairy (predominant), high rainfall beef and prime lambs
Wildcard	Panicum coloratum L.	Bambatsi	Perennial grass	Tropical	Beef
	Trifolium spumosum L.	Bladder clover	Annual legume	Mediterranean; Temperate, sub- humid	Sheep
	Cenchrus ciliaris	Buffel grass	Perennial grass	Widespread	Beef

Centrose pascuor	-	Centro	Perennial legume	Tropical; Sub- tropical	Beef
Chicoriu	m intybus L.	Chicory	Perennial herb	Temperate; Temperate, sub- humid	Dairy, sheep, beef
Vicia sat sativa L.	•	Common vetch	Annual legume	Mediterranean; Temperate, sub- humid	Sheep
Desman	thus spp.	Desmanthus	Perennial legume	Tropical	Beef
Brassica annua L	napus var.	Dual-purpose canola	Annual brassica	Temperate; Mediterranean	Sheep
Sorghun	n vulgare L.	Forage sorghum	Perennial grass	Sub-tropic, semi-humid	Beef
Penniset clandest Hochst.	-	Kikuyu	Perennial grass	Temperate	Dairy, beef
Plantage L.	o lanceolata	Plantain	Perennial herb	Temperate; Temperate, sub- humid	Dairy, sheep, beef
<i>Bromus</i> Kunth.	wildenowie	Prairie grass	Annual/short term perennial grass	Temperate; Temperate, sub- humid	Sheep, beef
Chloris g	layana	Rhodes grass	Perennial grass	Tropical; Sub- tropical	Beef and hay
<i>Medicag</i> Loisel.	o littoralis	Strand medic	Annual legume	Mediterranean	Sheep
(Shrubb Caribbea	an, Fine- Id Caatinga	Stylo	Biennial or short-lived perennial legume	Tropical	Beef
Bitumino	aria	Tedera	Perennial legume	Mediterranean	Sheep

4.2 Prioritising forages for future investment

Forages identified during the interview process were assessed for their:

- Suitability for future climates (section 4.2.1).
- Potential for commercialisation and adoption (section 4.2.2).
- Ease of transformation (section 4.2.3).

4.2.1 Suitability of forages for future climates

In general, the Australian climate is expected to become hotter and drier in the future. These changes are likely to result in a change in growing season, higher inter-annual variability in pasture/forage yields, impact water availability (including in irrigated systems), and challenge the persistence and sustainability of pasture/forage species that are best suited to temperate cool season wet environments. Specific climate challenges and implications for pasture/forages across the eight major National Resource Management (NRM) regions (Figure 3) are summarised in Table 9 below.

Based on future climate predictions, pasture/forage species were individually assessed on their suitability for future climates (Table 10). Both annual and perennial species with deeper-root systems, and high responsiveness to summer and/or autumn rainfall (e.g., French serradella, common vetch, Lucerne, Tedera, Phalaris, buffel grass, bambasti panic, and desmanthus) were highly favourable for future climates. In areas where increased summer rainfall is expected, deeprooted, summer active perennial species (e.g., Lucerne, Tedera, summer active fescues, kikuyu, Rhodes grass, chicory, and plantain) may become important. The use of summer-active forage crops like forage sorghum, is also likely to expand into other areas that are currently limited by low/unreliable summer rainfall. In monsoonal areas and wet tropic areas of Northern Australia, species such as Centro are still likely to serve an important role in grazing and hay production systems with some cultivars more tolerant to periods of waterlogging. Deep-rooted, drought tolerant Caatinga and fine-stem stylos are likely to remain relevant in sub-humid, sub-tropic areas within rangeland and east coast regions that are expected to experience shorter growing seasons and out-of-season rainfall events. However, in monsoonal north and wet tropic regions, the use of Shrubby and Caribbean stylos may be impacted by waterlogging. In these regions, there may also be greater reliance on stylo cultivars more tolerant to anthracnose (Colletotrichum gloeosporioides) (e.g., Siran, Amiga) to cope with potential fungal outbreaks and increased susceptibility of some cultivars under more wet, humid conditions.

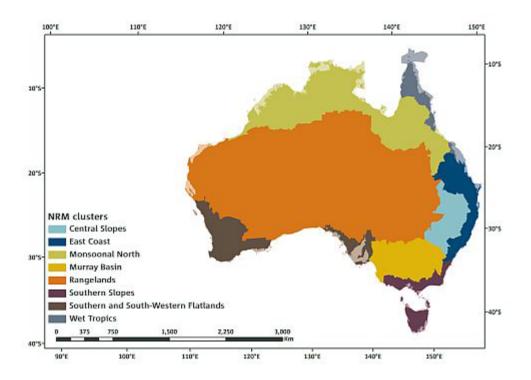


Figure 3. NRM regions in Australia (source: <u>Climate Change in Australia website</u>, CSIRO and Bureau of Meteorology).

Table 9. Climate projections and implications for pastures in major Natural Resource Management (NRM) regions. Information on current and future climates is taken from the <u>Climate Change in Australia website.</u> Land use information is based on the <u>Australian Land Use and Management</u> <u>Classification</u> (ABARES 2021)

NRM region	Livestock production systems	Current climate	Climate projections	Implications for pastures
Central slopes (Ekström et al. 2015)	Mostly beef and sheep, with some dairy production. Mixed crop-livestock systems are common. Livestock graze both native and modified pastures.	Temperate to sub-tropical Wetter summer/drier winter Annual rainfall: 150-500 mm/year	 Average temperatures will continue to increase in all seasons. More hot days and warm spells, and fewer frosts. Average winter and spring rainfall is projected to decrease. Changes in summer and autumn rainfall are possible but unclear. Increased intensity of extreme daily rainfall events. 	Shorter growing season, especially over summer and winter. More reliance on tropical pastures and deep-rooted perennials compared to shallow rooted temperate species.
East coast (Dowdy et al. 2015)	Mostly beef production with some sheep and dairy. Grazing of both native and modified pastures.	Predominantly sub-tropical Wet summer/dry winter Annual rainfall: ~500-900 mm/year	 Average temperatures will continue to increase in all seasons. More hot days and warm spells, and fewer frosts. Decreases in winter rainfall are projected for East Coast South. Other changes in rainfall are possible but unclear. Increased intensity of extreme daily rainfall events. 	Shorter growing season, especially over summer and winter. More reliance on tropical pastures and deep-rooted perennials compared to shallow rooted temperate species (<i>e.g.</i> , perennial ryegrass).
Monsoonal North (Moise et al. 2015)	Mostly extensive beef production. Livestock graze mostly native vegetation.	Tropical Very wet summer/dry winter Annual rainfall: 600-1600 mm/year	 Average temperatures will continue to increase in all seasons. More hot days and warm spells. Changes to rainfall are possible but unclear. Increased intensity of extreme daily rainfall events. Fewer but more intense tropical cyclones. 	Continued reliance on persistent tropical pasture/forages that can withstand waterlogging.

Murray Basin (Timbal et al. 2015)	Mostly sheep and beef production with some dairy. Mixed crop-livestock systems are common. Livestock graze both native and modified pastures, including some irrigated pasture.	Temperate Winter dominant rainfall Annual rainfall: 250-2000 mm/year	 Average temperatures will continue to increase in all seasons. More hot days and warm spells, and fewer frosts. Less rainfall is projected during the cool season. Rainfall may remain unchanged in the warm season. Increased intensity of extreme daily rainfall events. 	Shorter growing season in winter and spring with greater reliance on species with higher heat and drought tolerance, including deeper-rooted perennials.
Rangelands (Watterson et al. 2015)	Extensively grazed beef and sheep production. No dairy. Livestock mostly graze native vegetation.	Arid & semi-arid High inter-annual variation in rainfall Annual rainfall: 200-500 mm/year	 Average temperatures will continue to increase in all seasons. More hot days and warm spells, and fewer frosts. Changes to summer rainfall are possible but unclear. Less winter rainfall is projected in the south. Increased intensity of extreme daily rainfall events. 	Shorter growing season in winter and spring. More reliance on summer rainfall. Opportunities for deep-rooted pasture/forage species, that are highly responsive to out-of-season rainfall events.
Southern Slopes (Grose et al. 2015)	Sheep, beef, and dairy production. Grazing of modified pastures, including some irrigated pastures.	Temperate, maritime Winter-dominate rainfall Annual rainfall: 500-3000 mm/year	 Average temperatures will continue to increase in all seasons. More hot days and warm spells, and fewer frosts. Generally, less rainfall in the cool season (winter and spring) is projected but with strong regional differences. Changes to summer rainfall are possible but less clear. Increased intensity of extreme daily rainfall events. 	Shorter growing season in winter and spring. More reliance on summer rainfall. Opportunities for deep rooted summer active pasture/forage species.

