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Review

Harnessing plant bioactivity for enteric methane mitigation in Australia

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Abstract. This review provides examples of the utilisation of plant bioactivity to mitigate enteric methane (CH_4) emissions from the Australian ruminant production systems. Potential plant-based mitigation strategies that reduce CH_4 without major impacts on forage digestibility include the following: (i) low methanogenic tropical and temperate grass, legume and shrub forage species, which offer renewable and sustainable solutions and are easy to adopt, but may have restricted geographical distribution or relatively high costs of establishment and maintenance; (ii) plant-based agricultural by-products including grape marc, olive leaves and fruit, and distiller's grains that can mitigate CH_4 and provide relatively cheap high-nutrient supplements, while offsetting the impact of agricultural waste, but their use may be limited due to unfavourable characteristics such as high protein and water content or cost of transport; (iii) plant extracts, essential oils and pure compounds that are abundant in Australian flora and offer exciting opportunities on the basis of *in vitro* findings, but require verification in ruminant production systems. The greatest CH_4 mitigation potential based on *in vitro* assays come from the Australian shrubs *Eremophila* species, *Jasminum didymium* and *Lotus australis* (>80% CH_4 reduction), tropical forages *Desmanthus leptophyllus*, *Hetropogon contortus* and *Leucaena leucocephala* (~40% CH_4 reduction), temperate forages *Biserrula pelecinus* (70–90% CH_4 reduction), perennial ryegrass and white clover (~20% CH_4 reduction), and plant extracts or essential oils from *Melaleuca ericifolia*, *B. pelecinus* and *Leptospermum petersonii* (up to 80% CH_4 reduction). Further research is required to confirm effectiveness of these plant-based strategies *in vivo*, determine optimal doses, practical modes of delivery to livestock, analyse benefit–cost ratios and develop pathways to adoption.

Keywords: methane, mitigation, rumen, forages, plant-based feed additives, plant bioactive compounds.

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Introduction

Ruminants can consume and digest large amounts of plant material and convert it into high-quality products such as meat, milk and wool, by virtue of microbial fermentation in their gut, where volatile fatty acids (VFA) and methane (CH_4) are produced as end products of the fermentation. While VFA present a major energy source for the animal, the CH_4 serves as the main pathway of eliminating hydrogen in the rumen (Beijer 1952). In the past century, enteric CH_4 has been regarded simply as a loss of energy from ingested feed, but in more recent times, it has become of concern as a major greenhouse gas (GHG; Johnson and Johnson 1995). About 70% of the CH_4 produced on Earth is generated from anthropogenic sources, and ruminant livestock is the single most significant contributor (Moss *et al*. 2000). Each ruminant animal produces and eructates 5–130 kg of CH_4 year, depending on its size and the amount of fibrous material consumed in the diet (Johnson and Johnson 1995). Enteric CH_4 has been estimated to contribute half of the total GHG emissions from the agricultural sector (Swainson *et al*. 2018; Dong *et al*. 2019).

Methane is a highly potent GHG, being more effective than carbon dioxide (80 times over 10–20 years, or 28 times over 100 years from release), but has a shorter atmospheric lifetime of 12 years from release, compared with either carbon dioxide at 50–200 years from release, or nitrous oxide at 120 years from release (EPA 2003). Thus, enteric CH_4 is an attractive target for reducing overall GHG production. A reduction of 10% in enteric CH_4 production could be sufficient to prevent further accumulation of CH_4 in the atmosphere (Moss *et al*. 2000). In Australia, CH_4 emissions from livestock account for 60–70% of total GHG emissions from the agricultural sector (Cottle *et al*. 2011; Taylor *et al*. 2016). The size of this contribution is partly due to the large number of ruminants; Australia is a significant player in global food supply, accounting for ~4% of global beef production (Suybeng *et al*. 2019). Australian CH_4 emissions from livestock are also significant because most ruminants are managed under extensive conditions where the animals usually graze low-quality, fibrous diets that promote relatively high CH_4 production in the rumen compared with feedlot or dairy cattle offered high-grain diets (Charmley *et al*. 2008).

Early CH_4 mitigation strategies focussed on removal of protozoa from the rumen, because a portion of the methanogenic archaea in the rumen live in close association with them (Blaxter and Czerkawski 1966; Whitelaw *et al*. 1984). Subsequently, concepts and approaches broadened in the recognition that enteric CH_4 is a significant contributor to atmospheric pollution and global warming. The outcome was global development of solutions, including manipulation of animal physiology, genetics and management of the rumen microbiome by manipulation of diet composition and the use of feed additives (reviews: Moss *et al*. 2000; Busquet *et al*. 2006; Calsamiglia *et al*. 2007; McAllister and Newbold 2008; Hristov *et al*. 2013; Patra *et al*. 2017; Beauchemin *et al*. 2020; Min *et al*. 2020).

Early attempts to reduce enteric CH_4 emissions frequently relied on synthetic chemicals and antibiotics, but often with only modest benefits (reduction by only 10% *in vivo*), that were also variable (Johnson and Johnson 1995) and mostly ineffective in grazing animals (Grainger *et al*. 2010). Emerging concerns over the impact of antibiotics in feed on human, animal and environmental health led to legislation of restrictions on their use. The result was a worldwide search for natural, safe and sustainable alternatives, with a particular focus on plants and plant products.

The primary constituents of forage plants, including soluble and insoluble carbohydrates and oils, can drive the 'methanogenic potential', namely, amount of CH_4 produced when consumed and fermented by rumen microbes, but many plant secondary compounds (PSC) can also affect methanogenesis by acting directly and specifically on methanogens, or by acting indirectly on the overall processes of fermentation in the rumen (Bodas *et al*. 2012). The aim of a CH_4 mitigation strategy is to reduce CH_4 emissions, while not reducing overall digestibility/fermentability of the feed consumed. In that respect, a reduction in the concentration of VFA in the rumen can be used as an approximation for assessing negative effects of a strategy on overall feed digestibility/fermentability.

The present paper reviews current developments and strategies for the use of plant bioactivity to reduce enteric CH_4 emissions in Australia. We focussed on mitigation approaches based on the grazing of low methanogenic forages, the feeding of plant by-products as supplements, the use of whole plant extracts, essential oils and pure PSC, and also considered some commercially available plant-based products. We first illustrated each of these mitigation strategies with some of the work done globally, and then focussed on work in Australia, critically evaluating the prospects for local success. We compared the strategies in terms of national potential for CH_4 mitigation, agronomic and animal production benefits, and barriers and limitations to their use. This structured approach allowed us to finally analyse the options and prospects for practical applications in Australia.

Grazing low-methanogenic forages

Enteric CH_4 emissions from grazing animals can be targeted by manipulating the forage they consume, because there is significant variation among plant species in their methanogenic potential. There is worldwide interest in commonly used forages, but also in region-specific and novel grazing plants. Among the mainstream forages, the most prominent candidates with low-to-moderate methanogenic potential are lotus (*Lotus corniculatus*, *L. pedunculatus*), sulla (*Hedysarum coronarium*), lucerne (*Medicago sativa*), chicory (*Cichorium intybus*), white clover (*Trifolium repens*) and red clover (*Trifolium pratense*; Tavendale *et al*. 2005; Navarro-Villa *et al*. 2011; Hammond *et al*. 2013). Tropical forages have been found to be particularly effective at reducing rumen CH_4 production, *in vitro* and *in vivo*, with a wide variety of effective fodders from diverse plant families being reported (Hariadi and Santoso 2010; Silivong *et al*. 2013; Pal *et al*. 2015). The reduction in CH_4 production in these forages was generally attributed to the presence of PSC, particularly tannins, with reductions by up to 25% (kJ/MJ gross energy intake) seen with tannin-rich shrubs and trees (Soliva *et al*. 2008; Tiemann *et al*. 2008).

Work on low-methanogenic forages has been particularly strong in Australia, because feeding systems rely so heavily on pasture-based grazing, with minimal grain supplementation. In that respect, native woody perennial plants (trees and shrubs) seem able to play an important role, especially in low-rainfall areas, while tropical forages are utilised in the tropical and subtropical regions. There is also emerging evidence that grazing ruminants in Australia have a high methanogen diversity and harbour some unique methanogen populations (Wright *et al*. 2004; Rea *et al*. 2007; McSweeney and Tomkins 2015).

The search for variability in methanogenic properties in grazing plants in Australia began with an investigation of forage shrubs when 128 Australian native forage shrubs were assessed using *in vitro* 24-h batch culture (Durmic *et al*. 2010). Several highly potent candidates were revealed, with almost half of the species tested producing less CH_4 than with oaten chaff, a common supplementary feed. One plant in particular, commonly known as tar bush (*Eremophila glabra*), reduced CH_4 production by 81%. This CH_4 mitigation effect was subsequently confirmed using a continuous *in vitro* system (Li *et al*. 2014) and *in vivo* using sheep (Li 2013, K. Lund, pers. comm.). *Eremophila* species produce abundant terpenes and flavones that are potent CH_4 inhibitors (Oskoueian *et al*. 2013), and the effect in *E. glabra* was linked to direct inhibition of methanogenic populations (Li *et al*. 2014). However, *E. glabra* had a general inhibitory effect on fermentation, with a 15% reduction in VFA concentrations when it is used as the sole substrate *in vitro* (Durmic *et al*. 2010). However, the anti-methanogenic effect of *E. glabra* is sufficiently potent so that it can be used in a mix with other forages, thus moderating negative effects while still significantly reducing CH_4 (Li *et al*. 2014). *E. glabra* is well adapted to drought and infertile soils, two critical issues in our grazing systems, and it has an advantageous mineral profile, but it may be constrained by relatively low biomass production and poor palatability compared with some mainstream pastures (Revell *et al*. 2013). Such problems are likely to respond to plant improvement, or by just ensuring it is integrated as part of a mixed forage base in grazing systems; however, further research is needed for *E. glabra* to be widely adopted in grazing systems.

