



The Unseen Threat: Intensive Rabbit Farming's Potential Impact on GHG Emissions and the Need for Proactive Mitigation

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Abstract

As the global demand for animal protein continues to rise, intensive rabbit farming has emerged as a significant player in the livestock industry. This article explores the potential environmental impact of intensive commercial rabbit farming on greenhouse gas emissions and global warming. Despite the growing importance of rabbit farming, its contribution to methane emissions from manure and cecal fermentation remains poorly understood. As the industry intensifies and specializes, it may significantly contribute to environmental sustainability concerns. The article challenges the notion that rabbit farming is a minor threat to climate change and highlights the need for research on mitigation strategies. It suggests that rabbit farming's cumulative effect on greenhouse gas emissions may be substantial due to the large numbers of rabbits raised and their rapid reproductive rate. The article calls for further research to quantify rabbit farming's impact on global warming and develop effective mitigation strategies.

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1. Introduction

Rabbits produce cecal methane as part of their digestive process, contributing to greenhouse gas emissions and global warming. Rabbits, with an annual output exceeding 1.2 billion, contribute significantly to greenhouse gas emissions in livestock production that contribute to global warming (Liu *et al.* 2018) ^[135]. The global rabbit population is estimated to be approximately 2-3 billion; however, these figures are based on rough calculations and may not be entirely accurate due to the lack of records in smallholder farming sectors in developing countries. Rabbit livestock production has experienced an average growth of 2.6% in the past decade, with 770 million rabbits reared annually worldwide (FAO-STAT, 2016). Asia accounts for 83% of global rabbit production, followed by Europe at 14%. Africa and the Americas follow closely. Italy, the leading European producer and second world producer of rabbit meat, accounted for 68% of the 107 million rabbits in Europe in 2014. Rabbit farming has become an expanding sector in numerous developed countries, primarily due to increased demand for high-protein animal products driven by global population growth. Rabbit meat is gaining prominence as a highly nutritious and healthful protein source (Kumar *et al.* 2025) ^[127]. In 2017, the global production of rabbit meat was estimated to reach approximately 1,482,000 tons (FAOSTAT, 2019). Consequently, there is now widespread concern regarding the sustainability of rabbit production systems and their environmental impact due to the growth of rabbit populations worldwide and the quantity of rabbits farmed for human consumption (Dinuccio, *et al.* 2018) ^[53].

Intensive rabbit farming generates large amounts of manure, which, if not managed properly, can lead to significant environmental pollution. The expansion and specialization of rabbit farms have been identified as significant contributors to greenhouse gas emission release and environmental contamination (FAO, 2013). This has been attributed to several factors, including increased rabbit populations on farms, inadequate manure management, increased feed production and processing, as well as an increase in animal feces per unit of utilized agricultural area (Cesari, *et al.* 2018) ^[33]. Globally, the growing demand for sustainable land use to optimize animal production while reducing greenhouse gas emissions is now more critical than ever (Waghorn *et al.*, 2002).

This is particularly pertinent considering that human activities account for approximately two-thirds of global methane emissions (Saunois *et al.* 2016), with 41% attributed to agricultural activities such as ruminant enteric fermentation and manure management, among others. Within the agricultural sector, 73% of methane emissions originate from livestock (USEPA, 2013), with beef (35%) and dairy (30%) cattle, and 15% from small ruminants and buffalos (Opio *et al.* 2013). Global warming induced by anthropogenic greenhouse gas emissions has emerged as a significant threat to humanity in recent decades (Ripple *et al.* 2024) ^[193].

Rabbit breeding produces an average of 0.51 Gg of methane and 0.13 Gg of nitrous oxide annually, making it the third highest emitter after sheep (Tonhauzer *et al.* 2023) ^[216]. Free-range rabbit breeding contributes to 0.001 Gg of nitrous oxide, making it crucial to include this category in global emissions reports. Bragaglio *et al.*, (2017) assessed the environmental impact of raising cattle, pigs, chickens, and some fish species using a life cycle approach. In countries with advanced economies, extensive rabbit farming is a specialized form of agriculture. Numerous studies have already been conducted to examine rabbit management, dietary regimens, and genetic factors (Czech *et al.* 2024; Masara *et al.* 2024; El-Nagar *et al.* 2022; Nwachukwu *et al.* 2021; Kurchaeva *et al.* 2020) ^[154, 42, 61, 151, 173], nevertheless, the effects of intensive rabbit farming on carbon footprint are not fully understood. Specifically, there is a paucity of research on the impact of GHG emissions from rabbit farms (Hol *et al.*, 2004, Michl and Hoy, 1996) ^[160].

The intensification of rabbit (*Oryctolagus cuniculus*) husbandry should aim to meet the demand for high-quality protein while ensuring sustainable animal production practices (Paladan, 2022) ^[181]. FAO (2013) has identified the growth and specialization of rabbit farms as a significant contributor to greenhouse gas emissions and environmental contamination. Consequently, it is imperative to implement strategies to reduce these emissions and promote sustainability in rabbit farming. Researchers have posited that improved rabbit management could substantially decrease greenhouse gas emissions and mitigate future climate change. Methane reduction can be achieved through targeted breeding programs and improved feeding strategies. Utilizing natural variations in methane emissions through animal breeding offers a cost-effective, long-lasting, and cumulative solution for mitigation (de Hass *et al.* 2021). Among the various factors influencing total greenhouse gas production and production efficiency, animal feeding has been identified as one of the most significant (Hristov *et al.* 2018).