South and South-western Flatlands (Hope et al. 2015)	Mostly sheep production with some beef and dairy. Mixed crop-livestock systems are common. Grazing of modified pastures, including some irrigated pastures.	Mediterranean Winter-dominate rainfall Annual rainfall: 200-1200 mm/year	 Average temperatures will continue to increase in all seasons. More hot days and warm spells, and fewer frosts. A continuation of the trend of decreasing winter rainfall is projected. A decrease in spring rainfall is also projected. Changes in other seasons unclear. Increased intensity of extreme daily rainfall events. 	Shorter growing season in winter and spring. More reliance on deep rooted pasture/forage species with fast establishment and early grazing maturity.
Wet Tropics (McInnes et al. 2015)	Mostly extensive beef production based on native pastures.	Tropical Summer-dominant rainfall Annual rainfall: ~1450 mm/year	 Average temperatures will continue to increase in all seasons. More hot days and warm spells. Changes to rainfall possible but unclear. Increased intensity of extreme daily rainfall events. Fewer but more intense tropical cyclones. 	Continued reliance on persistent tropical pasture/forages that can withstand waterlogging.

Table 10. Assessment of the suitability of forages for future climates. The presence of a trait is indicated with X. Species with three or more traits were considered highly suitable, species with at least two traits were considered moderately suitable, and species with one or no traits were considered unsuitable.

Common name	Suitability for future climates	Traits	that enha	ance sui	tability f	for futur	e climate	S				Additional comments
		Hard seeded	Deep root system / high root density	Early flowering or maturing	Early plant vigour	Summer active	Drought/summer dormancy	Responsive to out of season rainfall	Low transpiration rate	Endophytic bacteria	Adapted to dry environments	
Annual ryegrass	Moderately suited							Х			Х	Can germinate in medium to high temperatures (10-30 °C)
Bambatsi panic	Highly suited		Х			Х		Х	Х		Х	
Bladder clover	Highly suited	X	Х	Х	Х						Х	
Buffel grass	Highly suited		Х			Х		Х	Х			Most drought-tolerant introduced grass
Centro	Moderately suited		X				X					Grows well in >700 mm rainfall or irrigation. Typically grown in higher rainfall monsoonal areas of North QLD and NT. Can survive periods of waterlogging.
Chicory	Highly suited		Х		Х	Х					Х	
Cocksfoot	Highly suited		Х				Х	Х			Х	
Common vetch	Highly suited		Х		Х			Х	Х		Х	Heat tolerant
Desmanthus	Highly suited	Х	Х			Х		Х				
Dual-purpose canola	Highly suited - forage production only		Х		Х						Х	Poor seed yield and quality under dry conditions that limits grain profitability.
Dual-purpose wheat	Highly suited		Х		Х						Х	Broad range of cultivars that are highly adapted to a range of environments
Fescue	Highly suited		Х			Х	X			Х		
Forage oats	Highly suited		Х		Х						Х	

Forage rape	Highly suited		X		X	X		X			X	Wide sowing window (autumn and spring planting possible).
Forage sorghum	Highly suited		X		X	X			X		X	Thick wax leaves that enhance drought tolerance.
French serradella	Highly suited	X	X	X	X			X			X	
Italian ryegrass	Not suitable							X				Shallower rooted annual; cool season growth; grows well in areas with >650 mm rainfall.
Kikuyu	Highly suited		X		X	X			X		X	Heat tolerance; peak production in warmer and drier summer months.
Lucerne	Highly suited		X			X		X			X	
Perennial ryegrass	Not suitable									X		Poor persistence and growth in hotter, drier climates that will impact growing season length; area of adaptation expected to decline with 30-50% of current area at risk.
Phalaris	Highly suited		X				X	X		X	X	Partial summer dormancy (persistent in summer drought);
Plantain	Highly suited		X		X	X						
Prairie grass	Moderately suited		X			X						
Rhodes grass	Moderately suited		X		X							Earlier maturing cultivars can have greater drought tolerance. Grows well in regions with >600 mm summer rainfall.
Strand medic	Highly suited	X			Х						X	
Stylo spp. (Shrubby, Caribbean, Fine- stem, and Caatinga stylo only)	Highly suited	X	X	X		X						Caatinga stylo grows well in areas with >500 mm annual rainfall. Shrubby and Caribbean stylos grow well in areas with >600 mm annual rainfall. Fine- stem stylo grows well in areas >700 mm annual rainfall. Poorly adapted to soils prone to seasonal flooding.

Subterranean	Moderately suited	X						Х	Failure to set seed in drought reducing
clover									persistence; some harder seeded
									cultivars.
Tedera	Highly suited		Х		x	Х		Х	Ability to survive hot dry summers and
									respond to out-of-season rainfall,
									though is most productive in medium-
									high rainfall environments (>300 mm
									annual rainfall)
White clover	Not suitable								

4.2.2 Likelihood of commercialisation and adoption

The willingness of seed companies to invest in improving specific forages was assessed based on information provided in the interviews (Table 11). While this assessment does not encompass all the seed companies operating within Australia, important insights are covered. Seed companies are a major player in forage improvement in Australia and play a key role in supporting the adoption of new species and cultivars. Seed companies will invest in species with the highest return on investment, and there is an increasing focus on improving the feeding value of drought tolerant species in anticipation of feed gaps that may arise in response to a changing climate, including those in southern Australia. However, investment by companies in species improvement is tempered by the willingness of producers to pay for new technologies. Historically, this has provided a focus on more intensive and high value production systems, especially dairy. Due to the limited domestic market, commercial investment in species more suited to beef and sheep production is more likely where there is also an international market. Company interest and investment for pasture/forage species is also influenced by whether they are classified as public vs. proprietary varieties since seed companies receive limited or no return on investment for public varieties that can be easily harvested and sold on-farm. Public varieties are more likely to receive interest from government or industry-owned research and development corporations with a primary focus on industry impact.

We also considered potential barriers such as the availability of locally relevant agronomic/grazing management guidelines, risks of animal health problems, how forages can be integrated into existing farming systems and potential weediness or other environmental considerations (Table 11). Many of these issues impact both the likelihood of adoption and investment by seed companies. In particular, we'd like to highlight the case of Buffel grass, which despite being an important and widely used species in pastoral systems across Australia presents several barriers to commercialisation and adoption in the context of this analysis. Most notably, it's potential as a serious environmental weed (Grice et al. 2012). State government agencies emphasise that buffel grass is associated with fire risks and competition with native species, leading to potential decreases in biodiversity and pasture productivity, with long-term economic impacts (Biosecurity SA 2019, Northern Territory Government 2020, DPIRD 2023). Buffel grass is a declared weed in South Australia and weed management plans exist in other states. Thus, breeders targeting improvements to buffel grass or other potentially weedy species may need to consider breeding sterile pasture varieties, increasing investment and propagation costs.

We recommend that initial investment in oil-enhanced forages focuses on species that are both climate resilient and have already been adopted in large areas. While newer/novel species may have important roles in the longer term, the value of this technology is more likely to be proven to industry by focusing on species that are currently widely used.

Species	Barriers to adoption	Potential interest from seed companies	Barriers to seed company investment
Annual ryegrass	-	Yes	• Limited suitability to future climates (becoming a lower priority for research and development).
Bambatsi panic	-	No	 Public variety precludes seed company involvement in supporting adoption and future research and development.
Bladder clover	-	No	 Not widely adopted currently. Bartolo (only current variety) is a public variety, precluding seed company involvement in supporting adoption and future research and development.
Buffel grass	 Difficult to sow. Potentially weedy and highly competitive with other sown and native species. Slow to establish – not suited to short term pasture. 	No	Already naturalised across large areas of Australia
Centro	 Area of adaptation limited to high rainfall/monsoonal areas of northern QLD and NT. 	Yes	Naturalised across large areas of northern Australia
Chicory	 Lacking detailed agronomic management guidelines for Australian systems. 	Yes	Not widely adopted currently.
Cocksfoot	_	Yes	-
Common vetch	-	No	Not currently widely adopted.
Desmanthus	 Agronomic management guidelines are still being developed. Woody stems are a disadvantage for use in crop rotations. 	Some	Not currently widely adopted.
Dual-purpose canola	 Photosensitisation and nitrate poisoning risk for livestock. 	Yes	-

Table 11. Factors that may	influence commercialisation and	d adoption of	improved forages.

Dual-purpose wheat	-	Yes	-
Fescue	Risk of fescue foot in livestock caused by endophytes.	Yes	-
Forage oats	-	Yes	-
Forage rape	 Poor understanding of agronomic management outside southern livestock only and cropping regions. Photosensitisation and nitrate poisoning risk to livestock. 	Yes	 Not currently widely adopted outside of higher rainfall livestock systems.
Forage sorghum	 Risk of poisoning to livestock from cyanogenic glycosides (especially in drought conditions). 	Yes	-
French serradella	-	No	Not widely adopted outside of Western Australia.
Italian ryegrass	-	Yes	Limited suitability to future climates.
Kikuyu	 Requires good management to maintain quality. High weed potential. 	Yes	-
Lucerne	 Poor tolerance of acidic soils and waterlogging. Requires rotational cutting/grazing for best yield and persistence. Limited use as a break crop in low-rainfall cropping systems. High risk for bloat, red gut in livestock. 	Yes	-
Perennial ryegrass	-	Yes	Limited suitability to future climates.
Phalaris	Potential to cause Phalaris poisoning in livestock.	Yes	Limited international market outside of Australia and Argentina.
Plantain	 Lacking detailed agronomic management guidelines for Australian systems. 	Yes	Not currently widely adopted.