Research has been conducted on the plants from the Australian tropics and subtropics. These species are particularly important because that region is home to half of Australia's beef cattle, so it is responsible for the majority of the nation's enteric CH_4 emissions (AGEIS 2017). Among those with low-to-moderate methanogenic potential are both grasses (*Andropogon gayanus*, *Brachiaria ruziziensis*, *Bothriochloa decipiens*, *Sorghum plumosum*, *Urochloa mosambicensis*) and leguminous forages (*Calliandra calothyrsus*, *Desmanthus leptophyllus*, *Gliricidia sepium*, *Stylosanthes scabra*, *Leucaena leucocephala*; Meale *et al*. 2012; Durmic *et al*. 2017; Suybeng *et al*. 2019). These species often contain tannins that can directly reduce the amount of CH_4 produced (Piñeiro-Vázquez *et al*. 2018), but these forages can also reduce CH_4 emission intensity because they improve growth rates and thus animal productivity (Taylor *et al*. 2016). There are some potential limitations, including eco-geographical constraints and some anti-nutritive or toxic PSC that impede feed intake or affect animal health (Dalzell *et al*. 2012), but as with all novel feedstuffs, it is important to complete a duty of care assessment (Revell and Revell 2006).

Concurrent with the investigations into tropical and subtropical forages was research focussed on temperate herbaceous forages. In our initial screening of 13 mainstream and alternative pasture species of southern Australia, using fermentation *in vitro*, we discovered that a legume biserrula (*Biserrula pelecinus*) produced 73% less CH_4 than did lucerne, and 90% less CH_4 than did the highest CH_4-producing species, bladder clover (*Trifolium spumosum*; Banik *et al*. 2013). Subsequently, other mainstream pasture species were investigated, and it was shown that when subterranean clover (*Trifolium subterraneum*) was fed to sheep, CH_4 production was reduced by 30% compared with feeding ryegrass (Muir *et al*. 2020). Moreover, the methanogenic potential of subterranean clover is found to be a heritable trait, so it can be manipulated by plant breeding (Kaur *et al*. 2017). Birds-foot trefoil (lotus) was also explored for its potential, and theoretical estimates suggest that it can reduce the CH_4 emission intensity for wool and prime lamb by increasing liveweight gain and fecundity (Doran-Browne *et al*. 2015). For some of the temperate forage species, the mitigation effect may be linked to their primary chemical composition and to enhancing productivity, thus reducing CH_4 emission intensity, whereas in others, such as biserrula, the effect may be linked to the presence of specific anti-methanogenic PSCs (Banik *et al*. 2016).

In addition to the mainstream species, some alternative forages that are aimed at filling seasonal feed gaps in temperate parts of Australia were also investigated. Local varieties of turnip (*Brassica rapa*), chicory or plantain (*Plantago lanceolata*) were found to produce ~25% less CH_4 (mL/g dry matter incubated) *in vitro* than did lucerne (Durmic *et al*. 2016). Feeding forage brassicas to cattle was found to reduce CH_4 yield (g CH_4/kg dry-matter intake) by 5%, and CH_4 emission intensity (g/kg energy-corrected milk) by 10% (Williams *et al*. 2016). The mechanism of these effects is largely unknown, but it is likely to be a combination of primary chemical constituents and their PSC.

During this period of exploration of plant bioactivity, it became evident that, while some variation in methanogenic potential was related to plant species, there was also within-species variation. Often due to environmental factors, the same species can differ in primary chemicals, PSC composition, or simply, in moisture, consequently resulting in differences in methanogenic potential (Durmic *et al*. 2017). As we examined a core collection of biserrula, we also demonstrated variation among cultivars, growth stages and cutting treatments that were not influenced by environmental factors (Banik *et al*. 2019).

In addition to reducing CH_4, most of the forages mentioned above presented fermentation profiles (as described by production of VFA and the acetate:propionate ratio) that were comparable or better than the respective controls (standard forages), implying that it is possible to target CH_4 without impeding microbial fermentation and thus compromising animal productivity. Figure 1 presents examples of low-methanogenic forages in Australia and their effect on CH_4, and outlines candidates that markedly reduce CH_4 production, while maintaining or even promoting VFA production.

Bioactivity in plants by-products fed as supplements

Horticulture generates huge amounts of organic waste (prunings, leaves, seeds, fruits, peels, pulp, stones) that is a loss of valuable biomass and is an environmental burden. There is a need for cost-effective, sustainable and environmentally friendly processes for the utilisation of these products. This issue is particularly important in developing countries where livestock industries are constrained by fodder shortages and the high costs of conventional feeds. Horticultural by-products often retain a high nutrient content, so they are attractive as a supplementary feed in animal production. They can also be rich in PSC (Sagar *et al*. 2018), so many of these materials have been investigated globally for their potential to modulate rumen fermentation and mitigate enteric CH_4 production (McGinn *et al*. 2009; Benchaar *et al*. 2013; Castillo-Lopez *et al*. 2017). The effects are mainly ascribed to increased intakes of fat and high-quality protein (Moate *et al*. 2011), or in some cases tannins or other PSC.

This concept is also relevant to Australia, where ~1500 kT of fruit and vegetable biomass is wasted each year during production, processing and packing stages, or is simply lost as food waste (ARCADIS 2019). The wine industry generates a by-product, grape marc, that is high in protein, fat, fibre and other nutrients and has been used as a feed supplement for cattle. It contains condensed tannins, which, in turn, can reduce enteric CH_4 production when fed to ruminants (Goel and Makkar 2012).

By-products can contain a substantial amount of crude protein, increasing the excretion of ammonia, or products

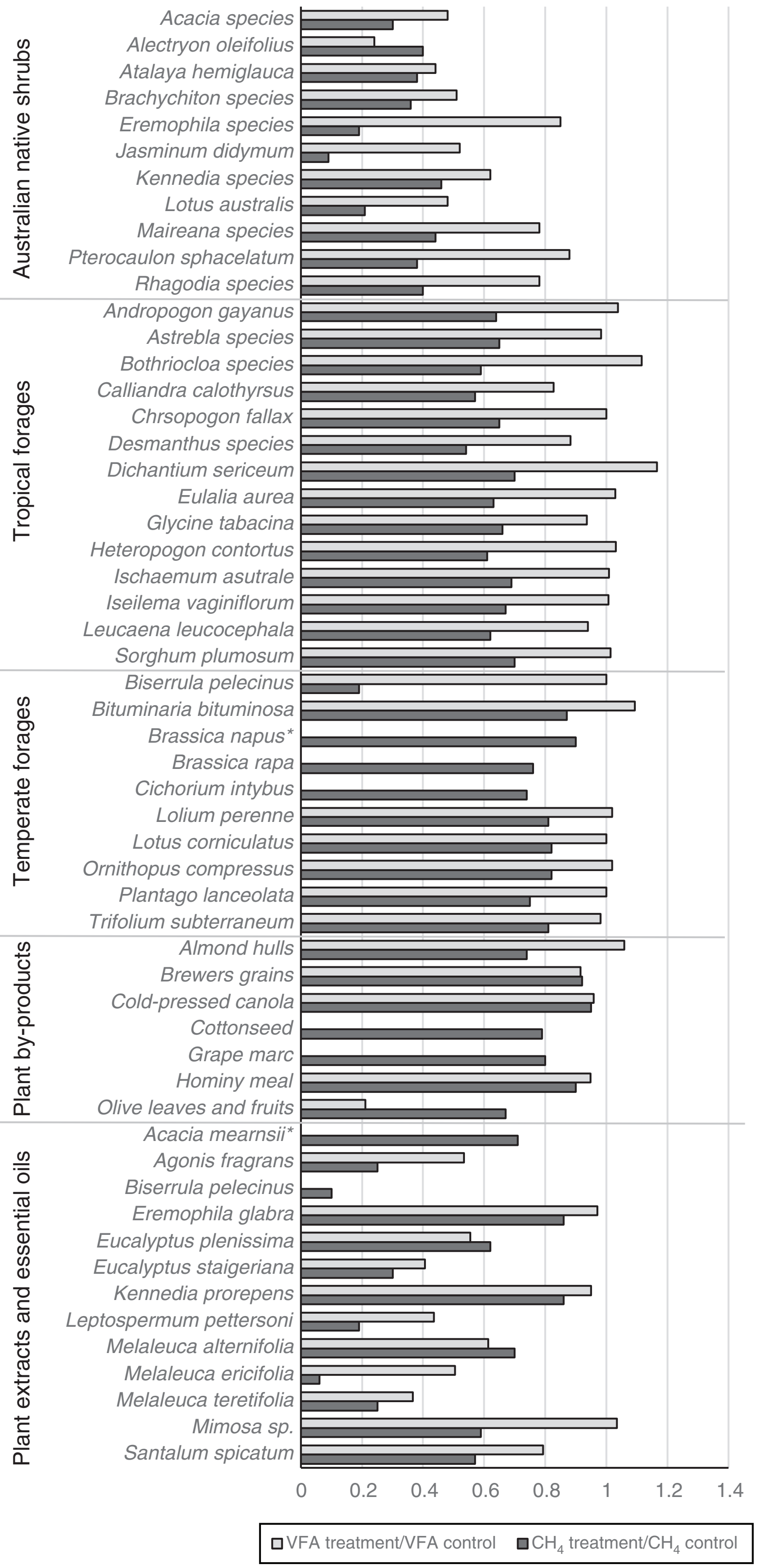

Fig. 1. The VFA and CH_4 values with different sources of plant bioactive compounds when fermented *in vitro* or *in vivo* (marked with *) by rumen microbes and compared with a control forage or diet used in the system. Data generated from: Durmic *et al*. (2010) (native shrubs); Durmic *et al*. (2017) (tropical forages); Banik *et al*. (2013), Durmic *et al*. (2016) and Williams *et al*. (2016) (temperate forages); Durmic *et al*. (2014), Shakeri *et al*. (2017), Moate *et al*. (2011), Russo *et al*. (2017) and Hixson *et al*. (2018) (by-products); and Grainger *et al*. (2009), Durmic *et al*. (2014), Banik *et al*. (2016) and Shakeri *et al*. (2017) (plant extracts and essential oils).

such grape marc have a high-water content, diluting out the bioactivity, causing product spoilage and increasing the cost of transport. Despite these issues, grape marc, in particular, continues to be a topic of interest because, as a major producer of wine, Australia generates ~200 kT of the by-product annually. However, there has been some inconsistencies in its mitigation potential, because only a limited reduction in CH_4 production was seen when extracts of grape marc were tested *in vitro* (Hixson *et al*. 2018), whereas it reduced CH_4 emissions by 20% when fed to lactating cows (Moate *et al*. 2014). This disagreement could be explained by the difference between *in vitro* and *in vivo* methodologies, or by variations in the chemical profiles of various types of grape marc (Russo *et al*. 2017). The high fibre content and low digestibility of grape marc reduces milk yield when fed to dairy cattle, when used to replace high energy supplements (Moate *et al*. 2020). However, when grape marc is substituted for feed with a similar energy value, CH_4 emissions are reduced, with little change in productivity (Black *et al*. 2021).