The global rabbit population's expansion, which stands at approximately 2-3 billion in commercial entities, has led to an increase in rabbit manure, potentially contributing to greenhouse gas emissions if not managed properly (Lukefahr *et al.* 2022) ^[135]. This is in addition to the cecal methane contribution. Livestock manure is crucial for animal agriculture's sustainability, climate change impact, and circular economy (Delin *et al.* 2024) ^[50]. However, improper management can lead to severe environmental issues such as greenhouse gas emissions, water quality impairment, and odor issues, despite its rich nutrient content for crop growth. Manure N loss in agriculture is primarily due to the emission of ammonia, nitrogen dioxide, and nitrate through soil, as well as the leaching of nitrate through the soil (Chadwick *et al.* 2011). Intensification and specialization in livestock production have increased the need for managing livestock manure, which is rich in carbon, nitrogen, and water (Somer

et al. 2013). This manure can release greenhouse gases such as methane and nitrous oxide, which are produced in anaerobic environments and low oxygen availability environments (Glover *et al.* 2023). The emissions from manure management depend on manure composition, oxygen availability, and temperature, with methane produced in strictly anaerobic environments and N₂O produced in environments with fluctuating oxygen availability (Owusu-Twum and Sharara, 2020) ^[175]. The growing global rabbit population and consumption demand necessitate a comprehensive assessment of the environmental impact and sustainability of rabbit production systems from a manure management perspective (Chadwick *et al.* 2015) ^[37]. Manure, which accounts for 31.4 and 64.5% of rabbit feed intake, contributes significantly to NH₃ and GHG emissions (Dinuccio *et al.* 2019) ^[53]. Storage and land application of manure result in cumulative NH₃ and N₂O losses, while manure-OM is lost as CO₂ and CH₄.

2. Uncovering the Role of Intensive Rabbit Farming in Greenhouse Gas Emissions and Global Warming Mitigation

Methane, the second most significant greenhouse gas, has a high global warming potential and emission rate. Herbivorous monogastric animals such as rabbits produce minimal methane, but their large numbers can result in significant production due to their large numbers impacting on environmental footprint. A recent review of CH₄ emissions across various mammals has been provided by Clauss *et al.* (2020) ^[41]. The populations of both ruminant and nonruminant animals are expected to grow in the coming years, leading to an increase in GHG emissions from animal agriculture (Misiukiewicz *et al.* 2021) ^[158]. Globally, approximately 770 million rabbits are raised annually, with production increasing by an average of 2.6% over the past decade (FAO-STAT, 2016). Currently, Asia accounts for about 83% of global rabbit production, followed by Europe at 14%, Africa at 2%, and the Americas at 1%. The worldwide rabbit meat output is estimated at 1,482,441 TEC, equivalent to 971,951 million rabbits slaughtered (Trocino *et al.* 2019) ^[214]. From 1998 to 2017, global rabbit meat production increased by 680,000 TEC (+85%). While ruminant animals are the primary contributors of methane (CH₄), significant GHG emissions, including CH₄, also come from large herbivorous non-ruminants and the substantial population of small farm animals like swine (Patra, 2014) ^[179].

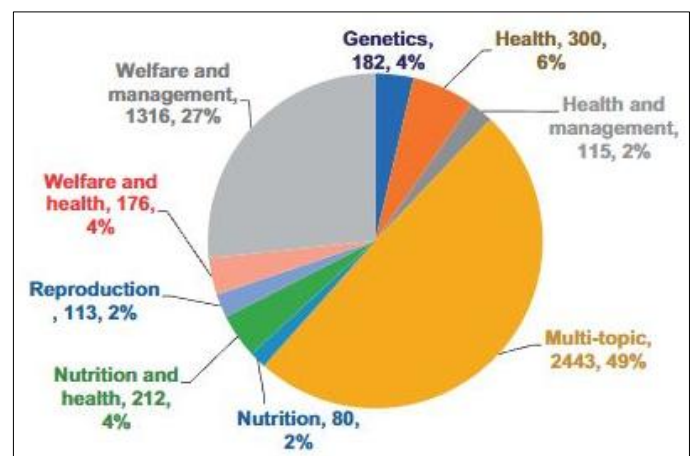


Fig 1: World distribution of research funding (k€ % of the total) for rabbit science by topic (years 2016–2020) (Trocino *et al.* 2018).

It has been observed that all mammals typically produce some amount of CH₄, with certain non-ruminants having CH₄ emission intensities comparable to ruminants (Clauss *et al.* 2020) ^[41]. However, the factors and parameters determining CH₄ production levels in different animal species are not fully understood. Herbivorous monogastric animals such as rabbits generate small amounts of methane, but their large population can result in substantial total production. Over the next three decades, a significant increase in global demand for animal-derived meals is anticipated, particularly in transitioning countries (Friel *et al.*, 2009) ^[80]. Falcon *et al.* (2022) ^[66] predict a 60-70% rise in demand for livestock products by 2050, primarily in developing and economically transitioning nations. Furthermore, the growing global population is increasing the demand for high-protein rabbit meat products, leading to the development of energy-intensive rabbit production technologies (Goswami *et al.* 2025) ^[91]. The study by Trocino *et al.* (2018) shows a global distribution of research funding for rabbit science by topic from 2016 to 2020. (Figure 1). Research funds have not been allocated to address GHG emissions linked to rabbit farming.

Monogastric animals produce small quantities of methane due to incidental fermentation during digestion. Non-ruminant herbivores generate methane at rates between those of monogastric and ruminant animals. Despite lacking a rumen, these animals experience significant fermentation in their large intestine, enabling considerable digestion and utilization of plant material (Murray *et al.* 1978) ^[166]. To address enteric gases and manure handling, it is crucial to understand the mechanisms of CH₄ production in farm animals and the variables affecting such gases. Additionally, comprehending available estimation methodologies and forecasting models for GHG monitoring is essential (Bellarby *et al.* 2013). Improved knowledge of the repercussions of different land use types will facilitate the establishment of efficient mitigation strategies to reduce CH₄ from farm animals (Malik 2015a). Climate change is putting pressure on global agriculture, which is expected to contribute 13.5% of anthropogenic GHG emissions (IPCC, 2007). Rabbit farming is expected to increase to meet the growing demand for animal products, which is expected to quadruple by 2050 due to urbanization, population growth, and rising wages in some areas (Sejian *et al.* 2016) ^[196].