Prairie grass	-	Yes	 Not currently widely adopted. Limited suitability to future climates.
Rhodes grass	 Rapid drop in feed quality upon flowering. High water and nitrogen inputs needed for high yield and quality (especially for hay production). 	Yes	-
Strand medic	Risk of bloat in cattle and red gut in sheep.	No	Not currently widely adopted.
Stylo <i>spp</i> . (Shrubby, Caribbean, Fine-stem, and Caatinga stylo only)	 Poor tolerance to waterlogging. Some cultivars are less tolerant to anthracnose fungus that may reduce the use of these cultivars in Monsoonal north and Wet Tropics where higher temperatures and more intense rainfall events are predicted. Reports of some species having lower palatability to livestock compared to grass- pastures. 	Some	
Subterranean clover	-	Yes	Limited suitability to future climates.
Tedera	 Some problems with persistence, especially in environments with cold wet winters. Seed is very expensive to purchase. 	No	Currently low levels of seed sales and adoption
White clover	Risk of bloat in livestock.	Yes	Limited suitability to future climates.

4.2.3 Ease of genetic transformation

There are no plant physiological barriers to oil-enhancement, and efficient genetic transformation methods are available for many of the potential species listed in Table 12. The only barriers to oil-enhancement, which can be overcome with time, funding and resources are the availability of: (1) a plant genome sequence, and (2) existing/non-protected transformation methods.

Species	Genome sequence available	Method available	Other comments	Timeframe for transformation
Annual ryegrass	No	No	A specific method for annual ryegrass is not available, but it is genetically similar to perennial ryegrass, and that method should be easily transferrable.	Mid-horizon
Bambatsi panic	No	No	Method not currently available, but there are groups in the USA working on <i>Panicum varigatum</i> for biodiesel	Late-horizon
Bladder clover	No	No		Late-horizon
Buffel grass	No	Yes		Mid-horizon
Centro	No	No		Late-horizon
Chicory	No	Yes		Mid-horizon
Cocksfoot	Yes	Yes		Early-horizon
Common vetch	Yes	Yes		Early-horizon
Desmanthus	No	No		Late-horizon
Dual-purpose canola	Yes	Yes	CSIRO has a method. Some very Early-ho preliminary data available.	
Dual-purpose wheat	Yes	Yes	CSIRO has a method	Early-horizon
Fescue	No	Yes		Mid-horizon
Forage oats	Yes	Yes		Early-horizon
Forage rape	Yes	Yes	CSIRO has a method. Some very preliminary data available.	Early-horizon
Forage Sorghum	Yes	Yes	CSIRO method has been used to provide 8% lipid content.	Early-horizon
French serradella	No	No		Late-horizon
Italian ryegrass	Yes	Yes	Methods available for 7 genotypes.	Early horizon
Kikuyu	No	No		Late-horizon
Lucerne	Yes	Yes	Multiple methods are available. Has been a focus of work in Canada and USA.	Early-horizon
Rhodes grass	No	Yes	Transcriptome Analysis is available	Mid-horizon
Perennial ryegrass	Yes	Yes	Method has been used extensively and can be applied to closely related grasses. 6-7% lipid content in leaves demonstrated by NZ researchers.	
Phalaris	No	No		Late-horizon
Plantain	No	No	Methods are currently being Mid-horizon developed	
Prairie grass	No	No		Late-horizon
Strand medic	No	No		Late-horizon
Stylo <i>spp.</i> (Shrubby, Caribbean, Fine-stem, and Caatinga stylo only)	No	No		Late-horizon
Subterranean clover	Yes	Yes		Early-horizon
Tedera	No	No	Late-horizon	
White clover				Early-horizon

Table 12. Readiness of transformation methods.
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High-quality reference genome sequences and extensive genomic resources are nowadays the keys to the discovery of genes and biological processes that are associated with traits of interest. Next generation sequencing platforms can produce millions of small DNA sequence reads in parallel, which need to be processed and compared to some references using bioinformatics pipelines. Therefore, an adequate reference genome sequence for the respective plant is a prerequisite for the analysis. A plant genome sequence cannot be protected by patents or IP, and where available, can be readily accessed for research and technology development. The reference genome sequence is available for many of the potential species listed in Table 12. In the absence of availability of genome sequence, transcriptome sequences can be generated by standard subcontracting to identify the potential target genes for alteration of oil content in various tissues.

Methods for oil transformation can be protected by companies and is the case with some high value crops. However, even where methods are protected, skilled research groups (such as those within the CSIRO Oils team) can develop new methods within ~6 months where a genome sequence is already available. For many of the potential species listed in Table 12, methods are available to deliver CRISPR/Cas9 genome editing components. The delivery of CRISPR/Cas9 components into plant cells and the regeneration of gene-edited lines is still challenging for some recalcitrant plant species. The CSIRO obtained a super binary/ternary vectors commercial license from JT Inc Japan for various monocots including sorghum. We are further negotiating a research licence for the use of very efficient morphogenic regulator gene(s) and transcription factors that enable the transformation of elite recalcitrant plant species. Our world-class plant genetic transformation expertise ranges from traditional Agrobacterium or phytoene desaturase (PDS)-mediated delivery to the use of Nanoparticles, Cell-Penetrating Peptides, Pollen magnetofection etc. In the past 10 years, the Synthetic Trait group at CSIRO has developed new plant varieties using genetic engineering technologies and de-regulated them for commercial use in Australia and many other countries. With its vast experience, CSIRO can quickly evaluate the most suitable method for the selected plant species and can deliver gene-edited lines.

The CSIRO Oils team reviewed the ranked species list and identified those that could be easily transformed based on existing transformation methods and an available genome sequence. Methods for the ranked species are not protected, except for sorghum, which is currently protected by CSIRO. This information allowed ranked species to be further classified into early-, mid- and late-horizon projects (Table 12).

Early-horizon projects have the potential to be completed within a relatively short time frame (~3 years) and at a lower cost. Transformation could be completed by the CSIRO Oils team using existing CSIRO methods (*e.g.*, forage rape, dual-purpose canola, dual-purpose wheat, sorghum, and white clover) or methods developed and published by other laboratories. Transformation of species with an existing genome sequence could be fast tracked further due to a greater understanding of genes to be targeted for oil-enhancing technology. Mid-horizon projects lack a genome sequence or available method but would take less time to complete than the late-horizon projects. Late-horizon projects would require significant time, funding and resources before transformation could be made possible.

There are other research groups throughout the world currently working on oil-enhancement in key pasture species (GM and non-GM). While the CSIRO Oils team is aware of work being conducted by other groups, the methods are not published or publicly available, so details cannot be included in this report. Although we have taken this into account in our selections, we cannot disclose any further information.

4.3 Ranking species for further evaluation

The list of forage species was ranked based on their suitability for future climates, potential for adoption, and then ease of transformation (Table 13). Interest from seed companies was not considered in this ranking since we only interviewed a small number of companies, and factors that influence their interest such as suitability for future climates and potential for adoption are already included. We used this ranking to inform the selection of species for further evaluation, but also considered the fit of species into farming systems and work being conducted by other research groups and seed companies internationally. Thus, the shortlist of species includes grasses, herbs, and legumes; species that are sown in monocultures and in mixes; and species that can be sown in permanent pastures as well as those used in crop rotations.

Table 13. Summary of species rankings. Species are scored from most (green, ***) to least (red, *) suitable, and then ranked first on suitability for future climates, followed by potential for adoption, and ease of transformation. Where multiple species were equally ranked, they are listed in alphabetical order.

