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# Best choice shrub and inter-row species for reducing methane emissions intensity

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# Executive summary

Grazing systems based on shrubs and pasture species that are selected for their nutritive value and anti-methanogenic bioactivity offer a practical means to reduce methane emissions and emissions intensity from grazing livestock. Our hypothesis was that the shrub-based system with the most potential to reduce methane emissions and emissions intensity will be one that takes into account the bioactivity and productivity of both the shrub and inter-row components of the plants being grazed. We showed that sheep grazing shrub-based systems without supplementary feeding during autumn gained at least twice the weight of sheep grazing pasture with conventional amounts of supplementary feeding with grain. The sheep grazing shrubs also had a lower methane emissions intensity and produced less methane per unit of energy intake. This result was replicated in a modelling analysis, which also demonstrated that over a 12-month period, sheep that graze shrubs in autumn, have a better body condition which enables them to require less supplementary feeding over the year. The results from our study support the concept of using perennial shrubs in a whole-farm system as a means of improving productivity whilst also improving methane emissions intensity, and also reducing the risk of inadequate feed during autumn.

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# 1. Background

A current challenge in extensive grazing systems is to reduce greenhouse gas (GHG) emissions without adversely affecting livestock productivity and profitability. A major limitation to improving animal productivity and methane emissions intensity in southern Australia is an annual ‘feed gap’ in autumn, when conventional annual-based pastures are typically in low supply and of poor nutritive value. There is therefore a reliance on expensive and labour-intensive supplementary feeding and/or acceptance of reduced and variable performance during a period of at least 2-3 months and, in some seasons, considerable longer. One strategy to address these constraints is to develop grazing systems that provide a mixture of plants in autumn that offer both good nutritive value and anti-methanogenic properties. The role of Australian perennial shrub species offers a feasible approach given their capacity to tolerate difficult conditions and provide green feed in autumn (Revell *et al.* 2013). Furthermore, if we select shrubs and pasture species based on biomass production and nutritive value in combination with those with anti-methanogenic bioactivity there is potential to reduce methane emissions and emissions intensity from grazing livestock. It may be possible for ‘anti-methanogenic’ plant-based grazing systems to be used in an Emissions Reduction Fund (ERF) - Methodology but, for this to occur, the impact of modified systems on livestock productivity and GHG emissions need to be quantified and long-term sustainability of the systems demonstrated.

Grazing systems in the low-medium rainfall zones of southern Australia face the challenge of regular feed shortages, particularly, but not limited to summer and autumn. This creates enormous challenges for livestock producers to deliver consistent and high quality products. Recent bio-economic modelling has indicated shrub-based systems over a modest 10-20% of a mixed farm could, depending on particular scenarios, increase whole-farm profit by up to 20% (Monjardino *et al.* 2010)or could maintain whole-farm profits with a reduced area allocated to cropping (thereby reducing risk;Monjardino *et al.* (2014)). The increase in whole-farm profit is achieved by reducing supplementary feeding during autumn, deferring the grazing of regenerating annual pastures on other parts of the farm leading to increased feed availability, and by increasing production on land classed as unsuitable and uneconomical for cropping. The modelling was based on grazing systems where perennial forage shrubs were incorporated with existing pasture, but the positive benefits could be enhanced if the choice of both shrub and inter-row pasture species were made with the dual purpose of reducing emissions directly (i.e. anti-methanogenic plants) and improving emissions intensity (i.e improving animal nutrition and growth). We have evidence from the Future Farm Industries (FFI) CRC-Meat & Livestock Australia (MLA) co-funded Enrich project (Eureka prize, 2013), projects within Reducing Emissions from Livestock Research Program (RELRP), and the Commonwealth Department of Agriculture ‘Drought hardy, carbon conscious shrub-based grazing systems’ project, that there are shrub and pasture species that could meet this goal. Importantly, in order for practice change to occur, there is a need to confirm modelling predictions and build the body of evidence that plants identified as anti-methanogenic from *in vitro* screening, actually provide the expected benefits under paddock conditions. In autumn 2012, we provided the first dataset in Australia on methane emissions intensity from sheep grazing a shrub-based forage system (in the Department of Agriculture project ‘Drought-hardy and carbon-conscious grazing systems’). We showed a 3- to 5-fold improvement in emissions intensity compared to sheep grazing a senesced autumn pasture with supplementary grain (Revell *et al*., manuscript in preparation). This result was achieved with just four species of shrubs, and did not include the most anti-methanogenic species that were included in the current study at the UWA Ridgefield Farm.

The research question addressed in this project was whether the provision of native shrubs with or without bioactive properties and inter-row pasture species with anti-methanogenic properties increase animal performance and reduce methane production compared to a ‘business as usual’ (BAU) management system during the ‘feed gap’ period in autumn. Our hypothesis was that the shrub-based system with the most potential to reduce methane emissions and/or emissions intensity will be one that takes into account the bioactivity and productivity of both the shrub and inter-row components of the plants being grazed. This hypothesis was tested by grazing commercially-available pasture species that we identified, using *in vitro* data, as having lower methanogenic potential in the inter-row of the shrub mixture, and comparing the use of these annual pastures on their own or in combination with perennial forage shrubs. We measured livestock productivity and methane emissions from animals over two years, during the autumn ‘feed gap’ period. All treatments were compared to a BAU management system to quantify the benefits of different combinations of pasture and/or shrubs on livestock productivity and methane emissions intensity. Scenarios investigated included the effect of adding high productivity shrubs or shrubs with high concentrations of bioactive compounds to either existing pasture or pasture containing bioactive compounds. The impact of the time at which the pasture system was grazed in relation to the autumn feed gap was also investigated. The data generated in this project is valuable for modelling the profitability of this type of system at a whole farm level.

Structure of the report

The KPIs and project outputs for the entire 3 year project have been addressed under 3 parts in this final report. In Part A we address the 2 main grazing experiments that were undertaken to examine whether sheep grazing shrubs and inter-row had lower emissions and emissions intensity than sheep fed under a business as usual scenario. Part B addresses the OPFTIR measurement of sheep grazing either shrubs or dry pasture with grain supplementation. It has been kept separate because the measurements were taken at the end of the 2$^{nd}$ grazing experiment and the treatments were simplified to be 'with or without shrubs'. Part C addresses a modelling component of the project, which was included towards the end of the program. It was a preliminary investigation of how the information generated in the grazing experiments could be used to look more generally at what impact shrubs may have on productivity and emissions at a farm scale over a longer time period. There is a general discussion section at the end to draw the outputs from the project together.

# 2. Part A. Field-based experimentation of grazing systems

## 2.1 Methodology

*Experimental design*

Shrubs and inter-row pasture species were chosen based on their potential to produce biomass (Enrich project database, FFI CRC) and or reduce methane production from grazing sheep. The shrubs and inter-row combinations were planted in a manner that would help address key questions of landholders who are interested in incorporating new species to the feedbase; for example, what combination of plants are required and in what spatial layout,and what are the relative contributions of shrubs and inter-row pasture to the overall goal of reducing emissions intensity. Sheep grazed the different forage systems in autumn over two consecutive years and methane emissions and animal productivity were measured. In year one, there were six treatments:

1. Conventional pasture (without shrubs) managed according to 'business as usual' with grazing in spring and again in autumn, with autumn grazing relying on supplementary feed to meet animal requirements (denoted as UG - unimproved pasture, grazed);

2. A conventional pasture (as for 1, also without shrubs) defer-grazed in autumn to maximise autumn biomass from the pasture, that is, spring grown pasture was not grazed until autumn (denoted as UD – unimproved pasture, deferred grazing);

3. Anti-methanogenic pasture (without shrubs) grazed in spring (denoted as IG – improved pasture, grazed);

4. Anti-methanogenic pasture (without shrubs) defer-grazed in autumn to maximise autumn biomass from the pasture (denoted as ID – improved pasture, deferred grazing);

5. Anti-methanogenic pasture with productive shrubs grazed in autumn and (denoted as 'Productive shrubs');

6. Anti-methanogenic pasture with anti-methanogenic shrubs grazed in autumn (denoted as 'Bioactive shrubs').

Treatments 1-4 were supplemented with 300 g/head/day of oat grain.

Modified based on the results from year one, there were five treatments in year two:

1. Oat grain fed as a supplement to achieve liveweight maintenance on a conventional anti-methanogenic pasture (without shrubs) grazed in spring (denoted as IG – improved pasture, grazed);
2. Oat grain fed at a level to achieve approximately 140 g/day of liveweight gain (the average liveweight gain of sheep grazing shrubs in the autumn 2014 grazing experiment);
3. Oat grain fed at a level to achieve up to 180 g/day (the highest rate of liveweight gain attained during the autumn 2014 experiment);
4. Anti-methanogenic pasture with productive shrubs grazed in autumn and;
5. Anti-methanogenic pasture with anti-methanogenic shrubs grazed in autumn.

Shrub survival was measured as the number of plants surviving at different times after establishment expressed as a percentage of the total number of seedlings of each species planted. In both the 2014 and 2015 grazing experiments, shrub biomass was measured based on the Adelaide technique (Andrew *et al.* 1979) and shrub samples were oven dried at 60°C to determine the weight of dry matter. The intake of shrub material by the sheep was estimated using a visual observation of leaf removal. A scale of 1 = 0% leaf material removed to 5 = 100% leaf material removed was used to score each species of shrub. Pasture biomass was estimated using the method based on Haydock and Shaw (1975), which involved the use of reference quadrats that covered the range of herbage mass in the paddocks. Pasture intake was estimated based on the difference in biomass before and after grazing. Samples of the shrub and pasture species on offer to grazing animals were collected, freeze dried, and analysed using wet chemistry for their nutritive value and an *in vitro* fermentation system to assess fermentation and anti-methanogenic characteristics. In-field methane production was determined using the portable polytunnel in year one and portable accumulation chambers (PAC) in year two. In year two, emissions from a flock of sheep over several days at the paddock scale were measured using the Open-Path Fourier Transformed Infra-red (OP-FTIR) method.

*Shrub and pasture site establishment*

We selected 10 shrub species based on their potential to provide biomass and/or reduce methane emissions and planted them at UWA Ridgefield Farm, Pingelly, WA (Table 1). Preliminary evidence of the *in vitro* methanogenic potential of 40 commercially available pasture species was used as part of the selection process (ELLE project 01200.042; B.CCH.6540). The most appropriate pasture species were selected based on the data generated from the ELLE project (and some earlier preliminary work) and their suitability for the region (Table 2). The selected species (*Biserrula pelecinus* Casbah and *Trifolium subterraneum ssp. Subterraneum* Dalkeith) were established separately and in the inter-row between shrubs to provide a forage system designed to reduce methane production. Biserrula was chosen because of the strength of its anti-methanogenic bioactivity from *in vitro* analyses in our laboratory in a previous project (B.CCH.1024) and confirmed in the first screening of plant material sampled from the ELLE nursery. Green (i.e. not senesced) biserrula can cause photosensitivity in animals when used as a monoculture and it is most likely to be used as part of a mixed pasture. We therefore also chose subterranean clover (sub-clover) as the second pasture species because of its established value in the grazing systems in the southwest WA. Although the autumn grazing period of our experimental work in this project was unlikely to be associated with photosensitivity issues with biserrula, we deliberately aimed for a mixed pasture to be relevant to how biserrula will most often be used in commercial farming situations.

Table 1. Species of shrubs planted at the experimental site on UWA Ridgefield Farm, Pingelly, WA.

| Species | Likely effects on methane based on current data | Other comments |
|---|---|---|
| **Anti-methanogenic shrubs** | | |
| **Small shrubs**<br>*Eremophil aglabra*<br>*Maireana brevifolia*<br>*Maireana georgei*<br>*Kennedia prostrata* | Marked reductions in methane although can reduce total rumen fermentation, so likely to be most useful as a 'medicinal' plant consumed as a modest proportion of the diet | *In vivo* data available on effects on rumen fermentation for *E. glabra*.<br>Nitrate in edible biomass of *M. brevifolia* may provide an additional mechanism to reduce methane production |
| **Moderate sized shrubs**<br>*Rhagodia candolleana &*<br>*Rhagodia preissii* | Moderate to marked reduction in methane with variable effects on total fermentation | *R. candolleana* has moderate preference values by sheep (better than most other Rhagodia species), whilst *R. preissii* has good biomass production and is adapted to a wide range of environments. |
| **Larger shrubs**<br>*Acacia ligulata* | Marked to moderate reduction in methane with variable reductions in total rumen fermentation | *Acacia ligulata*'s mode of action is likely to be via condensed tannins, unlike many of the other species in this list. It is a smaller acacia than many other species of this genus, making it an attractive proposition for grazing systems. |
| **Production shrub species (high biomass, not anti-methanogenic)** | | |
| *Atriplex nummularia* | Often associated with high methane production, but not across all provenances | This species is currently the main shrub of choice across southern Australia, primarily for its adaptation qualities and biomass production. A FFI CRC project is currently investigating the potential to select improved lines of this species. Good potential to improve productivity and reduce emissions intensity. |
| *Atriplex amnicola* | Associated with high methane production | One of the most preferred plants of the Atriplex genus, good adaptation traits, widely used. |
| *Enchylaena tomentosa* | Nil to moderate reduction | Excellent adaptation, easy to establish, reasonable biomass production and often shows bioactivity. |

Table 2. Shortlist of pasture species that were considered for planting as inter-row species at UWA Ridgefield Farm site. The species that were established for grazing in this project are delineated by an asterisk. The common and scientific names, cultivar/ accession and plant life cyclein Australia are also provided.

| Common name | Scientific name | Cultivar / accession | Plant life-cycle |
|---|---|---|---|
| *Biserrula | *Biserrula pelecinus* L. | Casbah | Annual |
| Tedera | *Bituminaria bituminosa* (L.) C. H. Stirt. | Accession | Perennial |
| Perennial ryegrass | *Lolium perenne* L. | Avalon | Perennial |
| Annual ryegrass | *Lolium rigidum* Guadin | Wimmera | Annual |
| Birdsfoot trefoil | *Lotus corniculatus* L. | San Gabriel | Perennial |
| Burr medic | *Medicago polymorpha* L. | Serena | Annual |
| Lucerne/Alfalfa | *Medicago sativa* L. | SARDI 10 | Perennial |
| Yellow serradella | *Ornithopus compressu s*L. | Santorini | Annual |
| French serradella | *Ornithopus sativus* Brot. | Cadiz | Annual |
| Red clover | *Trifolium pratense* L. | Redquin | Perennial |
| White clover | *Trifolium repens* L. | Haifa | Perennial |
| Bladder clover | *Trifolium spumosum* L. | Bartolo | Annual |
| Subterranean clover | *Trifolium subterraneum ssp. subterraneum* L. | Woogenellup | Annual |
| *Subterranean clover | *Trifolium subterraneum ssp. subterraneum L.* | Dalkeith | Annual |

The sites for the pasture treatments were planted in May 2013 (in accordance to the ‘break of season’ in the Pingelly area). Initial weed control occurred prior to summer but the plots were sprayed with glyphosate (540 g a.i./ha) two weeks after germinating rains and cultivated to produce a fine seed bed one week later. Based on soil tests, lime and basal levels of K and P were applied in April prior to sowing. Each plot was top-dressed with Superphosphate (100 kg/ha (9 % P)) prior to cultivation. The region experienced the driest June for 100 years and the establishment of some of the pasture plots was not optimal, however the growing season persisted for longer than normal so pasture growth reached acceptable levels in most areas within the plots.

A mixed plot of biserrula and sub-clover was sown at 6 kg/ha (biserrula) and 12 kg/ha (sub-clover). Biserrula Special was purchased to inoculate biserrula and a Group C inoculant was used for sub-clover. A ‘business as usual’ (BAU) pasture plot was grazed in spring 2013, ahead of the autumn grazing experiment in 2014.