3. Rabbit (*Oryctolagus cuniculus*), as herbivores and omnivores and its implications for methanogenesis.

The domestic rabbit (*Oryctolagus cuniculus*) belong to the leporid family (Leporidae), which comprises mid-sized herbivorous mammals that are hindgut fermenters and practitioners of caecotrophy (Mišta *et al.* 2015). Rabbits has a caecum which is the biggest compartment for fibre degradation and fermentation processes. Rabbits' caecum is the primary fermentation site, where microorganisms convert nutrients from the small intestine into methane, volatile fatty acids, ammonia, and microbial cell compounds (Belenguer *et al.* 2011) ^[20]. Methanogenesis is a process resulting from the metabolic interactions among various microorganism groups in the cecum (Mišta *et al.* 2015). Reductive acetogenesis, the synthesis of acetate from carbon dioxide and hydrogen, occurs primarily in young rabbits, but is partly replaced by methanogenesis with age (Piattoni *et al.* 1996) ^[181]. Caecal fermentation in rabbits has been extensively studied, but methanogenesis has only been reported in a few in vitro studies (Franz *et al.* 2010) ^[77]. In vivo measurements of methane production in adult rabbits showed that methane

production is almost absent before weaning (Marounek *et al.*, 2005; Piattoni *et al.*, 1996;) ^[181, 149]. Reductive acetogenesis is a major characteristic of caecal fermentation in kits, partially replaced by methanogenesis from 6 weeks of age, coinciding with increasing solid food intake.

Research on methanogenesis in rabbits (*Oryctolagus cuniculus*), which are herbivores and omnivores, has primarily relied on in vitro experiments using species-specific gut contents as inoculum (Marounek *et al.*, 2013; Marounek *et al.*, 2000; Tsukahara and Ushida, 2000; Piattoni *et al.*, 1997; Piattoni *et al.*, 1996; Yahav and Buffenstein, 1991; Prins and Lankhorst, 1977) ^[147, 148, 181, 217, 225]. In vivo studies are less frequent (Rodkey *et al.*, 1972; Belenguer *et al.*, 2008; Dufour-Lescoat *et al.*, 1995) ^[191, 59]. The establishment of methanogens in rabbits appears to be gradual. Marounek *et al.* (1999) ^[148] observed methanogenesis beginning at 6 weeks of age in one out of four rabbits. Frans *et al.* (2011) documented methane production and gross energy intake of (0.20 ± 0.10 L d⁻¹ and 0.22 ± 0.08 L d⁻¹) in rabbits and guinea pigs, respectively. As body mass increases, the relative methane production rises, resulting in a higher proportion of ingested gross energy lost. Rabbits, as herbivore monogastric animals, perform post-gastric fermentation due to their functional cecum, which contains microorganisms capable of efficiently digesting fiber components (cellulose and hemicellulose). The caecum hosts a rich community of microorganisms, mainly anaerobic bacteria, that process nutrients from the small intestine, producing short chain fatty acids (SCFA), gases (methane, carbon dioxide, hydrogen), ammonia, and compounds incorporated into microbial cells (Belenguer *et al.* 2011) ^[20]. Figure 2 shows rabbits release methane more than goats, poultry, and pigs, and more equally than sheep. The rabbit farming contribution to greenhouse gas emissions should be a significant concern, necessitating the development of effective mitigation strategies.

Methane emissions from rabbit breeding (enteric fermentation and manure management) averaged 0.51-0.63 Gg per year (Tonhauzer *et al.*, 2023) ^[216]. Nevertheless, not much research has been done on the methane potential of using other organic waste, such as rabbit, goat and horse dung (GM, RM, and HM), in AD (Carabeo Pérez, 2021). From rabbit farming activities, GHG emissions are primarily brought about by animal feed production operations, the fermentation of enteric matter, and the storing, going through processing, and subsequent application of rabbit manure to the soil (Biagini *et al.* 2021). Recently, Clauss *et al.* (2020) ^[41] published a review on the emissions of CH₄ in several animals and acknowledged that the intensity of non-ruminant CH₄ emissions is nonetheless similar to that of ruminant emissions. However, the criteria and variables that influence the amount of CH₄ produced by various animal species are not entirely known. Furthermore, there was no significant variation in CH₄ output (per DM intake of gross calorie intake) with body mass, whereas absolute CH₄ emissions (l/day) scaled linearly with DM consumption. The results indicated that rabbit breeding in households and farms in Slovakia generates an average of 0.51 Gg of methane and 0.13 Gg of nitrous oxide annually. Additionally, when free-range rabbit breeding is considered, emissions are 0.001 Gg of nitrous oxide. (Tonhauzer *et al.*, 2023) ^[216].

Methanogens are part of a microbiome that has developed a deep symbiotic relationship with animals. Clauss *et al.* (2020) ^[41] noted that absolute CH₄ emissions (l/day) correlated linearly with DM intake, while CH₄ yield (per DM intake of gross energy intake) showed no significant variation with

body mass. The differences in methanogenesis and methane release between rabbits and other livestock and poultry species may be attributed to variations in digestive tract microbiome composition (Jensen, 1996, Morvan *et al.*, 1996)^[110, 163] or digesta retention time (Goopy *et al.*, 2014)^[90]. Current understanding also considers the possibility that some herbivore species do not produce methane (Hackstein and Van Alen, 1996)^[94]. Mi *et al.* (2018) discovered relatively low methanogen abundances in the rabbit caecum, potentially due to its acidic pH of 5.8, which is unfavorable for methanogenic archaea but more suitable for acetogens. Abecia *et al.* (2013)^[7] reported almost double the concentration of total methanogens in goat rumens compared to rabbit caecums. The lower methanogen abundances and the availability of alternative H₂ disposal pathways, such as acetogenesis, may contribute to the comparatively low CH₄ emission levels observed in rabbits (Belenguer *et al.*, 2011)^[20].

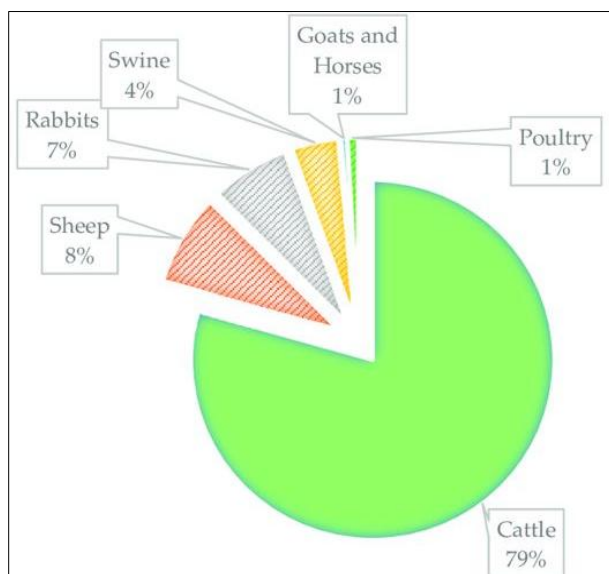


Fig 2: Comparison of greenhouse gas emissions with other livestock (Tonhauzer *et al.* 2023)^[216].