Species	Suitability for future climate	Potential for adoption	Ease of transformation
Cocksfoot	***	***	***
Dual-purpose canola	***	***	***
Dual-purpose wheat	***	***	***
Forage oats	***	***	***
Forage Sorghum	***	***	***
Lucerne	***	***	***
Fescue	***	***	**
Bambatsi panic	***	***	*
Kikuyu	***	***	*
Phalaris	***	***	*
Stylosanthes	***	***	*
Common vetch	***	**	***
Forage rape	***	**	***
Buffel grass	***	**	**
Chicory	***	**	**
Plantain	***	**	**
Bladder clover	***	**	*
French serradella	***	**	*
Strand medic	***	**	*
Desmanthus	***	*	*
Tedera	***	*	*
Subterranean clover	**	***	***
Annual ryegrass	**	***	**
Rhodes grass	**	***	**
Centro	**	***	*
Prairie grass	**	**	*
Italian ryegrass	*	***	***
Perennial ryegrass	*	***	***
White clover	*	***	***

The forage species prioritised for further evaluation included:

- **Forage Sorghum**: Scored highly in all criteria. Although not currently widely used within Australia, it is well suited to future climates and northern Australia, there are international markets for an oil-enhanced cultivar, and the transformation method developed by CSIRO is ready for application.
- Forage oats: Forage oats are suited to a wide range of climates, and widely used across Australia in the sheep, beef, and dairy sectors. Oaten hay has a high-value export market, so we consider that producers would be more likely to pay for this technology in oats than wheat.
- Phalaris: Phalaris, Cocksfoot and Fescue fill a similar gap in farming systems, so we recommend choosing only one of these species for further evaluation. While a transformation method is not currently available for Phalaris, it was highly ranked in the industry interviews, and occupies a broad region based on existing literature (Donald et al. 2012). Seed companies indicated strong interest in future research and development for Phalaris based on its current place and future role in farming systems. It is also likely to have a wider area of adaptation than the other deep-rooted perennial C4 grasses.
- **Chicory**: Not currently widely used within Australia, but adoption is expected to increase with a strong interest from seed companies. Furthermore, higher summer rainfall that is predicted in some regions in the future will favour deep-rooted summer active perennials like chicory; hence they are expected to have a wider role in those regions. Unlike the other shortlisted species, chicory is used as part of a mixed pasture sward, providing a useful comparison.
- **Buffel grass**: Widely used across northern Australia and well adapted to future climates. Results from modelling of Buffel grass would also be applicable to Rhodes grass.
- French serradella: Despite the lower ranking, we recommend further evaluation of French serradella rather than Lucerne to understand impact of increased oil content in legumes. There has already been lots of work by researchers in Canada and North America to increase the oil content of Lucerne, so French serradella provides a new opportunity for Australian seed companies. French serradella is also likely to play a more significant role in cropping systems in the future, especially low-rainfall cropping systems where Lucerne is less-well suited in rotations.

4.4 Impacts of increased oil content on animal performance metrics

4.4.1 Review of literature

4.4.1.1 Dietary lipids from plants

Lipids occur naturally in structural (*e.g.*, leaves) and storage (*e.g.*, seeds) plant organs (McDonald et al. 2002). Because ruminants are grazing animals, the majority of lipids they consume come from structural lipids made up predominately of glycolipids (70-80% of leaf lipids), phospholipids (Dewanckele et al., 2020; Harfoot, 1981) and very small amounts of triglycerides (< 1% of total leaf lipids) (Yang & Ohlrogge, 2009). On the other hand, storage lipids are made up predominately of triglycerides, which are major constituents of concentrate and oil-based supplements (Dewanckele et al., 2020).

Plant lipids are made up of hundreds of saturated and unsaturated fatty acids; however, there are seven that predominate (Table 14). The fatty acid profile of structural and storage lipids varies considerably. Polyunsaturated α -linolenic fatty acid (an omega-3 fatty acid) is the most abundant lipid in structural plant organs, and palmitic and oleic fatty acids are the most abundant saturated, and monosaturated fatty acids, respectively (McDonald et al. 2002). Palmitic, stearic, oleic, linoleic (an omega-6 fatty acid), and α -linolenic fatty acids are the most abundant fatty acids in storage plant organs (Weselake, 2005).

Table 14. Most abundant fatty acids present in plant structural and storage lipids. Adapted from
McDonald et al. (2002).

Carbon structure	Fatty acid	
Saturated		
14:0	Myristic	
16:0	Palmitic	
18:0	Stearic	
Unsaturated		
16:1	Palmitoleic (monosaturated)	
18:1	Oleic (monosaturated)	
18:2	Linoleic (polyunsaturated)	
18:3	α-linolenic (polyunsaturated)	

4.4.1.2 Enhancing triglycerides in the vegetative plant biomass via oil enhancement technology

Oil enhancing technologies have gained considerable attention in recent years due the increasing demand for plant oils (increase by 16%) by 2026 (FAO, 2017). Majority of current plant oils come from triglycerides derived from plant storage organs (*e.g.*, seeds). However, since plant leaves account for most of the above ground biomass, advances in technology for the oil enhancement of vegetative biomass would have significant implication to future plant oil production (Napier et al., 2014). In addition, oil enhancement in plant vegetative biomass offers opportunity for more energy-dense forages. Both genetically modified (GM; transgenic) and non-GM methods (genetic engineering) have been used to enhance oil content in plant vegetative biomass (i.e., diversion of carbon) with a more recent focus on producing energy-dense forages for livestock (Beechey-Gradwell, 2021; Winichayakul et al., 2020). Although triglycerides are found in lesser amounts in plant leaves, functional triglyceride biosynthesis pathways provide an opportunity to increase their concentration in structural plant organs (*e.g.*, leaves) (Vanhercke et al., 2019).

GM methods have been used to develop high metabolisable-energy (HME) perennial ryegrass containing leaf lipids up to 6-7% dry weight (approximately double the natural range) (Beechey-Gradwell et al., 2018). High metabolisable energy perennial ryegrass also has a very different fatty acid profile compared to wild-type plants, with significant elevations in linoleic, oleic, and palmitic fatty acids (common fatty acids found in triglycerides), and a moderate increase in α -linolenic acid (most common in plant leaves) (Winichayakul et al., 2020). The GM method used for HME perennial ryegrass uses the co-expression of diacylglycerol O-acyltransferase enzymes (DGAT1) and Cysoleosin, an oleosin protein engineered to enhance the stability of lipid droplets, and limiting triglyceride catabolism in vegetative plant tissue (Beechey-Gradwell, 2016; Beechey-Gradwell et al., 2022). DGAT1 has been of particular interest to scientists working in plant metabolic engineering and synthetic biology since it is the predominant catalyst in the final step of the triglyceride biosynthetic pathway, and is a limiting factor in the synthesis of triglycerides (Shockey et al., 2016).

Non-GM, genetic engineering methods have also been used to enhance triglycerides in vegetative plant biomass. Although not used as a forage crop, excessive enhancement of oil content in vegetative tissues of tobacco (Nicotiana tabacum) can impact plant growth and development and has been shown to reduce biomass yields in transgenic lines with extremely high levels of oil in their leaves (>30% on a DM basis) (Vanhercke et al., 2017). However, much lower oil levels (7-8% w/w) are targeted for high energy forages to reduce the negative impacts on biomass yield. There are also no known negative implications to general plant structure. Sorghum (Sorghum bicolor L.) plants have been successfully engineered to produce low-medium level oil content enhancements in their vegetative plant biomass of 3-8.4% (on a DM basis) depending on plant stage. These plants developed normally compared with a wild-type control (Vanhercke et al., 2019). In that study, the concentration of α -linolenic acid was reduced, whilst linoleic, palmitic, and oleic acid increased. The risk of selecting a negative plant phenotype using non-GM methods, can be mitigated by more intensive screening of larger populations of first-generation plants as well as testing a much broader range of gene-knock out combinations to alter both the total oil content and fatty acid profiles. This technology in sorghum has only been tested in glasshouse experiments and is yet to be tested more broadly in field studies under diverse climatic conditions.

4.4.1.3 Effects of dietary lipids on enteric methane and livestock productivity

Dietary lipids are broken down by ruminal microbes, with unsaturated fatty acids converted into saturated fatty acid end products via bacterial lipolysis and biohydrogenation (Lourenço et al., 2010). Rumen microbes can also synthesise lipids using short chain fatty acids produced during carbohydrate and protein metabolism (Harfoot, 1981). Most of the unsaturated fatty acids consumed undergo biohydrogenation, but some escape rumen bacteria and are readily absorbed into body fat and milk fat leading to higher nutritive feed from ruminant derived products for human consumption (Buccioni et al., 2012).

While ruminants generally consume a low lipid diet (< 5% total lipids in DM) (Beechey-Gradwell, 2021), lipid supplementation may improve productivity of ruminant livestock, reduce enteric CH₄ production, (Cosgrove et al., 2004; Klieve et al., 2012) and improve the nutritional quality of ruminant derived products for human health, especially omega-3 and omega-6 fatty acid content via higher supplementation of α -linolenic and linoleic fatty acids, respectively (Cosgrove et al., 2004; Ponnampalam et al., 2021). However, high intake of lipids, especially at concentrations above 10% DM, can limit fibre digestion (McGinn et al., 2004; Ørskov et al., 1978), feed intake and production outcomes (Maia et al., 2007; McDonald et al., 2002). As an example, a meta-analysis showed milk yield in dairy cows peaked between 3.9% and 6.0% lipid supplementation (R² = 0.41), and digestibility of dry matter (DM) (R² = 0.30) and NDF (R² = 0.51) in cattle peaked at 4.2% lipid supplementation and then decreased linearly with increasing lipid supplementation up to approximately 11.8% (Patra, 2013). In contrast to this, lipid concentrations between 6.3% and 8.8% had no adverse effect on lamb production (Cosgrove et al. 2004). For this reason, oil-enhancement in vegetative plant biomass is only targeting 'medium' level enhancement (maximum 7-8% plant lipid content) to mitigate any potential animal health and production issues.