*Grazing experiment autumn 2014*

In the autumn of 2014, Merino sheep (n = 180) were assigned to one of six treatments, as outlined above. The two shrub-based treatments had four replicates containing 10 sheep. The unimproved, defer-grazed treatment and the improved, grazed treatment had two replicate groups containing 10 sheep each. The unimproved, grazed and the improved, defer-grazed treatments had three replicate groups containing 10 sheep each.

The shrub plots were strip-grazed, that is each plot was divided into five sections using temporary electric fencing and the sheep were moved from one section to the next every week. This method, based on previous work by our group (e.g. Revell *et al*. 2013) ensured that the sheep selected a diet that contains the novel shrubs rather than only the familiar inter-row pasture.

The pre-treatment methane emissions from each group of 10 sheep were measured using the polytunnel technique (based on Murray *et al.* (2007)). The sheep grazed their assigned treatment plots for four weeks and methane emissions were measured again at the conclusion of the grazing period. Animal productivity was determined by measuring liveweight on a weekly basis and calculating liveweight gain over the treatment period.

Biomass measurements were completed prior to the commencement of the grazing experiment and at the end of the experiment. In light of the biomass measurements, we calculated that the animals grazing the pasture plots would require 300 g/head/day of oats as a supplement to meet their energy requirements and to ensure that the pasture was available until the end of the grazing period.

*Grazing experiment autumn 2015*

Based on some of our findings from the 2014 grazing experiment, we altered our experimental design for the 2015 autumn grazing experiment. In 2014, we measured animal productivity (i.e. liveweight and body condition score) as well as methane emissions using the in-field ‘polytunnel’ method. The polytunnel was used to measure the mean methane production from 10 sheep in a group and because there were only 2-4 replicates for each treatment, the statistical power of analyses was low. Therefore, in 2015, we used Portable Accumulation Chambers (PAC) where every animal in the group was a replicate, substantially increasing the statistical power of data analyses.

Merino sheep (n = 240) were stratified for liveweight and randomly assigned to one of five treatments. Each treatment group had three replicates of 16 sheep.

During this trial, we were not able to replicate the strip-grazing method that we used in 2014. The sheep, being weaners, were not experienced with electric fences and it was difficult to contain them in the strips. Therefore, the sheep grazed the whole plot and although the stocking rate over the entire grazing period was as intended, the stocking density during this trial was lower than we planned.

The pre-treatment methane emissions from each group of sheep were measured using the PAC technique (based onGoopy *et al.* (2011)). The sheep then grazed their assigned treatment plots for four weeks and methane emissions were measured again at the conclusion of the grazing period. Animal productivity was determined by measuring liveweight on a weekly basis and calculating liveweight gain over the treatment period.

After the final PAC measurement, 40 sheep that had grazed shrubs and 40 sheep that had grazed pastures were selected randomly and used to measure methane over seven days using the OPFTIR technique. The details pertaining to the OPFTIR methodology and the results from this part of the experiment have been presented in part B.

In vitro *fermentation, nutritive value and biomass*

It was important to estimate the productivity, nutritive value and anti-methanogenic bioactivity of the diet selected by grazing sheep to compare with expectations derived from screening plants sampled from a nursery (Project 01200-042; B.CCH.6540). We collected samples of the shrub and pasture species planted at UWA Ridgefield Farm, and used in-field techniques to assess biomass production of the pasture and shrub mix and we estimated diet selection using established methods (described above). We also used *in vitro* techniques to estimate bioactivity by measuring methane emissions (Durmic *et al*. 2010), nutritive value and dry matter digestibility of the selected components of the forages on offer.

*Statistical analysis*

A linear mixed model was fitted to the methane data from the pre-treatment measurements to determine the effects of starting liveweight, time in the PAC and the particular chamber used with each animal on the emissions. Since the purpose of the measurements in the pre-treatment period was to get an unbiased estimate of each sheep before the treatments were applied, the emissions data was adjusted to be used as a covariate for the analysis of post-treatment emissions.

A linear mixed model was used to analyse the post-treatment methane emissions. Methane intensity data underwent an inverse transformation before being used in the analysis.

We recorded abnormally high and clearly inaccurate values for the inter-row pasture intake, which could not be used in calculations for energy intake. Instead we have used data and the equation MEI (MJ/d) = 0.0586(LWG) + 7.43 from Thomas *et al.* (2009)to predict energy intake in our calculations below. An estimate of the inter-row pasture intake was back-calculated using the predicted total energy intake and the intake of the shrub portion of the diet.

## 2.2 Results

*Shrub establishment, survival and biomass*

Ten shrub species were chosen to be used in this study. Six species with anti-methanogenic potential were established in rows as and used as the 'bioactive' shrub treatment. Four species of shrub with the capacity to be highly productive were established in rows and used as the 'productive' shrub treatment (Figure 1). Seven months after establishment, survival of the shrubs ranged from 86% to 100% with an average across the whole site and all species of 96%. The survival rate of each shrub species after 14 months from establishment was not calculated due to the adjacent pasture covering some of the more prostrate shrub species. At the time of the autumn 2014 grazing event (20 months after establishment), the average survival rate of all species of shrubs was 70%. At the conclusion of this grazing event, the shrubs were allowed to regenerate. At the time of the 2015 grazing event (32 months after establishment), the average survival rate of the shrubs still remaining at that time was 89%, which indicates that the shrubs recovered well after the heavy grazing of the previous year.

The average dry weight of edible biomass per shrub for each shrub species at several points in time since the shrubs were established is reported in Table 3. During the autumn 2014 grazing, the shrubs were grazed heavily and little edible biomass remained on the plants when the sheep were removed from the paddocks. At the commencement of the autumn 2015 grazing experiment, there was a similar total of edible biomass available compared to the same season of the previous year (Table 3).

Table 3. Biomass estimates of 10 shrub species 7, 14, 20 (start of autumn 2014 grazing event) and 32 (start of autumn 2015 grazing event) months after establishment at the experimental site on UWA Ridgefield Farm, Pingelly, WA. It was not possible to estimate the biomass of *K. prostrata* at 14 and 32 months because the inter-row had established well and covered the *K. prostrata* (because of the prostrate form of plant)

| | Edible biomass available (g DM per shrub) | | | |
|---|---|---|---|---|
| Shrub species | 7 mths | 14 mths | 20 mths | 32 mths |
| *E. glabra* | 27 | 78 | 24 | 40 |
| *R. preissii* | 411 | 1174 | 740 | 775 |
| *M. brevifolia* | 378 | 422 | 203 | 72 |
| *R. candolleana* | 151 | 374 | 166 | 268 |
| *A. ligulata* | 193 | 418 | 349 | 410 |
| *K. prostrata* | 70 | N/A | 16 | N/A |
| *A. amnicola* | 367 | 163 | 278 | 160 |
| *A. nummularia* | 328 | 545 | 231 | 428 |
| *E. tomentosa* | 130 | 79 | 91 | 85 |
| *M. georgii* | 267 | 297 | 270 | 222 |

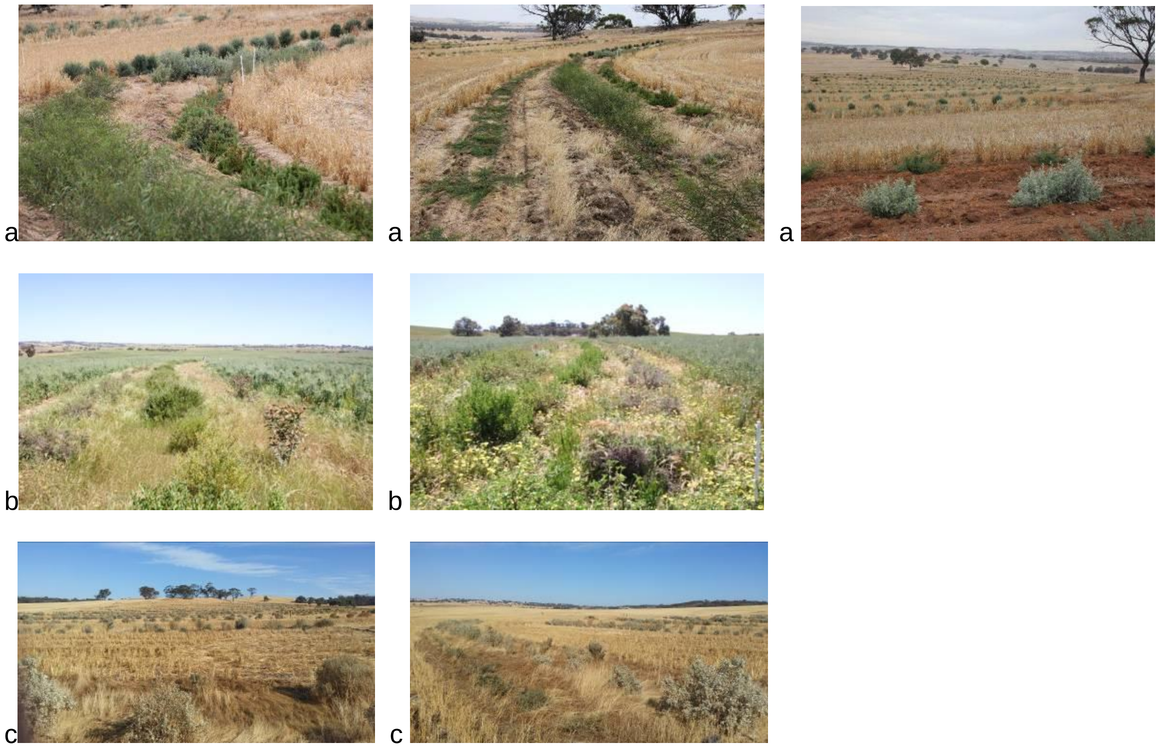


Figure 1. Examples of the forage shrubs at the research site (a - photographs taken 14 March 2013, b – photographs taken 14 October 2013, c – photographs taken 7 March 2014).

The biomass of the pasture treatments (without shrubs) at the commencement and conclusion of the autumn 2014 grazing experiment is reported in Table 4. At the end of the autumn 2014 grazing experiment, biserrula seed was re-applied to the improved pasture plots at 10 kg/ha with the aim of improving biserrula biomass for the autumn 2015 grazing experiment. Visual observations of these plots indicated that this practice had little effect on the dry matter yield of biserrula.

Table 4. Pasture biomass measurements from autumn 2014 and 2015 grazing experiments.

| Treatment | Pre-grazing biomass (kg DM) | Post-grazing biomass (kg DM) |
|---|---|---|
| **Autumn 2014 grazing** | | |
| Unimproved, deferred (UD) | 688 | 478 |
| Unimproved, grazed (UG) | 412 | 331 |
| Improved, deferred (ID) | 1195 | 481 |
| Improved, grazed (IG) | 826 | 603 |
| Inter-row pasture (production shrub plots) | N/A* | 1793 |
| Inter-row pasture (bioactive shrub plots) | N/A* | 1514 |
| **Autumn 2015 grazing** | | |
| Pasture + grain M | 1241 | 691 |
| Pasture + grain 140 g LWG | 1216 | 847 |
| Pasture + grain 180 g LWG | 1391 | 979 |
| Inter-row pasture (productive shrub plots) | 6495 | 3965* |
| Inter-row pasture (bioactive shrub plots) | 7602 | 3770* |

*Calculated biomass was very high and considered to be inaccurate. Consequently, energy intake was predicted for all treatments from live weight gain (LWG) using the equation of Thomas *et al*. (2009) MEI (MJ/d) = 0.0586(LWG) + 7.43.

***Autumn 2014 grazing experiment***

*Feed intake*

After the first week of grazing, the removal of edible leaf material provided clear evidence that the sheep were consuming the shrub and pasture mix (Figure 2). Over the four week grazing period, the sheep consumed an average of 240 g/head/day of shrub material. The shrub intake was complemented by approximately 1.3 kg/head/day of inter-row pasture intake. This biomass intake equates to approximately 15 MJ ME/day (Table 5). The four pasture treatment groups (without shrubs) consumed an average of 0.5 kg/head/day of pasture biomass and 300 g/head/day of oat grain (approximately 11 MJ ME/day).

Figure 2. An example of the effects of one week of grazing in the 'Bioactive shrubs' treatment (Photo taken by Nathan Phillips on March 31, 2014).

Table 5. Mean (±SE) estimated energy intake (MJ ME/day) of sheep consuming one of six grazing systems (Predictions based on live weight gain using the model of Thomas *et al*. (2009)).

| | Total energy intake (MJ ME/day) |
|---|---|
| Unimproved, deferred (UD) | 11.6 ± 0.37 |
| Unimproved, grazed (UG) | 11.5 ± 1.58 |
| Improved, deferred (ID) | 11.6 ± 1.09 |
| Improved, grazed (IG) | 11.3 ± 0.79 |
| Productive shrubs | 15.8 ± 0.62 |
| Bioactive shrubs | 15.6 ± 1.29 |

*Liveweight*

The sheep that grazed shrubs gained twice the amount of liveweight compared to sheep grazing the pasture treatments ($p = 0.024$; Figure 3). The sheep grazing the productive shrubs gained 143 g/head/day and the sheep grazing the bioactive shrubs gained 140 g/head/day. The sheep grazing the shrub systems finished the four-week grazing period at an average weight of 43.6 kg, which was 4.5 kg heavier than their starting weight. The sheep that grazed pastures were, on average, 2.3 kg heavier than their starting weight.

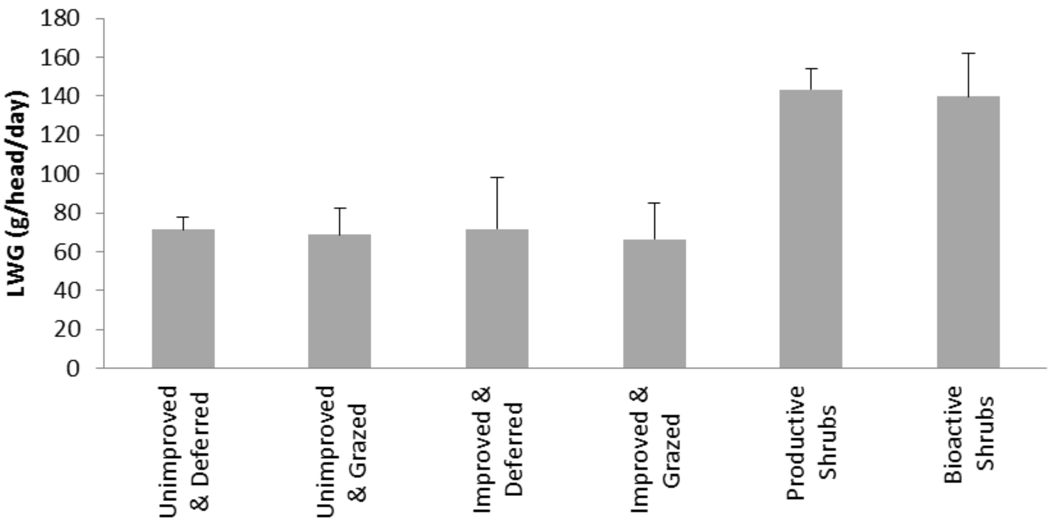


Figure 3. Mean (±SE) daily liveweight gain (g/head/day) of sheep grazing one of six grazing systems for four weeks.

*Methane*

Sheep that grazed the shrub/inter-row systems tended to have a lower methane intensity (i.e. produced less methane per unit of average daily liveweight gain) than the sheep that grazed pastures ($p = 0.073$; Table 6 and Figure 4). The lower methane intensity produced by these sheep was largely attributed to their higher liveweight gain as a result of their greater energy intake. However, the sheep grazing the shrub treatments also produced less methane per unit of calculated energy intake ($p = 0.011$) than the sheep grazing the pasture (without shrubs) treatments (Table 6 and Figure 5). There were no differences in methane intensity between sheep offered the bioactive shrubs compared with the productive shrubs.

