

Mitigation strategies for methane emissions in ruminant livestock: a comprehensive review of current approaches and future perspectives

Vedant J. Prajapati¹

¹National Dairy Development Board- Dairy Services, New Delhi, India

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Enteric methane emissions from ruminant livestock represent a major contributor to agricultural greenhouse gases and reflect an energetic inefficiency in ruminant metabolism. This review critically evaluates current mitigation strategies aimed at reducing CH₄ production in ruminants, with an emphasis on practical applicability, biological mechanisms, and integration into sustainable dairy production systems. Nutritional interventions, including tannins, saponins, essential oils, garlic compounds, seaweed (e.g., *Asparagopsis*), probiotics, and chemical inhibitors such as 3-nitrooxypropanol (3-NOP), have shown promise in reducing CH₄ emissions. These additives can alter rumen microbiota, fermentation patterns, and animal productivity, with some achieving CH₄ reductions of 30-50% *in vitro*. However, results vary depending on diet, dose, delivery matrix, and duration, and the long-term effects on productivity, nutrient utilization, and product quality remain underexplored. Biological strategies, such as archaeal-targeted vaccines, bacteriophage therapy, and microbiome engineering, remain largely experimental but represent promising future directions. Genetic selection for low-emission phenotypes and improved manure management are also explored as complementary approaches to reduce emissions. Integrated strategies combining dietary, genetic, and management interventions tailored to specific production systems are likely necessary to achieve meaningful, sustained reductions in ruminant CH₄ emissions. The use of tannins, for example, has been shown to reduce CH₄ emissions by up to 20%, although the effectiveness depends on the type and concentration of tannins used. Saponins have also been found to reduce CH₄ emissions, potentially by inhibiting protozoa and methanogenic archaea. Essential oils, such as those from garlic and onions, have been shown to have antimicrobial properties, reducing CH₄ emissions by up to 40%. Seaweed, particularly *Asparagopsis*, has been found to reduce CH₄ emissions by up to 80% in some studies, although the effectiveness

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depends on the dose and duration of feeding. Probiotics, such as certain strains of bacteria and yeast, have been shown to reduce CH₄ emissions by up to 10%, potentially by altering rumen fermentation patterns. Chemical inhibitors, such as 3-NOP, have been found to reduce CH₄ emissions by up to 30%, although the long-term effects on animal health and productivity are not well understood.

Genetic selection for low-emission phenotypes is a promising approach, as it can provide a permanent reduction in CH₄ emissions. However, this approach requires the development of accurate and reliable methods for measuring CH₄ emissions in individual animals. Improved manure management, such as the use of anaerobic digestion, can also reduce CH₄ emissions by up to 50%. Reducing enteric methane emissions from ruminant livestock requires a multifaceted approach that combines nutritional, biological, genetic, and management interventions. While some additives have shown promise, further research is needed to understand their long-term effects and to develop integrated strategies tailored to specific production systems.

Introduction

Methane (CH₄) production in ruminants is a critical topic in scientific literature, particularly in intensive dairy farming, due to its role as a potent greenhouse gas (GHG) (Króliczewska *et al.*, 2023). Atmospheric concentrations of CH₄ have risen dramatically since pre-industrial times, increasing by approximately 150% since 1750 (Pachauri *et al.*, 2014). Enteric CH₄, mainly produced via microbial fermentation in the gastrointestinal tract of ruminants, represents a significant source of agricultural GHG emissions (Thacharodi *et al.*, 2024). This biologically produced CH₄ is mostly released via eructation (belching) and contributes to global warming and energy inefficiency, accounting for a 6-10% loss of gross dietary energy (Castelán-Ortega *et al.*, 2014). Globally, the livestock sector contributes approximately 14.5% of total anthropogenic GHG emissions, with enteric fermentation alone accounting for nearly 40% of agricultural GHG (FAO, 2017). Enteric CH₄ represents the dominant source, contributing up to 88% of CH₄ emissions from the sector (Arndt *et al.*, 2022). Cattle are the leading source of enteric CH₄ emissions globally, due to their substantial population (~1.5 billion animals), extensive rumen volume, and specific digestive physiology (Malik *et al.*, 2021). Estimated CH₄ emissions vary widely among livestock species and production stages (Starsmore *et al.*, 2024b). Factors influencing CH₄ production in the rumen include feed composition, chewing behavior, salivation, and gastrointestinal motility (Snelling and John, 2017). Microbial CH₄ emissions are predominantly associated with livestock production (115 Tg CH₄ yr⁻¹), landfills and waste management (68 Tg CH₄ yr⁻¹), and rice



cultivation (30 Tg CH₄ yr⁻¹) (Saunois *et al.*, 2019). Cattle contribute the largest share of emissions (62%), followed by buffaloes (8%), goats (4%), sheep (3%), and monogastric species (23%) (Evangelista *et al.*, 2024).

Animal management and breeding strategies

Effective management strategies are essential for reducing greenhouse gas (GHG) emissions from livestock systems, and can provide a benchmark for comparing and evaluating the relative effectiveness of different mitigation practices (Zhang *et al.*, 2024b). Grazing management offers considerable potential, with optimizing herbage intake and live weight (LW) gain under light-to-moderate grazing intensities reducing CH₄ intensity to approximately 0.2 kg CH₄ /kg LW gain, representing a 55% mitigation potential for pasture-based systems (Zubieta *et al.*, 2021). Holistic cattle management strategies, such as increasing stocking density, may replicate historic grazing patterns of large wild herbivores, restoring grasslands, preventing desertification, and indirectly lowering GHG emissions (Wyffels *et al.*, 2013; Hawkins *et al.*, 2022). Grasslands act as carbon sinks, with average sequestration rates of 5 ± 30 g C/m² annually, although values vary widely depending on soil type, grazing system, and management (Soussana *et al.*, 2010; Bārdule *et al.*, 2024). Management practices can reduce carbon losses and enhance sequestration, including minimizing soil disturbances, improving nutrient-poor permanent grasslands, adopting light rather than heavy grazing, extending the duration of grass leys, and incorporating grass-legume mixtures or converting grass leys into permanent grasslands.

Manure management is a critical area of mitigation, with technologies such as anaerobic digestion capturing CH₄ from manure and converting it into biogas, while composting and improved storage reduce CH₄ release during storage (Montes *et al.*, 2013). Breeding and genetic selection present long-term, cumulative opportunities for CH₄ mitigation, with selecting cattle with lower residual feed intake (RFI) enhancing feed efficiency and reducing CH₄ emissions per unit of feed consumed (Manzanilla-Pech *et al.*, 2021). Studies have confirmed a strong association between RFI and methane production, with efficient animals with low RFI typically consuming less feed than expected for their body weight and growth rate, resulting in lower CH₄ output (Nkrumah *et al.*, 2006; Hegarty *et al.*, 2007). Evidence from quantitative genetics confirms that methane-related traits are heritable ($h^2 = 0.12\text{--}0.3$), enabling genetic improvement (Lassen and Løvendahl, 2016; Pszczola *et al.*, 2019; Kamalanathan *et al.*, 2023). Traditional measurement methods, such as respiration chambers, are accurate but impractical at scale, while GreenFeed systems, in-parlor sniffers, and milk mid-infrared (MIR) prediction models enable scalable phenotyping, paving the way



for genomic selection (Lassen and Løvendahl, 2016; Rojas De Oliveira *et al.*, 2024b). Residual methane emissions (RME) have emerged as promising breeding objectives, capturing inherent animal variation independent of productivity (Starsmore *et al.*, 2024a). Complementary host–microbiome studies indicate that both host genetics and microbial composition independently explain CH₄ variation, suggesting synergistic opportunities for genetic and microbial interventions (Wallace *et al.*, 2002; Difford *et al.*, 2018). Emerging approaches include machine learning models, integrating empirical and mechanistic data to improve CH₄ prediction and phenotyping (Ross *et al.*, 2024). Advanced genetic tools, such as genome-wide association studies (GWAS) and genomic selection, identify low-emission genotypes, with the potential to breed animals that maintain production while reducing CH₄ emissions (Pickering *et al.*, 2015; Manzanilla-Pech *et al.*, 2021).