There is a large focus in the literature on the potential of lipid supplementation to reduce enteric CH₄, particularly through the use of storage lipids. Increasing lipid supplementation has been reported to linearly decrease enteric CH₄ (R² = 0.38-0.63) in cattle; though higher levels of lauric (C12:0), myristic, oleic, and α -linolenic fatty acids reduce CH₄ to a greater degree compared to other fatty acids (Patra, 2013). Often the positive effects on reduced CH₄ are offset by the negative effects on fibre fermentation and DMI, which are fundamental drivers of animal production. These fatty acids have been found to be inhibitory to methanogens and cellulolytic bacteria, especially for diets low in structural carbohydrates and calcium (Machmüller et al., 2003a). The major supplementary lipid sources known to negatively affect methanogens and/or ruminal protozoa populations and reduce enteric CH₄ production both in *in vitro* and *in vivo* include canola oil (high in lauric and oleic fatty acids) (Dohme et al., 2000), linseed oil (predominately α -linolenic) (Martin et al., 2008) and coconut oil (high in lauric and myristic fatty acids) (Dong et al., 1997; Hristov et al., 2009; Machmüller et al., 2001, 2003b). For example, in vitro, a grass hay diet supplemented with coconut oil (10% lipids w/w) produced nearly 2.5 times less CH_4 per unit of DM than the hay only diet, though DM and NDF digestibility was reduced 1.5 and 1.9 times, respectively, coinciding with a significant reduction in methanogenic microbe populations (Dong et al., 1997). Supplementing beef cattle with sunflower oil (predominately oleic and linoleic fatty acid) at 5% of the diet (on a DM basis) has been reported to reduce enteric CH₄ production by 22%; though NDF and ADF digestibility was reduced by 23% and 29%, respectively, with no impact on DMI (McGinn et al., 2004).

Other supplementary lipids known to effectively reduce CH₄, but with minimal-no effects on fibre digestion and ruminal microbes (notably methanogens) include marine algal oil (Algamac 3050; predominately palmitic and docosahexaenoic fatty acids (22:6)), and safflower oil (predominately linoleic fatty acid) (Klieve et al., 2012). Generally, these lipids work to reduce CH₄ by altering the proportion of propionic acid produced (i.e., lower acetic: propionic acid), which acts as an alternative H₂ sink to methanogenesis in the rumen (Klieve et al., 2012; McGinn et al., 2004). Brahman crossbred steers fed a basal diet of Rhodes grass (*Chloris gayana*) hay that was supplemented with marine algal meal (Algamac 3050) at 50 g lipid/kg DMI over an 11-week period increased liveweight and reduced enteric CH₄ as a function of average daily gain (ADG) by more than four-fold compared to steers fed the Rhodes grass-only basal diet; though there were palatability issues with the algal meal supplement (Klieve et al., 2012).

More recently, perennial ryegrass has been genetically modified to produce higher leaf lipid concentrations (6-7% DM; 59-66% higher total fatty acid content than wild types), particularly oleic and linoleic fatty acids (Beechey-Gradwell et al., 2018; Winichayakul et al., 2020). *In vitro* incubation with rumen fluid showed fresh genetically modified perennial ryegrass, hereafter referred to as high metabolisable energy (HME) perennial ryegrass, reduced CH₄ production, and the proportion of CH₄ in total gas by 30% and 10%, respectively (Winichayakul et al., 2020). There are no *in vivo* studies that have been carried out on HME perennial ryegrass and no other similar studies carried out for other oil-enhanced forage crops (*e.g.,* forage sorghum). However, medium (6.3% total lipids) and high (8.8% total lipids) lipid perennial ryegrass that was 'simulated' via supplementation with linseed and sunflower oils, increased feed conversion efficiency of lambs by 17% and 32%, respectively, compared to standard perennial ryegrass (4% total lipids) (Cosgrove et al., 2004).

No studies have quantified the effect of feeding HME perennial ryegrass on livestock production, enteric CH₄ formation, and changes to the fatty acid profile in animal products (i.e., meat and milk)

in vivo. However, *in vitro* studies indicate fresh HME perennial ryegrass led to a 6% increase in metabolisable energy (ME) content (equivalent to ~ 1.1 MJ ME/kg DM) and NDF content (equivalent to ~2% DM), 30% decrease in CH₄ production, and 22% decrease in total gas production compared to wild types (Winichayakul et al., 2020). In the same study, CP and ADF content remained unchanged with GM, and HME perennial ryegrass also showed potential for use also as ensilaged forage but further research in field experiments is needed (Winichayakul et al., 2020).

4.4.2 Animal modelling

4.4.2.1 Nutritive value of pastures/forages

Our modelling analysis assumed no change in plant biomass yields or general plant structure in response to increases in lipid content, as supported by previous studies (Vanhercke et al., 2017) and discussed in section 4.4.1.2 above. Increasing the lipid content of pasture/forage species to 8% DM increased the ME content of all species modelled (Table 15). The largest increase in ME was in buffel grass (14% increase, equivalent to 1.2 MJ ME/kg DM), followed by French serradella, Phalaris, forage oats and forage sorghum, with the differences due to variations in baseline nutritive value. Much smaller improvements in ME content were observed for the chicory-grass/clover mix (4%) due to the dilution effect of the non-oil enhanced grass and clover species in the mix.

	Lipid content ME (% DM) (MJ/kg DM)		Predicted DMI (kg/head.day)		
			Lambs	Ewes	Steers
Buffel grass					
Baseline	3.0	8.5	-	-	5.30
Oil-enhanced	8.0	9.7	-	-	5.60
Chicory-grass/clover mix (50:50) ¹					
Baseline	3.2	10.8	1.08	-	-
Oil-enhanced	6.0	11.2	1.11	-	-
Forage oats					
Baseline	2.5	10.2	1.06	-	5.40
Oil-enhanced	8.0	11.1	1.11	-	5.50
Forage sorghum					
Baseline	3.9	10.1	-	-	5.40
Oil-enhanced	8.0	10.9	-	-	5.50
French serradella					
Baseline	2.3	10.8	1.07	1.30	-
Oil-enhanced	8.0	11.8	1.13	1.30	-
Phalaris					
Baseline	3.4	10.8	1.09	1.30	5.50
Oil-enhanced	8.0	11.7	1.13	1.30	5.50

Table 15. Modelled metabolisable energy (ME) content and dry matter intake (DMI) of baseline and oil-enhanced pasture/forage species.

¹Nutritive value for chicory-grass/clover mix accounts for proportion of species in mix. Total fat lipid content was only adjusted for the chicory.

For prime lambs, increasing lipid levels to 8% DM always improved predicted DMI by ~4%, which was both a function of the increasing energy content of the feed, and the high energy requirements of the growing lamb. This was not the case for dry ewes as the French serradella and Phalaris pastures were already of high nutritive value and were able to satisfy maintenance level requirements. For beef steers, improvements in predicted DMI were marginal and only occurred when grazing either buffel grass, forage oats, or forage sorghum that were of generally lower nutritive value (higher NDF and lower CP content) compared to Phalaris (Table 2).

4.4.2.2 Livestock productivity and methane efficiency modelling

For all pasture/forage species and livestock classes (i.e., prime lambs, dry ewes, and beef steers), increasing lipid levels from baseline levels to a maximum 8% DM resulted in linear improvements in modelled average daily LWG and CH₄ efficiency (Figures 4-5). Average daily LWG and CH₄ efficiency of prime lambs grazing Phalaris, forage oats, or French serradella, with 8% lipid levels were improved on average by ~17% and ~12%, respectively, compared to those grazing the same pastures/forages at baseline lipid levels (Table 16). Average daily LWG and CH₄ efficiency of dry ewes grazing Phalaris or French serradella with 8% lipid levels were improved on average by ~19% for both these metrics compared to baseline lipid levels (Table 16).

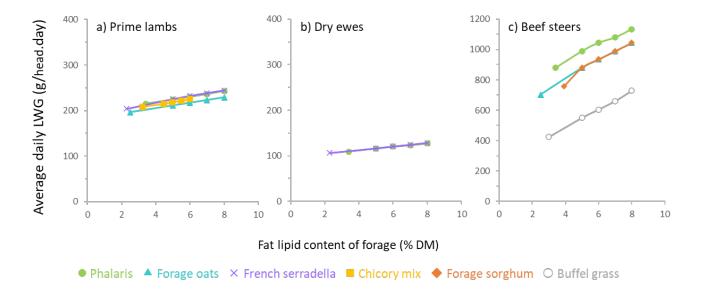


Figure 4. Average daily LWG of a) prime lambs, b) dry ewes and c) beef steers grazing different pasture/forage species with increasing lipid content from baseline to 8% DM. Note the different y-axis values for sheep and cattle.