Table 6. Mean (SE) liveweight gain, methane production and methane intensity of sheep fed one of six grazing treatment diets for four weeks. Note that methane production and methane intensity are representative of an estimated 15 hours of grazing/ruminating activity per day.

| | Liveweight gain (g/head/day) | Methane production (g/head/day) | Methane intensity (g/g LWG) | Estimated energy intake (MJ ME/d) | Methane production per unit intake (g/MJ ME) |
|---|---|---|---|---|---|
| Unimproved, deferred (UD) | 71 ± 6.3 | 11.3 ± 0.24 | 0.16 ± 0.010 | 11.6 ± 0.37 | 0.98 ± 0.01 |
| Unimproved, grazed (UG) | 69 ± 13.5 | 13.6 ± 0.51 | 0.21 ± 0.035 | 11.5 ± 1.58 | 0.94 ± 0.14 |
| Improved, deferred (ID) | 72 ± 26.9 | 11.5 ± 1.03 | 0.17 ± 0.074 | 11.6 ± 1.09 | 0.98 ± 0.02 |
| Improved, grazed (IG) | 66 ± 18.6 | 9.9 ± 1.32 | 0.21 ± 0.028 | 11.3 ± 0.79 | 1.20 ± 0.04 |
| Productive shrubs | 143 ± 10.6 | 12.4 ± 1.49 | 0.09 ± 0.007 | 15.8 ± 0.62 | 0.78 ± 0.07 |
| Bioactive shrubs | 140 ± 22.0 | 11.1 ± 1.37 | 0.09 ± 0.019 | 15.6 ± 1.29 | 0.73 ± 0.13 |

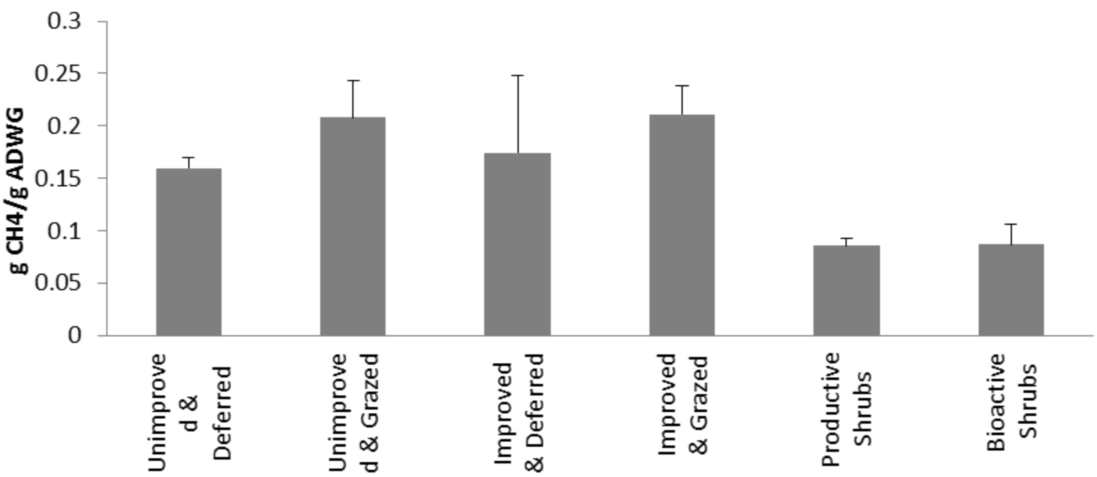


Figure 4. Methane intensity (g $CH_4$/g average daily liveweight gain, ADWG) for sheep grazing one of six shrub/pasture treatments for four weeks.

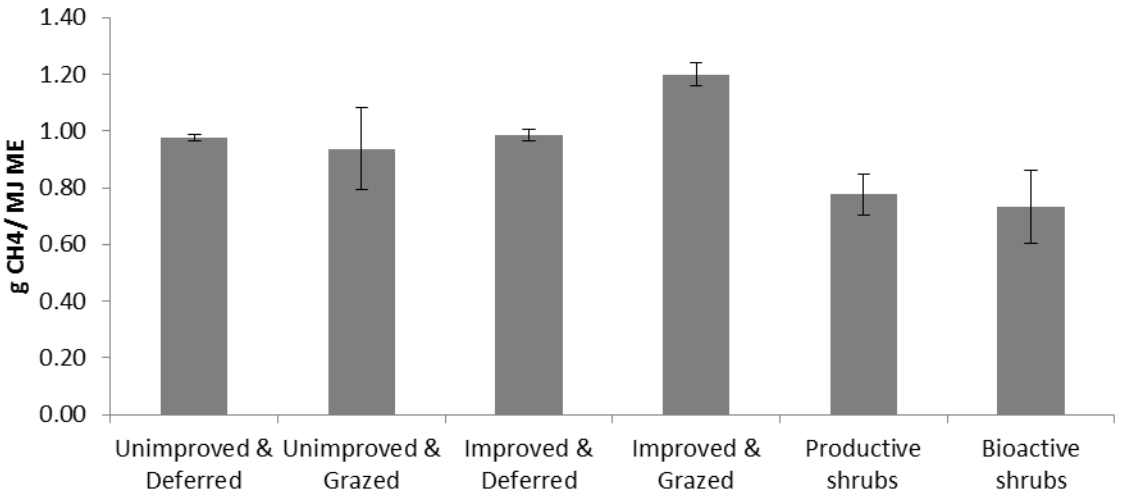


Figure 5. Mean methane emissions per unit of energy intake (g $CH_4$/ MJ ME) of sheep grazing one of six shrub/pasture systems (energy intake based on model predictions)

*In vitro methane analysis*

Results from the *in vitro* methane analysis indicated that *E. glabra* produced less methane than the other shrub species (Figure 6). This result is consistent with other work that has demonstrated that *E. glabra* modifies rumen fermentation and has an anti-methanogenic effect (Li *et al.* 2010). *A. ligulata* and *K. prostrata*, species are also known have an anti-methanogenic effect and produced low levels of methane. A similar pattern was evident in the ruminal ammonia ($NH_3$) concentration for these three species (Figure 7). However, volatile fatty acid (VFA) production was similar to the other shrub species (Figure 8).The increase in ruminal $NH_3$ at the final sample collection for some of the shrub species coincided with the break-of-season rains. *A. ligulata* produced the lowest volume of gas, although only by 5-15% (Figure 9).

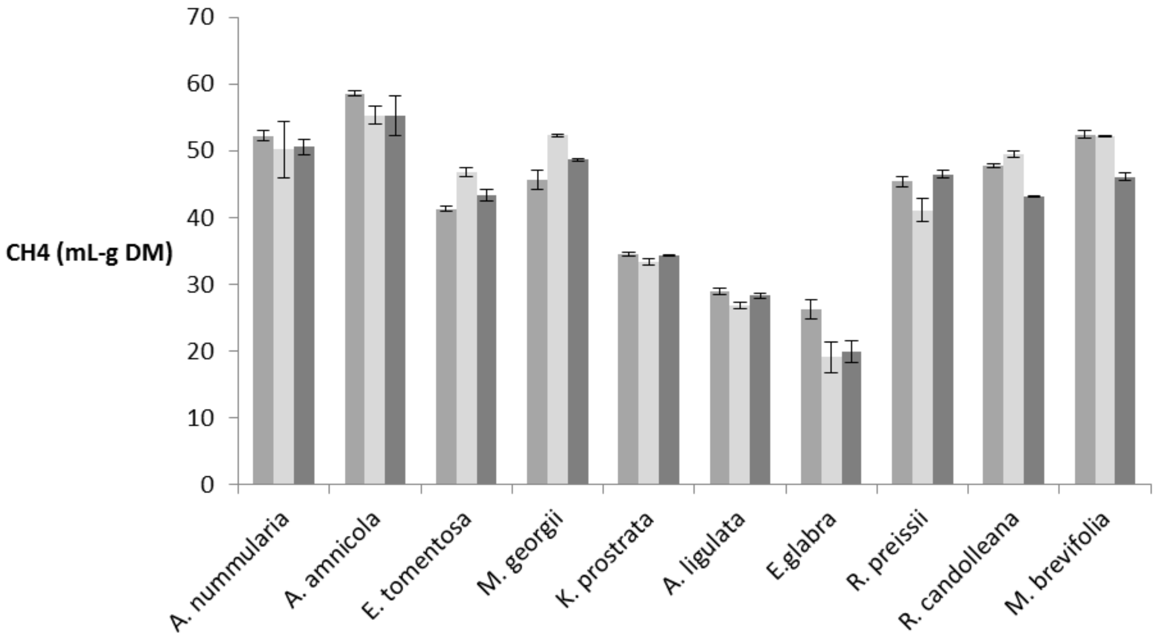


Figure 6. Mean (±SE) methane (mL/g DM) produced in an *in vitro* assay by shrub species at three different time points during the autumn 2014 grazing experiment. Mid-grey bars represent data collected on 7/3/14, light grey bars represent data from 14/4/14 and dark grey bars represent data from 12/5/14.

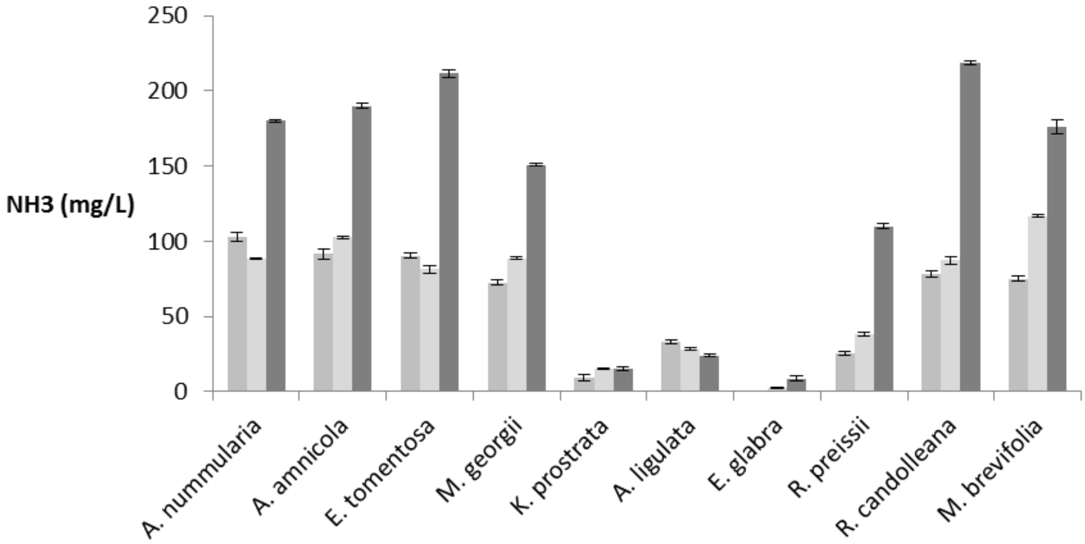


Figure 7. Mean (±SE) ammonia ($NH_3$) concentration (mL/L) produced in an *in vitro* assay by shrub species at three different time points during the autumn 2014 grazing experiment. Mid-grey bars represent data collected on 7/3/14, light grey bars represent data from 14/4/14 and dark grey bars represent data from 12/5/14.

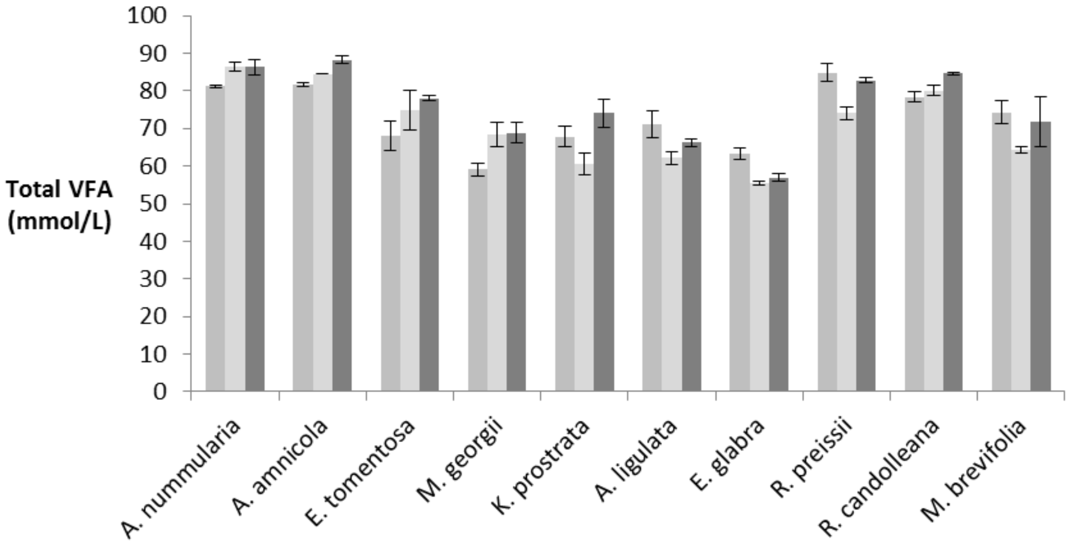


Figure 8. Mean (±SE) total volatile fatty acids (VFA) concentration (mmol/L) produced in an *in vitro* assay by shrub species at three different time points during the autumn 2014 grazing experiment. Mid-grey bars represent data collected on 7/3/14, light grey bars represent data from 14/4/14 and dark grey bars represent data from 12/5/14.

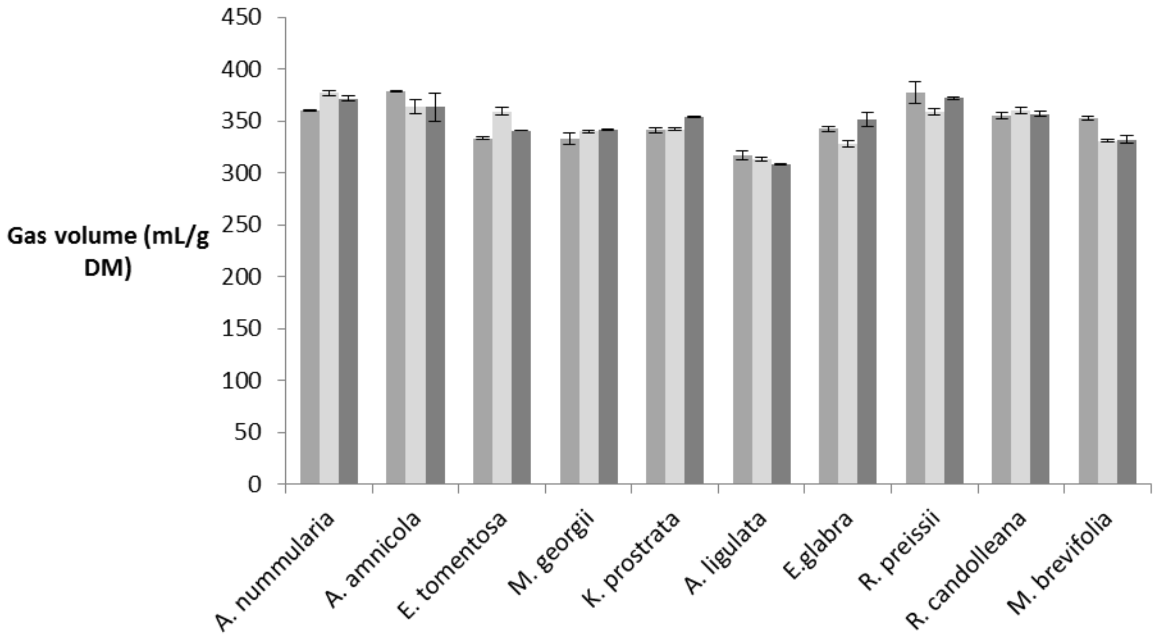


Figure 9. Mean (±SE) volume of gas (mL/g DM) produced in an *in vitro* assay by shrub species at three different time points during the autumn 2014 grazing experiment. Mid-grey bars represent data collected on 7/3/14, light grey bars represent data from 14/4/14 and dark grey bars represent data from 12/5/14.

*Nutritive value analysis*

The crude protein content varied between the shrub species (Table 7). *M. georgii* had the highest protein content (17.7%), while *E. glabra* had the lowest (8.7%). On average, the productive shrubs had a higher crude protein content (15.4%) than the bioactive shrubs (13.0%). Conversely, the bioactive shrubs had a higher ADF and NDF proportion than the productive shrubs (Table 7).