Hare caecal methanogenesis is lower than rabbit caecal methanogenesis, with a lower level (0.25 mmol/kg) and no significant impact of substrate presence (Mišta *et al.* 2015). However, substrate presence increases total gas production during fermentation. The study found differences in microbial activity between hare and rabbit caecum, particularly in methanogenic Archaea. Methanogenic microorganisms are diverse in rabbit caecum and cow rumen, while differences exist in soft and hard feces of rabbits. These findings highlight the importance of understanding microbial populations in fermentation processes. Piattoni *et al.* (1996)^[181] found that CH₄ production in rabbits was almost absent until 32 days of age, then increased significantly with age. Methane is produced during food fermentation in the gastrointestinal tract by methanogenic archaea, known as methanogens. These methanogenic archaea belong to the phylum Euryarchaeota (Moissl-Eichinger *et al.*, 2018)^[160]. Higher yields of methane in horse and rabbit manure may be due to simpler metabolism in the monogastric system, allowing unassimilated material to pass into the excreta. This analysis aligns with previous research on the relationship between animal microbiota dynamics and methane potential. Studies have shown that methanogens are present in the digestive tract of various animals, including rabbits. It is assumed that small herbivores produce negligible amounts of

methane (Miller *et al.* 1986; Danielsson *et al.* 2012; Luo *et al.* 2012)^[157, 45, 137], but it is unclear whether this is a physiological peculiarity or simply a scaling effect (Frans *et al.* 2011). The results confirmed the published data on the presence of methanogens in the digestive tract of rabbits. The study of methanogen composition in the digestive tract and control strategies in nonruminant farm animals is scarce (Misiukiewicz, *et al.* 2020)^[158]. To understand the methanogens, present in domestic animals and their implications in non-ruminant livestock and poultry, further research is needed.

Despite the limited studies on rabbit caecal methanogenesis, some studies have been conducted using in vivo methods (Yang *et al.* 2010; Marounek *et al.* 2005, 2002; Piattoni *et al.* 1996)^[181, 150]. Enteric fermentation in farm animals is an important source of methane, representing 27% of anthropogenic methane emissions (Bousquet *et al.* 2006)^[27]. Therefore, understanding the complexity of the methanogenesis process in the gut and methane-producing archaea is crucial for developing cost-effective methane-mitigation strategies (Buddle *et al.* 2011). Both foregut and hindgut fermenters produce methane (CH₄) as an inevitable by-product during feed fermentation. Franz *et al.* (2011) found that methane production in rabbits and guinea pigs is similar, representing 0.69 ± 0.32 and $1.03 \pm 0.29\%$ of gross energy intake, respectively. However, guinea pigs produced significantly more methane in relation to body mass (BM). The regression indicates linear scaling of methane output, resulting in increasing energetic losses at increasing BM. This leads to an increasing proportion of ingested gross energy being lost due to relative methane production increasing with BM. Although these losses appear too small in non-ruminant herbivores, this relationship may represent a physiological disadvantage with increasing herbivore body size.

4. Digestive System Architecture and Methane Production: A Comparative Study Across Animal Groups

Animals differ in many characteristics of digestive physiology, including the amount of methane (CH₄) they emit per day (Miller and Wolin, 1986, Jensen, 1996)^[110]. Animal digestion types are classified as herbivores (ruminants and hindgut fermenters), carnivores, and omnivores (Figure 2). Many ruminant microbiota profiling studies have focused on cattle because of their importance in the beef and dairy industry (Brulc *et al.*, 2009; Callaway *et al.*, 2010; Jami & Mizrahi, 2012; Welkie, *et al.* 2010)^[31, 34, 109, 209]. The rumen and the hindgut represent two different fermentation organs in herbivorous mammals, with the former producing much more methane than the latter. The anatomy and gastrointestinal physiology of rabbits, guinea pigs, and chinchillas are different than other exotic companion mammals (Kohles, 2023)^[120]. These species are strict herbivores, classified as hindgut (cecum and colon) fermenters, concentrate selectors, and are designed to ingest large amounts of high fibrous food. Their unique gastrointestinal system (Figure 3), with its rapid transit time and ability to differentiate particulates of fiber, allows them to remain small and active, while surviving on high-fiber, low-energy density plant materials. Rabbit breeding produces an average of 0.51 Gg of methane and 0.13 Gg of nitrous oxide annually, making it the third highest emitter after sheep (Tonhauzer *et al.* 2023)^[216]. Free-range rabbit breeding contributes to 0.001 Gg of nitrous oxide, making it crucial to include this category in global emissions report.

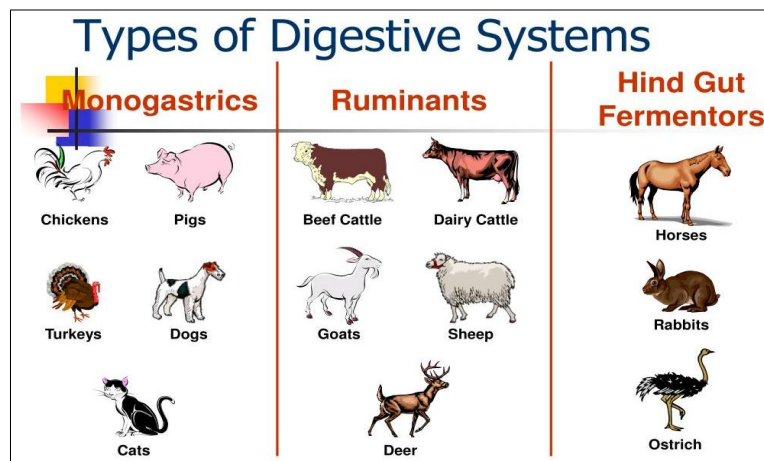


Fig 3: Animal Digestion. AGR Veterinary Science (Credit the University of Kentucky Jul 29, 2017).