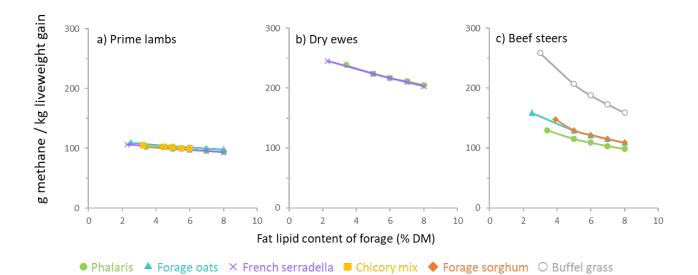


Figure 5. Methane efficiency of a) prime lambs, b) dry ewes and c) beef steers grazing different pasture/forage species with increasing lipid content from baseline to 8% DM.

In sheep grazing systems, the greatest improvement in modelled average daily LWG and CH₄ efficiency was for both prime lambs and dry ewes grazing French serradella, reflecting the lower baseline fat lipid levels for this species compared to other pasture/forage species simulated (Table 16). However, LWG and CH₄ efficiency were similar across all species at 8% lipid content.

In beef grazing systems, oil-enhancement provided the greatest benefit in buffel grass compared to other species (Table 16). However, modelled average daily LWG and CH₄ efficiency of beef steers consuming buffel grass with 8% lipid content was similar to that of the other species at baseline lipid values.

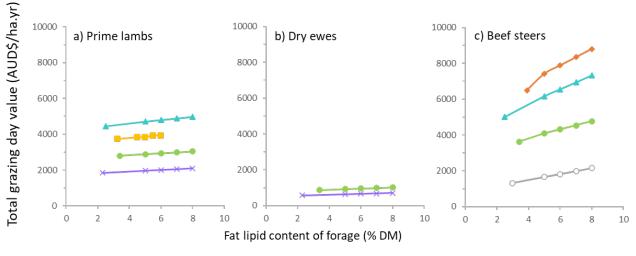
Table 16. Average daily LWG, methane (CH₄) efficiency, and total grazing day value of prime lambs, dry ewes and beef steers grazing various pasture/forage species at baseline fat lipid levels and following oil-enhancement with 8% fat lipid levels.

Desture (farage Livestock		Average daily LWG (g/head.day)		Methane efficiency (g CH₄/kg LWG)		Total grazing day value (\$/ha.year)				
Pasture/Torage	asture/forage type/class		Oil- enhanced	Change	Baseline	Oil- enhanced	Change	Baseline	Oil- enhanced	Change
Buffel grass	Beef steers	424	728	72% 个	259	159	63% 🗸	1322	2148	62% 个
Chicory-grass/clover mix (50:50)	Prime lambs	208	225	8% 个	105	100	5% 🗸	3744	3941	5% 个
Forage oats	Prime lambs	196	229	17% 个	110	98	12% 🗸	4456	4971	12% 个
	Beef steers	702	1045	49% 个	159	109	46% ↓	5012	7326	46% 个
Forage sorghum	Beef steers	756	1045	38% 个	148	109	36% 🗸	6486	8803	36% 个
French serradella	Prime lambs	204	244	20% 个	106	94	14% ↓	1853	2099	13% 个
	Dry ewes	106	128	21% 个	246	203	21% 🗸	581	702	21% 个
Phalaris	Prime lambs	215	243	13% 个	103	94	9% ↓	2796	3048	9% 个
	Dry ewes	109	127	17% 个	239	205	16% 🗸	872	1016	17% 个
	Beef steers	880	1134	29% 个	129	99	31% 🗸	3629	4763	31% 个

¹Fat lipid content of the chicory/grass-clover mix was only adjusted for the chicory.

4.4.2.3 Total grazing day value

Overall, forage crops such as forage oats, and forage sorghum achieved higher total grazing day value (Table 16) because of their much higher grazable biomass yields relative to the pasture species (Table 2), irrespective of average daily LWG. For example, both prime lambs and beef steers grazing Phalaris had marginally higher average daily LWG than those grazing forage oats, yet total grazing day value was higher for forage oats because it produced 41% more grazable biomass (Table 2; Figure 6).



● Phalaris 🔺 Forage oats 🛛 French serradella 📕 Chicory mix 🔶 Forage sorghum 🔿 Buffel grass

Figure 6. Linear change in total grazing day value for a) prime lambs, b) dry ewes and c) beef steers grazing different pasture/forage species with increasing fat lipid content from baseline to 8% DM.

Total grazing day value also varied between the livestock types/classes due to the differences in liveweight prices and daily growth rate potential, irrespective of total grazing days as calculated from grazable biomass and DMI. Hence, beef steers always achieved higher total grazing day value (and change in total grazing day value) than prime lambs and dry ewes when grazing the same pasture/forage species (*e.g.*, Phalaris and forage oats) at 8% fat lipid levels (Table 16). Although dry ewes achieved lower total grazing day value than prime lambs when grazing Phalaris or French serradella pastures, the change in total grazing day value for dry ewes was always greater (*e.g.*, $^{\sim}16\%$ vs. $^{\sim}19\%$) at 8% fat lipid levels.

The greatest improvement in total grazing day value for the different pasture/forage species was in the order of buffel grass > forage oats > forage sorghum > Phalaris > French serradella > chicory > chicory-grass/clover mix. Buffel grass, forage oats, forage sorghum and Phalaris grazed by beef steers had considerable improvement (> 30%) in total grazing day value at 8% fat lipid levels (Table 16).

Although the SRNS and LRNS can indicate relative changes to daily LW change in response to increasing fat lipid levels, the model is not able to simulate potential negative changes to NDF digestibility or DMI. Furthermore, the model is not able to simulate possible changes to rumen microbial populations or relative proportions of key volatile fatty acids that may otherwise reduce

enteric CH₄ production as described in the literature for lipid supplements. Thus, estimates for enteric CH₄ and CH₄ efficiency could only be estimated from predicted DMI based on calculations of Howden and White (1994) and Charmley et al. (2016). These are limitations with desktop analyses compared to *in vitro* or *in vivo* studies.

4.5 Cost-benefit analysis

4.5.1 Profitability

Profitability is presented as net present value per ha, which is the sum of discounted net benefits over a 10-year period (*i.e.,* not per annum). Of the six pasture/forage species evaluated, forage sorghum and forage oats had the highest overall profitability with 8% oil-enhancement (Table 17). This was directly related to their higher total grazing day value (Table 16). Relative to baseline lipid levels, buffel grass had the highest gain in net present value from 8% oil-enhancement, followed by forage oats, forage sorghum, and Phalaris, all of them in a beef steer grazing system (Table 17 and Figure 6). This aligned with the results from the animal modelling.

Pasture/forage	Livestock type/class	NPV with baseline lipid (\$/ha)	NPV with 8% oil-enhanced (\$/ha)	Change
Buffel grass	Beef steers	8,333	13,585	63% 个
Chicory-grass/clover mix (50:50)	Prime lambs	30,046	33,190	10% 个
Forage oats	Prime lambs Beef steers	34,829 39,342	38,935 58,030	12% 个 47% 个
Forage sorghum	Beef steers	51,369	70,090	36% 个
French serradella	Prime lambs Dry ewes	14,686 4,368	16,664 5,332	13% 个 22% 个
Phalaris	Prime lambs Dry ewes Beef steers	22,601 6,992 29,356	24,642 8,154 37,846	9% 个 17% 个 29% 个

Table 17. Net present value (NPV) over 10 years for the 10 scenarios based on total grazing day
value for standard year biomass

For prime lambs and dry ewes, Phalaris had higher net present value from 8% oil enhancement compared to French serradella, but French serradella had the highest gain in net present value relative to baseline fat lipid levels (Table 17).

This result trades off improvements in average daily LWG and CH₄ efficiency of sheep grazing on oilenhanced French serradella, forage oats, chicory mixture and/or Phalaris with total biomass production and the cost of seeding, *e.g.*, \$79/ha every 10 years for Phalaris, \$139/ha every 4 years for serradella versus \$165/ha every year for forage oats (Table 4).

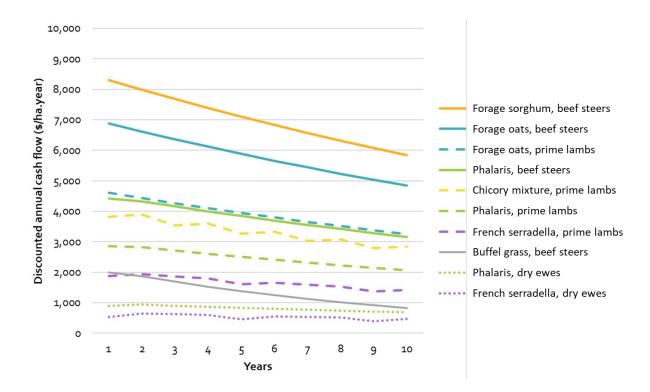


Figure 6. Discounted annual cash flow (\$/ha.year) of 10 scenarios for different pastures/forages with 8% oil-enhancement over 10 years based on total grazing day value for standard year biomass.