Feeding ruminants plant oils is another effective way of reducing enteric CH_4 production, while utilising oil-rich waste products generated during the oil-extraction process. Olive cake, cashew nut shell, hazelnut pericarps, and the seeds from sunflower, flax and canola have been identified as CH_4 mitigators (Beauchemin *et al*. 2009; Watanabe *et al*. 2010; Niderkorn *et al*. 2020). The anti-methanogenic action in these involves direct removal of hydrogen during fermentation (Eugène *et al*. 2008; Rasmussen and Harrison 2011). The products considered in Australia for CH_4 mitigation include brewers' grains, cold-pressed canola, hominy meal, pequi oil, almond hulls and cottonseed (Moate *et al*. 2011; Durmic *et al*. 2014, Duarte *et al*. 2017; Williams *et al*. 2018). Among these, almond hulls have been shown to reduce CH_4 production by 25% (Durmic *et al*. 2014), while preliminary investigations into olive leaves and fruits suggested a 50% reduction in CH_4 production *in vitro* (Shakeri *et al*. 2017), and a recent study linked the effect to polyphenol content and shift in bacterial populations (Lee *et al*. 2021).

Whole plant extracts and essential oils

The exploration of CH_4 mitigation strategies extended from feeding whole plants or plant products, to a quest for specific bioactive molecules of plant origin. Early reports from Europe showed that extracts from flavouring oils, particularly garlic, reduced CH_4 emissions (Busquet *et al*. 2005*a*, 2005*b*; Chaves *et al*. 2008).

In Australia, extracts from two native forage plants (i.e. Tar Bush or *Kennedia prorepens*, as well as biserrula), significantly reduced CH_4 production *in vitro* (Durmic *et al*. 2014; Banik *et al*. 2016). In biserrula, selected fractions were tested against methanogens in pure culture and found to inhibit some key ones, including those found in Australian grazing sheep (Banik *et al*. 2016). More research is needed to identify specific anti-methanogenic compounds from a variety of candidate plants that can be tested *in vivo*.

Significant research has been dedicated to studying the anti-methanogenic effects of essential oils, and, globally, those from clove, white thyme, citronella, peppermint, anise and cinnamon have been reported to reduce CH_4 emissions (Patra and Yu 2012; Benchaar 2016; Günal *et al*. 2017). Essential oils from Australian plants gained attention in the late 1990s as potent antimicrobials for human pathogens (Hammer *et al*. 1999), leading to assessment of their value for CH_4 mitigation. Essential oils from swamp paperbark (*Melaleuca ericifolia*), honey myrtle (*M. teretifolia*) and lemon-scented teatree (*Leptospermum petersonii*) were found to be very potent and inhibiting CH_4 production by up to 75% (*in vitro*), although they also inhibited microbial fermentation (VFA) at the doses tested (Durmic *et al*. 2014). Subsequently, we identified optimal doses that did not affect overall rumen fermentation, but were still effective at reducing CH_4 production (Jahani-Azizabadi *et al*. 2019). *In vitro* work is continuing to identify the mechanisms that explain the effect and to optimise the doses of pure active ingredient, after which we will move to *in vivo* testing.

Pure plant compounds: tannins, saponins and other PSC

Tannins and saponins are often abundant in plants of low methanogenic potential, with condensed tannins becoming a major focus for anti-methanogenic compound research (Waghorn 2008; Patra and Saxena 2011; Rira *et al*. 2013). *In vivo* studies confirmed the potency of condensed tannins, with emission reductions of more than 50% having been reported (Carulla *et al*. 2005; Lima *et al*. 2019). Tannins can act directly, affecting methanogens and protozoa, and preventing methanogens from attaching to protozoa, or act indirectly by inhibiting overall rumen microbial activity, with the subsequent consequence of reducing animal productivity (Kumar and Vaithiyanathan 1990; Ku-Vera *et al*. 2020).

Saponins have been also considered for CH_4 mitigation because they can control ruminal protozoa and thus reduce the number of methanogens that are directly associated with them (Patra and Saxena 2009; Jayanegara *et al*. 2012). However, the results with saponin-rich plant sources have been variable, with effects ranging from no CH_4 reduction to moderate reduction *in vitro* and *in vivo* (Goel and Makkar 2012; Liu *et al*. 2019; Molina-Botero *et al*. 2019). These disagreements could be explained by variation in the saponin source. The most promising candidates reported around the world appear to be *Yucca schidigera*, *Saponaria officinalis*, *Medicago sativa*, *Camellia sinensis*, *Enterolobium cyclocarpum* and *Quillaja Saponaria*, inhibiting CH_4 production by up to 40% (Rodríguez and Fondevila 2012; Patra *et al*. 2017).

In Australia, Ramírez-Restrepo *et al*. (2016) reported an 18% reduction in total daily CH_4 emissions (g/day) and a 22% reduction in yield (g/kg dry-matter intake) after feeding steers with tea-seed saponins in combination with Rhodes grass and grain concentrate

Investigation of the active components of essential oils has progressed, also leading to the discovery of some potent pure compounds from these that inhibit CH_4 production *in vitro* and *in vivo*, including thymol, carvacrol, cinnamaldehyde, garlic organosulfur compounds, citral, limonene, linalool, α- and β-pinene (Busquet *et al*. 2005*a*; Cardozo *et al*. 2006; Macheboeuf *et al*. 2008; Joch *et al*. 2016; Ma *et al*. 2016).

Similar work is currently being conducted on compounds found in Australian plant essential oils.

There are advantages in working with pure compounds. The structure is well known and the effects on CH_4 production can be attributed specifically to the compound itself, presenting opportunities to investigate mechanisms of action. They are more attractive than mixed compounds or whole plants from a commercialisation perspective, because purity can be verified for drug acceptability and efficacy. Once identified, they can be obtained from natural sources, using scaled-up extraction processes, or even synthesised. However, anti-methanogenic effects might be less efficient with a single compound than with a combination of compounds, some of which may not even be identified (Patra *et al*. 2017).

Commercial plant-based products

Plant bioactivity has been explored globally, with a view to the development of commercial products. One of the first, based on essential oil compounds, was Crina® Ruminant (Akzo Nobel Ltd, Netherlands) that had positive effects on animal production, but with limited effects on CH_4 mitigation (Beauchemin and McGinn 2006; Tomkins *et al*. 2015; Patra *et al*. 2017). Another essential oil product, Agolin® Ruminant (Agolin SA, Switzerland), was more effective and became the first feed additive certified for CH_4 mitigation in ruminants (Carbon Trust Assurance Ltd, https://agolin.ch/certifications/). It contains compounds from coriander seed, eugenol, geranyl acetate and geraniol. *In vitro*, Agolin® Ruminant reduced CH_4 production by 30% (Durmic *et al*. 2014), and when fed to dairy cattle at a rate of 1 g/head daily, it decreased CH_4 production by more than 10% without affecting animal productivity (Belanche *et al*. 2020). A recently emerged product that is showing promise, Mootral™ (Mootral, Switzerland), a combination of extracts from garlic and bitter orange, persistently reduced CH_4 production *in vitro* by 70% (Eger *et al*. 2018). Activo® Premium (EW Nutrition, Germany), a mix of microencapsulated PSC, has been reported to reduce enteric CH_4 production in sheep by 26%, while improving rumen fermentation, digestibility and protein synthesis (Soltan *et al*. 2018). All of these products are commercially available as feed additives in Australia, but, in parallel, the work is progressing towards developing local products.

There are some obvious advantages when using a commercial product for CH_4 mitigation, including the following: it is backed up by extensive research and development; it has passed regulatory requirements; and it is easy to adopt and apply. However, these products do come at a cost, so their use is often limited to intensive industries that can reliably incorporate such additives in the feedlots.

Advantages and limitations of plant bioactivity

Plant-based approaches to CH_4 mitigation in ruminants may offer several benefits and advantages. Many of the plant sources under consideration are already a major part of ruminant diets, so there is little imposition on the animal and they qualify as ‘natural’. The anti-methanogenic PSC are present, abundant and diverse in some of these plants. In addition to reducing CH_4 production, some bioactive plants and plant-based products have other beneficial properties. They can improve animal feed intake and utilisation, enhance fermentability and digestibility, reduce protein degradability, and increase animal productivity (Aerts *et al*. 1999; Akanmu and Hassen 2018). A wide variety of these plants and PSC are also effective in managing animal digestive disorders, such as lactic acidosis, controlling animal diseases (i.e. worms), or enhancing animal reproduction (Kotze *et al*. 2009; Hutton *et al*. 2010; Durmic and Blache 2012). Many of the bioactive plants that have been investigated in Australia are native (unbred) plants, grown locally; so, they contribute to the preservation of biodiversity and thus a more ‘ecologically friendly’ animal production system. Adding these native forages to the production system has also been reported to add value to the feed-base (Vercoe *et al*. 2009; Revell *et al*. 2013) and improve overall farm profitability (Monjardino *et al*. 2010). Further, if plant-based bioactive compounds are derived from organic waste that would otherwise end up as landfill, then the CH_4 mitigation achieved by feeding them to livestock is accompanied by a reduction in environmental pollution and gas emissions from the secondary fermentation.