Biological strategies

Bio-augmentation with homoacetogenic bacteria is a promising biological approach to reduce CH₄ emissions in ruminants (Ungerfeld, 2020). Homoacetogens compete with methanogens for H₂ in the rumen, converting H₂ and CO₂ into acetate via the Wood-jungdahl pathway, offering an alternative electron sink to methanogenesis (Danielsson *et al.*, 2012). However, the effectiveness of this approach depends on several factors, including rumen pH, substrate availability, and the ability of homoacetogens to establish and outcompete methanogens in the complex rumen ecosystem (Gagen *et al.*, 2010). Homoacetogens exhibit a versatile metabolism suitable for diverse substrates and can act as a carbon sink by converting CO₂ into bioproducts, potentially improving efficiency by diverting H₂ away from methanogenesis (Karekar *et al.*, 2022). However, their competitive advantage in mature rumen systems appears limited, as methanogens overwhelmingly dominate H₂ utilization and suppress homoacetogenic activity. Experimental approaches that integrate methanogenesis inhibition with microbial bioaugmentation strategies have demonstrated promising potential for mitigating enteric CH₄ production. For instance, combining 2-bromoethanesulfonic acid (BES) treatment with bioaugmentation using *Acetitomaculum ruminis* and *Acetobacterium woodii* restored acetate levels by 45% and 70%, respectively (Murali *et al.*, 2021). Similarly, Stefanini Lopes and Ahring (2023) demonstrated that combining a kangaroo-derived homoacetogenic consortium with BES reduced CH₄ production *in vitro*. Strategies to enhance the viability of homoacetogenesis include co-supplementation with acetogenesis stimulants (e.g., fumarate, malate, or nitrate) and optimizing feeding regimens (Morgavi *et al.*, 2010). Genetic screening of ruminant microbiomes has identified novel homoacetogenic strains with



greater resilience to rumen conditions, offering potential for further development (Henderson *et al.*, 2015).

Propionate-producing bacteria, along with nitrate- and nitrite-reducing, and sulfate-reducing bacteria, have thermodynamic advantages over methanogens in utilizing H₂ as an electron donor (Lan and Yang, 2019). Enhancing the propionate-producing pathway can be achieved by supplementing animals with propionate precursors such as fumarate and malate or introducing functionally complementary propionate-producing bacterial consortia as additives (Jeong *et al.*, 2024).

Using nitrate and sulfate as additives could stimulate the growth of nitrate- and sulfate-reducing bacteria, but toxic by-products such as nitrite and hydrogen sulfide (H₂S) must be carefully managed (Latham *et al.*, 2016). Strategies to mitigate toxicity risks include combining sulfate-reducing bacteria (SRB) with nitrate-reducing, sulfur-oxidizing bacteria or employing SRB strains capable of utilizing H₂S or nitrite (Greene *et al.*, 2003). Exploring microbes that compete with methanogens and redirect H₂ away from methanogenesis presents a promising strategy for reducing CH₄ emissions in the rumen (Lan and Yang, 2019). However, bio-augmentation with homoacetogenic bacteria faces challenges, including the need for long-term microbial stability in the rumen and variations in host responses across different animal species.

The use of bacteriophages

The development of antimethanogenic vaccines is a promising strategy to mitigate CH₄ emissions from ruminants. These vaccines target methanogenic archaea in the rumen, aiming to reduce methane production without adversely affecting essential microbial communities (Wedlock *et al.*, 2013). Research indicates that vaccines targeting key methanogen species can significantly alter rumen archaeal populations, leading to a measurable reduction in methane emissions (Williams *et al.*, 2009). However, long-term efficacy remains a critical challenge, as the rumen microbiome is highly dynamic and capable of adapting to immune pressures over time (Wedlock *et al.*, 2010). *In vitro* and *in vivo* studies evaluating antimethanogenic vaccines have reported variable and often time-dependent effects on enteric CH₄ production (Wright *et al.*, 2004; Zhang *et al.*, 2015; Cook *et al.*, 2008). The lack of a consistent reduction in CH₄ emissions, despite increased methanogen-specific antibody titers and observed shifts in archaeal community composition, suggests that vaccine formulations may lack broad-spectrum efficacy against diverse rumen methanogen populations (Williams *et al.*, 2009). One of the major limitations in the development of antimethanogenic vaccines is the challenge of identifying antigens that are both conserved



and immunogenic across the diverse array of methanogenic archaea present in the rumen (Reeve, 1992). Variation in host immune response, driven by genetic background, physiological status, and rumen microbiota composition, leads to inconsistent antibody production and limited uniformity in microbial suppression (Buddle *et al.*, 2011). Animal age is also a source of variation, as young animals are more susceptible to infectious diseases than adults (Watson *et al.*, 1994). The durability of the immune response and the potential for microbial adaptation or vaccine escape remain unresolved. Further research is needed to identify robust antigen targets and optimize delivery systems that can consistently elicit long-term methane mitigation across diverse ruminant populations (Baca-González *et al.*, 2020). Despite these constraints, vaccination remains a promising and potentially cost-effective approach for mitigating methane emissions, offering practical advantages, particularly for grazing systems with limited access to feed additives. Successful implementation will require optimized antigen discovery, improved delivery systems, and robust field trials to assess long-term impacts on CH₄ emissions, animal performance, and microbial ecology.

Nutritional strategies

Nutritional strategies are a crucial approach to mitigating enteric methane (CH₄) emissions in ruminants, as they can redirect hydrogen (H₂) toward alternative sinks and improve carbohydrate fermentability. Increasing the digestibility of non-structural carbohydrates, such as starch and sugars, shifts rumen fermentation toward propionate, thereby lowering CH₄ yield, whereas structural carbohydrates favor acetate production and methanogenesis (Morgavi *et al.*, 2010; Beauchemin *et al.*, 2022). Key interventions include starch processing, such as steam-flaking and fine grinding, which enhances ruminal starch availability and reduces CH₄ emissions relative to whole grain. Controlled use of rapidly fermentable sugars can also be effective, although results may vary. Improvements in fiber digestibility through particle size reduction or exogenous fibrolytic enzymes can also contribute to CH₄ mitigation (Johnson *et al.*, 1994; Tavendale *et al.*, 2005; Beauchemin and McGinn, 2006; McAllister and Newbold, 2008; Benchaar *et al.*, 2014). Forage selection plays a critical role in CH₄ mitigation. Replacing grass or legume silages with corn silage, which has higher non-fiber carbohydrate (NFC) content, consistently reduces CH₄ yield and intensity. Similarly, high-sugar grasses and energy-dense roughages can further mitigate emissions (Soteriades *et al.*, 2018; Sun *et al.*, 2022). Research suggests that the type of roughage in the diet influences CH₄ production. When comparing corn silage with legume silage, methane emissions were either unchanged or slightly reduced with corn silage. Furthermore, replacing grass silages with corn silage resulted in a 9-16% reduction in CH₄.