The best known example is the general difference between ruminants and non-ruminant herbivores (Franz *et al.*, 2010, 2011)^[77], but why ruminants produce generally more CH₄ is not completely understood. The emissions of methane are determined by the sort of digestive system in alongside the quantity of feed and animal population. Misiukiewicz *et al.* (2020)^[158] alluded to the fact that while ruminants are considered the main source of CH₄ from animal agriculture, non-ruminant animals also contribute substantially, and the CH₄ emission intensity of non-ruminants' species remain comparable to that of ruminants (Figure 3). Means of mitigating CH₄ emissions from enteric fermentation have therefore been sought. Emissions of GHG, including CH₄, produced by both large herbivorous non-ruminants and the large population of small farm animals, such as swine, remain substantial (Patra, 2014)^[179]. Differences in digestive strategies between the two presented species suggest that microbial populations in the caecum could present dissimilar activity, and these distinctions could affect methanogenesis and caecal fermentation patterns.

The longest mean retention times are found in herbivores, in

which digesta are retained and fermented by dense microbial populations in one or more regions of relative stasis (Hume, 2002)^[107]. However, not all herbivores have digestive systems that maximize fiber digestibility; only ruminants, and very large hindgut fermenters achieve this. Instead, many other herbivores (foregut fermenters such as kangaroos and small hindgut fermenters such as rabbits) have digestive systems that sacrifice maximal fibre digestibility for a capacity to process large amounts of forage, even when forage fibre content becomes very high. These different digestive strategies result in the wide range of nutritional niches found among mammals.

In non-ruminants, the caecum and colon are the most important chambers, where structural carbohydrates, which are not digested in the animal stomach and small intestine, are fermented by the local microflora (Místa *et al.* 2016). Rabbits have a large colon and/or cecum are often present in monogastric herbivores in order to facilitate the bacterial fermentation of fiber (Clauss *et al.*, 2003)^[42]. Figure 4 gives a comparative digestive anatomical structure of different classes of animals.

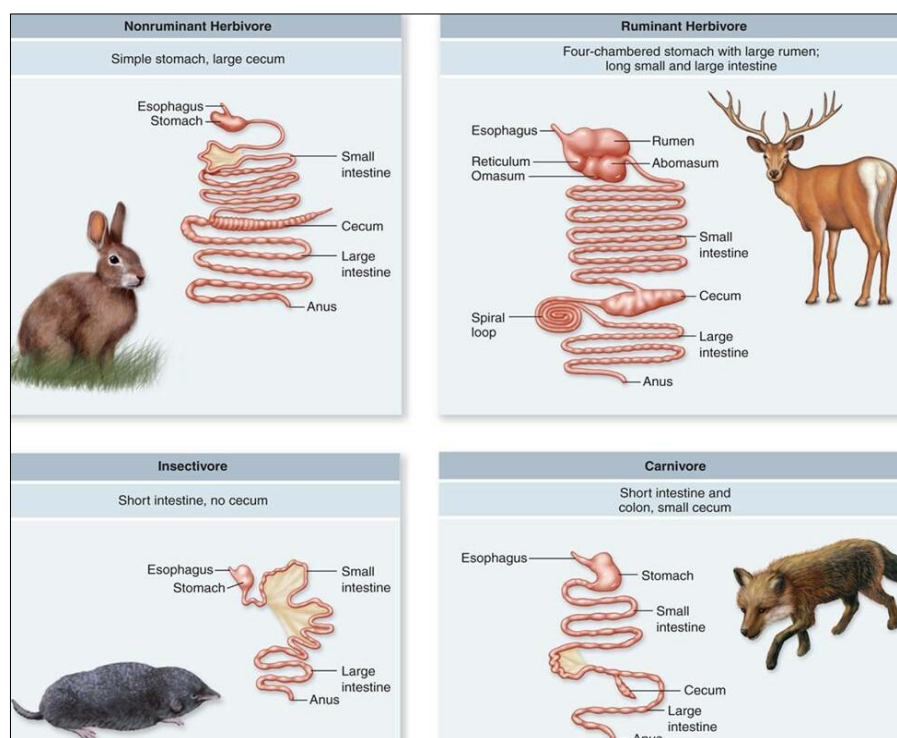


Fig 4: Digestive System Architecture: A Comparative Study Across Animal Groups (Mi *et al.* 2018).

The rumen and the hindgut represent two different fermentation organs in herbivorous mammals, with the former producing much more methane than the latter (Mi *et al.* 2018). Methanogenic microorganisms have been described in the caecum of adult rabbits and are diverse. Upcoming studies employing metatranscriptomics and metabolomics will likely validate the hypothesis and elucidate the elements influencing the distinct microbiota in various rabbit digestive organs. Additionally, the research strongly indicated the existence of novel fibrolytic bacteria in the rabbit cecum, potentially explaining the enhanced fibrolytic activities observed there. Herbivorous mammals lack the ability to produce enzymes necessary for cellulose or hemicellulose digestion. They rely on a symbiotic relationship with a microbial community, primarily bacteria, possessing fibrolytic capabilities in either their foregut (e.g., rumen of ruminants and pseudo-ruminants) or hindgut (e.g., cecum and colon of non-ruminant herbivores) for fiber breakdown (Furness *et al.*, 2015). Microbial fermentation in animal gastrointestinal tracts is a species-specific physiological adaptation, largely determined by the animal's size and dietary habits (Pagan 2011). Given the significance of microbial activity, fermentation chambers often comprise a substantial portion of animal digestive systems, with some small herbivorous mammals having enlarged ceca as their primary fermentation site, leading to their classification as hindgut fermenters. Microbial fermentation led to increased gas production in rabbit cecal samples compared to those of hares. While methanogenesis tended to be lower in hares than in rabbits, considerable individual variation was noted, particularly in hares. These findings suggest that the observed differences in fermentation patterns may not be diet-related but rather stem from species-specific characteristics.