Table 7. Nutritive value parameters of shrub species during the autumn 2014 grazing experiment

| Shrub species | Crude protein (%) | Neutral Detergent Fibre (%) | Acid Detergent Fibre (%) | Ash (%) | Organic matter fraction (%) |
|---|---|---|---|---|---|
| **Bioactive shrubs** | | | | | |
| *E. glabra* | 8.7 | 32.5 | 24.6 | 10.0 | 90 |
| *R. preissii* | 10.7 | 30.1 | 18.5 | 9.7 | 90 |
| *M. brevifolia* | 16.8 | 36.0 | 19.3 | 20.4 | 80 |
| *R. candolleana* | 14.8 | 36.6 | 20.6 | 11.9 | 88 |
| *A. ligulata* | 11.4 | 42.2 | 33.7 | 5.6 | 94 |
| *K. prostrata* | 15.5 | 35.2 | 25.4 | 4.9 | 95 |
| Mean | 13.0 ± 1.29 | 35.4 ± 1.68 | 23.7 ± 2.31 | 10.4 ± 2.28 | 89.5 ± 2.19 |
| **Productive shrubs** | | | | | |
| *A. amnicola* | 14.8 | 30.1 | 15.3 | 20.0 | 80 |
| *A. nummularia* | 14.4 | 32.2 | 17.6 | 18.3 | 82 |
| *E. tomentosa* | 14.4 | 25.7 | 13.4 | 20.5 | 80 |
| *M. georgii* | 17.8 | 38.4 | 22.3 | 14.2 | 86 |
| Mean | 15.3 ± 0.81 | 31.6 ± 2.63 | 17.2 ± 1.92 | 18.3 ± 1.43 | 82.0 ± 1.41 |

***Autumn 2015 grazing experiment***

*Feed intake*

Over the four week grazing period, the sheep consumed an average of 75 g/head/day of shrub material. Using the formula from Thomas *et al*. (2009), sheep in the shrub treatments had an energy intake of approximately 19 MJ ME/day (Table 8). The estimated pasture intake of the sheep grazing the shrub systems was 2.7 kg/head/d. This estimate of pasture intake was used in a calculation for total energy intake and compared to the predicted energy intake (Table 8). The three pasture treatment groups (without shrubs) consumed an average of 1 kg DM/head/day of biomass, in addition to their allocated grain ration (Table 8).

Table 8. Mean intake of dietary components (±SE), mean predicted energy intake (MJ ME/day) (using data from model predictions) and total energy intake (MJ ME/day) (using data back-calculated for shrub treatments) of sheep consuming one of five grazing systems.

| | Total shrub intake (g DM/h/d) | Total pasture intake (g DM/h/d) [a] | Total grain intake (g DM/h/d) | Predicted energy intake (MJ ME/d)[b] | Total energy intake (MJ ME/d) |
|---|---|---|---|---|---|
| Pasture + grain M | | 1235 ± 157.3 | 77.6 ± 0.55 | 8.3 ± 0.53 | 9.5 ± 1.10 |
| Pasture + grain 140 g LWG | | 840 ± 269.3 | 198.6 ± 0.96 | 10.2 ± 0.91 | 8.2 ± 1.89 |
| Pasture + grain 180 g LWG | | 922 ± 156.7 | 348.7 ± 0.96 | 12.7 ± 1.08 | 10.5 ± 1.11 |
| Productive shrubs | 79.5 ± 10.07 | 2695 ± 234.0 | | 19.4 ± 1.57 | 19.4 ± 1.57 |
| Bioactive shrubs | 70.2 ± 13.16 | 2691 ± 104.3 | | 19.3 ± 0.80 | 19.3 ± 0.80 |

[a]Estimated pasture intake was calculated using predicted energy intake (for shrub groups only); [b]Predicted from liveweight gain using the equation of Thomas *et al*. (2009) .

*Liveweight*

The sheep that grazed the shrub-pasture mixture gained at least twice the amount of live weight compared to sheep that grazed pasture only ($p < 0.001$; Figure 10). The sheep grazing the productive shrubs gained 204 g/head/day and the sheep grazing the bioactive shrubs gained 203 g/head/day. The sheep grazing the shrub systems finished the four-week grazing period at an average weight of 40.6 kg, which was 5.7 kg heavier than their starting weight. The sheep grazing only pasture were supplemented with oat grain at levels to achieve different weight gains (maintenance, 140 g/head/d and 180 g/head/d). Although these sheep did not reach the desired liveweight gains during the four weeks, there was a linear effect of the amount of grain supplement on liveweight ($p = 0.018$). The sheep in the maintenance treatment finished the four-week grazing period at an average weight of 35.3 kg, which was 400 g heavier than their starting weight. The sheep that were fed grain to achieve a gain of 140 g/head/d had an average liveweight of 36.2 kg, which was 1.3 kg heavier than their starting weight. The sheep fed the highest level of grain (which aimed to achieve 180 g/head/d liveweight gain) had an average liveweight of 37.4 kg, which was 2.5 kg heavier than their starting weight. The lower than expected growth rates of the sheep in the pasture treatments were due to pasture intake being less than expected.

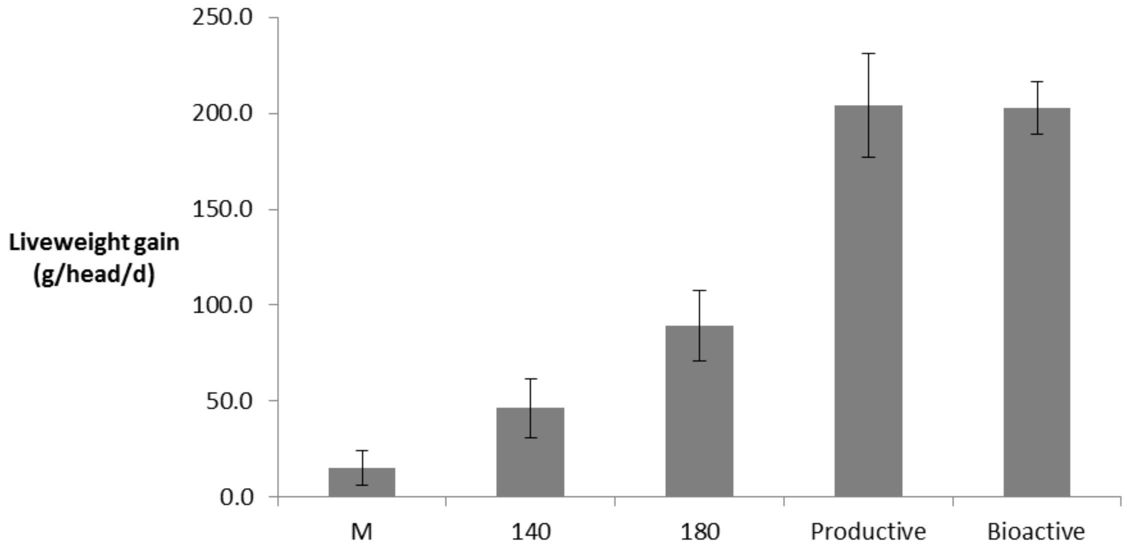


Figure 10. Mean (±SE) liveweight gain (LWG; g/head/d) of sheep in one of five grazing systems: pasture with oat grain fed at a level to maintain weight (M), pasture with oat grain fed at a level estimated to achieve 140 g/head/d LWG (140), pasture with oat grain fed at a level estimated to achieve 180 g/head/d LWG (180), shrubs with productive characteristics with an anti-methanogenic pasture inter-row (Productive) and shrubs with anti-methanogenic characteristics with an anti-methanogenic pasture inter-row (Bioactive).

*Methane*

Sheep that grazed the shrub/inter-row systems had a lower methane intensity (i.e. produced less methane per unit of average daily liveweight gain) than the sheep that grazed pastures ($p < 0.001$; Table 9). The lower methane intensity produced by these sheep was largely attributed to their higher liveweight gain. Likewise, methane intensity was also different between the three pasture treatments ($p = 0.005$), where the sheep fed the highest level of grain had a lower methane intensity than the sheep fed at a level to maintain weight. Again, this improvement in methane intensity is a result of improved liveweight gain. There were no differences in methane intensity between sheep offered the bioactive shrubs compared with the productive shrubs. The sheep grazing the shrub systems produced numerically less methane per unit of calculated ME intake, but this was not significantly different to that of the pasture groups (Table 10).

Table 9. Mean (SE) methane production (g/day) and methane intensity of sheep fed one of five grazing treatment diets for four weeks. Note that the data for methane intensity underwent an inverse transformation to meet the assumptions of analysis of variance, and the means of the re-transformed data are presented here.

| | Methane production (g/hd/day) | Methane intensity ($g\ CH_4/g$ LWG) |
|---|---|---|
| Pasture + grain M | $9.9 \pm 0.43^a$ | $0.54 \pm 0.085^a$ |
| Pasture + grain 140 g LWG | $11.1 \pm 1.37^a$ | $0.36 \pm 0.058^a$ |
| Pasture + grain 180 g LWG | $12.5 \pm 1.11^a$ | $0.14 \pm 0.005^b$ |
| Productive shrubs | $19.3 \pm 1.03^b$ | $0.093 \pm 0.015^c$ |
| Bioactive shrubs | $18.0 \pm 1.65^b$ | $0.089 \pm 0.007^c$ |

Table 10. Mean methane production per unit of energy intake (g/MJ ME intake) of sheep grazing one of five grazing treatment diets for four weeks. Note that this was calculated using the 'total energy intake' presented in Table 8.

| | Methane production per unit intake ($g\ CH_4$/MJ ME) |
|---|---|
| Pasture + grain M | $1.10 \pm 0.158^a$ |
| Pasture + grain 140 g LWG | $1.62 \pm 0.567^a$ |
| Pasture + grain 180 g LWG | $1.24 \pm 0.226^a$ |
| Productive shrubs | $0.98 \pm 0.041^a$ |
| Bioactive shrubs | $0.93 \pm 0.063^a$ |

*In vitro methane analysis*

The *in vitro* methane analysis indicated that *E. glabra, K. prostrata* and *A. ligulata* produced less methane than the other shrub species (Figure 11). *A. ligulata* also produced the lowest volume of gas (Figure 14). Overall, the bioactive shrubs produced less methane and had a lower gas volume than the productive shrubs. The productive shrubs produced almost twice the ammonia concentration than the bioactive shrubs (Figure 12). The volatile fatty acid concentration of the two shrub groups was similar (Figure 13).

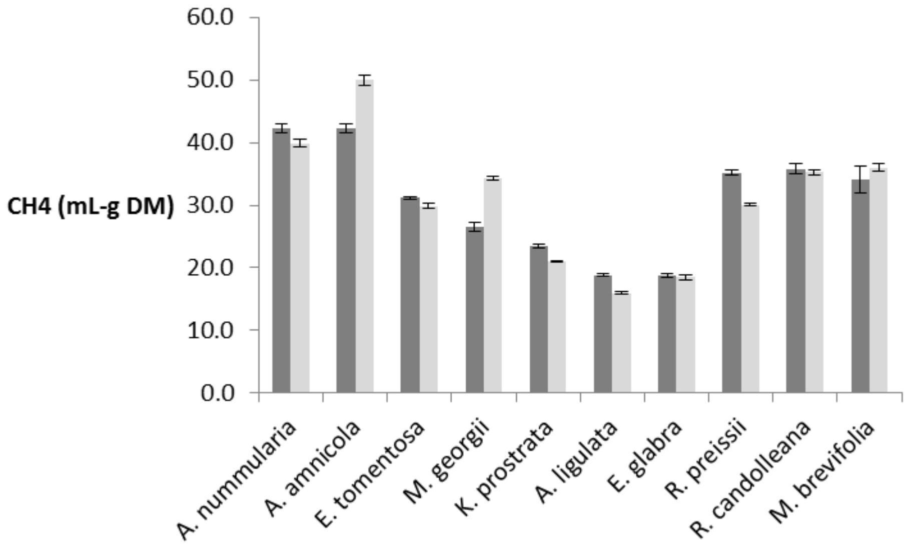


Figure 11. Mean (±SE) methane (mL/g DM) produced in an *in vitro* assay by shrub species at start (dark grey bars) and end (light grey bars) of the autumn 2015 grazing experiment.

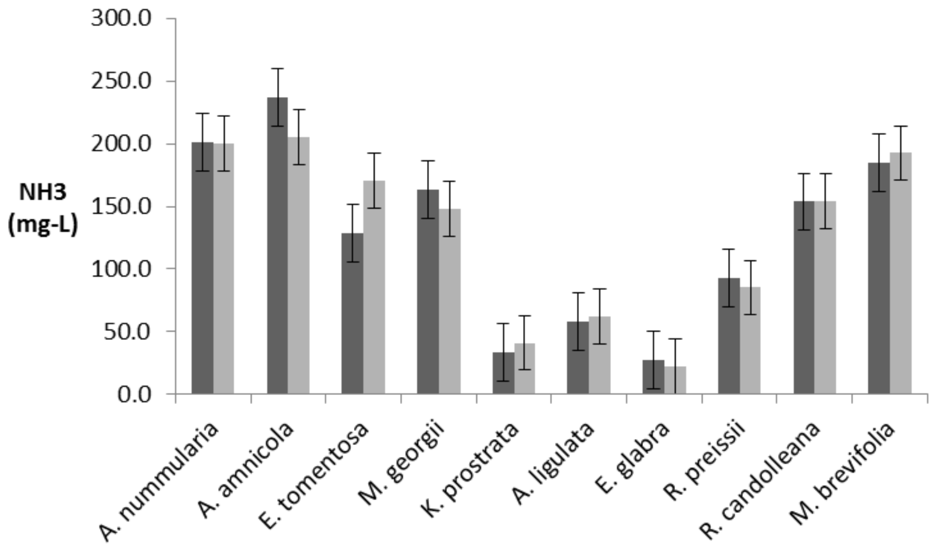


Figure 12. Mean (±SE) total ammonia ($NH_3$) concentration (mg/L) produced in an *in vitro* assay by shrub species at the start (dark grey bars) and end (light grey bars) of the autumn 2015 grazing experiment.

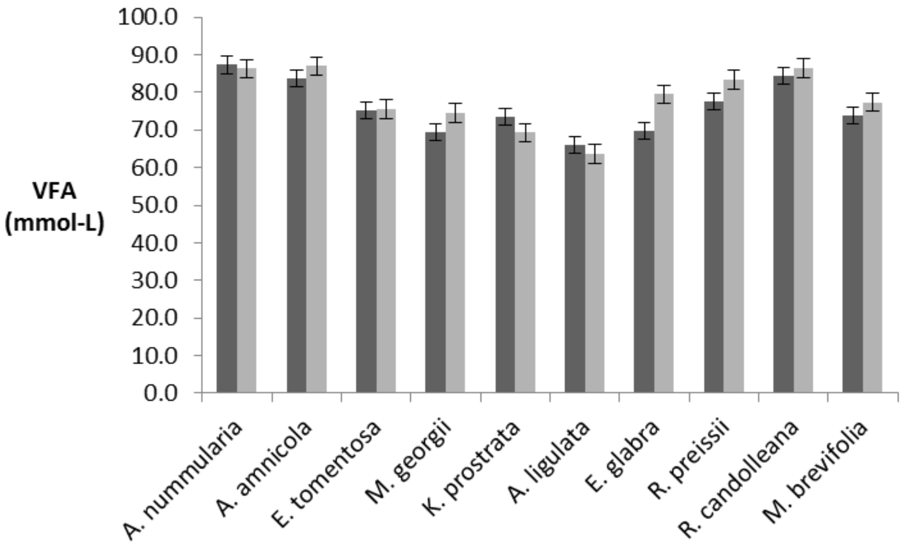


Figure 13. Mean (±SE) total volatile fatty acids (VFA) concentration (mmol/L) produced in an *in vitro* assay by shrub species at the start (dark grey bars) and end (light grey bars) of the autumn 2015 grazing experiment.