Plant-based CH_4 mitigation strategies, while having many advantages, also have certain limitations and present challenges. Propagation and utilisation of low methanogenic forages may be restricted to a single geographical location, climate or season, with constraints in their nutritional profile, supply, biomass, or because the strategy to incorporate them is not feasible for all animal production systems. Tannin- and saponin-rich browse and extracts are too often restricted in their application due to the depression of feed intake, fermentation and milk yield (Busquet *et al*. 2005*b*; Hess *et al*. 2006; Tan *et al*. 2011; Castro-Montoya *et al*. 2012). The use of industry by-products is often limited because their high water content can lead to spoilage, as well as increased cost of transport and processing. High content of protein, tannin, sugar or lignin may also affect the animal directly, by inhibiting rumen function, or lead to increased GHG emissions from manure of animals fed these by-products (Hünerberg *et al*. 2014). Vegetable oils and fats that remain in oil by-products can also have negative effects on milk production (Martin *et al*. 2008), whereas tannins and terpenes may leave residues and taint the animal products (Mason *et al*. 2017). Although essential oils are considered natural, with a history of use in traditional medicine, some toxic effects have been recorded in livestock (Horky *et al*. 2019).

However, the main problems of progressing plant-based approaches for CH_4 mitigation are confirmation of effectiveness *in vivo* and finding the optimal dose and mode of delivery. Many reports have failed to demonstrate *in vivo* efficacy of promising candidates that have emerged from *in vitro* screening (Meale *et al*. 2014; Benchaar 2016). For those that are shown to be effective *in vivo*, doses are often too high, so there are adverse effects on rumen microbes, or the feeding requirements are simply impractical (Benchaar *et al*. 2008; Macheboeuf *et al*. 2008; Grainger *et al*. 2009). There is also the effect of the interactions among host species, genotype, rumen conditions and animal diet, limiting extrapolation to specific production systems and situations

(Calsamiglia *et al*. 2007; Patra and Saxena 2009; Castro-Montoya *et al*. 2012). For example, some plant-based feed additives are most effective when combined with a high-fibre diet (Shakeri *et al*. 2017), whereas others are better suited to combination with concentrate rations (Calsamiglia *et al*. 2007). Furthermore, most *in vivo* data in the literature are derived from short-term trials. As a result, extrapolation to production systems becomes difficult because the rumen microbes can adapt to the PSC (Moss *et al*. 2000; Busquet *et al*. 2005*a*; Pellikaan *et al*. 2011), or degrade them into metabolites with less bioactivity (Malecky *et al*. 2012; Ghaffari *et al*. 2015). These adaptations can explain reduced efficacity over time of candidate PSC *in vivo* (Benchaar *et al*. 2008). We also need to take testing beyond laboratory-based or animal house-type *in vivo* studies and assess candidates and plant-based strategies under commercial conditions. This issue becomes evident when we consider extensive grazing systems in which the amount of bioactive compound that an animal ingests is unpredictable. Moreover, in some production systems, the application of plant bioactivity may not be practical or cost-effective.

Options for plant bioactivity-based CH_4 mitigation in Australia

Table 1 and Figs 1 and 2 present an overview of benefits and limitations of plant-based mitigation strategies for Australian ruminant industries, summarised from information provided by Black *et al*. (2015, 2021) and other literature cited in the present review. Briefly, in Fig. 1, we have summarised the information on the level of CH_4 reduction, as well as the effect on rumen fermentation (VFA production) gathered in Australia and for different sources of plant bioactivity. In Table 1, we have then summarised the extent (moderate–high) and the type (specific–nonspecific) of effect on CH_4 for each category. In there, we have also evaluated the other benefits to animal production, such as good agronomic properties and nutritive value of the plant material fed, effect on fermentation and animal health, and, consequently, on animal productivity and welfare. We have also considered the barriers and limitations to adoption for each practice. Finally, we sourced information from Black *et al*. (2021) to plot these mitigation categories according the predicted time to practical application, barrier/cost to implement, and the national CH_4 mitigation potential in Australia based on 25% reduction in CH_4 emissions across all Australian ruminants and 10% adoption. We then used the information on the barriers and limitations, whether the methodology is something that producers are familiar with, whether the plant used in the strategy has good agronomic properties, biomass and NV, so as to estimate and present 'likelihood of adoption' (low–high).

Given the Australian focus on grazing livestock, changing forage species available for consumption seems the obvious first option. The wide range of eco-climatic zones, from tropical to temperate, to hostile, dry environments, will also dictate the strategies that are most applicable. The natural, sustainable, cost-effective solutions for CH_4 mitigation in grazing ruminants are therefore likely to be low-methanogenic browse, i.e. native forage shrubs in low- to medium-rainfall zones; mainstream/alternative herbaceous plants for temperate climates; tropical or rangeland plants for the northern regions of Australia.

Temperate forages such as subterranean clover and lucerne are highly ranked in terms of national mitigation potential and practicality; they achieve only a moderate reduction in CH_4 production, but they offer a significant reduction in CH_4 emission intensity due to their high nutritive value and positive effect on other fermentation pathways. They are familiar to producers, and despite requiring high inputs for sourcing, establishment, cultivation and maintenance, they are likely to have a high adoption rate, and as such can contribute significantly to national CH_4 mitigation overall (Fig. 2). By contrast, the current estimates for Australian native forage shrubs predict that these have a smaller role in national mitigation, as they are geographically contained and need to be grazed in a mix to offset any deficiencies in nutritive profile or negative effects on the rumen, animal health and productivity. Greater implementation is also limited by our incomplete agronomic knowledge of the species and insufficient analysis of the anti-methanogenic properties in a wider range of native plants that are naturally present in the feedbase. These limitations extend the timeline of more widespread adoption, which is currently limited to areas of marginal land and as a drought reserve (Fig. 2). Research is required to overcome these knowledge gaps, so the shrubs are contributing more to the feedbase in a variety of regions. As some of them have a strong, direct anti-methanogenic effect, and many are found to be naturally present and already grazed in Australian rangelands, predictions of ruminant emissions from Australian rangelands, and the value of our native plants, may need to be revisited and altered when more information becomes available.

Tropical forages (e.g. *Leucaena*) generally elicit moderate reductions in enteric CH_4 production, but have the potential to increase animal productivity by 20% and therefore significantly reduce CH_4 emission intensity, when compared with the standard practice of grazing Rhodes grass pastures (Harrison *et al*. 2015). Despite barriers due to high establishment costs, complex management, and issues with anti-nutritive factors and toxicity (Table 1), they have the advantage of providing good biomass and nutritive profiles (Taylor *et al*. 2016), resulting in moderate prospects for practical application.

Industry by-products are already valued as a feed supplement in Australia, and some of these also bring a desired reduction in CH_4 production (Table 1). However, the distribution and use of the by-products is limited to farms that are in relatively close proximity to the site of generation, resulting in a relatively low likelihood of adoption (Fig. 2). As a high-energy supplement, their most obvious application is intensive systems (feedlot) and high-performance animals (dairy cattle). Moreover, the anti-methanogenic effect is often non-specific and some of the by-products may have negative effects on the animal; so, further research is needed to find the optimal inclusion levels that balance these positive and negative effects. In this category, by-products of oil extraction processes or brewing industries have been reported to have some

Table 1. Summary of potential benefits and limitations for Australian ruminant industries from mitigation strategies involving plant bioactive compounds

Values for CH_4 mitigation potential (Mt/year) are national estimates

Category	Effect on CH_4	Animal production and agronomic benefits	Barriers and limitations
Native shrubs	Moderate to high effect in reduction of CH_4. Some with specific effect on CH_4. Anti-methanogenic effect for some confirmed *in vivo*	5% animal productivity gain. Some with benefits on animal health: anthelmintic and preventing rumen disorders. Drought tolerance, eliminating the need for supplementary feeding. Already part of feedbase for sheep in south-western Australia	Geographically contained, full agronomy unknown, poor germination, low biomass, palatability issues, some negatively affecting fermentation. Need to be grazed in a mix
Tropical forages	Moderate and non-specific effect in reduction of CH_4, but significant impact on CH_4 emission intensity. Anti-methanogenic effect for some confirmed *in vivo*	Good nutritive value, 20% animal productivity gain. Some with good biomass and known agronomic properties	Some result in reduced intake, toxicity, have long establishment and management time and many need to be grazed in a mix. Some considered weed in parts of Australia. Some negatively affecting fermentation
Temperate forages	Moderate to high effect in reduction of CH_4. Some with specific effect on CH_4, significant impact on CH_4 emission intensity. Anti-methanogenic effect for some confirmed *in vivo*	Good nutritive value, fermentability, 15% animal productivity gain. Good biomass, agronomic properties well known	Limited range of agro-ecological environments. Many sensitive to drought, prone to infestation and diseases, need enhanced soils
Plant by-products	Moderate and non-specific effect in reduction of CH_4. Anti-methanogenic effect for some confirmed *in vivo*	Source of energy, protein and fibre. Valuable for enterprises in proximity to sources and when other feed supply is low	Possible negative effect on productivity and milk yield. Effect variable depending on types of product and interaction with diet. Restricted by cost of processing or proximity to source
Plant extracts and essential oils	High effect in reduction of CH_4. Some with specific effect on CH_4	Benefits in fermentation and animal health (anthelmintic, preventing rumen disorders, gut pathogens). Some already commercially available, can be incorporated as feed additive	Limited testing performed on animals; doses, delivery systems and cost unknown. Use may be restricted in extensive systems

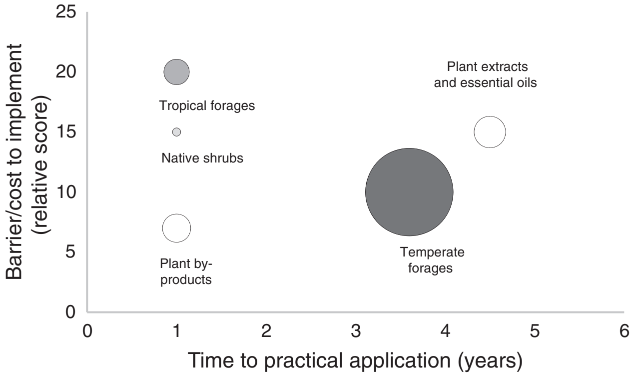

Fig. 2. Distribution of plant-based strategies for enteric CH_4 mitigation according to predicted time to practical application (years), barrier/cost to implement (relative score: 1 (low) to 20 (high)). The bubble size illustrates the predicted national CH_4 mitigation potential from each strategy in Australia, based on 25% reduction in CH_4 emissions across all Australian ruminants and 10% adoption, while the shade of the bubble (white (low) to dark (high)) illustrates predicted likely level of adoption. Information from modified from Black *et al*. (2021).

potential for practical application in intensive farming systems and have been shown to reduce CH_4 emission by 15–20% (Moate *et al*. 2011). Other by-products should be assessed further for their benefits and their potential to address feed shortages and contribute to agricultural waste management in Australia.