5. Gut Microbiome and Diet Interplay: Implications for Methane Emissions in Monogastric Herbivores and Beyond

In rabbits, the relationship between diet, cecum microbes, and methane production is complex and interconnected (Marounek *et al.* 2013; Mišta *et al.* 2011; Franz *et al.* 2011; Falcão-e-Cunha *et al.* 2004; Garcia *et al.* 2002)^[149, 65, 82]. Diet is considered a key factor shaping microbial populations in all animals, including hindgut fermenters like rabbits (Muegge *et al.*, 2011)^[165]. Rabbits have a digestive system designed to extract nutrients from plant-based foods, leading to high-fiber diets rich in cellulose, hemicellulose, and lignin. These diets are fermented by cecum microbes, producing volatile fatty acids and methane. Conversely, low-fiber diets high in starch or sugars can cause an imbalance in the cecum microbiota, favoring methane production. Herbivores typically exhibit greater gut microbial diversity compared to omnivores or carnivores (Ley *et al.*, 2008)^[130]. Bacterial symbionts enhance host digestion by fermenting cellulose and hemicellulose (Tilg, 2010)^[209]. Both intrinsic factors such as age, sex, genetics, and host phylogeny, as well as extrinsic factors like diet and environmental conditions, can influence the rabbit gut microbiome (Funosas *et al.*, 2021; North, *et al.* 2019)^[81, 168]. Dietary changes can rapidly and significantly affect gut microbiota (Human Microbiome Project Consortium, 2012). As herbivores, rabbits depend on fiber for gastrointestinal health due to their nutritional and physiological characteristics (Ley, *et al.*, 2008)^[130]. Fibrous feeds provide energy through short-chain fatty acids produced by microorganisms in the rabbit cecum.

The cecum of adult rabbits harbors a diverse array of methanogenic microorganisms. The rabbit gut microbial

community consists of 100 1,000 billion microorganisms per gram of cecal content, representing 1,000 different species (Combes *et al.*, 2011)^[43]. Bacteria from the Lachnospiraceae, Clostridiaceae, and Ruminococcaceae families play a crucial role in cellulose and hemicellulose digestion, producing short-chain fatty acids (Biddle *et al.*, 2013). Other families, including Desulfovibrionaceae, Eubacteriaceae, Bacteroidaceae, Christensenellaceae, Erysipelotrichaceae, Rikenellaceae and Spirochaetaceae, have been identified in lagomorphs (North, *et al.* 2019; Shanmuganandam *et al.*, 2020; Stalder *et al.*, 2019)^[168, 205, 197]. The cecum of rabbits is home to a diverse community of microbes, including bacteria, archaea, and protozoa, which are essential for fermenting dietary fiber, producing VFAs, and methane (North, *et al.* 2019)^[168]. Methanogenic archaea, like *Methanobrevibacter* and *Methanosphaera*, are responsible for methane production using hydrogen and carbon dioxide.

Rabbits produce methane as a byproduct of microbial fermentation in the cecum, contributing to greenhouse gas emissions (Hackstein and Van Alen, 1996)^[94]. Dietary factors like fiber content, carbohydrate source, and nutrient levels can also influence methane production, with diets high in fiber producing more methane than those low in fiber (Liu *et al.* (2018)^[135]. The digestion of plant biomass by symbiotic microbial communities in the gut of herbivore hosts also results in the production of methane, a greenhouse gas that is released into the environment where it contributes to climate change (Mi *et al.* 2018). Muegge *et al.* (2011)^[165] compared the microbiota of 33 mammalian species and demonstrated the adaptation of gut microbiota to diet. This obvious will influence the amount of methane released (Alvarenga *et al.* 2017). In addition to hay or forage in the diet rabbits are commonly fed a supplementary food had an effect on the diets total tract digestibility and cecal fermentation patterns. Methanogenic archaea are the exclusive producers of methane, and numerous research teams have examined these symbiotic organisms in herbivore digestive systems.

Methane production has been observed in the feces of most herbivores and some omnivores and carnivores among terrestrial vertebrates (Hackstein and Van Alen, 1996)^[94]. Methanogenesis is considered a primitive trait shared by reptiles, birds, and mammals (Mackie *et al.*, 1999)^[140]. The breakdown of plant matter by symbiotic microbial communities in herbivore guts also generates methane, a greenhouse gas that contributes to climate change when released into the atmosphere (Mi *et al.* 2018). Liu *et al.* (2018)^[135] discovered that decreasing alfalfa particle size led to reduced archaea diversity, increased abundance of *Methanobrevibacter*, and decreased *Methanosphaera*. Finer particle sizes favored *Methanobrevibacter* growth, promoting rabbit growth performance. Since methanogenic archaea are the sole source of methane, various research groups have focused on studying the population dynamics of these symbiotic organisms in herbivore digestive tracts. Microbial fermentation of carbohydrates produces gases such as carbon dioxide, hydrogen, and methane, with methanogenic Archaea being one of the microbial groups competing for metabolic hydrogen uptake (Belenguer *et al.* 2011; Abecia *et al.* 2013)^[20, 7]. The extent of methanogenesis in an animal's gastrointestinal system is influenced by several interconnected factors specific to each species' intestinal ecosystem, as well as individual characteristics, current physiological state, and diet (Christl *et al.* 1992; Franz *et al.* 2010; Hook *et al.* 2010;)^[77, 40, 101].

The diversity of animal microbiota communities is thought to be influenced by various factors, including habitat, diet, and

potential genetic and immunological differences (Ley *et al.*, 2008) ^[130]. The mutually beneficial relationship between gut microbiota and the host is essential for survival and plays a significant role in methane production. Methanogens, which are archaea found in animal digestive systems, contribute to fermentation processes and generate methane (Vogel *et al.* 1980). Since the variety of methanogen communities is believed to explain CH₄ production differences among nonruminant animals, it is crucial to examine the archaeal composition in specific species. The type of diet (herbivorous, carnivorous, or omnivorous) and host phylogeny affect microbiota diversity, with herbivores showing the most diverse microbiota (Mackie, 2002) ^[140].