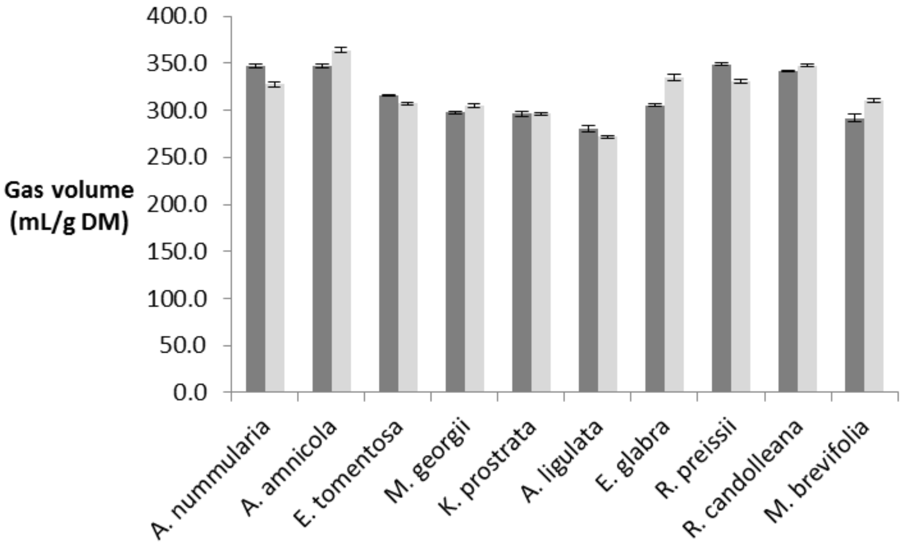


Figure 14. Mean (±SE) volume of gas (mL/g DM) produced in an *in vitro* assay by shrub species at the start (dark grey bars) and end (light grey bars) of the autumn 2015 grazing experiment.

*Nutritive value analysis*

The crude protein content varied between the shrub species (Table 11). *M. brevifolia* had the highest protein content (21.5%), while *E. glabra* had the lowest (8.6%). On average, the bioactive shrubs had a similar NDF proportion than the productive shrubs, yet the bioactive shrubs had a slightly higher ADF proportion (Table 11).

Table 11. Nutritive value parameters of shrub species during the autumn 2015 grazing experiment

| Shrub species | Crude protein (%) | Dry Matter (%) | Neutral Detergent Fibre (%) | Acid Detergent Fibre (%) | Ash (%) | Organic matter fraction (%) |
|---|---|---|---|---|---|---|
| **Bioactive shrubs** | | | | | | |
| *E. glabra* | 8.6 | 70 | 30.0 | 20.1 | 4.6 | 95 |
| *R. preissii* | 12.9 | 38 | 23.0 | 9.9 | 13.6 | 86 |
| *M. brevifolia* | 21.5 | 42 | 32.9 | 16.0 | 23.3 | 77 |
| *R. candolleana* | 18.4 | 40 | 27.8 | 13.5 | 17.1 | 83 |
| *A. ligulata* | 11.3 | 55 | 39.6 | 32.1 | 10.6 | 89 |
| *K. prostrata* | 14.6 | 81 | 36.5 | 25.1 | 5.3 | 95 |
| Mean | 14.6 ± 1.93 | 54 ± 7.3 | 31.6 ± 2.45 | 19.5 ± 3.32 | 12.4 ± 2.93 | 88 ± 2.9 |
| **Productive shrubs** | | | | | | |
| *A. amnicola* | 17.0 | 24 | 34.9 | 16.8 | 21.3 | 79 |
| *A. nummularia* | 19.8 | 38 | 27.0 | 14.0 | 23.5 | 76 |
| *E. tomentosa* | 19.2 | 30 | 27.5 | 11.3 | 22.4 | 78 |
| *M. georgii* | 17.6 | 33 | 39.0 | 23.6 | 20.1 | 78 |
| Mean | 14.6 ± 0.66 | 31 ± 2.9 | 32.1 ± 2.92 | 16.4 ± 2.64 | 21.8 ± 0.73 | 78 ± 0.6 |

# 3. Part B. Open-path methane measurements

## 3.1 Introduction

This report details the in-field methane emission measurements from sheep grazing two grazing systems, conventional pasture and shrubs plus inter-row pasture chosen for anti-methanogenic potential. Methane emissions were measured at the paddock scale using open-path FTIR spectroscopy (OP-FTIR) in conjunction with a tracer gas to infer the emission strength. Data on animal productivity (feed intake, live weight and condition) were also collected to provide emissions intensity estimates from the two treatments.

The open-path FTIR plus tracer gas (OPFTIR-tracer gas) technique is a technique used to estimate emissions of methane from ruminants in their normal, undisturbed free-grazing environment. The technique releases a tracer-gas at a known, controlled rate close to the mouth of the animal with both the tracer-gas and the emitted $CH_4$ measured simultaneously by OP-FTIR spectroscopy downwind from the animals. Generally $N_2O$ is used as the tracer gas as it is safe and non-toxic, can be readily released in sufficient quantities for enhancements above background to dominate natural fluctuations, and can be measured simultaneously with $CH_4$ and $CO_2$ by FTIR spectroscopy.

The technique measures herd-averaged emission strength from the animals. Emissions are measured continuously over 24 hours for multiple days, and with a temporal resolution of minutes (typically 5), provides information on the distribution of emission over the day to highlight changes in emissions with animal behaviour.

The technique is a micrometeorological technique and relies on wind to transport the animal produced methane and tracer-gas to the measurement paths of the instrument, and is subject to loss of data under non-favourable wind conditions. This can lead to discrimination to, most often, daytime data. Using a tracer-gas allows the minimum wind criteria to be less restrictive, with minimum wind speed criteria reduced from the typical 1.5 m/s to 0.5m/s.

## 3.2 Methodology

*Site*

Measurements were made at the University of Western Australia Future Farm, Ridgefield, located near Pingelly, east of Perth.

Emissions were measured from sheep on the treatments, grazing conventional pasture and shrubs from 40 sheep in each treatment, with the sheep sourced at random from 3 of the replicates of each treatment (16 sheep per replicate). Emissions were measured following the final portable accumulation chambers (PAC) measurements over 7 days for each treatment. Emissions from the two treatments were measured sequentially with equipment moved between sites. The paddock where sheep grazed conventional pasture was lacking in pasture with any nutritional value, and sheep were supplemented with 350 g/h/day of oat grain and 250 g/h/day of oaten hay. The paddock where the sheep grazed shrubs was chosen as it provided the range of shrubs from both the productive and bioactive treatments that were grazed by the sheep for the PAC measurements and the terrain was more suitable to the measurement technique. A sub paddock was fenced within the larger paddock to provide an area suitable for the measurements with a similar area to the pasture paddock. The species included within the sub plot are detailed in Table 2. The dimensions of the paddocks for both treatments were ~ 100 x 40 m.

Table 2 Species available in the sub-paddock of the nursery paddock.

Note K. prostrate was not available

| Species | Species |
|---|---|
| E. glabra | A. nummularia |
| R. preissii | A. amnicola |
| M. brevifolia | M. georgii |
| R. candolleana | E. tomentosa |
| A. ligulata | |

*Experimental Design*

Three measurements paths were positioned outside, and parallel to, the two long paddock boundaries and one shorter boundary. One OP-FTIR instrument formed 2 measurement paths at 90 degrees. The instrument either rotated between the two paths on a 5 min duty cycle, or remained orientated to one path, dependent on predicted wind directions. A second instrument, orientated to one reflector provided the 3$^{rd}$ measurement path. Due to the geometry of the trial paddocks, a 4$^{th}$ measurement path on the remaining boundary could not be used.

The $CH_4$ emitted from the animals and the $N_2O$ from the canisters is carried by the same wind turbulence to the measurement path, downwind from the animals, and through the infrared beam of the open-path system, where the concentration of $N_2O$ tracer-gas and $CH_4$ emitted by the animals is measured simultaneously by the spectrometer. The upwind path measured any $CH_4$ or $N_2O$ entering the experimental site from other local sources. The concentration due to the experimental animals is assumed to be the difference in concentration entering and leaving the paddock Figure 1.

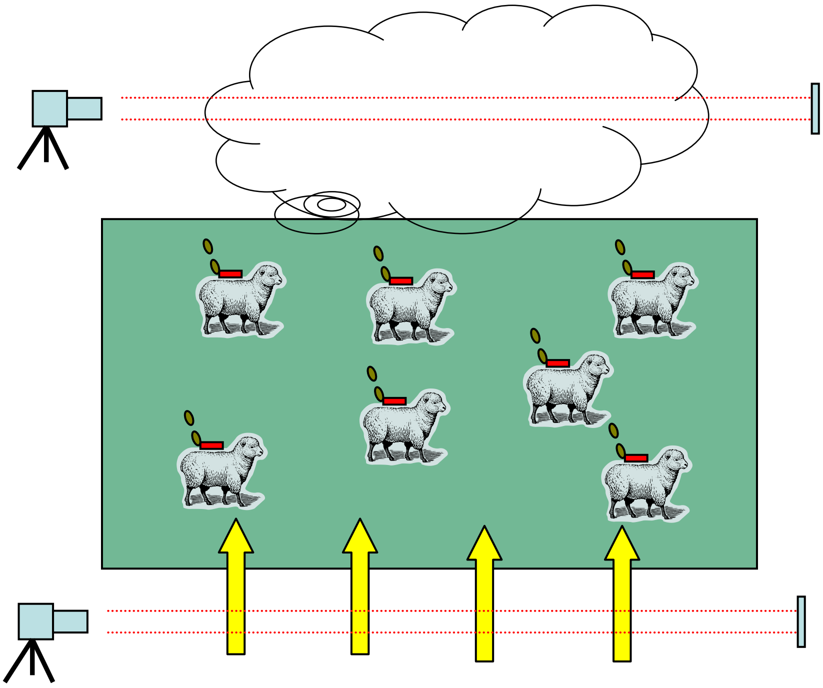

Figure 1: Schematic showing placement of instruments I1 and I2 in relation to experimental enclosure for animals and the predominant wind direction; tracer gas canisters located on animals.

*Open-Path FTIR Spectrometer*

The OP-FTIR instrument consists of an FTIR spectrometer, (Matrix IR-Cube, Bruker Optik GmbH, Ettlingen, Germany) equipped with a mechanically cooled (-196°C, RicorK508) MCT detector (Infrared Associates Inc., Florida, USA, or Judson Industries, Montgomeryville, PA, USA) coupled to a 250 mm Schmidt-Cassegrain telescope (LX 200ACF, Meade Instruments Corporation, Irvine California, USA). The telescope has been modified to function as a parallel beam expander, expanding the beam from 25 to 250 mm diameter and reducing beam divergence by a factor of ten to 2 mradians. The system is mounted onto a heavy duty tripod (Gibralter model 4-60450-OA, Quickset International Inc., Illinois, USA) on a computer controlled automated instrument mount (AIM; IAAC; Unanderra, Australia) to allow automated alignment of the beam between spectrometer and multiple retro-reflectors. The spectrometer scans continuously and, in typical operation, records a time-averaged (nominally every 3 to 5 minutes) infrared absorption spectrum of the open atmospheric path between spectrometer and retro-reflector located 100 to 130 m from the instrument. Each spectrum is analysed immediately after collection using the MALT analysis program to provide path-averaged concentrations of $NH_3$, $N_2O$, $CO_2$, $CH_4$, CO and water vapour (Griffith, 1996). Operation of the system is fully automated under the control of a laptop computer running a program written at the University of Wollongong (OSCAR, G. Kettlewell).

*Tracer-gas*

The tracer-gas canisters are 240 x 60 mm diameter aluminium canisters commonly used as “paint ball” canisters fitted with a head encompassing a capillary tube (PEEKsil HPLC capillary tubing, 0.025 mm inner diameter, SGE Analytical Science Pty Ltd, Ringwood, Vic. Australia) to limit the flow-rate of tracer gas to around 10 $gh^{-1}$. Laboratory tests showed an increase in flow rate with the canister in a horizontal position, as on the sheep’s back, compared to vertical as attached to cattle, and the length of the capillary was increased from 25 to 35 mm to ensure a flow rate of ~10 $gh^{-1}$. Each canister is filled with approximately 300 g of $N_2O$ (liquid nitrous oxide, engine boost grade, product code 624, BOC Australia, Sydney, NSW, Australia) as the tracer gas, with canisters replaced after 24 hours. Canisters are exchanged each 24 hours, with animals moved into a portable yard adjacent to the paddock to facilitate the exchange (Figure 2).

Canisters were mounted on the sheep’s back using a purpose built canvas backpack, attached via Velcro strips glued onto the sheep’s back (Figure2). A layer of insulation provides protection from solar radiation.

The wool on the back of sheep was clipped to limit movement of the canister (Figure 2). A full gas canister weighs between 900 and 1100 g, and once the animal is trained to the canister, appears to have minimal detrimental effect on animal behaviour. The sheep were trained to the backpack and the weight of the canisters using water bottles for 24-48 hours before the start of measurements. Canisters were mounted on 20 sheep for the measurements, with 25 sheep initially fitted and trained to backpacks. If a sheep appeared distressed by the backpack, the backpack was removed. In this experiment, 1 sheep showed signs of agitation and the backpack removed. Clipping the wool is important as any movement of the canister on the sheep's back is more likely to cause stress to the animal than the weight of the canister.

Figure 2 Sheep's wool being clipped ready for backpacks; with backpack and gas canisters in the race to have the canisters exchanged, and in the shrubs plus inter-rows paddock.

The average tracer-gas flow-rate for each canister is determined from the weight loss of gas and the release time. However, as the instantaneous flow-rate of the gas varies with temperature the canister temperature is monitored using temperature logging buttons (logging interval 5 minutes; Thermocron eTemperature model TCS, OnSolutions, Baulkham Hills, NSW, Australia) attached to each canister.

The time (t, hours) and temperature (T, °C) dependent flow rate of the $N_2O$ from a canister, *F*(t), ($gh^{-1}$) can be calculated from the relationship:

$$F(t) = F_0 + \frac{dF}{dT}\left(T(t) - T_0\right) \quad \text{Eq. 1}$$

Where $\frac{dF}{dT}$ is the temperature dependence of the flow rate, determined in the laboratory by monitoring the flow rate at a range of temperatures as 0.184 ± 0.036g $h^{-1}°C^{-1}$,

$F_0$ is the canister flow rate ($gh^{-1}$) at $T_0$=0°C.

As the integrated flow rate over the release time, tr, is equal to the mass of gas lost $\Delta m$ (g),

$$\int_0^{t_r} F(t)dt = \Delta m \quad \text{Eq. 2}$$

$F_0$ can be calculated from Eq. 1 and 2 such that:

$$F_0 = \frac{\Delta m}{t_{t_r}} - \frac{dF}{dT}\int_0^{t_r}\left(T(t) - T_0\right)dt \quad \text{Eq. 3}$$

$F_0$ is calculated for each canister from Eq. 3, allowing F(t) to be calculated at temperature T and time t from Equation 1. The time-temperature dependent $N_2O$ emission rate is the sum of the flow from the total number of canisters, n

$$Q_{N_2O(t)} = \sum_{i=1}^{n} F_i(t) \quad \text{Eq. 4}$$

$Q_{N2O(t)}$ is interpolated from the time resolution of the temperature buttons to that of the $CH_4$ and N2O volume mixing ratio data.