At the industry level, profitability of oil-enhanced forages will be influenced by the total area sown, whether they are grazed by sheep or cattle, and the scale and rate of adoption. Data on area sown was only available for two of our six shortlisted species (Table 7) and not disaggregated by livestock system or region. We were also unable to estimate adoption rates using ADOPT, but some generalisations can be made. French serradella, chicory, and forage sorghum are not currently widely sown in Australia, and industry-wide value of an oil-enhanced cultivar would be small at this time. However, we would expect the area sown to these species to increase in the future due to their suitability for future climates, and investment could be justified on the basis of local importance (*e.g.*, while French serradella is not widely sown outside of WA, it is a valuable alternative to subterranean clover on sandy soils and in low rainfall years). In comparison, Phalaris and forage oats are both already widely used, and could be expected to provide substantial benefit to industry based on potential adoption rates and estimated profit. Finally, buffel grass occurs in large areas across Australia, with estimates ranging from 5 to >50 M ha (Friedal et al. 2006). However, the majority of this area is naturalised pastures, with low likelihood of land managers investing in an improved cultivar.

4.5.2 Profit-risk trade-offs

The potential economic implications of variability in grazable biomass; thus, total grazing day value was explored. An annual profit benchmark of \$2000/ha.year was assumed for all scenarios and was dependent on the grazable biomass achieved and livestock system in place.

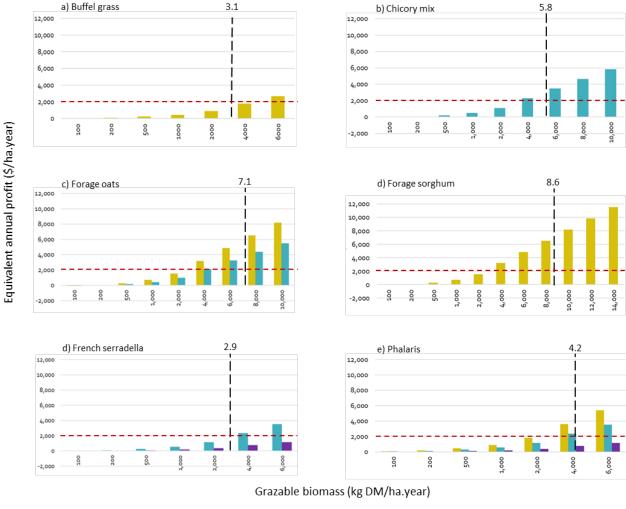
For beef steers, grazable biomass required to reach the annual profit benchmark was 3.0 t DM/ha.year for Phalaris, forage oats, and forage sorghum. This target would be considerably more achievable in more years (including more lower production years) for forage oats and forage

sorghum, with grazable biomass in a standard year on average ~60% higher than the 3.0 t/ha.year benchmark target, compared to only ~30% higher for Phalaris. Over 4.0 t DM/ha.year was needed for buffel grass to meet the benchmark target, which was 24% higher than the grazable biomass achievable under standard year conditions (Figure 7).

For prime lambs, grazable biomass required to reach the benchmark target was ~4.0 t DM/ha.year for Phalaris, forage oats, French serradella, and the chicory-grass/clover mix. This benchmark target would again be more achievable for forage oats in more years, and the chicory-grass/clover mix. However, this would be harder to achieve for French serradella (standard year grazable biomass ~ 30% less than benchmark target), unless under good year conditions (Figure 7).

Significantly more grazable biomass is required for dry ewes to reach this profit benchmark, though this does not take into consideration the value of lambs born from ewes.

This profit-risk analysis suggests the investment in oil-enhanced pastures/forages may be riskier for pastures, compared to forage crops, especially in drier/lower production environments.



Prime lambs Dry ewes Beef steers

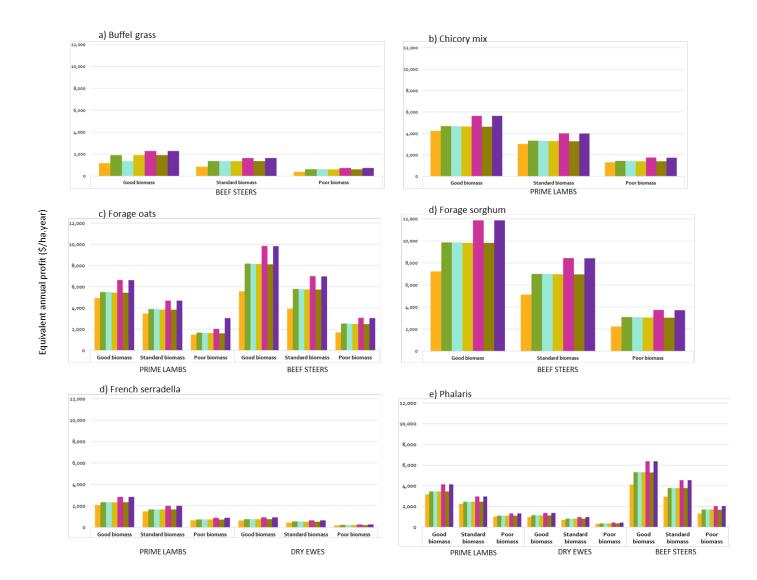
Figure 7. Profit-risk return as equivalent annual profit across a range of grazable biomass yields for the 10 scenarios with 8% lipid content. Benchmark target of \$2000/ha.year is indicated by the red dashed horizontal line, and grazable biomass in a standard year is indicated by the black dashed vertical line.

4.5.3 Sensitivity to seed cost, market changes and grazable biomass

In all scenarios, improving pasture/forage species with 8% lipid levels led to higher annual profits compared to baseline levels, even with the additional costs of seed technology premium and general seed cost (Figure 8). Buffel grass grazed by beef steers was most sensitive to the additional costs of seed technology premiums and general seed cost, with little change in profit gains compared to baseline fat lipid levels, even under good year conditions (Figure 8). Annual profits for buffel grass and French serradella were also relatively low (< \$2000/ha.year) under standard and good year conditions compared to the other pasture/forage species, especially forage oats and forage sorghum that made up to \$8000/ha.year under standard year conditions (Figure 8).

In all scenarios, poor year conditions significantly reduced annual profits. This resulted in very low annual profits (~500-\$2000/ha.year) for the pasture species (*e.g.,* Phalaris, buffel grass and French serradella), whilst the forage crops (*e.g.,* forage oats and forage sorghum) remained quite profitable under these poor conditions (~2000-4000/ha.year).

In all scenarios, the 20% increase in meat price was an important driver of higher annual profits and was not impacted by the additional cost of seed technology premiums (Figure 8).



Baseline lipid content

8% lipid content

- 8% lipid content, +25% tech premium
- 8% lipid content, +50% seed cost
- 8% lipid content, +20% meat price
- 8% lipid content, +25% tech premium, +50% seed cost
- 8% lipid content, +25% tech premium, +20% meat price

Figure 8. Sensitivity of equivalent annual profit of the 10 scenarios to changes in technology cost, seed cost and meat price for grazable biomass under good, standard, and poor year conditions.

5 Discussion/Conclusion

For all six pasture/forage species evaluated, oil-enhancement from baseline to 8% fat lipid levels led to higher density feed (increased ME content) for livestock, resulting in greater livestock productivity and CH₄ efficiency outcomes.

While the sectoral analysis indicated that improved forages were most likely to be adopted by the sheep and dairy industries, our modelling showed that the greatest productivity and economic gains for the red meat industry would come from feeding oil-enhanced forages to growing beef cattle. Feeding oil-enhanced forages to growing sheep (prime lambs) was also profitable in most scenarios, with profits likely to increase if improvements in wool yields were considered. The use of high-value forages was not considered economic for dry ewes, whose maintenance energy requirements are already satisfied by currently available forages.

At the paddock level, oil-enhanced forage oats and forage sorghum returned the highest net present value and were also more likely to remain highly profitable under poor seasonal conditions (i.e., drier years) reducing investment risk, compared to the other pasture/forage species evaluated. Both these species are also likely to attract interest from seed companies as they are very commonly used annual crops that can be grown across a wide range of environments and used for both sheep and cattle systems. They are also early-horizon options for oil-enhancement.

Oil-enhanced Phalaris and chicory (sown in a mixture) were also both profitable at the paddock level under standard year conditions, but more sensitive to poor years than the higher-yielding forage crops. However, Phalaris is widely sown across temperate Australia, so at industry level, there is potential for oil-enhancement to have large impacts. Phalaris was also identified as a species of interest by several seed companies. In comparison, chicory is still relatively new in Australia, and the potential for widespread adoption of an oil-enhanced cultivar is considered low at this time.