While we already have access to some of the imported commercial plant-based products, our products are yet to be developed, and are yet to be investigated for our conditions and systems. Also, we do not know how much they could realistically reduce CH_4 and what the cost of this strategy would be.

Future work

The outcomes from the research undertaken have moved us closer towards practical solutions to mitigating methane. While some strategies for enteric CH_4 mitigation in Australia exist, more are yet to be researched and established. Focusing on plant bioactivity is clearly an option worthy of further investigation and investment. Australia is well positioned to explore and exploit its own plant resources as a means of balancing out the environmental effects of its livestock. While doing so, it may also address some other issues, including animal productivity, farm profitability, animal health, the security of plant biodiversity and the management of organic waste. While there seems to be an enormous potential for Australian plants and PSC, most of the exciting candidates are yet to be investigated in detail *in vivo* and under commercial settings; so, their potential for commercialisation is not clear. Also, the long-term impacts on palatability, intake, performance, and the quality of animal products, need to be investigated. Moreover, they need to be carefully assessed with regards to seasonal availability, total CH_4 emission and to rule out any toxicity to the animals and humans needs. Balancing these issues will almost universally depend on finding optimal doses and delivery methods, and then developing and adopting plant-based mitigation strategies for whole-farm enterprises. Clearly, Australia must align with global efforts to find effective mitigation strategies, and start developing locally relevant approaches tailored to our animals, climate, national profile, capacities and needs.

Data availability statement

Data sharing is not applicable as no new data were generated or analysed during this study.

Conflicts of interest

Zoey Durmic was Associate Editors of Animal Production Science at the time of submission, but was blinded from the peer-review process for this paper.

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References

Aerts RJ, Barry TN, McNabb WC (1999) Polyphenols and agriculture: beneficial effects of proanthocyanidins in forages. *Agriculture, Ecosystems & Environment* **75**, 1–12. doi:10.1016/S0167-8809(99)00062-6

AGEIS (2017) 'Australian Greenhouse Emissions Information System National Greenhouse Gas Inventory: Kyoto Protocol Classifications.' Available at http://ageis.climatechange.gov.au/ [Verified May 2021]

Akanmu AM, Hassen A (2018) The use of certain medicinal plant extracts reduced *in vitro* methane production while improving *in vitro* organic matter digestibility. *Animal Production Science* **58**, 900–908. doi:10.1071/AN16291

ARCADIS (2019) National Food Waste Assessment – final report. Available at https://www.environment.gov.au/protection/waste/publications/national-food-waste-assessment-final-report [Verified October 2020]

Banik BK, Durmic Z, Erskine W, Ghamkhar K, Revell C (2013) *In vitro* ruminal fermentation characteristics and methane production differ in selected key pasture species in Australia. *Crop and Pasture Science* **64**, 935–942. doi:10.1071/CP13149

Banik BK, Durmic Z, Erskine W, Revell CK, Vadhanabhuti J, McSweeney CS, Padmanabha J, Flematti GR, Algreiby AA, Vercoe PE (2016) Bioactive fractions from the pasture legume *Biserrula pelecinus* L. have an anti-methanogenic effect against key rumen methanogens. *Anaerobe* **39**, 173–182. doi:10.1016/j.anaerobe.2016.04.004

Banik BK, Durmic Z, Erskine W, Revell C (2019) Anti-methanogenic advantage of biserrula (*Biserrula pelecinus*) over subterranean clover (*Trifolium subterraneum*) from *in vitro* fermentation is maintained across growth stages and cutting treatments. *Crop and Pasture Science* **70**, 263–272. doi:10.1071/CP18069

Beauchemin KA, McGinn SM (2006) Methane emissions from beef cattle: effects of fumaric acid, essential oil, and canola oil. *Journal of Animal Science* **84**, 1489–1496. doi:10.2527/2006.8461489x

Beauchemin KA, McGinn SM, Benchaar C, Holtshausen L (2009) Crushed sunflower, flax, or canola seeds in lactating dairy cow diets: effects on methane production, rumen fermentation, and milk production. *Journal of Dairy Science* **92**, 2118–2127. doi:10.3168/jds.2008-1903

Beauchemin KA, Ungerfeld EM, Eckard RJ, Wang M (2020) Review: fifty years of research on rumen methanogenesis: lessons learned and future challenges for mitigation. *Animal* **14**, s2–s16. doi:10.1017/S1751731119003100

Beijer WH (1952) Methane fermentation in the rumen of cattle. *Nature* **170**, 576–577. doi:10.1038/170576a0

Belanche A, Newbold CJ, Morgavi DP, Bach A, Zweifel B, Yáñez-Ruiz DR (2020) A meta-analysis describing the effects of the essential oils blend Agolin ruminant on performance, rumen fermentation and methane emissions in dairy cows. *Animals* **10**, 620. doi:10.3390/ani10040620

Benchaar C (2016) Diet supplementation with cinnamon oil, cinnamaldehyde, or monensin does not reduce enteric methane production of dairy cows. *Animal* **10**, 418–425. doi:10.1017/S175173111500230X

Benchaar C, McAllister TA, Chouinard PY (2008) Digestion, ruminal fermentation, ciliate protozoal populations, and milk production from dairy cows fed cinnamaldehyde, quebracho condensed tannin, or *Yucca schidigera* saponin extracts. *Journal of Dairy Science* **91**, 4765–4777. doi:10.3168/jds.2008-1338

Benchaar C, Hassanat F, Gervais R, Chouinard PY, Julien C, Petit HV, Massé DI (2013) Effects of increasing amounts of corn dried distillers grains with solubles in dairy cow diets on methane production, ruminal fermentation, digestion, N balance, and milk production. *Journal of Dairy Science* **96**, 2413–2427. doi:10.3168/jds.2012-6037

Black J, Davison T, Fennessy P, Cohn P, Sedger A, Empson M (2015) National Livestock Methane Program National Needs and Gaps Analysis. B.CCH.6000 final report. Available at https://www.mla.com.au/research-and-development/search-rd-reports/final-report-details/Environment-On-Farm/National-Livestock-Methane-Program-National-Needs-and-Gaps-Analysis/3196 [Verified October 2020]

Black JL, Davison TM, Box I (2021) Methane emissions from ruminants in Australia: mitigation potential and applicability of mitigation strategies. *Animals (Basel)* **11**, 951. doi:10.3390/ani11040951

Blaxter KL, Czerkawski J (1966) Modifications of the methane production of the sheep by supplementation of its diet. *Journal of the Science of Food and Agriculture* **17**, 417–421. doi:10.1002/jsfa.2740170907

Bodas R, Prieto N, García-González R, Andrés S, Giráldez FJ, López S (2012) Manipulation of rumen fermentation and methane production with plant secondary metabolites. *Animal Feed Science and Technology* **176**, 78–93. doi:10.1016/j.anifeedsci.2012.07.010

Busquet M, Calsamiglia S, Ferret A, Cardozo PW, Kamel C (2005*a*) Effects of cinnamaldehyde and garlic oil on rumen microbial fermentation in a dual flow continuous culture. *Journal of Dairy Science* **88**, 2508–2516. doi:10.3168/jds.S0022-0302(05)72928-3

Busquet M, Calsamiglia S, Ferret A, Carro MD, Kamel C (2005*b*) Effect of garlic oil and four of its compounds on rumen microbial fermentation. *Journal of Dairy Science* **88**, 4393–4404. doi:10.3168/jds.S0022-0302(05)73126-X

Busquet M, Calsamiglia S, Ferret A, Kamel C (2006) Plant extracts affect *in vitro* rumen microbial fermentation. *Journal of Dairy Science* **89**, 761–771. doi:10.3168/jds.S0022-0302(06)72137-3

Calsamiglia S, Busquet M, Cardozo PW, Castillejos L, Ferret A (2007) Invited review: essential oils as modifiers of rumen microbial fermentation. *Journal of Dairy Science* **90**, 2580–2595. doi:10.3168/jds.2006-644

Cardozo PW, Calsamiglia S, Ferret A, Kamel C (2006) Effects of alfalfa extract, anise, capsicum, and a mixture of cinnamaldehyde and eugenol on ruminal fermentation and protein degradation in beef heifers fed a high-concentrate diet. *Journal of Animal Science* **84**, 2801–2808. doi:10.2527/jas.2005-593

Carulla JE, Kreuzer M, Machmüller A, Hess HD (2005) Supplementation of *Acacia mearnsii* tannins decreases methanogenesis and urinary nitrogen in forage-fed sheep. *Australian Journal of Agricultural Research* **56**, 961–970. doi:10.1071/AR05022

Castillo-Lopez E, Jenkins CJR, Aluthge ND, Tom W, Kononoff PJ, Fernando SC (2017) The effect of regular or reduced-fat distillers grains with solubles on rumen methanogenesis and the rumen bacterial community. *Journal of Applied Microbiology* **123**, 1381–1395. doi:10.1111/jam.13583