The CH4 emission, QCH4, at time t is calculated from the relationship:

$$Q_{CH_4} = \frac{\Delta[CH_4]}{\Delta[N_2O]} Q_{N_2O} * \frac{MWt_{CH_4}}{MWt_{N_2O}} * \frac{24}{n_{animals}} \quad \text{Eq. 5}$$

where: $Q_{CH_4}$ = flux $CH_4$ at time t (g $animal^{-1}$ $day^{-1}$), $Q_{N_2O}$ = time-temperature emission of the tracer gas, $N_2O$ at time t as calculated above ($gh^{-1}$), $\Delta[CH_4]$ = enhancement in $CH_4$ mixing ratio over local background mixing ratio (ppbv) and $\Delta[N_2O]$ = enhancement in $N_2O$ mixing ratio over local background mixing ratio (ppbv) both at time t, $MWt_{CH4}$ and $MWt_{N2O}$ are the molecular weights of $CH_4$ (16.0 $gmol^{-1}$) and $N_2O$ (44.0 $gmol^{-1}$), $n_{animals}$ is the number of animals, and 24 ($h.day^{-1}$) converts the flux from per hour to per day.

*Weather Station and Meteorological Criteria*

A weather station installed close to the measurement site provided 3-dimensional wind speed and wind direction data at 10 Hz resolution and averaged to 15 minutes (sonic anemometer, CSAT3, Campbell Scientific Inc, Logan Utah, USA). A wind sentry and cup anemometer (03001 RM Young Wind Sentry set, Campbell Scientific Inc, Logan Utah, USA) provide additional data on wind direction and speed, in conjunction with air temperature (T107, Campbell Scientific Inc, Logan Utah, USA) and humidity (HMP55C, Campbell Scientific Inc, Logan Utah, USA) measured each minute and averaged to 5 minutes.

All data are recorded to a data logger (CR5000, Campbell Scientific Inc, Logan Utah, USA) and downloaded hourly via an internet connection.

## 3.3 Results and discussion

Animal productivity data are detailed in Table 2. Sheep on both treatments lost weight over the trial period, with the weight loss by sheep on the shrubs significantly less, reflecting a higher intake.

Within the experimental area of the Shrubs treatment, the sheep had grazed the majority of the available shrubs by day 4. On day 4, 5 and 6 the sheep were released into the greater area of the larger paddock to graze for several hours coinciding with the typical afternoon grazing period (~ 15:00 to 18:00), relieving the pressure on the remaining shrub biomass in the experimental enclosure for the morning grazing period. On day 4 and 5 the sheep remained close to the experimental enclosure boundaries and quality emissions data were successfully collected. On day 6 the sheep moved further into the main plot, and measured $CH_4$ and $N_2O$ mixing ratios were below the minimum criteria.

Feed was supplied to the sheep on the conventional pasture each day after the canisters were exchanged (~10:00).

Table 2 Summary of sheep productivity data

| Treatment | Number Sheep | No Days | LWG (g/head/day) | Total ME intake (MJ/kg DM) |
|---|---|---|---|---|
| Pasture | 40 | 7 | -120 | 6.31 |
| Shrubs | 40 | 7 | -22.9 | 6.68 |

*Methane Emissions*

$CH_4$ and $N_2O$ mixing ratios were measured in the gas plume from the sheep and gas canisters at a temporal resolution of 5 minutes. Prior to the calculation of $CH_4$ emissions, (Equation 5) filtering criteria were imposed to remove data collected under poor conditions, which included wind speeds < 0.5 m/s and a mixing-ratio above local background levels, $\Delta CH_4$ and $\Delta N_2O$, < 10 ppbv. On day 2 and 3 of the Shrubs trial, data were removed with the minimum wind speed filtering criteria. On day 6 of the Conventional pasture trial ~ 6 hours data, between 2:00 and 12:00 data were removed again due to low wind speeds. Data was also lost each day for 30 to 45 min when the tracer-gas canisters were exchanged. Of the possible 2016 5-min data points over 7 days, during the shrubs trial 1218 (60%) 5-min $CH_4$ emissions were calculated and during the conventional pasture trial 1339 (66%) emissions were calculated (see Table 3).

$CH_4$ emissions (5-min temporal resolution) were calculated from the mixing-ratio data according to Equation 5. These data were reduced to an hourly average (Figure 3 and 4), with the uncertainty given as 1 standard error of the mean (n = 4 to 12). An hourly average was only accepted when the number of data points > 4 (maximum possible 12 points).

Emission strengths vary over the day, and while there isn't a clear pattern in emissions with time-of-day, with sheep grazing the shrubs and inter-rows, maximum emissions are typically ~ 18:00 and increasing again in the early morning ~ 6:00. Emissions were comparable over the 7 days of measurements from the shrub treatment, with limited variation in emissions as available biomass reduced in the experimental area. The biomass remaining at the end of the 7 days was dominated by species with high anti-methanogenic potential. With emissions from the sheep grazing the conventional pasture, (hay and oats supplied ~ 10:00) any pattern with time-of-day is less clear, but emissions do appear to increase in the afternoon, after 12:00 and into the evening, with lowest emissions in the early hours of the morning.

Daily $CH_4$ emissions (average of hourly emissions) measured from the sheep over 7 days from the two treatments are detailed in Table 3. Prior to calculating the average, missing hourly data were replaced using a cubic spline interpolation, with time (Igor Pro V6.3, WaveMetrics, Oregon USA; pre-averaging =100). The uncertainty is the standard error (stderr) of the average.

Emissions over the 7 days of the measurement trial (Table 4) is the average of the daily emissions, with the uncertainty as 1 stderr of the average (n=7). The $CH_4$ emissions, as g-$CH_4$/head/day, measured from sheep grazing the shrubs plus inter-rows were less than that measured from the sheep grazing the conventional pasture, with the difference being significant, based on the standard error in the two values. The difference in emissions intensity (Table 4), as measured against live weight gain (loss) or total ME intake, is increased with the lower loss of weight and associated higher intake for the sheep on shrubs, and also significant.

Table 3. Daily $CH_4$ emissions from sheep grazing conventional pasture and shrubs and inter-rows chosen for anti-methanogenic potential. The uncertainty is 1 standard error (stderr) of the average. Average (g/head/day) is the average emissions over the 7 days of measurement, with the uncertainty as 1 stderr of the average

| Day No | Conventional Pasture | | Shrubs and Inter-rows | |
|---|---|---|---|---|
| | Daily Emissions g/head/day | Number points | Daily Emissions g/head/day | Number points |
| 1 | 24.6±2.2 | 168 | 17.1±0.9 | 261 |
| 2 | 14.7±1.5 | 152 | 16.6±1.3 | 101 |
| 3 | 15.4±1.2 | 252 | 16.1±1.5 | 165 |
| 4 | 22.0±1.3 | 246 | 16.3±1.4 | 200 |
| 5 | 17.7±1.6 | 169 | 17.7±1.4 | 249 |
| 6 | 22.3±1.2 | 183 | 15.2±1.2 | 194 |
| 7 | 21.6±2.5 | 169 | 19.5±1.7 | 48 |

Table 4 Average Daily Methane Emissions from 40 sheep grazing the Pasture Treatment and Shrubs Treatment. The stderr of the emissions intensity is based on the relative error in the $CH_4$ emissions estimate.

| Treatment | No Days | Emissions $CH_4$ g/head/day | * Methane intensity ($g\text{-}CH_4/g\text{-}LWG$) | $CH_4$/ME intake ($g\text{-}CH_4/MJ/kgDM$) |
|---|---|---|---|---|
| Pasture | 7 | 19.8±1.4 | -0.165±0.012 | 3.13±0.23 |
| Shrubs | 7 | 16.9±0.5 | -0.741±0.023 | 2.53±0.078 |

* As LWG was negative, the more negative number represents the lower $CH_4$ intensity

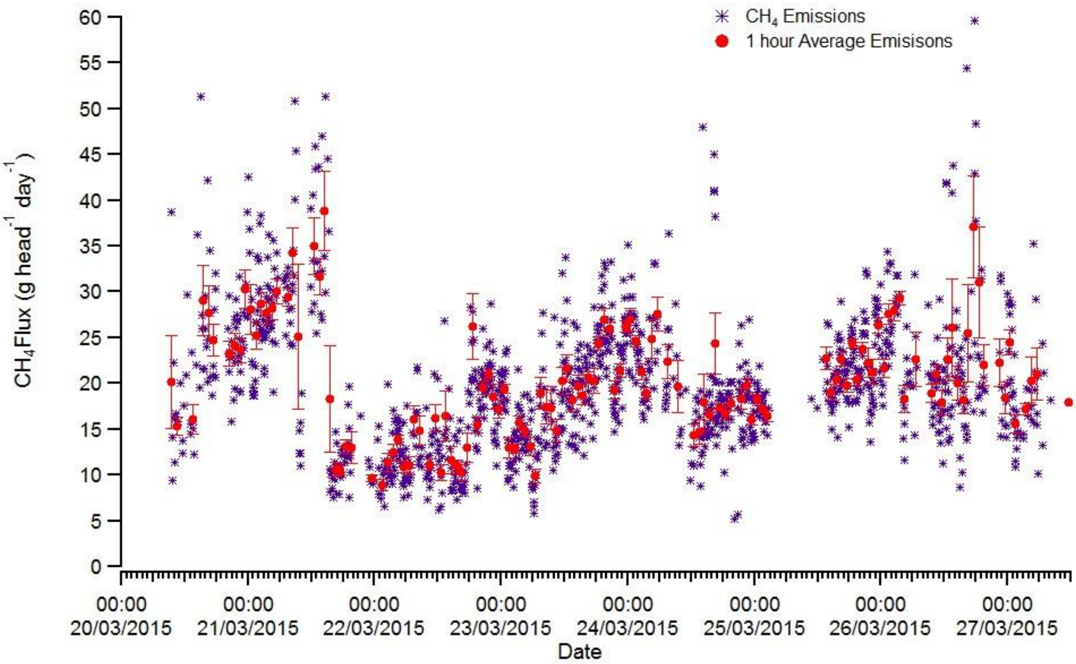


Figure 3. $CH_4$ emissions measured from 40 sheep grazing conventional pasture, and the 1 hour average of those emissions. Emissions were measured by OP-FTIR. The error bars are 1 standard error of the average.

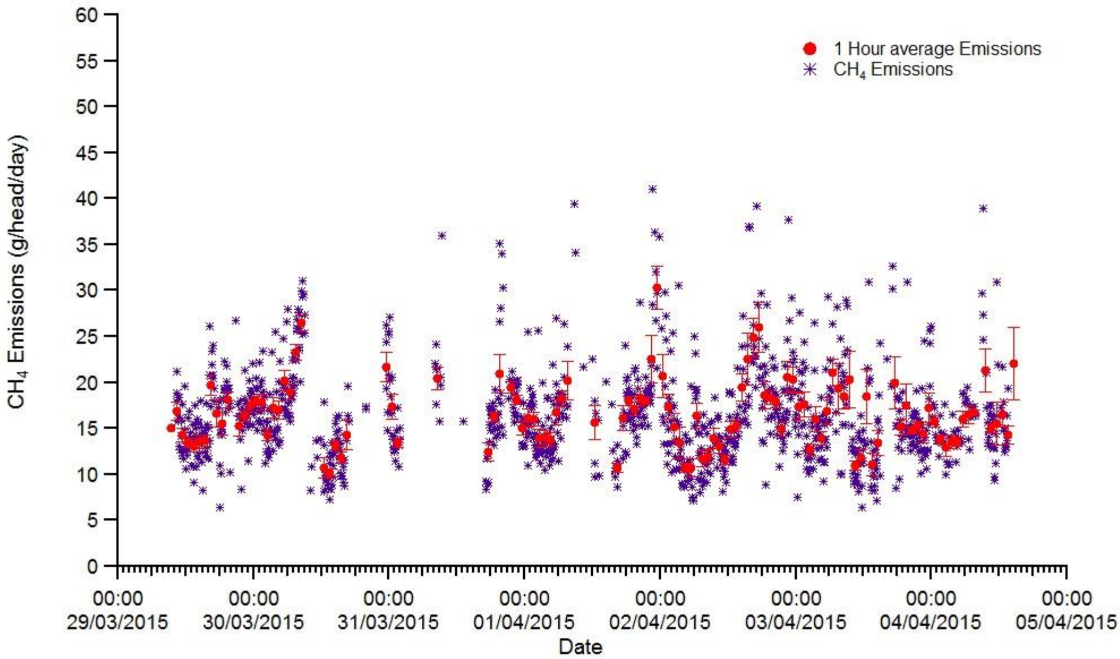


Figure 4. $CH_4$ emissions measured from 40 sheep grazing shrubs and inter-rows, chosen for anti-methanogenic potential and the 1 hour average of those emissions. Emissions were measured by OP-FTIR. The error bars are 1 standard error of the average.

## 3.4 Conclusion

$CH_4$ Emissions were measured from 2 groups of sheep (40 per group) grazing conventional pasture and shrubs plus inter-rows chosen for anti-methanogenic potential using open-path FTIR spectroscopy and a tracer-gas ($N_2O$). Emissions were measured continuously over 7 days, while the sheep were free grazing.

Some data were removed from the analysis due to poor micrometeorological conditions (wind speed < 0.5 m/s or increase in levels of $CH_4$ and $N_2O$ < 10 ppbv) with 66% of the data retained from the conventional pasture and 60% from the shrubs measurements.

With the sheep grazing shrubs plus inter-row species there was some indication of increased emissions in the evening and again in the early morning. Emissions from sheep grazing conventional pasture were highest in the afternoon to evening and lowest in the early morning.

Daily $CH_4$ emissions from the sheep grazing conventional pasture ranged from 14.4±1.2 to 24.6±2.2 g/head/day, and averaged 19.8±1.4 g/head/day over the 7 days. Emissions from the sheep grazing shrubs plus inter-rows ranged from 15.2±1.2 to 19.5±1.7 g/head/day and averaged 16.9±0.5 g/head/day over the 7 days. The emissions intensity, based on total ME intake from the conventional pasture was 3.13±0.23/ g-$CH_4$/MJ/kgDM and 2.53±0.078 g-$CH_4$/MJ/kgDM from the shrubs and inter-rows species.

# 4. Part C. Preliminary investigation of shrub-based pasture using a livestock systems model

## 4.1 Methodology

The paddock-scale studies undertaken in this project provide the data that enables us to investigate the outcome of establishing a shrub-based pasture within a traditional livestock enterprise on a mixed farm. We used GrassGro biophysical modelling software (version 3.3.3.) to simulate a typical self-replacing Merino enterprise within a mixed crop and livestock farm in order to compare systems with (Shrub) and without (Control) access to a shrub based pasture. The feedbase of the Control system comprised a subterranean clover and annual ryegrass-based simulated pasture, combined with simulated crop stubble available during December and January. The feedbase of the Shrub system was identical, except that livestock were rotated into a 'Shrub paddock', where feed equivalent to that of the shrub system in this project was supplied. Due to limitations in the existing plant models in GrassGro, the stubble and shrub pastures were simulated using supplementary feeding components that had a metabolisable energy and protein concentration similar to experimental values from the two experiments in this project (for shrubs) and another livestock model for the stubbles (Thomas *et al*, 2012).

The GrassGro model is a grazing systems model, comprised of components that each describe a portion of the biophysical (climate, soils and land management units (paddocks), pastures, livestock), managerial (e.g., stocking rate, soil fertility, pasture grazing rotations and animal reproductive management) and financial subsystems, which form the 'farm system' under consideration. These components combine to simulate biophysical and economic performance within the farm system at daily time steps for the chosen time interval (years), and from this data output summaries are generated using reporting templates, which can be customised. The simulation experiment was based on the Pingelly location in Western Australia (32°30'S, 117°00'E). The farming enterprises in this region are characterised by a mixture of predominantly annual pastures and cropping. Models were built for two farm systems:

1. "Control", without the establishment of shrub system.

2. "Shrub", feedbase the same as the control except for the inclusion of a simulated shrub-based pasture. No additional reduction in methane associated with shrub-based forage was assumed in the model.