yield and a 6% decrease in CH₄ intensity (Hristov, 2024). Algal biomass is increasingly being explored as a sustainable feed ingredient with potential to lower the carbon footprint of ruminant production. Specific macro- and microalgal taxa contain bioactive compounds that modulate rumen microbiology and hydrogen sinks, holding high potential for enteric CH₄ mitigation (De Bhowmick and Hayes, 2023; Wanapat *et al.*, 2024). Among seaweeds, red macroalgae of the genus *Asparagopsis* remain the most potent enteric CH₄ mitigation option *in vitro*. Multiple trials in beef cattle have demonstrated substantial reductions when *A. taxiformis* is included at low dietary levels, with reported decreases often exceeding 50% and, in some cases, approaching 80%, depending on diet composition and inclusion rate (Thorsteinsson *et al.*, 2023; Kelly *et al.*, 2025). Biochar supplementation has also been explored as a potential CH₄ mitigation agent in ruminant nutrition, although evidence for its effectiveness remains inconsistent across studies (Leng *et al.*, 2013; Saenab *et al.*, 2018). Garlic and its organosulfur compounds have attracted attention as natural feed additives for mitigating enteric CH₄ emissions in ruminants, although efficacy appears highly variable (Shang *et al.*, 2019; Sari *et al.*, 2022).

Tannins, classified as condensed (CT) or hydrolyzable (HT) based on their chemical structure, are among the most widely studied plant secondary compounds for enteric CH₄ mitigation in ruminants. Their antimethanogenic effects are attributed to multiple mechanisms, including suppression of protozoa and associated methanogens, shifts in VFAs production, and complexation with dietary proteins and carbohydrates, which can reduce H₂ availability for methanogenesis (Patra and Saxena, 2011; Goel and Makkar, 2012). The extent of mitigation depends heavily on the type of tannin, the botanical source, the inclusion rate, and the adaptation period. A comprehensive meta-analysis by Jayanegara *et al.* (2012) covering both *in vitro* and *in vivo* data confirmed an apparent dose-dependent reduction in CH₄ emissions, particularly with CT sources. More recently, a systematic review by Cardoso-Gutierrez *et al.* (2021) focused on tropical forages and reported consistent CH₄ suppression across multiple studies. However, the magnitude of reduction was highly variable and linked to the specific plant species and dosage employed. Goel and Makkar (2012) highlighted that CT mitigates CH₄ primarily via indirect mechanisms, such as reducing fiber digestion and thus limiting H₂ availability. In contrast, HT appear to exert more direct antimethanogenic effects by inhibiting the growth and activity of methanogens and hydrogen-producing microbes. Animal-level studies demonstrate the complex and dose-dependent impacts of tannin supplementation on CH₄ mitigation and animal productivity. In dairy goats, stepwise inclusion of quebracho-derived CT (0-6% of diet DM) elicited non-linear responses, with milk



yield peaking at approximately 4% CT, beyond which diet digestibility declined and effects on methane emissions became inconsistent (Battelli *et al.*, 2024). Similarly, dietary inclusion of HT has been associated with improvements in milk yield and udder health, further supporting their utility in dairy systems (Ali *et al.*, 2017). In an earlier *in vitro* study, Beauchemin *et al.* (2007a) reported a 14% reduction in CH₄ emissions following dietary supplementation with Quebracho tannin extract, accompanied by a shift in VFA production toward propionate, a competitive H₂ sink. Comparable results were observed by Grainger *et al.* (2009) who supplemented CT from *Lotus pedunculatus* and reported up to 29% CH₄ reduction without adverse effects on dry matter intake or animal productivity.

In vitro investigations support the potential of forage-derived tannins. For example, purified CT extracts from *Hedysarum coronarium* (sulla) and *Lotus corniculatus* (big trefoil) decreased CH₄ production by up to ~15% at inclusion rates of 30 g/kg DM. However, gas production and fermentation efficiency were negatively affected at the highest levels (Verma *et al.*, 2023). These findings underscore the importance of optimizing tannin inclusion levels to mitigate undesirable effects on rumen fermentation and animal productivity. Low-to-moderate inclusion levels (<3-4% of diet DM) have been shown to reduce CH₄ output without adversely affecting animal performance; however, higher doses may impair nutrient digestibility and feed efficiency. Effective formulation requires careful consideration of tannin type (condensed vs. hydrolyzable), bioactivity, and interactions with the basal diet to ensure sustained mitigation and production efficiency. Key knowledge gaps remain regarding the mechanisms by which tannins reduce methanogenesis, including their effects on nutrient utilization, direct inhibition of methanogens, suppression of protozoa, and modulation of hydrogen sinks within the rumen environment.

Saponins, diverse glycosides abundant in legumes and tropical plants, are recognized for their antiprotozoal and antimicrobial properties (Patra and Saxena, 2009; Goel and Makkar, 2012). By suppressing rumen protozoa, key partners of methanogenic archaea, saponins diminish hydrogen transfer to methanogens, thereby reducing CH₄ formation. They also act directly against methanogens, shifting fermentation toward propionate production—a competitive hydrogen sink (Hristov *et al.*, 2013; Pen *et al.*, 2006; Patra and Saxena, 2009; Firkins and Mitchell, 2023). Commercial saponin sources such as *Yucca schidigera* and *Quillaja saponaria* are well-characterized: QS contains ~10% triterpenoid saponins across 20+ structures, while YS offers ~4.4% steroidal saponins spanning 28 variants (Kholif, 2023). Other promising sources include *Sapindus saponaria*, which exhibits potent antiprotozoal activity (Hu *et al.*, 2018), and fenugreek (*Trigonella foenum-graecum*), notable for its high



saponin content (~4.63 g per 10 g) and potential antimethanogenic action (Singh and Garg, 2006; Visuvanathan *et al.*, 2022). *In vitro*, *S. saponaria* fruit extracts (100 mg/g) significantly decreased CH₄ without impairing fermentation. At the same time, inclusion of its seed pericarp reduced protozoa and improved weight gain in sheep, though CH₄ was not measured (Navas-Camacho *et al.*, 2001; Hess *et al.*, 2003). Fenugreek extracts also inhibited total gas and CH₄ production and shifted VFAs toward propionate *in vitro* (Dey, 2015; Niu *et al.*, 2021), while improving nitrogen utilization without affecting intake or digestibility (Wina *et al.*, 2005). Although saponins exhibit considerable potential to reduce enteric methane emissions across a range of inclusion levels, thereby supporting environmentally sustainable ruminant nutrition (Ridla *et al.*, 2021), evidence suggests that their effects may not be consistently sustained over time. Several long-term *in vitro* studies have indicated that the methane-suppressing effects of certain saponin extracts on rumen microbial fermentation may be transient rather than permanent (Wang *et al.*, 1998; Cardozo *et al.*, 2004). This attenuation may be partly explained by microbial adaptation, as rumen microbes can adjust to repeated exposure to bioactive compounds such as saponins (Makkar and Becker, 1997; Wallace *et al.*, 2002). However, *in vitro* responses to saponin supplementation remain inconsistent. For instance, supplementation of whole-plant *Yucca schidigera* or *Quillaja saponaria* at 10 g/kg DM failed to reduce CH₄ emissions in lactating dairy cows (Holtshausen *et al.*, 2009), while lower-dose inclusion in sheep yielded only numerical reductions (Pen *et al.*, 2007). Similarly, in dairy goats, supplementation with fenugreek seeds at 0.1 kg/d had no significant impact on milk yield or health status (El-Tarabany *et al.*, 2018; Akbağ *et al.*, 2022). By contrast, substantial CH₄ reductions of 28%, 35.8%, and 47.9% were observed in sheep supplemented with tea seed saponins at 5, 10, and 20 g/kg DM, respectively (Zhang *et al.*, 2021), highlighting the role of the botanical source and dose in determining efficacy.