Castro-Montoya J, De Campeneere S, Van Ranst G, Fievez V (2012) Interactions between methane mitigation additives and basal substrates on *in vitro* methane and VFA production. *Animal Feed Science and Technology* **176**, 47–60. doi:10.1016/j.anifeedsci.2012.07.007

Charmley E, Stephens ML, Kennedy PM (2008) Predicting livestock productivity and methane emissions in northern Australia: development of a bio-economic modelling approach. *Australian Journal of Experimental Agriculture* **48**, 109–113. doi:10.1071/EA07264

Chaves AV, He ML, Yang WZ, Hristov AN, McAllister TA, Benchaar C (2008) Effects of essential oils on proteolytic, deaminative and methanogenic activities of mixed ruminal bacteria. *Canadian Journal of Animal Science* **88**, 117–122. doi:10.4141/CJAS07061

Cottle DJ, Nolan JV, Wiedemann SG (2011) Ruminant enteric methane mitigation: a review. *Animal Production Science* **51**, 491–514. doi:10.1071/AN10163

Dalzell SA, Burnett DJ, Dowsett JE, Forbes VE, Shelton HM (2012) Prevalence of mimosine and DHP toxicity in cattle grazing *Leucaena leucocephala* pastures in Queensland, Australia. *Animal Production Science* **52**, 365–372. doi:10.1071/AN11236

Dong H, MacDonald JD, Ogle SM, Sanchez MJS, Rocha MT (2019) Volume 4. Agriculture, forestry and other land use. In 'Refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories'. (Eds E Calvo Buendia, K Tanabe, A Kranjc, J Baasansuren, M Fukuda, S Ngarize, A Osako, Y Pyrozhenko, P Shermanau, S Federici) (IPCC: Switzerland)

Doran-Browne N, Behrendt R, Kingwell R, Eckard R (2015) Modelling the potential of birdsfoot trefoil (*Lotus corniculatus*) to reduce methane emissions and increase production on wool and prime lamb farm enterprises. *Animal Production Science* **55**, 1097–1105. doi:10.1071/AN13543

Duarte AC, Durmic Z, Vercoe PE, Chaves AV (2017) Dose-response effects of dietary pequi oil on fermentation characteristics and microbial population using a rumen simulation technique (Rusitec). *Anaerobe* **48**, 59–65. doi:10.1016/j.anaerobe.2017.06.013

Durmic Z, Blache D (2012) Bioactive plants and plant products: effects on animal function, health and welfare. *Animal Feed Science and Technology* **176**, 150–162. doi:10.1016/j.anifeedsci.2012.07.018

Durmic Z, Hutton P, Revell DK, Emms J, Hughes S, Vercoe PE (2010) *In vitro* fermentative traits of Australian woody perennial plant species that may be considered as potential sources of feed for grazing ruminants. *Animal Feed Science and Technology* **160**, 98–109. doi:10.1016/j.anifeedsci.2010.07.006

Durmic Z, Moate PJ, Eckard R, Revell DK, Williams R, Vercoe PE (2014) *In vitro* screening of selected feed additives, plant essential oils and plant extracts for rumen methane mitigation. *Journal of the Science of Food and Agriculture* **94**, 1191–1196. doi:10.1002/jsfa.6396

Durmic Z, Moate PJ, Jacobs JL, Vadhanabhuti J, Vercoe PE (2016) *In vitro* fermentability and methane production of some alternative forages in Australia. *Animal Production Science* **56**, 641–645. doi:10.1071/AN15486

Durmic Z, Ramirez-Restrepo CA, Gardiner C, O'Neill CJ, Hussein E, Vercoe PE (2017) Differences in the nutrient concentrations, *in vitro* methanogenic potential and other fermentative traits of tropical grasses and legumes for beef production systems in northern Australia. *Journal of the Science of Food and Agriculture* **97**, 4075–4086. doi:10.1002/jsfa.8274

Eger M, Graz M, Riede S, Breves G (2018) Application of Mootral™ reduces methane production by altering the archaea community in the rumen simulation technique. *Frontiers in Microbiology* **9**, 2094. doi:10.3389/fmicb.2018.02094

EPA (2003) Inventory of U.S. Greenhouse Gas Emissions and Sinks: 1990–2001. Available at https://www.epa.gov/ghgemissions/inventory-us-greenhouse-gas-emissions-and-sinks-1990-2001 [Verified October 2020]

Eugène M, Massé D, Chiquette J, Benchaar C (2008) Meta-analysis on the effects of lipid supplementation on methane production in lactating dairy cows. *Canadian Journal of Animal Science* **88**, 331–337. doi:10.4141/CJAS07112

Ghaffari MH, Durmic Z, Real D, Vercoe P, Smith G, Oldham C (2015) Furanocoumarins in tedera do not affect ruminal fermentation in continuous culture. *Animal Production Science* **55**, 544–550. doi:10.1071/AN13335

Goel G, Makkar HP (2012) Methane mitigation from ruminants using tannins and saponins. *Tropical Animal Health and Production* **44**, 729–739. doi:10.1007/s11250-011-9966-2

Grainger C, Clarke T, Auldist MJ, Beauchemin KA, McGinn SM, Waghorn GC, Eckard RJ (2009) Potential use of *Acacia mearnsii* condensed tannins to reduce methane emissions and nitrogen excretion from grazing dairy cows. *Canadian Journal of Animal Science* **89**, 241–251. doi:10.4141/CJAS08110

Grainger C, Williams R, Eckard RJ, Hannah MC (2010) A high dose of monensin does not reduce methane emissions of dairy cows offered pasture supplemented with grain. *Journal of Dairy Science* **93**, 5300–5308. doi:10.3168/jds.2010-3154

Günal M, Pinski B, AbuGhazaleh AA (2017) Evaluating the effects of essential oils on methane production and fermentation under *in vitro* conditions. *Italian Journal of Animal Science* **16**, 500–506. doi:10.1080/1828051X.2017.1291283

Hammer KA, Carson CF, Riley TV (1999) Antimicrobial activity of essential oils and other plant extracts. *Journal of Applied Microbiology* **86**, 985–990. doi:10.1046/j.1365-2672.1999.00780.x

Hammond KJ, Burke JL, Koolaard JP, Muetzel S, Pinares-Patiño CS, Waghorn GC (2013) Effects of feed intake on enteric methane emissions from sheep fed fresh white clover (*Trifolium repens*) and perennial ryegrass (*Lolium perenne*) forages. *Animal Feed Science and Technology* **179**, 121–132. doi:10.1016/j.anifeedsci.2012.11.004

Hariadi BT, Santoso B (2010) Evaluation of tropical plants containing tannin on *in vitro* methanogenesis and fermentation parameters using rumen fluid. *Journal of the Science of Food and Agriculture* **90**, 456–461. doi:10.1002/jsfa.3839

Harrison MT, McSweeney C, Tomkins NW, Eckard RJ (2015) Improving greenhouse gas emissions intensities of subtropical and tropical beef farming systems using *Leucaena leucocephala*. *Agricultural Systems* **136**, 138–146. doi:10.1016/j.agsy.2015.03.003

Hess HD, Tiemann TT, Noto F, Carulla JE, Kreuzer M (2006) Strategic use of tannins as means to limit methane emission from ruminant livestock. *International Congress Series* **1293**, 164–167. doi:10.1016/j.ics.2006.01.010

Hixson JL, Durmic Z, Vadhanabhuti J, Vercoe PE, Smith PA, Wilkes EN (2018) Exploiting compositionally similar grape marc samples to achieve gradients of condensed tannin and fatty acids for modulating *in vitro* methanogenesis. *Molecules (Basel, Switzerland)* **23**, 1793. doi:10.3390/molecules23071793

Horky P, Skalickova S, Smerkova K, Skladanka J (2019) Essential oils as a feed additives: pharmacokinetics and potential toxicity in monogastric animals. *Animals (Basel)* **9**, 352. doi:10.3390/ani9060352

Hristov AN, Ott T, Tricarico J, Rotz A, Waghorn G, Adesogan A, Dijkstra J, Montes F, Oh J, Kebreab E, Oosting SJ, Gerber PJ, Henderson B, Makkar HPS, Firkins JL (2013) SPECIAL TOPICS: mitigation of methane and nitrous oxide emissions from animal operations: III. A review of animal management mitigation options. *Journal of Animal Science* **91**, 5095–5113. doi:10.2527/jas.2013-6585

Hünerberg M, Little SM, Beauchemin KA, McGinn SM, O'Connor D, Okine EK, Harstad OM, Kröbel R, McAllister TA (2014) Feeding high concentrations of corn dried distillers' grains decreases methane, but increases nitrous oxide emissions from beef cattle production. *Agricultural Systems* **127**, 19–27. doi:10.1016/j.agsy.2014.01.005

Hutton PG, Durmic Z, Vercoe PE (2010) Investigating *Eremophila glabra* as a bioactive agent for preventing lactic acidosis in sheep. *Animal Production Science* **50**, 449–453. doi:10.1071/AN09191

Jahani-Azizabadi J, Durmic Z, Vadhanabhuti J, Vercoe PE (2019) Effect of some Australian native shrubs essential oils on *in vitro* rumen microbial fermentation of a high-concentrate diet. *The Journal of Animal & Plant Sciences* **29**, 8–15.