These model scenarios were mainly based on production and management parameters for a mixed cropping and self-replacing Merino sheep enterprise in the UWA Future Farm, Pingelly in Western Australia. All simulations were run over the years 1984–2013 using historical weather data for the experimental location obtained as Patched Point Datasets from the SILO database. The "Control" farm scenario was set up in GrassGro with 2 paddocks available for grazing, a 850 ha annual pasture paddock and another paddock to simulate a crop stubble on the farm suitable for feeding the sheep during December and January. The "Shrub" farm system was selected for 3 paddocks, 850 ha of annual pasture, a paddock simulating crop stubble available for grazing during December and January, and another paddock simulating 150 ha of shrubs available for grazing during March based on field data. Stocking rate for the two farm models was 8 ewes per ha winter-grazed pasture area for both Control and Shrubs scenarios. Winter-grazed pasture area refers to the area of the farm that is retained for grazing (not cropped) during the winter/spring growing season. A combination of annual ryegrass and subterranean clover Dalkeith was selected from the plant component for annual pastures.

The GrassGro simulations paddocks were assigned a similar sandy soil type (Binnu-Uc5.22). The simulated paddocks were based on the “Medium Merino” breed. The weight of a mature ewe in average condition was set at 50 kg, with an annual greasy fleece production of 5.0 kg and average fibre diameter 20.0 µm. Ewe lambing occurred on 1 May each year and lambs were weaned on 13 July. Lambs were fed a ‘production supplement’ of 70:30 oats crushed and lupin grain ration as required to reach a marketable weight of 45 kg by 1 November, and were sold between 7 September and 1 November when they reached the required weight. Flock ewes were fed a grain ration consisting of 30% lupins and 70% crushed oats in feedlot paddocks to maintain their body condition, when the condition of the lightest animals reached CS 1. The low threshold was selected in order to reduced the very high condition that sheep were being maintained at, on average, when the simulation was being tested. Mature ewes were sold on 1 December when they reached age 5-6 years. The shearing date for all mature sheep was on 1 December and weaners were shorn on 29 October each year.

## 4.2 Results

In the Control simulation, the body condition of the animals tracked the seasonal supply of feed and was influenced by the reproductive cycle as would be expected for the highly Mediterranean-type climate in south-western Western Australia (Figure 1). By comparison, the relatively high feeding value during March for the Shrub scenario produced an increase in body condition during March, and higher body condition score values thereafter in Autumn and early Winter (Figure 2).

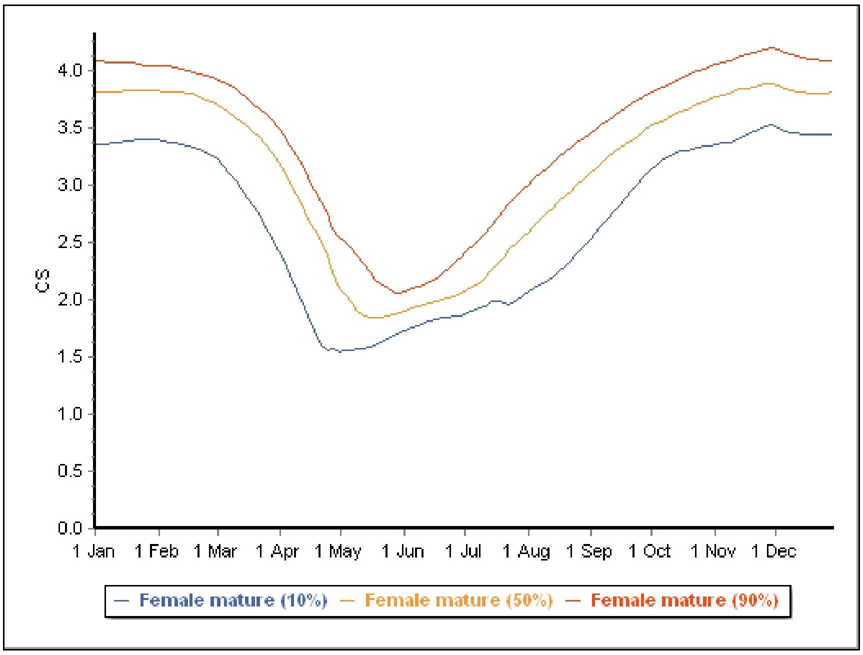


Figure 1. The seasonal condition score of adult ewes for 10% (blue), 50% (orange) and 90% (red) decile seasons for the simulation scenario with only annual pasture feedbase components.

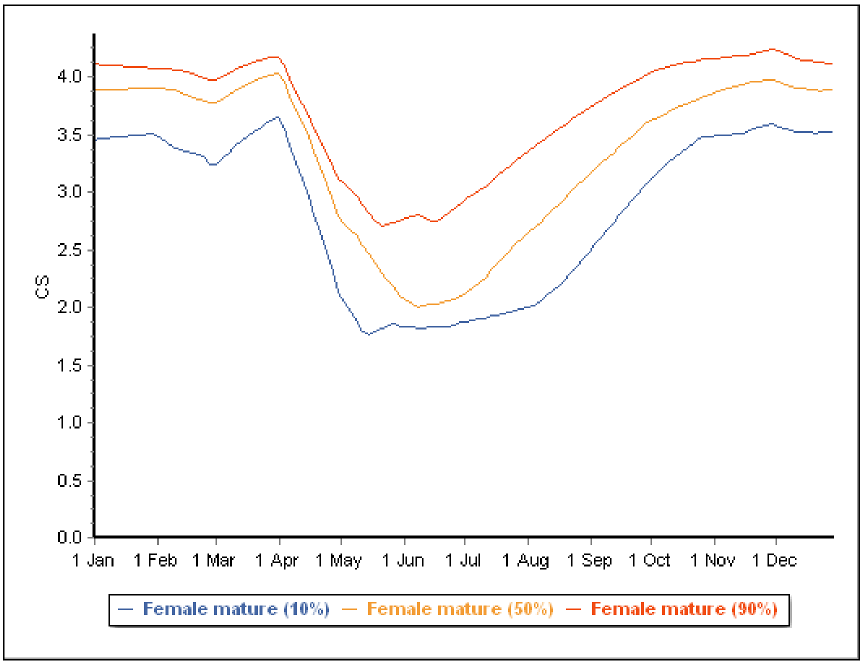


Figure 2. The seasonal condition score of adult ewes for 10% (blue), 50% (orange) and 90% (red) decile seasons when a shrub-based pasture was added to the annual feedbase.

The inclusion of shrubs in the feedbase decreased supplement fed to sheep to almost one third, from 94 to 36 kg/ha each year (Figure 3). Furthermore, the proportion of years where supplementation was no longer required increased from 13% to 43%. This sharp decrease in supplementation was both due to the increase body condition score when sheep grazed shrubs, so the sheep were subsequently less likely to reach a threshold where supplement was needed, but also the deferment of dry annual pasture so it was available during later autumn and early winter. The cost of supplementary feeding decreased by $61/ha.

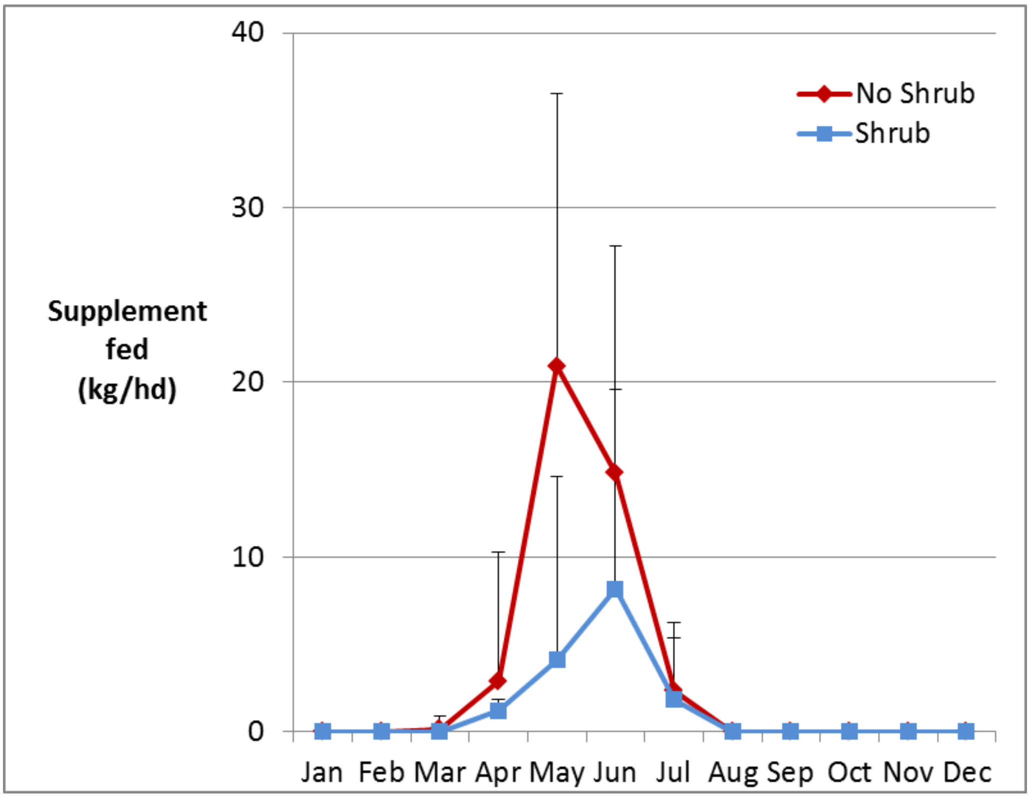


Figure 3. Monthly supplement fed (kg/hd) to Merino ewes with or without access to shrub-based pasture.

Consistent with the field data, the total methane emissions increased when ewes were grazing shrubs (Figure 4) but emissions intensity decreased by about 30% (2.8 v 2.0 g $CH_4$/MJ ME; Figure 5). There appears to be a reversal of these results over subsequent months, which was likely due to the Shrub group eating more of the dry annual pasture that was deferred, whereas the control group were generally being fed a grain-based supplement so had higher overall emissions but lower emissions intensity during this period (higher nutritive value improves emissions intensity).

Cumulative probability curves for gross margin ($/ha) in both Shrub and Control scenarios are presented in Figure 6. The For the simulated location, Pingelly, Western Australia, economic gains from establishing are predicted to exist in both good and poor seasons.

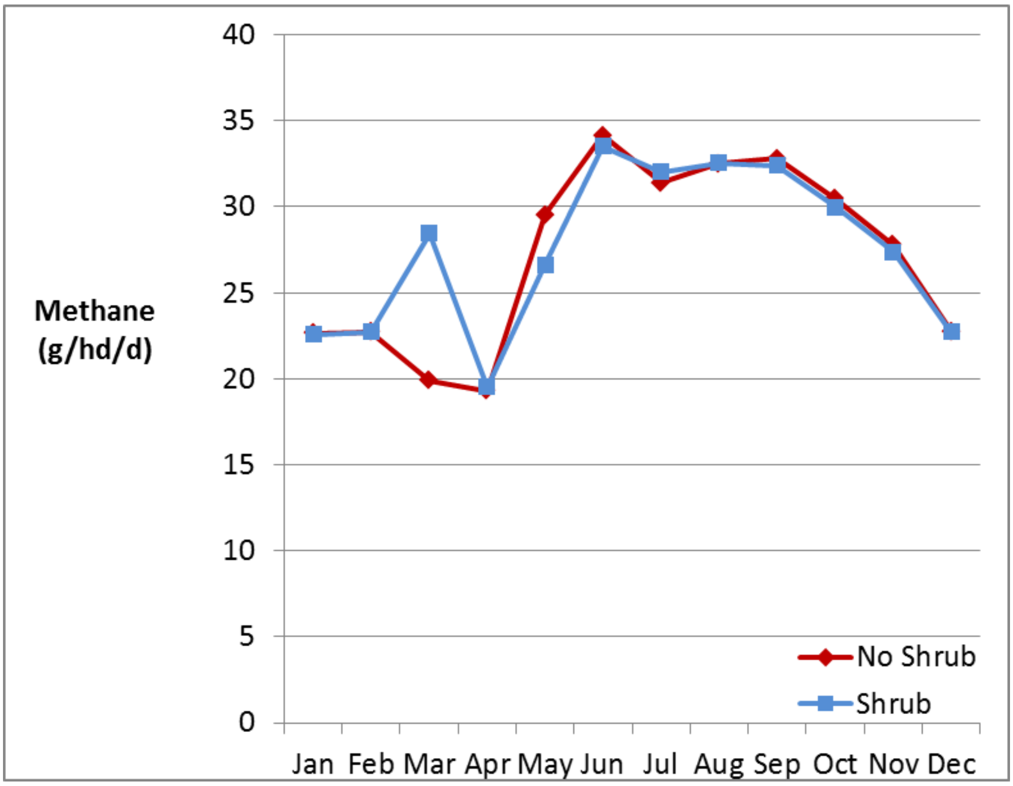


Figure 4. Methane emissions (g/hd/d) by Merino ewes with or without access to shrub-based pasture.

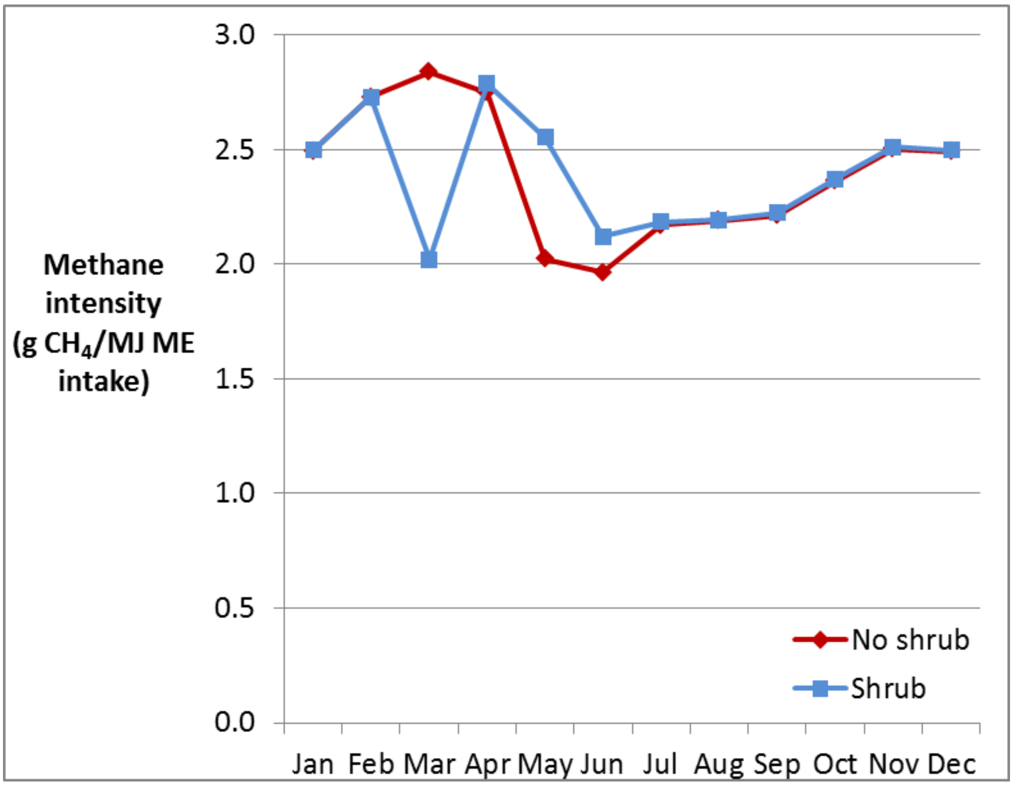


Figure 5. Methane intensity (g $CH_4$/MJ ME intake) by Merino ewes with or without access to shrub-based pasture.