Studies have shown that methanogenic Archaea and acetogenic bacteria compete for metabolic hydrogen in animal digestive tracts, resulting in increased methane production as animals age (De Graeve *et al.* 1994) ^[46]. In hindgut fermenters, bacterial populations are more abundant in the hindgut than the foregut, as microbial fermentation primarily occurs in the hindgut (Stevens & Hume, 1998) ^[221]. Hydrogen produced in the intestine through anaerobic fermentation of undigested carbohydrate polysaccharides serves as a substrate for intestinal methanogens to produce methane (Triantafyllou *et al.* 2014) ^[213]. Differences in microbial activity may explain the observed variations in hindgut fermentation patterns among monogastric herbivores (Mišta *et al.* 2016). Studies have revealed that the rabbit cecum contains a higher proportion of acetogens, including those from the genus *Blautia*, order Clostridiales, and family Ruminococcaceae (Mi *et al.* 2018). While bacteria dominate the microbial population in the rabbit caecum, methanogenic Archaea also play a vital role, contributing up to 22% of total microbial RNA.

The varying presence of hydrogen-metabolizing bacteria likely accounts for the differences in methane output between the rumen and cecum. Kušar and Avguštin (2010) ^[226] suggested that the methanogenic community in the rabbit's caecum is unique, with low complexity and few dominant species, primarily dominated by *Methanobrevibacter* sp. In the equine hindgut, *Ruminococcus*, *Sporobacter*, and *Treponema* are common genera (O'Donnell *et al.*, 2013) ^[170], which are also prevalent in other hindgut fermenters such as chinchillas, rabbits, miniature ponies, and donkeys. *Fibrobacter* has been identified as an important genus in hindgut-fermenting equids, consistent with previous findings (Shepherd *et al.*, 2012) ^[198]. To date, no research has used next-generation sequencing techniques to compare the fecal microbiota across various common domesticated ruminants and hindgut fermenters. Michelland *et al.* (2010) ^[155] demonstrated differences between the archaeal communities in cow rumens and rabbit soft and hard feces. In rabbits, the ratio of dietary starch to fiber did not impact methanogen diversity or density (Zhu *et al.*, 2015) ^[3], but smaller fiber particle sizes enhanced CH₄ production (Liu *et al.*, 2018) ^[135]. The interplay between diet, cecum microorganisms, and methane production in rabbits is intricate and reciprocal. Dietary factors shape the makeup and function of cecum microbes, which subsequently influence methane generation. Grasping these connections can guide efforts to reduce methane emissions in rabbit farming. Dietary strategies, such as incorporating methane-inhibiting substances or adjusting fiber levels, may contribute to lowering methane output. Additional studies are necessary to elucidate these interactions and create effective methane mitigation approaches.

6. Intensive Rabbit Production and Manure Generation: Implications for Greenhouse Gas Emissions and Climate Change

The production of manure in commercial rabbit farming is associated with increased greenhouse gas emissions, potentially contributing to global warming. Manure storage, processing, and application to soil, along with animal feed production and enteric fermentation, are the main sources of gaseous emissions (Tonhauzer *et al.* 2023) ^[216]. These emissions include unpleasant odors and gases such as ammonia (NH₃), methane (CH₄), and nitrous oxide (N₂O) (Biagini *et al.* 2021). While manures are used for plant nutrition, they also provide substrates for microbial production of nitrogen dioxide, carbon dioxide, and methane. Manure is composed of animal excrement, bedding materials, feed, wash water, and other waste products from animal operations (Liu *et al.* 2012). A study by Calvet *et al.* (2011) on greenhouse emissions in rabbitry revealed average ammonia emissions of 55.9 and 10.2 mg/h per animal, influenced by temperature and relative humidity.

Carbon dioxide emissions were measured at 12588 mg/h per animal for does and 3341 mg/h for fattening rabbits. Nitrous oxide emissions were 10.3 mg/h per animal, with negligible emissions from fattening rabbits. The characteristics of fresh manure determine methane production levels, with rabbit manure generating 2.70 mg g⁻¹ of methane. Biogas production is considered the most effective method for reducing methane from manure (Mazurkiewicz, *et al.* 2023) ^[152]. The IPCC methodology 2021 Refinement (IPCC.2021) establishes a default emission factor for manure management at 0.08 kg CH₄ per animal, surpassing the nationally specific emission factor by 99%. Globally, livestock farming systems produce significant amounts of manure, with an estimated annual nitrogen content ranging from 81.5 to 128.3 Tg (FAO, 2020).

Intensive farming practices pose significant challenges due to inadequate manure and sewage management, potentially leading to climate change, ecosystem eutrophication and acidification, biodiversity loss, and human health risks (Schmeller, *et al.* 2020) ^[194]. Although manure management systems and mitigation strategies have reduced nitrogen emissions, assessing their impact in intensive rabbit farming is challenging due to a lack of official published data. Greenhouse gases are generated and emitted at various stages of the manure management continuum, including rabbitry, manure stores, treatment, and manure spreading. In some countries, manure management can account for over 50% of total national agricultural emissions of nitrogen dioxide and carbon dioxide (UNFCCC, 2021). The intensification and specialization of rabbit production increase the volume of manure to be managed, resulting in greenhouse gases like methane and nitrous oxide (Sommer and Feilberg, 2013) ^[202]. Rabbit farm management decisions interact with environmental factors such as temperature and water availability, affecting key microbial processes and emissions throughout the manure management continuum. These emissions are influenced by microbial activities in the manure environment and depend on manure composition, local management practices, and ambient climatic conditions. There are differentiated approaches to the diversity of ruminant and non-ruminant production systems and their manure management with increasing intensification levels (Malomo *et al.* 2018) ^[142]. Research has examined greenhouse gas (GHG) reduction strategies for solid and liquid manure management systems across various animal species, noting potential interactions between pollutants and

management practices. Accurately modeling gas emissions from these systems remains challenging due to the diversity in farming methods that affect manure's physical and chemical properties (Owen and Silver, 2015)^[174]. In animal slurry storage, a complex interplay of physical, chemical, and microbial processes influences the release of NH₃, N₂O, and NO. Higher temperatures significantly increase ammonia and methane emissions (Wang *et al.* 2021). As rabbit farming becomes more intensive, concerns arise about manure management and the adoption of emission reduction techniques by farmers and households in breeding operations. This data is crucial for national entities preparing emission inventory reports, improving inputs for GHG and air pollutant inventories, and addressing other regulatory issues such as veterinary considerations and animal welfare ((UNFCCC, 2021). The animal feeding and health strategy aims to increase production per animal to meet market demand while reducing the total number of animals bred, thus minimizing waste generation (Steinfeld and Gerber, 2010)^[85]. This approach involves creating diets tailored to animals' specific nutritional requirements for optimal growth and productivity. The impact of synthetic amino acid supplements on gas emissions from animal manure, especially in developing nations, has not been thoroughly studied. A study found that these supplements reduced ammonia and total nitrogen levels in freshly excreted manure (Xiao *et al.* 2023)^[223].