Oil-enhancement of buffel grass and French serradella were the least profitable options evaluated at the paddock level, primarily due to low pasture yields, though this does not account for differences in area sown. While increasing the oil content of buffel grass or other tropical species could have significant impact for the northern Australian cattle industry if widely adopted, modelled annual profits were still very low, especially under standard and poor seasonal conditions. Previous reviews (*e.g.*, Bell et al. 2016) highlight difficulties in adoption of new pastures in extensive beef systems of northern Australia, including high costs, poor establishment, and variable persistence under challenging climatic conditions. There are also reservations on how this technology may persist in long-term buffel pastures (20-30 years), as buffel grass is not commonly re-sown and often exists in a naturalised state.

In summary, the results of this desktop analysis highlight the potential opportunities for oilenhanced forages in the Australian red meat industry. We find that feeding oil-enhanced forages could increase liveweight gain and decrease methane emissions intensity in growing animals, with economic benefits in beef cattle and prime lambs. At the industry scale, the greatest benefits are likely to come from the application of this technology to forages which are already widely adopted and adapted to future climate challenges.

5.1 Key findings

• Use of improved pastures is highest in southern Australia, and improved forages are most likely to be adopted by the sheep and dairy industries.

- Oil-enhanced forages are most likely to provide productivity and economic benefits to growing beef cattle and sheep.
- Adoption of oil-enhanced high-yielding forage crops such as forage oats and forage sorghum are most likely to provide economic benefits for individual farmers.
- At the industry scale, oil-enhancement of species such as Phalaris, which are already widely adopted, would provide the greatest benefit.

5.2 Benefits to industry

This initial desktop analysis provides an indication of how oil-enhanced forages could benefit Australian livestock industries, and acts as a guide to further investment. While the productivity and economic analysis focused on just six species, they represent a range of pasture types, and the results may be extended to other similar species.

A lack of publicly available data on area sown to specific species limits our ability to quantify potential economic benefits at the industry scale.

6 Future research and recommendations

Since the inception of this project, CSIRO has divested its interests in this technology, and we are unable to provide a business case for future investment. Future research or commercialisation will be subject to a licence. However, our results are relevant to the application of other technologies that may increase the ME content or other aspects of feeding value. Generally, we recommend that initial investments into improved forages should target species that are climate resilient and already widely adopted by industry. This investment could come from seed companies, government agencies, or industry-owned research and development corporations.

Although our modelling showed productivity benefits from feeding oil-enhanced forages, these results need to be confirmed in pen-feeding or grazing trials. Similarly, although the modelling showed improvements in CH₄ efficiency, we were unable to quantify the potential benefits of oil-enhancement on enteric CH₄ production, as observed in *in vitro* and *in vivo* studies for GM high metabolisable energy perennial ryegrass (Cosgrove et al. 2004; Winichayakul et al. 2020).

To mitigate risk to ruminant production and the environment, appropriate duty of care needs to be considered during the creation and selection process of first-generation plants before they can be tested *in vivo* and commercially released. The ability to tailor the fatty acid profile of oil-enhanced forages will also become important in mitigating these risks to ruminant livestock and enhancing livestock production and environmental benefits.

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8 Appendix

Industry (seed companie	es)
Question 1.	What do you consider the most important forage/pasture species for
	your business? What makes these forage/pasture species important
	to your business? (<i>e.g.,</i> profits from annual seed sales, profits from
	royalties, area sown/industry adoption)
Question 2.	Can you provide any details on the volume of seed sold for these
	species on a state-by-state basis? (<i>e.g.,</i> tonnes seed sold/year or
	hectares sown)
	 Sub-question: Where hectares sown is not given - Can you
	provide details on the sowing rate/ha for those species?
Question 3.	What animal/agricultural sector makes up the majority of these seed
	sales, and does it vary on a state-by-state basis? (<i>e.g.</i> , sheep-beef
	grazing high/medium rainfall, dairy, fodder production – export vs
	domestic use)
Question 4.	Do you have any seed sale/industry-based statistics you would be
	willing to share with us? (<i>e.g.,</i> National or state-by-state distribution
	summary of seed sales for major products). These will be kept
	confidential.
Question 5.	Are these forage/pasture species you mentioned suitable for future
	climates? (e.g., suitability by 2030) If so, what makes them suitable?
	(<i>e.g.,</i> deep root system, high WUE).
	• See General question 5 for further details of climate change
	predictions on a state-by-state basis.
Question 6.	Is your business targeting future R&D at any specific forage/pasture
	species? If so, why? (<i>e.g.,</i> traits like drought tolerance (suitability to a
	future climate), environmental benefits, regrowth potential, nutritive
	value, pest and disease resistance/tolerance, animal health, or
	targeting other marketing opportunities)
General questions (state	e departments/universities and practitioners)
General question 1.	What agroecological zone do you work in? What is the main
	forage/pasture species sown in that zone?
General question 2.	Can you estimate the area sown to these key forage/pasture species?
General question 3.	What animal/agricultural sector utilises these forage/pasture species
	the most? (<i>e.g.</i> , sheep/beef grazing, dairy, fodder production – export
	vs. domestic use).
General question 4.	What are the key traits that makes these forage/pasture species
	valuable to the industry? (<i>e.g.</i> , drought tolerance, high nutritive value
	and/or biomass production, public variety vs proprietary, or other)
General question 5.	What is the suitability of these forage/pasture species for future
	climates (<i>e.g.</i> , suitability by 2030)?

8.1 Appendix 1: Sectoral analysis – participant questions

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General question 6.	What do you consider key traits for future forage/pasture species?		
	(<i>e.g.</i> , drought tolerance (suitability to future climates), environmental		
	benefits, regrowth potential, high nutritive value, pest and disease		
	resistance/tolerance, animal health, other).		
State Departments/Un	iversities		
Question 7.	Has your organisation invested in the research of any particular		
	forage/pasture species in the last 5-10 years? Why?		
Question 8.	Have these forage/pasture species been readily adopted by industry?		
	Do you have a measure for this?		
Practitioner (research	+ systems fit)		
Question 7.	What are the key traits that growers are looking for in forage/pasture		
	species?		
Question 8.	Are there any particular forage/pasture species you consider being		
	important in the future when considering a changing climate?		
Question 9.	How do these forage/pasture species fit in a 'systems-context'? (e.g.,		
	fill feed gaps, provide additional income from grazing and fodder		
	production, complement existing feed base, crop rotation benefits,		
	other)		

8.2 Appendix 2: ADOPT tool questions (Source: Kuehne et al. 2017)

Quadrant	ADOPT variable	Question asked in ADOPT		
	1. Profit orientation	What proportion of the target population has maximising profit as a strong motivation?		
	2. Environmental orientation	What proportion of the target population has protecting the natural environment as a strong motivation?		
	3. Risk orientation	What proportion of the target population has risk minimization as a strong motivation?		
Relative advantage for the population	4. Enterprise scale	On what proportion of the target farms is there a major enterprise that could benefit from the practice?		
	5. Management horizon	What proportion of the target population has a long-term (greater than 10 years) management horizon for their farm?		
	6. Short-term constraints	What proportion of the target population is under conditions of severe short-term financial constraints?		
Learnability characteristics of the practice	7. Trialling ease	How easily can the practice (or significant components of it) be trialled on a limited basis before a decision is made to adopt it on a larger scale?		
	8. Practice complexity	Does the complexity of the practice allow the effects of its use to be easily evaluated when it is used?		
	9. Observability	To what extent would the practice be observable to farmers who are yet to adopt it when it is used in their district?		
Population-specific influences on the ability to learn about the practice	10. Advisory support	What proportion of the target population uses paid advisors capable of providing advice relevant to the practice?		
	11. Group involvement	What proportion of the target population participates in farmer-based groups that discuss farming?		

Quadrant	ADOPT variable	Question asked in ADOPT		
	12. Relevant existing skills & knowledge	What proportion of the target population will need to develop substantial new skills and knowledge to use the practice?		
	13. Practice awareness	What proportion of the target population would be aware of the use or trialling of the practice in their region?		
	14. Relative upfront cost of the practice	What is the size of the up-front cost of the investment relative to the potential annual benefit from using the practice?		
	15. Reversibility of the practice	To what extent is the adoption of the practice able to be reversed?		
	16. Profit benefit in years that it is used	To what extent is the use of the practice likely to affect the profitability of the farm business in the years that it is used?		
Relative advantage of the practice	17. Profit benefit in future	To what extent is the use of the practice likely to have additional effects on the future profitability of the farm business?		
	18. Time for profit benefit to be realised	How long after the practice is first adopted would it take for effects on future profitability to be realized?		
	19. Environmental impact	To what extent would the use of the practice have net environmental benefits or costs?		
	20. Time for environmental impacts to be realised	How long after the practice is first adopted would it take for the expected environmental benefits or costs to be realized?		
	21. Risk	To what extent would the use of the practice affect the net exposure of the farm business to risk?		
	22. Ease and convenience	To what extent would the use of the practice affect the ease and convenience of the management of the farm in the years that it is used?		