Jayanegara A, Leiber F, Kreuzer M (2012) Meta-analysis of the relationship between dietary tannin level and methane formation in ruminants from *in vivo* and *in vitro* experiments. *Journal of Animal Physiology and Animal Nutrition* **96**, 365–375. doi:10.1111/j.1439-0396.2011.01172.x

Joch M, Cermak L, Hakl J, Hucko B, Duskova D, Marounek M (2016) *In vitro* screening of essential oil active compounds for manipulation of rumen fermentation and methane mitigation. *Asian–Australasian Journal of Animal Sciences* **29**, 952–959. doi:10.5713/ajas.15.0474

Johnson KA, Johnson DE (1995) Methane emissions from cattle. *Journal of Animal Science* **73**, 2483–2492. doi:10.2527/1995.7382483x

Kaur P, Appels R, Bayer PE, Keeble-Gagnere G, Wang J, Hirakawa H, Shirasawa K, Vercoe P, Stefanova K, Durmic Z, Nichols P, Revell C, Isobe SN, Edwards D, Erskine W (2017) Climate clever clovers: new paradigm to reduce the environmental footprint of ruminants by breeding low methanogenic forages utilizing haplotype variation. *Frontiers in Plant Science* **8**, 1463. doi:10.3389/fpls.2017.01463

Kotze AC, O'Grady J, Emms J, Toovey AF, Hughes S, Jessop P, Bennell M, Vercoe PE, Revell DK (2009) Exploring the anthelmintic properties of Australian native shrubs with respect to their potential role in livestock grazing systems. *Parasitology* **136**, 1065–1080. doi:10.1017/S0031182009006386

Ku-Vera JC, Jiménez-Ocampo R, Valencia-Salazar SS, Montoya-Flores MD, Molina-Botero IC, Arango J, Gómez-Bravo CA, Aguilar-Pérez CF, Solorio-Sánchez FJ (2020) Role of Secondary Plant Metabolites on Enteric Methane Mitigation in Ruminants. *Frontiers in Veterinary Science* **7**, doi:10.3389/fvets.2020.00584

Kumar R, Vaithiyanathan S (1990) Occurrence, nutritional significance and effect on animal productivity of tannins in tree leaves. *Animal Feed Science and Technology* **30**, 21–38. doi:10.1016/0377-8401(90)90049-E

Lee SJ, Kim HS, Eom JS, Choi YY, Jo SU, Chu GM, Lee Y, Seo J, Kim KH, Lee SS (2021) Effects of Olive (*Olea europaea* L.) Leaves with Antioxidant and Antimicrobial Activities on *In Vitro* Ruminal Fermentation and Methane Emission. *Animals (Basel)* **11**, 2008. doi:10.3390/ani11072008

Li X (2013) *Eremophila glabra* reduces methane production in sheep. PhD, The University of Western Australia, Perth, WA, Australia.

Li X, Durmic Z, Liu S, McSweeney CS, Vercoe PE (2014) *Eremophila glabra* reduces methane production and methanogen populations when fermented in a Rusitec. *Anaerobe* **29**, 100–107. doi:10.1016/j.anaerobe.2013.10.008

Lima PR, Apdini T, Freire AS, Santana AS, Moura LML, Nascimento JCS, Rodrigues RTS, Dijkstra J, Garcez Neto AF, Queiroz MAÁ, Menezes DR (2019) Dietary supplementation with tannin and soybean oil on intake, digestibility, feeding behavior, ruminal protozoa and methane emission in sheep. *Animal Feed Science and Technology* **249**, 10–17. doi:10.1016/j.anifeedsci.2019.01.017

Liu Y, Ma T, Chen D, Zhang N, Si B, Deng K, Tu Y, Diao Q (2019) Effects of tea saponin supplementation on nutrient digestibility, methanogenesis, and ruminal microbial flora in dorper crossbred ewe. *Animals (Basel)* **9**, 29. doi:10.3390/ani9010029

Ma T, Chen D, Tu Y, Zhang N, Si B, Deng K, Diao Q (2016) Effect of supplementation of allicin on methanogenesis and ruminal microbial flora in Dorper crossbred ewes. *Journal of Animal Science and Biotechnology* **7**, 1. doi:10.1186/s40104-015-0057-5

Macheboeuf D, Morgavi DP, Papon Y, Mousset JL, Arturo-Schaan M (2008) Dose–response effects of essential oils on *in vitro* fermentation activity of the rumen microbial population. *Animal Feed Science and Technology* **145**, 335–350. doi:10.1016/j.anifeedsci.2007.05.044

Malecky M, Albarello H, Broudiscou LP (2012) Degradation of terpenes and terpenoids from Mediterranean rangelands by mixed rumen bacteria *in vitro*. *Animal* **6**, 612–616. doi:10.1017/S1751731111001947

Martin C, Rouel J, Jouany JP, Doreau M, Chilliard Y (2008) Methane output and diet digestibility in response to feeding dairy cows crude linseed, extruded linseed, or linseed oil. *Journal of Animal Science* **86**, 2642–2650. doi:10.2527/jas.2007-0774

Mason SE, Mullen KAE, Anderson KL, Washburn SP, Yeatts JL, Baynes RE (2017) Pharmacokinetic analysis of thymol, carvacrol and diallyl disulfide after intramammary and topical applications in healthy organic dairy cattle. *Food Additives & Contaminants: Part A* **34**, 740–749. doi:10.1080/19440049.2017.1285056

McAllister TA, Newbold CJ (2008) Redirecting rumen fermentation to reduce methanogenesis. *Australian Journal of Experimental Agriculture* **48**, 7–13. doi:10.1071/EA07218

McGinn SM, Chung Y-H, Beauchemin KA, Iwaasa AD, Grainger C (2009) Use of corn distillers' dried grains to reduce enteric methane loss from beef cattle. *Canadian Journal of Animal Science* **89**, 409–413. doi:10.4141/CJAS08133

McSweeney C, Tomkins N (2015) B.CCH.6510_Final_Report. Available at https://www.mla.com.au/research-and-development/search-rd-reports/final-report-details/Environment-On-Farm/Impacts-of-Leucaena-plantations-on-greenhouse-gas-emissions-in-northern-Australian-cattle-production-systems/3039

Meale SJ, Chaves AV, Baah J, McAllister TA (2012) Methane production of different forages in *in vitro* ruminal fermentation. *Asian-Australasian Journal of Animal Sciences* **25**, 86–91. doi:10.5713/ajas.2011.11249

Meale SJ, Chaves AV, McAllister TA, Iwaasa AD, Yang WZ, Benchaar C (2014) Including essential oils in lactating dairy cow diets: effects on methane emissions. *Animal Production Science* **54**, 1215–1218. doi:10.1071/AN14152

Min BR, Solaiman S, Waldrip HM, Parker D, Todd RW, Brauer D (2020) Dietary mitigation of enteric methane emissions from ruminants: a review of plant tannin mitigation options. *Animal Nutrition* doi:10.1016/j.aninu.2020.05.002

Moate PJ, Williams SRO, Grainger C, Hannah MC, Ponnampalam EN, Eckard RJ (2011) Influence of cold-pressed canola, brewers grains and hominy meal as dietary supplements suitable for reducing enteric methane emissions from lactating dairy cows. *Animal Feed Science and Technology* **166–167**, 254–264. doi:10.1016/j.anifeedsci.2011.04.069

Moate PJ, Williams SRO, Torok VA, Hannah MC, Ribaux BE, Tavendale MH, Eckard RJ, Jacobs JL, Auldist MJ, Wales WJ (2014) Grape marc reduces methane emissions when fed to dairy cows. *Journal of Dairy Science* **97**, 5073–5087. doi:10.3168/jds.2013-7588

Moate PJ, Jacobs JL, Hixson JL, Deighton MH, Hannah MC, Morris GL, Ribaux BE, Wales WJ, Williams SRO (2020) Effects of feeding either red or white grape marc on milk production and methane emissions from early-lactation dairy cows. *Animals* **10**, 976. doi:10.3390/ani10060976

Molina-Botero IC, Arroyave-Jaramillo J, Valencia-Salazar S, Barahona-Rosales R, Aguilar-Pérez CF, Ayala Burgos A, Arango J, Ku-Vera JC (2019) Effects of tannins and saponins contained in foliage of *Gliricidia sepium* and pods of *Enterolobium cyclocarpum* on fermentation, methane emissions and rumen microbial population in crossbred heifers. *Animal Feed Science and Technology* **251**, 1–11. doi:10.1016/j.anifeedsci.2019.01.011

Monjardino M, Revell D, Pannell DJ (2010) The potential contribution of forage shrubs to economic returns and environmental management in Australian dryland agricultural systems. *Agricultural Systems* **103**, 187–197. doi:10.1016/j.agsy.2009.12.007

Moss AR, Jouany J-P, Newbold J (2000) Methane production by ruminants: its contribution to global warming. *Annales de Zootechnie* **49**, 231–253. doi:10.1051/animres:2000119

Muir SK, Kennedy AJ, Kearney G, Hutton P, Thompson AN, Vercoe P, Hill J (2020) Offering subterranean clover can reduce methane emissions compared with perennial ryegrass pastures during late spring and summer in sheep. *Animal Production Science* **60**, 1449–1458. doi:10.1071/AN18624

Navarro-Villa A, O'Brien M, López S, Boland TM, O'Kiely P (2011) *In vitro* rumen methane output of red clover and perennial ryegrass assayed using the gas production technique (GPT). *Animal Feed Science and Technology* **168**, 152–164. doi:10.1016/j.anifeedsci.2011.04.091

Niderkorn V, Barbier E, Macheboeuf D, Torrent A, Mueller-Harvey I, Hoste H (2020) *In vitro* rumen fermentation of diets with different types of condensed tannins derived from sainfoin (*Onobrychis viciifolia* Scop.) pellets and hazelnut (*Corylus avellana* L.) pericarps. *Animal Feed Science and Technology* **259**, 114357. doi:10.1016/j.anifeedsci.2019.114357

Oskoueian E, Abdullah N, Oskoueian A (2013) Effects of flavonoids on rumen fermentation activity, methane production, and microbial population. *BioMed Research International* **2013**, 349129. doi:10.1155/2013/349129

Pal K, Patra AK, Sahoo A, Kumawat PK (2015) Evaluation of several tropical tree leaves for methane production potential, degradability and rumen fermentation *in vitro*. *Livestock Science* **180**, 98–105. doi:10.1016/j.livsci.2015.07.011

Patra AK, Saxena J (2009) The effect and mode of action of saponins on the microbial populations and fermentation in the rumen and ruminant production. *Nutrition Research Reviews* **22**, 204–219. doi:10.1017/S0954422409990163