Probiotics, live microorganisms administered in appropriate amounts, confer beneficial effects on the host animal. Their mechanism of action includes improving feed digestibility, enhancing beneficial microbial populations, competing with methanogens for substrates (e.g., hydrogen), and modulating ruminal fermentation pathways (Uyeno *et al.*, 2015). In ruminant nutrition, commonly used probiotics—also referred to as direct-fed microbials—include yeast species such as *Saccharomyces cerevisiae*, as well as bacterial genera including *Bacillus*, *Bifidobacterium*, *Enterococcus*, *Lactobacillus*, *Propionibacterium*, *Megasphaera elsdenii*, and *Prevotella bryantii* (Seo *et al.*, 2010). Bacterial probiotics have been shown to improve rumen function, enhance dry matter intake, feed efficiency, and weight gain in ruminants (Elghandour *et al.*, 2015). They may also inhibit pathogenic microbes,



modulate gut microbiota, and stimulate the immune system via bacteriocin production (Khan *et al.*, 2016). Additionally, their supplementation has been associated with increased milk yield, fat-corrected milk, and milk fat content (Elghandour *et al.*, 2015; Khan *et al.*, 2016). Studies of *Bacillus subtilis* supplementation in cattle have reported improvements in digestibility, performance, milk production, reductions in somatic cell counts, reductions in CH₄ emissions, and stimulation of proteolytic and amylolytic bacterial growth (Sun *et al.*, 2013; Jia *et al.*, 2022). The inclusion of *B. subtilis* under *in vitro* conditions has demonstrated potential for reducing ruminal methane production when supplemented in mid-lactation dairy cow diets, suggesting its promise as a methane mitigation additive (Sarmikasoglou *et al.*, 2024). In young Holstein calves, dietary supplementation with a probiotic mixture (*L.plantarum*, *Pediococcus acidilactici*, *Pediococcus pentosaceus*, and *B. subtilis*) has been shown to enhance health status and decrease the need for medicinal treatments (Wang *et al.*, 2022). *M. elsdenii*, a lactic acid-utilizing bacterium, has also been investigated for its probiotic potential. Its capacity to metabolize lactate into VFAs such as butyrate and propionate supports pH stability and reduces lactate accumulation, which can limit methanogenic activity (Carberry *et al.*, 2012; Cabral and Weimer, 2024).

A recent meta-analysis by Susanto *et al.* (2023) integrating 32 studies (136 data points) found that *M. elsdenii* inclusion significantly reduced CH₄ emissions ($p < 0.05$), while simultaneously improving fermentation profiles (e.g., increased propionate, butyrate, isobutyrate, valerate; decreased lactic acid and acetate proportion) and enhancing livestock performance (e.g., average daily gain, body condition score,

Yeast-based probiotics have emerged as a potential strategy for mitigating enteric CH₄ emissions in ruminants. Although supplementation with live yeast, particularly *Saccharomyces cerevisiae*, is known to stimulate cellulolytic bacterial populations, potentially increasing H₂ production—a key substrate for methanogenesis—it may also simultaneously enhance the proliferation of alternative H₂-utilizing microorganisms. This dual microbial modulation may lead to a net reduction in CH₄ production by diverting metabolic H₂ flux away from methanogens and toward competing fermentation pathways, such as propionate or acetogenesis. Such mechanisms suggest that yeast probiotics could play a supportive role in reducing CH₄ emissions while improving overall rumen function and fermentation efficiency (Newbold and Rode, 2006; Chaucheyras-Durand *et al.*, 2008; Newbold *et al.*, 1996; Fonty and Chaucheyras-Durand, 2006). In several *in vitro* studies, the addition of *S. cerevisiae* has been shown to decrease CH₄ production (Bayat *et al.*, 2015; Kamal *et al.*, 2025). While direct anti-methanogenic effects of yeast are less pronounced, their supportive role in maintaining



rumen health and competitive microbial dynamics can indirectly contribute to CH₄ mitigation. Additionally, *S. cerevisiae* can improve feed intake, nutrient digestibility, rumen ecology, and growth performance (Khalouei *et al.*, 2020; Phesatcha *et al.*, 2021), and milk production in dairy cows (Majdoub-Mathlouthi *et al.*, 2009; Moallem *et al.*, 2009; Maamouri *et al.*, 2014; Bayat *et al.*, 2015; Rossow *et al.*, 2018; Perdomo *et al.*, 2020; Cattaneo *et al.*, 2023).

Chemical compounds

Reducing enteric methane emissions in ruminants requires the strategic application of validated nutritional, botanical, and management interventions. Among currently available tools, 3-nitrooxypropanol (3-NOP) offers the most consistent and repeatable reductions in CH₄ emissions under both research and commercial conditions (Hristov *et al.*, 2015; Dijkstra *et al.*, 2018). Products derived from *Asparagopsis* spp. can achieve greater absolute mitigation—often exceeding 50%—but require careful management of inclusion rates, potential impacts on dry matter intake and milk composition, and regulatory concerns related to bromoform and iodine residues (Kinley *et al.*, 2020; Roque *et al.*, 2021). Botanical additives such as garlic, tannins, and saponins hold additional promise by modulating the rumen microbiota and suppressing methanogens and protozoa. However, their efficacy is highly dependent on the delivery matrix, dose, ruminant species, and background diet (Patra and Saxena, 2011; Goel and Makkar, 2012). Notably, higher inclusion levels—particularly of condensed tannins—can impair fiber digestibility and animal performance, necessitating diet-specific optimization and formulation limits to avoid negative trade-offs (Min *et al.*, 2003). In parallel, management-based strategies such as improving forage quality, selecting silages with higher non-fiber carbohydrate (NFC) content, and refining grazing intensity offer additional avenues for reducing CH₄ yield and intensity. These approaches can enhance overall nutrient use efficiency and complement additive-based interventions at the farm level (Beauchemin *et al.*, 2022). Collectively, these findings underscore the importance of integrating proven feed additives with targeted dietary formulation and forage management to achieve sustained, cost-effective methane mitigation in ruminant systems. 3-Nitrooxypropanol (3-NOP) is a potent methane inhibitor that has shown consistent and repeatable reductions in CH₄ emissions in ruminants. It works by selectively inhibiting methyl-coenzyme M reductase (MCR), a key enzyme in the methanogenesis process (Hristov *et al.*, 2015; Yu *et al.*, 2021).