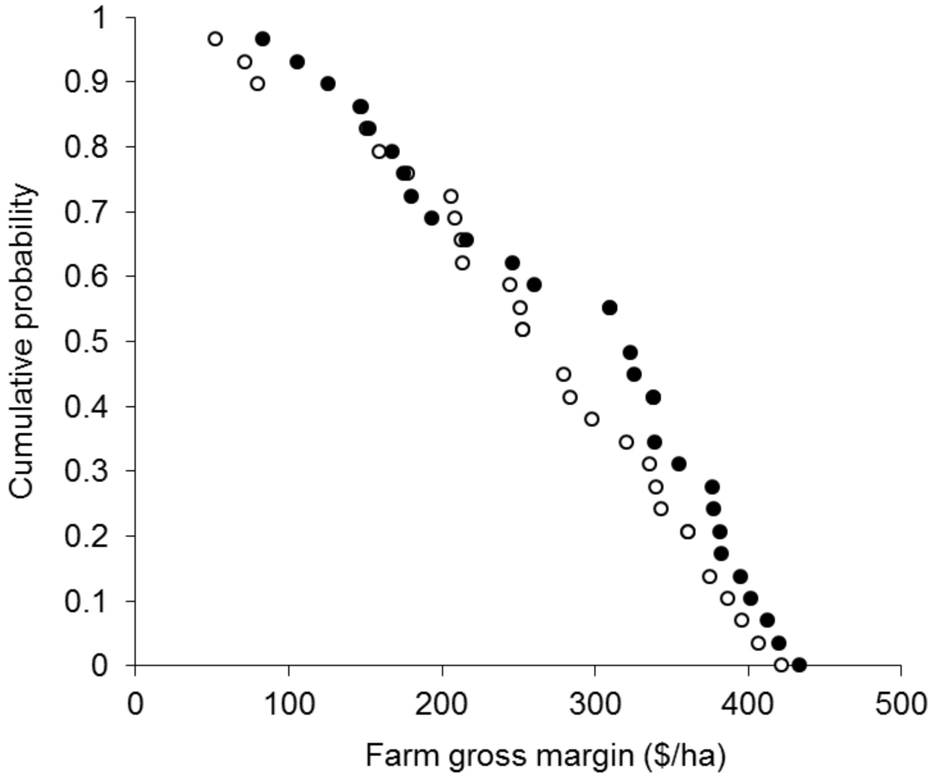


Figure 6. Simulated cumulative distribution for farm gross margin ($/ha) with (closed symbol) or without (open symbol) shrubs included as a component of the farm feedbase. Each curve is composed of 30 seasons.

## 4.3 Conclusion

Based on the field data, the simulation modelling indicates that having access to shrub based pastures at the time when they were grazed in the field experiments has substantial benefits for seasonal feed supply and associated reductions in supplementary feeding. However, we did not find substantial evidence of methane mitigation in our modelling scenarios, in which no antimethanogenic effects of shrubs were assumed. While methane emissions intensity decreased during the period when the shrubs were grazed, the higher feeding of grain-based supplements in the Control scenario resulted in relatively higher emissions intensity in the Shrub system in the period after the shrubs were grazed. However, our modelling considered only one potential scenario for feedbase management and it may be possible to design feeding systems where overall methane emissions intensity is reduced, such as longer intervals of shrub grazing or use of dual-purpose crops. The study demonstrates the benefits of high feeding value options to reduce methane emissions intensity in livestock systems with a Mediterranean-type climate.

# 5. General discussion

The provision of native shrubs, with or without bioactive properties, together with inter-row pasture species with anti-methanogenic properties increased animal performance and reduced methane production compared to ‘business as usual’ (BAU) management systems during the ‘feed gap’ period in autumn. Although we hypothesised that methane production would be reduced when bioactive shrubs were incorporated into a shrub grazing system, there was no significant improvement compared to shrub species selected for their biomass production. It was evident, however, a feeding system that allows animals to gain weight, at a time of year when it is normally difficult to achieve, will have benefits to methane emissions intensity.

The improvements in methane intensity in both years of grazing trials were primarily a result of improved liveweight gain. In 2015, the sheep grazing shrubs achieved liveweight gains that were more than double the gains achieved by the best performing sheep grazing pasture with grain supplementation, and they had a lower methane intensity. There was a linear effect between the three pasture-based grazing systems where methane intensity decreased as liveweight gain increased. In 2014, a similar outcome occurred where sheep grazing shrubs had twice the weight gain of sheep grazing a pasture system. Other studies have also demonstrated that the addition of shrubs to the feed base can be productive under commercial conditions and, in southern Australia, offer a particularly attractive option for areas of land that are not economically suitable for cropping (Revell *et al.* 2013). Modelling predictions using data from the current autumn grazing trials also support the economic and productive benefits to a whole-farm system, over a 12 month period.

The results from our study lend support to this concept of perennial shrubs benefiting the whole-farm system. A producer could obtain extra liveweight gain in livestock by grain supplementation, but the advantage of using a shrub-pasture system instead is that it reduces labour and direct costs of supplementary feed each year. Implementing a shrub-based system, on the other hand, does incur a single, up-front cost of establishment, which can be substantial, but it can be amortised over 15 to 20 years.

In addition to improving liveweight gain of sheep during autumn, shrub grazing systems were associated with less methane emission per unit of ME intake, regardless of the measurement system, implying a greater efficiency of production. This suggests that the shrub grazing systems played some role in altering rumen function and enteric methane production. These findings, in combination with the direct effects of shrubs on livestock nutrition and the additional benefits that shrubs can have in a whole-farm system, for example the productive use of otherwise marginal soils, reducing the risk of wind erosion, and provision of shelter for livestock, offer a strong case for the inclusion of shrubs in farming systems.

It would be worthwhile to investigate the influence of shrub-based systems on wool growth and, consequently, methane emissions per unit of wool produced. It is well known that wool growth is low in autumn in southern Australia (Robards 1978). Many producers try to reduce the impact of low quality pasture on wool growth with supplemental feeding, usually with grain. Shrubs could provide a valuable source of crude protein (nitrogen) and sulphur, which are typically the limiting nutrients for wool growth in autumn. Adding this extra performance indicator to our emissions intensity calculation could help build an even stronger case for the use of shrubs in a whole-farm system that is designed to improve livestock performance and efficiency.

At the start of this project, we expected that bioactive shrub species would provide an anti-methanogenic effect and improve methane production. The results from the *in vitro* fermentation analysis supported previous work on the fermentative traits of these species (Durmic *et al.* 2010), where lower methane production was observed as a result of plant chemistry. The clear difference in methane production between the bioactive shrubs species and the productive shrub species was not translated to the *in vivo* grazing trials. It is possible that because the shrubs were a relatively small component of the total intake by the sheep, any anti-methanogenic effects due to plant chemistry were obscured by the large intake of inter-row pasture. It is possible that we may have seen a greater difference between the two shrub treatments had the biserrula been more productive and a greater proportion of inter-row biomass. There is likely to be different compounds responsible for the antimethanogenic effects in the shrubs and biserrula that reduce methane through different targets. There could be complementarity between the range of compounds and targets that could increase the degree of antimethanogenic activity as well as the persistence of the effect. The diluting effect of the inter-row pasture could perhaps have been ameliorated if the biserrula that was sown in the inter-row had been more productive. However, although there were no significant differences between the shrub treatments, the methane production per unit of ME intake was 5% lower with sheep grazing the bioactive shrubs than those grazing productive shrubs, which could tentatively suggest an anti-methanogenic effect of the bioactive shrubs, or, alternatively, a slight methanogenic effect associated with *Atriplex* species in the productive shrub system (Mayberry *et al.* 2009). The *in vitro* results presented here indicate that the methanogenic potential in the Atriplex are higher than other shrub species and that the average methanogenic potential of the productive shrubs is at least numerically higher than that of the cohort of bioactive shrubs. In some shrub-based systems, the proportion of shrub intake in the sheep's diet may be greater than 50% (Norman *et al.* 2010), which is much higher than the current experiments. Therefore, in an overall farming system perspective, if a producer wanted to improve liveweight gain in autumn by using a shrub-based system and avoid any possible elevation in methane from their livestock, they should select plants that have demonstrated bioactive properties and avoid using shrubs from the 'productive' category.

This study also highlighted the suitability of using the polytunnel and the Portable Accumulation Chamber (PAC) techniques to measure methane of grazing animals in the field. Although the polytunnel method measures the production of methane by a group of sheep, rather than an individual animal's emissions, we found similar patterns of methane production by sheep grazing shrubs versus pasture when measured with the polytunnel or with the PACs. Likewise the OPFTIR technique yielded similar results, with the sheep grazing shrubs producing less methane than sheep grazing a pasture-only system (Part B, Table 4).

The standard error of measurements for the PACs and polytunnel were similar, but slightly higher in the PACs. If we compare the estimates of methane production per predicted MJ ME consumed between the three approaches averaged for pasture and shrub treatments, the ratio of shrub to pasture estimates are similar (74% polytunnel, 72% PAC and 80% OPFTIR). The average emissions intensity for pastures compared with shrub treatments estimated using the polytunnel and the PACS were consistent in the sense that the shrubs had a lower emissions intensity. However, the PAC estimate of emission intensity for the pasture treatments was double that of the polytunnel estimate. This is probably due, in part, because it is an average across different levels of grain fed in the pasture treatments in experiment 2 compared with experiment 1 where only one level of grain was fed. All animals grew at similar rates in the pasture treatments in experiment 1, whereas the animals grew at different rates in experiment 2, which affects the emission intensity estimates. If we restrict our comparison to the Pasture + grain 180 g treatment in experiment 2 where the liveweight gains were similar to the average of the pasture treatments in experiment 1, the emissions intensity were similar (0.18 v 0.14). The emissions intensity estimates for the sheep grazing the shrub treatments were identical for both measurement systems. It is difficult to compare the emissions intensity for the OPFTIR with the other two methods because of the weight loss over the 7 days the measurements were taken.

Regardless of the type of shrubs that are incorporated into a farming system, there is evidence to justify that native shrubs can be beneficial. The results from this study, could lend support to the concept of bioactive shrubs becoming a part of a CFI methodology. Since both shrub-based systems investigated in this study improved liveweight gains of sheep, the inclusion of bioactive shrubs in a grazing system would be a distinct management decision with the end goal being to hopefully improve emissions intensity.

*Value of research to farmers*

This results from this research has demonstrated that native shrubs can improve animal productivity by offering an alternative to supplemental feeding with grain during autumn. Sheep that grazed a shrub-based system achieved greater liveweight gains than sheep that grazed conventional pasture with grain. This research is also encouraging for farmers who not only want to improve productivity, but are conscious of the amount of methane produced by their enterprise because a feeding system that allows animals to gain weight, at a time of year when it is normally difficult to achieve, will have benefits to methane emissions intensity. The modelling aspect of this research also demonstrated that the inclusion of shrubs in a grazing system reduces the yearly costs associated with traditional supplementary feeding practises. The animals that grazed shrubs had a better body condition and were less likely to reach a threshold where supplement was needed. Also, the grazing of dry annual pasture can be deferred so it is available during later autumn and early winter. Importantly, the methane predictions from the modelling were based solely on efficiency of feed use with no direct effect of the 'shrub' intake on methane. There is evidence that there are direct effects of shrubs and biserrula on methane production and so the results from our modelling have only captured part of the potential benefits.

# 6. Future research needs

One of the gaps that needs to be addressed, and builds on this study, is to examine the emissions and emissions intensity of the annual grazing cycle. Grazing is a year round activity and in this project we have only examined the value of a shrub and inter-row system during the summer autumn feed gap. Normally the shrubs and inter-row would have a second grazing during the winter and spring. Importantly, the inter-row pasture would be green and most productive at this time and the impact of the biserrula and subclover mix may have a significant impact on methane production during this time. The antimethanogenic effect of biserrula alone is greater when it is fresh compared to when it has dried off and senesced. For the remainder of the year, animals graze pastures where there are no shrubs, and these pastures could be based on antimethanogenic (eg. biserrula) and/or more productive species (eg. high biomass production and hold nutritive value for longer), and managed to reduce emissions. The overall improvement in annual emissions with this system in place would be greater than the small, but critical, window of the year we have examined in this project, but it needs to be tested.

There are a number of modelling projects that could be undertaken. An obvious one is an exercise that best captures both the direct and indirect effects of a system designed around shrub and antimethanogenic inter-row combined with innovative pastures and grazing systems on the rest of the farm. The issue of methane intensity as a function of both liveweight gain and wool growth could be considered with both a field-based experiment and incorporated into GrassGro modelling analysis. Shrubs could provide a valuable source of crude protein (nitrogen) and sulphur that are typically the limiting nutrients for wool growth in autumn. It would be beneficial to understand how shrub-pasture systems influence wool production over different seasons. It would also be interesting to gain a better understanding of how prolonged the effect of shrubs is, and therefore how long a shrub-based system would need to be grazed before there is any evident change in production.

Nutritive value analysis using NIR is efficient and effective, yet the current NIR database for shrubs is limited. It would be beneficial to build the shrub database so that there are sufficient standards that can be used to analyse shrubs using this technique.

There is evidence to suggest that some shrub species can improve aspects of animal health, such as provide particular vitamins or minimise worm burdens. Little is known about the health impacts a shrub-pasture system can have on livestock, which may also improve productivity and production efficiency because healthy animals are usually considered more productive. It would be beneficial to understand more about the health status of sheep when they graze such a system and the follow-on impacts this has on overall productivity. At the same time, measurements of the full value of shrubs including below and above ground biomass, shade and shelter, nutrient and water flow in those systems would add to our picture of the overall benefit shrubs can have on productivity but also on carbon balance.

# 7. Publications

One paper that is directly related to this project has been published in a peer reviewed journal and was presented at the Recent Advances in Animal Nutrition conference. We anticipate further publications from both grazing experiments in this project.

Revell D. K., Norman, H. C., Vercoe, P. E., Phillips, N, Toovey, A., Bickell, S., Hulm, E., Hughes, S. and Emms, J (2013) Australian perennial shrub species add value to the feed base of grazing livestock in low- to medium-rainfall zones. Animal Production Science 53:1221-1230.

N. Phillips, A.F. Toovey, S. Bickell, P.E. Vercoe and D.K. Revell (2014) Technical note: A method to measure methane production of sheep in the field using a polyvinyl inflatable tunnel (polytunnel). Journal of Animal Science (requires revision to meet page restrictions for a technical note).

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# 9. Appendix:

## 9.1 Institute of Agriculture Newsletter - http://www.ioa.uwa.edu.au/publications/newsletters

## 9.2 Press release in Narrogin Observer

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### UWA Pingelly farm research ‘impressive’

■ Lisa Morrison

Minister for Agriculture Ken Baston has described scientific research being carried out on a Pingelly farm as “impressive” and “meaningful for the future of the agriculture industry”.

UWA scientists led Mr Baston on a tour of the 1600ha Ridgefield Future Farm property, located next to the Boyagin Nature Reserve in the Shire of Pingelly, last Friday afternoon to brief the minister on their plans.

The Future Farm 2050 project is a testing site for developing best-practice agriculture methods. Plans include conservation cropping, ethical animal production, ecosystem restoration, as well as carbon and water management systems.

Mr Baston said he was pleased local farmers could see researchers directing science into a profitable and practical agricultural enterprise on the property.

“I have always believed research needs to be relevant to farmers’ needs, and for that, trials need to be conducted over a longer time frame, rather than on a year-to-year basis,” Mr Baston said.

“UWA is able to achieve that through this farm.

“The work at Ridgefield not only involves other disciplines from the university, such as architecture and life sciences, but also the local community.

“It is important local growers and the wider community benefit from this investment in their local area, and also have the opportunity to share knowledge with scientists to ensure research stays relevant to the industry’s needs.”

Mr Baston said a major benefit of the project was hosting students from metropolitan schools in an effort to “bridge the country-city divide”.

“One of the major benefits is that the next generation of scientists is getting exposure to agriculture,” he said.

“Engaging with students from urban schools and bringing them down to Pingelly demonstrates that agriculture is not just about farming, but also about science at a high level.

“Additionally, what I heard from local farmers today, particularly in reference to fodder shrub research, is they were constrained because they simply did not have the time or money to push further.

“What UWA has been able to offer is the time and resources needed to extend research and help growers get results on the ground sooner.”

**CSIRO livestock researcher Dean Revell, UWA school of animal biology Associate Professor Dr Phil Vercoe, Agriculture Minister Ken Baston and Pingelly farmer Gary Page tour Ridgefield Future Farm on Friday.**
Picture: David Stacey