Patra AK, Saxena J (2011) Exploitation of dietary tannins to improve rumen metabolism and ruminant nutrition. *Journal of the Science of Food and Agriculture* **91**, 24–37. doi:10.1002/jsfa.4152

Patra AK, Yu Z (2012) Effects of essential oils on methane production and fermentation by, and abundance and diversity of, rumen microbial populations. *Applied and Environmental Microbiology* **78**, 4271–4280. doi:10.1128/AEM.00309-12

Patra A, Park T, Kim M, Yu Z (2017) Rumen methanogens and mitigation of methane emission by anti-methanogenic compounds and substances. *Journal of Animal Science and Biotechnology* **8**, 13. doi:10.1186/s40104-017-0145-9

Pellikaan WF, Stringano E, Leenaars J, Bongers DJGM, van Laar-van Schuppen S, Plant J, Mueller-Harvey I (2011) Evaluating effects of tannins on extent and rate of *in vitro gas* and CH_4 production using an automated pressure evaluation system (APES). *Animal Feed Science and Technology* **166–167**, 377–390. doi:10.1016/j.anifeedsci.2011.04.072

Piñeiro-Vázquez AT, Canul-Solis JR, Jiménez-Ferrer GO, Alayón-Gamboa JA, Chay-Canul AJ, Ayala-Burgos AJ, Aguilar-Pérez CF, Ku-Vera JC (2018) Effect of condensed tannins from *Leucaena leucocephala* on rumen fermentation, methane production and population of rumen protozoa in heifers fed low-quality forage. *Asian-Australasian Journal of Animal Sciences* **31**, 1738–1746. doi:10.5713/ajas.17.0192

Ramírez-Restrepo CA, Tan C, O'Neill CJ, López-Villalobos N, Padmanabha J, Wang J, McSweeney CS (2016) Methane production, fermentation characteristics, and microbial profiles in the rumen of tropical cattle fed tea seed saponin supplementation.

Animal Feed Science and Technology **216**, 58–67. doi:10.1016/j.anifeedsci.2016.03.005

Rasmussen J, Harrison A (2011) The benefits of supplementary fat in feed rations for ruminants with particular focus on reducing levels of methane production. *ISRN Veterinary Science* **2011**, 613172. doi:10.5402/2011/613172

Rea S, Bowman JP, Popovski S, Pimm C, Wright AG (2007) *Methanobrevibacter millerae* sp. nov. and *Methanobrevibacter olleyae* sp. nov., methanogens from the ovine and bovine rumen that can utilize formate for growth. *International Journal of Systematic and Evolutionary Microbiology* **57**, 450–456. doi:10.1099/ijs.0.63984-0

Revell D, Revell C (2006) Implications of 'duty of care' for the development of new pasture species. In 'Groundbreaking Stuff. Proceedings of the 13th Australian Agronomy Conference', 10–14 September 2006, Perth, WA, Australia. (Eds N Turner, T Acuna) pp. 1–9.

Revell DK, Norman HC, Vercoe PE, Phillips N, Toovey A, Bickell S, Hulm E, Hughes S, 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. doi:10.1071/AN13238

Rira M, Marie-Magdeleine C, Archimède H, Morgavi DP, Doreau M (2013) Effect of condensed tannins on methane emission and ruminal microbial populations. In 'Energy and protein metabolism and nutrition in sustainable animal production, Vol. 134'. (Eds JW Oltjen, ELH Kebreab) pp. 501–502. (Wageningen Academic Publishers: Wageningen, The Netherlands)

Rodríguez R, Fondevila M (2012) Effect of saponins from *Enterolobium cyclocarpum* on *in vitro* microbial fermentation of the tropical grass *Pennisetum purpureum. Journal of Animal Physiology and Animal Nutrition* **96**, 762–769. doi:10.1111/j.1439-0396.2011.01161.x

Russo VM, Jacobs JL, Hannah MC, Moate PJ, Dunshea FR, Leury BJ (2017) *In vitro* evaluation of the methane mitigation potential of a range of grape marc products. *Animal Production Science* **57**, 1437–1444. doi:10.1071/AN16495

Sagar NA, Pareek S, Sharma S, Yahia EM, Lobo MG (2018) Fruit and vegetable waste: bioactive compounds, their extraction, and possible utilization. *Comprehensive Reviews in Food Science and Food Safety* **17**, 512–531. doi:10.1111/1541-4337.12330

Shakeri P, Durmic Z, Vadhanabhuti J, Vercoe PE (2017) Products derived from olive leaves and fruits can alter *in vitro* ruminal fermentation and methane production. *Journal of the Science of Food and Agriculture* **97**, 1367–1372. doi:10.1002/jsfa.7876

Silivong P, Hervaseng B, Preston TR (2013) Methane production from Jack fruit, *Muntingia*, *Leucaena*, Gliricidia (*Gliricidia sepium*), Mimosa (*Mimosa pigra*) and *Acacia auriculoformis* foliages in an *in vitro* incubation with potassium nitrate as source of NPN. *Livestock Research for Rural Development* **25**, 15

Soliva CR, Zeleke AB, Clément C, Hess HD, Fievez V, Kreuzer M (2008) *In vitro* screening of various tropical foliages, seeds, fruits and medicinal plants for low methane and high ammonia generating potentials in the rumen. *Animal Feed Science and Technology* **147**, 53–71. doi:10.1016/j.anifeedsci.2007.09.009

Soltan YA, Natel AS, Araujo RC, Morsy AS, Abdalla AL (2018) Progressive adaptation of sheep to a microencapsulated blend of essential oils: ruminal fermentation, methane emission, nutrient digestibility, and microbial protein synthesis. *Animal Feed Science and Technology* **237**, 8–18. doi:10.1016/j.anifeedsci.2018.01.004

Suybeng B, Charmley E, Gardiner CP, Malau-Aduli BS, Malau-Aduli AEO (2019) Methane emissions and the use of *Desmanthus* in beef cattle production in northern Australia. *Animals (Basel)* **9**, 542. doi:10.3390/ani9080542

Swainson N, Muetzel S, Clark H (2018) Updated predictions of enteric methane emissions from sheep suitable for use in the New Zealand national greenhouse gas inventory. *Animal Production Science* **58**, 973–979. doi:10.1071/AN15766

Tan HY, Sieo CC, Abdullah N, Liang JB, Huang XD, Ho YW (2011) Effects of condensed tannins from *Leucaena* on methane production, rumen fermentation and populations of methanogens and protozoa *in vitro*. *Animal Feed Science and Technology* **169**, 185–193. doi:10.1016/j.anifeedsci.2011.07.004

Tavendale MH, Meagher LP, Pacheco D, Walker N, Attwood GT, Sivakumaran S (2005) Methane production from *in vitro* rumen incubations with *Lotus pedunculatus* and *Medicago sativa*, and effects of extractable condensed tannin fractions on methanogenesis. *Animal Feed Science and Technology* **123–124**, 403–419. doi:10.1016/j.anifeedsci.2005.04.037

Taylor CA, Harrison MT, Telfer M, Eckard R (2016) Modelled greenhouse gas emissions from beef cattle grazing irrigated leucaena in northern Australia. *Animal Production Science* **56**, 594–604. doi:10.1071/AN15575

Tiemann TT, Lascano CE, Wettstein HR, Mayer AC, Kreuzer M, Hess HD (2008) Effect of the tropical tannin-rich shrub legumes *Calliandra calothyrsus* and *Flemingia macrophylla* on methane emission and nitrogen and energy balance in growing lambs. *Animal* **2**, 790–799. doi:10.1017/S1751731108001791

Tomkins NW, Denman SE, Pilajun R, Wanapat M, McSweeney CS, Elliott R (2015) Manipulating rumen fermentation and methanogenesis using an essential oil and monensin in beef cattle fed a tropical grass hay. *Animal Feed Science and Technology* **200**, 25–34. doi:10.1016/j.anifeedsci.2014.11.013

Vercoe PE, Durmic Z, Revell DK (2009) Rumen microbial ecology: helping to change landscapes. *Optiones Mediterraneennes. A85* **225–236**, 20103102279

Waghorn G (2008) Beneficial and detrimental effects of dietary condensed tannins for sustainable sheep and goat production: progress and challenges. *Animal Feed Science and Technology* **147**, 116–139. doi:10.1016/j.anifeedsci.2007.09.013

Watanabe Y, Suzuki R, Koike S, Nagashima K, Mochizuki M, Forster RJ, Kobayashi Y (2010) *In vitro* evaluation of cashew nut shell liquid as a methane-inhibiting and propionate-enhancing agent for ruminants. *Journal of Dairy Science* **93**, 5258–5267. doi:10.3168/jds.2009-2754

Whitelaw FG, Eadie JM, Bruce LA, Shand WJ (1984) Methane formation in faunated and ciliate-free cattle and its relationship with tureen volatile fatty acid proportions. *British Journal of Nutrition* **52**, 261–275. doi:10.1079/BJN19840094

Williams SRO, Moate PJ, Deighton MH, Hannah MC, Wales WJ, Jacobs JL (2016) Milk production and composition, and methane emissions from dairy cows fed lucerne hay with forage brassica or chicory. *Animal Production Science* **56**, 304–311. doi:10.1071/AN15528

Williams SRO, Chaves AV, Deighton MH, Jacobs JL, Hannah MC, Ribaux BE, Morris GL, Wales WJ, Moate PJ (2018) Influence of feeding supplements of almond hulls and ensiled citrus pulp on the milk production, milk composition, and methane emissions of dairy cows. *Journal of Dairy Science* **101**, 2072–2083. doi:10.3168/jds.2017-13440

Wright AD, Williams AJ, Winder B, Christophersen CT, Rodgers SL, Smith KD (2004) Molecular diversity of rumen methanogens from sheep in Western Australia. *Applied and Environmental Microbiology* **70**, 1263–1270. doi:10.1128/AEM.70.3.1263-1270.2004

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