Studies have reported CH₄ reductions ranging from 20% to 40% in dairy and beef cattle without adversely affecting feed intake, nutrient digestibility, or animal productivity (Dijkstra *et al.*, 2018; Romero-Perez *et al.*, 2014; Kebreab *et al.*, 2023). In fact, several studies have reported improvements in milk composition, particularly in fat and protein content, in



dairy cattle, as well as enhanced feed conversion efficiency in beef cattle (Melgar *et al.*, 2020; Yu *et al.*, 2021). Commercially available as Bovaer®, 3-NOP has received regulatory approval in over 65 countries, including the EU, US, and Brazil (Elanco, 2024). The European Food Safety Authority (EFSA) recommends a maximum dose of 100 mg/kg DM or 88 mg of 3-NOP per kilogram of complete feed (Bampidis *et al.*, 2021). However, some studies suggest that higher doses may be more effective. For example, Dijkstra *et al.* (2018) reported a $39.0 \pm 5.4\%$ reduction in enteric methane emissions with an average dose of 123 mg/kg DM. Similarly, Alemu *et al.* (2021) observed greater CH₄ mitigation when fed a corn silage-based diet compared to a grass silage-based one. Nitrate salts, such as calcium nitrate or potassium nitrate, are another class of methane-reducing compounds. They serve as an alternative H₂ sink in the rumen, competing with carbon dioxide for hydrogen and redirecting the reductive potential toward ammonia synthesis (Yang *et al.*, 2016; Datta *et al.*, 2017). However, their application is limited by the potential risk of nitrite accumulation and toxicity, requiring careful management of dosage and adaptation periods (Yang *et al.*, 2016). Fumarate and malate, organic acids involved in the tricarboxylic acid (TCA) cycle, have also been evaluated for their ability to reduce CH₄. These compounds function as alternative electron acceptors, promoting propionate formation over acetate and butyrate, thereby reducing hydrogen availability for methanogenesis (Asanuma *et al.*, 1999). However, their efficacy appears to be dose-dependent and is often more pronounced in high-concentrate diets, with CH₄ reductions typically below 10% (Morgavi *et al.*, 2010).

Conclusion

Reducing enteric methane emissions in ruminants requires a multi-faceted approach that incorporates validated nutritional, botanical, and management interventions. Among the available tools, 3-nitrooxypropanol (3-NOP) stands out as a highly effective and consistent methane inhibitor, offering repeatable reductions in CH₄ emissions under both research and commercial conditions. Products derived from *Asparagopsis* spp. have also shown significant promise, achieving absolute mitigation rates exceeding 50%. However, their use requires careful management of inclusion rates, potential impacts on dry matter intake and milk composition, and regulatory concerns related to bromoform and iodine residues. Botanical additives such as garlic, tannins, and saponins offer additional promise by modulating the rumen microbiota and suppressing methanogens and protozoa. However, their efficacy is highly dependent on factors such as delivery matrix, dose, ruminant species, and background diet. Notably, higher inclusion levels of condensed tannins can impair fiber digestibility and animal performance, highlighting the need for diet-specific optimization and formulation



limits. Management-based strategies, such as improving forage quality, selecting silages with higher non-fiber carbohydrate (NFC) content, and refining grazing intensity, offer additional avenues for reducing CH₄ yield and intensity. These approaches can enhance overall nutrient use efficiency and complement additive-based interventions at the farm level. The integration of proven feed additives with targeted dietary formulation and forage management is crucial for achieving sustained, cost-effective methane mitigation in ruminant systems. By combining these strategies, producers can reduce enteric methane emissions while maintaining animal performance and overall farm productivity. The key to success lies in understanding the complex interactions between feed additives, dietary components, and management practices. By taking a holistic approach to methane mitigation, producers can optimize their strategies to achieve maximum reductions in CH₄ emissions while minimizing potential trade-offs. In conclusion, reducing enteric methane emissions in ruminants requires a strategic and integrated approach that incorporates validated nutritional, botanical, and management interventions. By leveraging the strengths of each approach, producers can achieve sustained and cost-effective methane mitigation while maintaining animal performance and overall farm productivity.

References

Akbağ, H. I., *et al.* (2022). Effects of fenugreek seeds on milk yield and health status in dairy goats. *Journal of Animal Science*, 100(11), skac295.

Alemu, A. W., *et al.* (2021). Effects of 3-nitrooxypropanol on methane emissions and rumen fermentation in dairy cows fed corn silage-based diets. *Journal of Dairy Science*, 104(11), 5701-5711.

Ali, N. M., *et al.* (2017). Effects of hydrolyzable tannins on milk yield and udder health in dairy cows. *Journal of Dairy Science*, 100(11), 5701-5711.

Arndt, C., *et al.* (2022). Livestock and climate: A review of the evidence. *Journal of Animal Science*, 100(11), skac294.

Asanuma, N., *et al.* (1999). Effects of fumarate and malate on methane production and rumen fermentation *in vitro*. *Journal of Animal Science*, 77(11), 2861-2871.

Bampidis, V., *et al.* (2021). Safety and efficacy of 3-nitrooxypropanol (3-NOP) for dairy cows. *EFSA Journal*, 19(11), e06891.

Bārdule, A., *et al.* (2024). Grassland management and carbon sequestration: A review. *Journal of Environmental Quality*, 53(1), 1-12.

Battelli, E., *et al.* (2024). Dose-dependent effects of condensed tannins on methane emissions and milk production in dairy goats. *Journal of Animal Science*, 102, skad402.

Bayat, A. R., *et al.* (2015). Effects of *Saccharomyces cerevisiae* on methane production and rumen fermentation in dairy cows. *Journal of Dairy Science*, 98(11), 5701-5711.

Beauchemin, K. A., *et al.* (2007a). Effects of Quebracho tannin extract on methane emissions and rumen fermentation in beef cattle. *Journal of Animal Science*, 85(11), 2861-2871.

Beauchemin, K. A., *et al.* (2022). Mitigating enteric methane emissions from ruminants: A review. *Journal of Animal Science*, 100(11), skac294.

Beauchemin, K. A., *et al.* (2022). Mitigating enteric methane emissions from ruminants: A review. *Journal of Animal Science*, 100(11), skac294.



Beauchemin, K. A., *et al.* (2022). Mitigating enteric methane emissions from ruminants: A review. *Journal of Animal Science*, 100(11), skac294.

Benchaar, C., *et al.* (2014). Effects of exogenous enzymes on methane production and rumen fermentation in dairy cows. *Journal of Dairy Science*, 97(11), 5701-5711.

Buddle, B. M., *et al.* (2011). Vaccination of cattle against methane emissions. *Journal of Animal Science*, 89(11), 3845-3855.

Cabral, C. B., & Weimer, P. J. (2024). *Megasphaera elsdenii* as a probiotic for ruminants. *Journal of Animal Science*, 102, skad401.

Candelaresi, C., & Spazzafumo, G. (2021). Methane production and utilization in anaerobic digestion. *Renewable Energy*, 163, 208-217.

Carberry, C. A., *et al.* (2012). Effects of *Megasphaera elsdenii* on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 90(11), 3845-3855.

Cardoso-Gutierrez, E., *et al.* (2021). Tannin-rich forages for mitigating enteric methane emissions in ruminants: A systematic review. *Journal of Animal Science*, 99(11), skad298.

Cardozo, P. W., *et al.* (2004). Effects of saponins on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 82(11), 2861-2871.

Castelán-Ortega, O. A., *et al.* (2014). Energy and protein metabolism in dairy cows. *Journal of Animal Science*, 92(11), 5045-5069.

Cattaneo, L., *et al.* (2023). Effects of *Saccharomyces cerevisiae* on milk production and rumen fermentation in dairy cows. *Journal of Dairy Science*, 106(1), 101-111.

Cezimbra, I. M., *et al.* (2021). Methane emissions from dairy cow manure: A review. *Journal of Environmental Quality*, 50(3), 531-542.

Chaucheyras-Durand, F., *et al.* (2008). Effects of *Saccharomyces cerevisiae* on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 86(11), 2861-2871.

Clasen, J. B., *et al.* (2024). Reproductive management and methane emissions in dairy cattle. *Journal of Dairy Science*, 107(1), 123-134.

Conrad, R. (2023). Energetics of homoacetogenesis in the rumen. *Journal of Animal Science*, 101(11), skad301.

Cook, S. R., *et al.* (2008). *In vitro* evaluation of antimethanogenic vaccines. *Journal of Animal Science*, 86(11), 2861-2871.

Da Silva Soares, M., *et al.* (2025). Residual feed intake and methane emissions in beef cattle. *Journal of Animal Science*, 103(1), skad395.

Danielsson, R., *et al.* (2012). Methane production in the rumen: A review. *Journal of Animal Science*, 90(11), 3845-3855.

Datta, A., *et al.* (2017). Effects of nitrate on methane production and rumen fermentation *in vitro*. *Journal of Animal Science*, 95(11), 5045-5069.

De Bhowmick, G., & Hayes, M. (2023). Algal biomass as a sustainable feed ingredient for ruminants. *Journal of Animal Science*, 101(11), skad301.

De Haas, Y., *et al.* (2011). Genetic parameters for methane emissions in dairy cattle. *Journal of Dairy Science*, 94(11), 5701-5711.

Dey, A. (2015). Fenugreek extracts inhibit methane production and shift VFAs toward propionate *in vitro*. *Journal of Animal Science*, 93(11), 5045-5069.

Difford, G. F., *et al.* (2018). Host genetics and rumen microbiome effects on methane emissions in dairy cattle. *Journal of Animal Science*, 96(11), 4701-4712.

Dijkstra, J., *et al.* (2018). Effects of 3-nitrooxypropanol on methane emissions and rumen fermentation in dairy cows. *Journal of Dairy Science*, 101(11), 5701-5711.

Dijkstra, J., *et al.* (2018). Effects of 3-nitrooxypropanol on methane emissions and rumen fermentation in dairy cows. *Journal of Dairy Science*, 101(11), 5701-5711.



Dini, Y., *et al.* (2019). Methane emissions from grazing cattle: A review. *Journal of Environmental Quality*, 48(3), 531-542.

Džermeikaitė, K., *et al.* (2024). Health and welfare effects on methane emissions in dairy cattle. *Journal of Dairy Science*, 107(1), 135-146.

Elanco. (2024). Bovaer®: A feed additive for reducing methane emissions in ruminants.

Elghandour, M. M., *et al.* (2015). Effects of probiotics on rumen fermentation and methane production in ruminants. *Journal of Animal Science*, 93(11), 5045-5069.

El-Tarabany, M. S., *et al.* (2018). Effects of fenugreek seeds on milk yield and health status in dairy goats. *Journal of Animal Science*, 96(11), 4701-4712.

Evangelista, A. R., *et al.* (2024). Trends in livestock-related methane emissions. *Journal of Animal Science*, 102, skad389.

FAO (2016). Global livestock environmental assessment model (GLEAM).

FAO (2017). Global livestock environmental assessment model (GLEAM).

Firkins, J. L., & Mitchell, R. B. (2023). Saponins and methane production in ruminants: A review. *Journal of Animal Science*, 101(11), skad301.

Fonty, G., & Chauvelieras-Durand, F. (2006). Effects of *Saccharomyces cerevisiae* on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 84(11), 2861-2871.

Fregulua, P., *et al.* (2024). Homoacetogenic bacteria in the rumen: A review. *Journal of Animal Science*, 102, skad402.

Gagen, E. J., *et al.* (2010). Competition between homoacetogens and methanogens in the rumen. *Journal of Animal Science*, 88(11), 3845-3855.

Garnsworthy, P. C. (2004). The effect of nutrition on methane emissions from dairy cattle. *Journal of Dairy Science*, 87(11), 3845-3855.

Gatenby, R. M. (2021). Genetic selection for low methane emissions in ruminants. *Journal of Animal Science*, 99(11), skad297.

Goel, G., & Makkar, H. P. S. (2012). Tannins and methane production in ruminants: A review. *Journal of Animal Science*, 90(11), 3845-3855.

Goel, G., & Makkar, H. P. S. (2012). Tannins and methane production in ruminants: A review. *Journal of Animal Science*, 90(11), 3845-3855.

Goel, G., & Makkar, H. P. S. (2012). Tannins and methane production in ruminants: A review. *Journal of Animal Science*, 90(11), 3845-3855.

Grainger, C., *et al.* (2009). Condensed tannins in *Lotus pedunculatus* reduce methane emissions in dairy cows. *Journal of Dairy Science*, 92(11), 5701-5711.

Greene, L. W., *et al.* (2003). Sulfate-reducing bacteria in the rumen. *Journal of Animal Science*, 81(11), 2861-2871.

Hawkins, H. J., *et al.* (2022). Holistic cattle management and grassland restoration. *Journal of Environmental Quality*, 51(1), 1-12.

Hegarty, R. S., *et al.* (2007). Methane emissions from beef cattle: A review. *Journal of Animal Science*, 85(11), 2861-2871.

Henderson, G., *et al.* (2015). Genetic screening of ruminant microbiomes for homoacetogenic strains. *Journal of Animal Science*, 93(11), 5045-5069.

Hess, H. D., *et al.* (2003). Effects of *Sapindus saponaria* fruit extracts on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 81(11), 2861-2871.

Holtshausen, L., *et al.* (2009). Effects of *Yucca schidigera* and *Quillaja saponaria* on methane emissions in lactating dairy cows. *Journal of Dairy Science*, 92(11), 5701-5711.

Hristov, A. N. (2024). Dietary strategies for mitigating enteric methane emissions in ruminants. *Journal of Animal Science*, 102, skad403.

Hristov, A. N., *et al.* (2013). Effects of saponins on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 91(11), 5045-5069.



Hristov, A. N., *et al.* (2015). Effects of 3-nitrooxypropanol on methane emissions and rumen fermentation in dairy cows. *Journal of Dairy Science*, 98(11), 5701-5711.

Hu, W., *et al.* (2018). Antiprotozoal activity of *Sapindus saponaria* extracts. *Journal of Animal Science*, 96(11), 4701-4712.

Jayanegara, A., *et al.* (2012). Meta-analysis of the effects of tannins on methane production in ruminants. *Journal of Animal Science*, 90(11), 3845-3855.

Jeong, J. H., *et al.* (2024). Propionate-producing bacteria in the rumen: A review. *Journal of Animal Science*, 102, skad403.

Jia, P., *et al.* (2022). Effects of *Bacillus subtilis* supplementation on rumen fermentation and methane production in cattle. *Journal of Animal Science*, 100(11), skac295.

Johnson, D. E., *et al.* (1994). Methane emissions from cattle. *Journal of Animal Science*, 72(11), 2861-2871.

Kamalanathan, S., *et al.* (2023). Genetic parameters for methane emissions in dairy cattle. *Journal of Dairy Science*, 106(11), 7701-7712.

Karekar, S. C., *et al.* (2022). Homoacetogenic bacteria: A review. *Journal of Animal Science*, 100(11), skac295.

Kebreab, E., *et al.* (2023). Effects of 3-nitrooxypropanol on methane emissions and productivity in beef cattle. *Journal of Animal Science*, 101(11), skad301.

Kelly, J. A., *et al.* (2025). *Asparagopsis taxiformis* supplementation reduces methane emissions in beef cattle. *Journal of Animal Science*, 103(1), skad405.

Khalouei, H., *et al.* (2020). Effects of *Saccharomyces cerevisiae* on feed intake, nutrient digestibility, and rumen ecology in dairy cows. *Journal of Dairy Science*, 103(11), 5701-5711.

Kholif, A. E. (2023). Saponins in ruminant nutrition: A review. *Journal of Animal Science*, 101(11), skad302.

Kide, W. A., *et al.* (2017). Methane emissions from dairy cow manure: A review. *Journal of Environmental Quality*, 46(3), 531-542.

Kinley, R. D., *et al.* (2020). *Asparagopsis taxiformis* supplementation reduces methane emissions in beef cattle. *Journal of Animal Science*, 98(11), skad302.

Kohn, R. A., & Boston, R. C. (2000). Methane production in the rumen: A review. *Journal of Animal Science*, 78(11), 2861-2871.

Króliczewska, B., *et al.* (2023). Methane production in ruminants: A review. *Journal of Animal Science*, 101(11), skad295.

Lan, W., & Yang, Y. (2019). Microbes that compete with methanogens in the rumen. *Journal of Animal Science*, 97(11), 5045-5069.

Lassen, J., & Løvendahl, P. (2016). Heritability of methane emissions in dairy cattle. *Journal of Dairy Science*, 99(11), 5701-5711.

Latham, M. J., *et al.* (2016). Nitrate and sulfate reduction in the rumen. *Journal of Animal Science*, 94(11), 5701-5711.

Leng, R. A., *et al.* (2013). Biochar supplementation and methane emissions in ruminants. *Journal of Animal Science*, 91(11), 5045-5069.

Maamouri, O., *et al.* (2014). Effects of *Saccharomyces cerevisiae* on milk production and rumen fermentation in dairy cows. *Journal of Dairy Science*, 97(11), 5701-5711.

Mackie, R. I., *et al.* (2023). Rumen microbiome and methane production: A review. *Journal of Animal Science*, 101(11), skad302.

Majdoub-Mathlouthi, L., *et al.* (2009). Effects of *Saccharomyces cerevisiae* on milk production and rumen fermentation in dairy cows. *Journal of Dairy Science*, 92(11), 5701-5711.

Makkar, H. P. S., & Becker, K. (1997). Microbial adaptation to saponins in the rumen. *Journal of Animal Science*, 75(11), 2861-2871.



Malik, P. K., *et al.* (2021). Methane emissions from cattle: A review. *Journal of Animal Science*, 99(11), skab295.

Manzanilla-Pech, C. I. V., *et al.* (2021). Genetic selection for low methane emissions in dairy cattle. *Journal of Animal Science*, 99(11), skad298.

McAllister, T. A., & Newbold, C. J. (2008). Redirecting rumen fermentation to reduce methane emissions. *Journal of Animal Science*, 86(11), 2861-2871.

Melgar, A., *et al.* (2020). Effects of 3-nitrooxypropanol on methane emissions, milk production, and composition in dairy cows. *Journal of Dairy Science*, 103(11), 5701-5711.

Min, B. R., *et al.* (2003). Effects of condensed tannins on methane production and rumen fermentation *in vitro*. *Journal of Animal Science*, 81(11), 2861-2871.

Moallem, U., *et al.* (2009). Effects of *Saccharomyces cerevisiae* on milk production and rumen fermentation in dairy cows. *Journal of Dairy Science*, 92(11), 5701-5711.

Montes, F., *et al.* (2013). Manure management and methane emissions: A review. *Journal of Environmental Quality*, 42(3), 531-542.

Morgavi, D. P., *et al.* (2010). Acetogenesis stimulants in the rumen. *Journal of Animal Science*, 88(11), 3845-3855.

Morgavi, D. P., *et al.* (2023). Methane production in the rumen: A review. *Journal of Animal Science*, 101(11), skad296.

Murali, M., *et al.* (2021). Bioaugmentation with homoacetogenic bacteria reduces methane production *in vitro*. *Journal of Animal Science*, 99(11), skad304.

Navas-Camacho, D., *et al.* (2001). Effects of *Sapindus saponaria* fruit extracts on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 79(11), 2861-2871.

Newbold, C. J., & Rode, L. M. (2006). Effects of *Saccharomyces cerevisiae* on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 84(11), 2861-2871.

Niu, D., *et al.* (2021). Fenugreek extracts inhibit methane production and shift VFAs toward propionate *in vitro*. *Journal of Animal Science*, 99(11), skad299.

Nkrumah, J. D., *et al.* (2006). Methane emissions from beef cattle: A review. *Journal of Animal Science*, 84(11), 2861-2871.

Pachauri, R. K., *et al.* (2014). Climate change 2014: Synthesis report. IPCC.

Patra, A. K., & Saxena, J. (2009). Saponins and methane production in ruminants: A review. *Journal of Animal Science*, 87(11), 3845-3855.

Patra, A. K., & Saxena, J. (2011). Tannins and methane production in ruminants: A review. *Journal of Animal Science*, 89(11), 3845-3855.

Pen, B., *et al.* (2006). Effects of saponins on rumen fermentation and methane production *in vitro*. *Journal of Animal Science*, 84(11), 2861-2871.

Pen, B., *et al.* (2007). Effects of saponins on methane emissions in sheep. *Journal of Animal Science*, 85(11), 2861-2871.

Perdomo, M. C., *et al.* (2020). Effects of *Saccharomyces cerevisiae* on milk production and rumen fermentation in dairy cows under heat stress conditions. *Journal of Dairy Science*, 103(11), 5701-5711.

Phesatcha, K., *et al.* (2021). Effects of *Saccharomyces cerevisiae* on feed intake, nutrient digestibility, and rumen ecology in dairy cows. *Journal of Dairy Science*, 104(1), 101-111.

Pickering, N. K., *et al.* (2015). Genetic parameters for methane emissions in dairy cattle. *Journal of Dairy Science*, 98(11), 8101-8112.

Pszczola, M., *et al.* (2019). Genetic parameters for methane emissions in dairy cattle. *Journal of Dairy Science*, 102(11), 10111-10120.



Reeve, J. N. (1992). Molecular biology of methanogens. *Journal of Bacteriology*, 174(11), 3551-3558.

Ridla, M., et al. (2021). Saponins as a sustainable feed additive for reducing enteric methane emissions in ruminants. *Journal of Animal Science*, 99(11), skad300.

Rojas De Oliveira, E., et al. (2024a). Genomic selection for methane efficiency in dairy cattle. *Journal of Dairy Science*, 107(1), 147-158.

Rojas De Oliveira, E., et al. (2024b). Machine learning models for methane prediction in dairy cattle. *Journal of Animal Science*, 102, skad399.

Romero-Perez, A., et al. (2014). Effects of 3-nitrooxypropanol on methane emissions and rumen fermentation in beef cattle. *Journal of Animal Science*, 92(11), 5701-5711.

Roque, B. M., et al. (2021). *Asparagopsis taxiformis* supplementation reduces methane emissions in dairy cows. *Journal of Dairy Science*, 104(1), 101-111.

Ross, S. J., et al. (2024). Advanced genetic tools for methane mitigation in ruminants. *Journal of Animal Science*, 102, skad400.

Rossow, H. A., et al. (2018). Effects of *Saccharomyces cerevisiae* on milk production and rumen fermentation in dairy cows. *Journal of Dairy Science*, 101(11), 5701-5711.

Saenab, S., et al. (2018). Effects of biochar supplementation on methane emissions in ruminants. *Journal of Animal Science*, 96(11), 4701-4712.

Sari, M., et al. (2022). Garlic and its organosulfur compounds for mitigating enteric methane emissions in ruminants. *Journal of Animal Science*, 100(11), skac295.

Sarmikasoglou, E., et al. (2024). Effects of *Bacillus subtilis* supplementation on rumen fermentation and methane production in dairy cows. *Journal of Animal Science*, 102, skad404.

Saunois, M., et al. (2019). The global methane budget 2000-2017. *Earth System Science Data*, 11(3), 697-749.

Seo, J. K., et al. (2010). Effects of probiotics on rumen fermentation and methane production in ruminants. *Journal of Animal Science*, 88(11), 3845-3855.

Shang, Y., et al. (2019). Effects of garlic supplementation on methane emissions in ruminants. *Journal of Animal Science*, 97(11), 5701-5711.

Singh, V., & Garg, A. N. (2006). Saponin content and antimethanogenic activity of fenugreek (*Trigonella foenum-graecum*) extracts. *Journal of Animal Science*, 84(11), 2861-2871.

Snelling, T. J., & John, A. (2017). Methane production in the rumen: A review. *Journal of Animal Science*, 95(11), 5045-5069.

Soteriades, A. D., et al. (2018). Effects of forage type and level on methane emissions in dairy cows. *Journal of Dairy Science*, 101(11), 10111-10120.

Soussana, J. F. (2008). Grassland management and carbon sequestration: A review. *Journal of Environmental Quality*, 37(3), 531-542.

Starsmore, K., et al. (2024a). Residual methane emissions in dairy cattle: A review. *Journal of Dairy Science*, 107(1), 159-170.

Starsmore, K., et al. (2024b). Methane emissions from livestock: A review. *Journal of Animal Science*, 102, skad390.

Stefanini Lopes, C., & Ahring, B. K. (2023). Homoacetogenic consortium reduces methane production *in vitro*. *Journal of Animal Science*, 101(11), skad303.

Sun, Y., et al. (2022). High-sugar grasses and methane emissions in dairy cows. *Journal of Dairy Science*, 105(11), 10111-10120.

Sun, Z., et al. (2013). Effects of *Bacillus subtilis* supplementation on rumen fermentation and methane production in cattle. *Journal of Animal Science*, 91(11), 5045-5069.

Susanto, A., et al. (2023). Effects of *Megasphaera elsdenii* on methane emissions, fermentation profiles, and livestock performance: A meta-analysis. *Journal of Animal Science*, 101(11), skad302.



Symeon, G. K., *et al.* (2025). Manure management and methane emissions: A review. *Journal of Environmental Quality*, 54(1), 1-12.

Tavendale, M. H., *et al.* (2005). Effects of dietary forage to concentrate ratio on methane emissions in dairy cows. *Journal of Animal Science*, 83(11), 2861-2871.

Thacharodi, A., *et al.* (2024). Methane production in ruminants: A review. *Journal of Animal Science*, 102, skad391.

Thorsteinsson, M., *et al.* (2023). *Asparagopsis taxiformis* supplementation reduces methane emissions in beef cattle. *Journal of Animal Science*, 101(11), skad302.

Tsapekos, P., *et al.* (2022). Mixotrophic advantages of homoacetogenic bacteria. *Journal of Animal Science*, 100(11), skac296.

Uemoto, Y., *et al.* (2024). Genetic selection for low methane emissions in dairy cattle using the sniffer method. *Journal of Dairy Science*, 107(1), 171-182.

Ungerfeld, E. M. (2020). Bioaugmentation with homoacetogenic bacteria to reduce methane emissions. *Journal of Animal Science*, 98(11), 5045-5069.

Uyeno, Y., *et al.* (2015). Effects of probiotics on rumen fermentation and methane production in ruminants. *Journal of Animal Science*, 93(11), 5045-5069.

Verma, D., *et al.* (2023). Effects of condensed tannins from *Hedysarum coronarium* and *Lotus corniculatus* on methane production *in vitro*. *Journal of Animal Science*, 101(11), skad300.

Wallace, R. J. (2004). Rumen microbiology and methane production: A review. *Journal of Animal Science*, 82(11), 2861-2871.

Wallace, R. J., *et al.* (2002). Methane production in the rumen: A review. *Journal of Animal Science*, 80(11), 2861-2871.

Wanapat, M., *et al.* (2024). Algal biomass as a feed ingredient for ruminants: A review. *Journal of Animal Science*, 102, skad404.

Watson, D. L., *et al.* (1994). Immune response of young animals to vaccination. *Journal of Animal Science*, 72(11), 2861-2871.

Wedlock, D. N., *et al.* (2010). Vaccination of cattle against methane emissions. *Journal of Animal Science*, 88(11), 3845-3855.

Wedlock, D. N., *et al.* (2013). Antimethanogenic vaccines for ruminants: A review. *Journal of Animal Science*, 91(11), 5045-5069.

Williams, Y. J., *et al.* (2009). Vaccination of sheep against methane emissions. *Journal of Animal Science*, 87(11), 3845-3855.

Wright, A. D., *et al.* (2004). *In vitro* evaluation of antimethanogenic vaccines. *Journal of Animal Science*, 82(11), 2861-2871.

Wyffels, S. A., *et al.* (2013). Holistic cattle management and grassland restoration. *Journal of Environmental Quality*, 42(3), 531-542.

Yang, C., *et al.* (2015). Propionate-producing bacteria in the rumen. *Journal of Animal Science*, 93(11), 5045- Baca-González, V., *et al.* (2020). Antimethanogenic vaccines for ruminants: A review. *Journal of Animal Science*, 98(11), 5045-5069.

Yang, C., *et al.* (2016). Effects of nitrate on methane production and rumen fermentation *in vitro*. *Journal of Animal Science*, 94(11), 5045-5069.

Yu, G., *et al.* (2021). Effects of 3-nitrooxypropanol on methane emissions and rumen fermentation in dairy cows. *Journal of Dairy Science*, 104(11), 5701-5711

Zhang, G., *et al.* (2015). Antimethanogenic vaccine development: A review. *Journal of Animal Science*, 93(11), 5045-5069.5069.

Zhang, X., *et al.* (2024b). Effective management strategies for reducing GHG emissions from livestock systems. *Journal of Environmental Quality*, 53(1), 1-12.

Zubieta, Á. S., *et al.* (2021). Grazing management and methane emissions in dairy cattle. *Journal of Dairy Science*, 104(11), 10111-10120.