7. Minimizing Methane Emissions in Intensive Rabbit Production: A Review of Promising Approaches in both cecal fermentation and manure management.

In intensive rabbit production, it is more important more than ever to investigate and put into practice efficient methane mitigation techniques, such as feed manipulation, use of feed additives and breeding programs. For rabbits cecal fermentation is the source of methane emissions, necessitating strategies to reduce methane output for ecological balance. Various approaches have been tested for ruminants, but their applicability to rabbits remains unexplored. These strategies include vaccination, enzyme inhibitors, phages, feed supplements, animal breeding, feed processing, ration alteration, defaunation, supplementation of fatty acids, halogenated methane analogues, ionophores, microbial feed additives, non-ionic surfactants, and sulphates (Gerber *et al.* 2013; Hammond *et al.* 2011; Singh, 2010; Agrawal and Kamra, 2010; Chagunda *et al.*, 2009; Puchala *et al.*, 2005; Kamra *et al.*, 2004; Hess *et al.*, 2003; Johnson *et al.* 2000)^[85, 95, 200, 9, 99, 112, 38]. Research has demonstrated the potential of plant secondary metabolites (PSM) as alternatives to chemical feed additives, as they can modify rumen fermentation and decrease CH₄ production. Methane reduction from rabbit manure is also achievable, with promising studies conducted on rabbits. As methane emission from rabbits diminishes nutrient utilization efficiency, manipulating rumen microbial ecosystems presents a potential strategy to minimize cecal fermentation.

7.1 Dietary manipulation and feed additive as possible mitigation strategy in rabbit nutrition to minimize methane production

Franz *et al.* (2011) in their study involving rabbits and guinea pigs found that while small herbivores produce negligible amounts of methane, it is unclear if this is a physiological peculiarity or a scaling effect. The rabbits consumed a higher amount of dry matter (DM) and digestible DM, while the guinea pigs consumed a higher amount. Methane production

was similar for both species, representing 0.69 ± 0.32 and $1.03 \pm 0.29\%$ of gross energy intake, respectively. However, guinea pigs produced significantly more methane compared to rabbits. Khan *et al.* (2017) studied the nutrient composition, digestibility, and methane emission potential of tropical forages used in rabbits. They evaluated seven fodder species and seven grass species at different maturity stages. The study found that fodder species, particularly T. alexandrinum, had higher fiber digestibility and less CH₄ production compared to grass species. Harvest maturity significantly impacts rabbit nutrient supply and CH₄ emission to the environment, making these fodder species ideal for rabbit feed as forage sources. Therefore, it is encouraged to produce high quality fodder with high digestibility to minimize methane gas emission from intensive rabbit production.

To reduce methane nutritionally, choose feed ingredients that shift volatile fatty acid (VFA) production from acetate to propionate (Tseten *et al.* 2022)^[216]. The nutritional intervention method or nature could potentially reduce up to 70% of ruminant methane emissions. Increase passage rate to change rumen microbial population and change VFA production. Feed better-quality diets to increase production and dilute methane-per-animal total. Research on increasing passage rate and improving feed efficiency can help identify suitable feed additives for methane mitigation (Sara *et al.* 2022). Changes in feed composition can be a simple and cost-effective method for reducing enteric methane levels through dietary manipulation.

For ruminants feed additives have proved to reduce methane production from the rumen (Dijkstra, *et al.* 2025; Hristov, 2024; Hegarty *et al.* 2021)^[103]. The use of natural feed additives like prebiotics, probiotics, organic acids, bacteriocins, and phytobiotics as dietary antibiotic alternatives in rabbit nutrition is being explored to meet customer demand. The focus has not been largely on the use of feed additives to reduce rabbit gas emissions. Most effort in feed additive research was targeted at enhancing growth, intestinal health, and carcass and meat quality, and also improving intestinal health (Belhassen, 2023; Abdel-Wareth *et al.* 2021; Mancini and Paci, 2021; Pogány Simonová *et al.* 2020)^[18, 183, 21, 6].

Some studies suggest that additives can enhance feed intake and conversion efficiency in rabbits, this alone potentially leading to a reduction in methane levels. Research indicates that specific acids can enhance feed conversion in rabbits. Kamal *et al.* (2008)^[114] discovered that malic, fumaric, and citric acids improved feed conversion. Amber *et al.* (2004)^[10] found that probiotic supplementation positively impacted feed conversion but had no effect on mortality rates. Kritas *et al.* (2008)^[122] noted higher gains and better feed conversion ratios in rabbits given probiotics. Bhatt *et al.* (2017)^[24] observed superior feed conversion ratios in probiotic-supplemented rabbits. However, Yamani *et al.* (1992)^[226] found that feeding a pelleted diet with probiotics did not improve feed conversion in New Zealand White rabbits.

Sara *et al.* (2028) reported that feed additives (enzymes, organic acids and probiotics) led to improved feed conversion, while Fathi *et al.* (2019)^[74] observed that dietary supplementation with probiotic (400 g/ton) significantly increased feed intake compared to the control group. Ezema and Eze (2010)^[63] and Bhatt *et al.* (2017)^[24] noted that probiotics positively affected feed intake in growing rabbits. Oso *et al.* (2013)^[172] found that rabbits fed probiotic-supplemented diets had significantly higher feed intake than their control counterparts. They proposed that the

improvement in feed efficiency of rabbits receiving probiotic-supplemented diets could be attributed to increased beneficial gut microflora and enhanced nutrient digestion and absorption. Cachaldora *et al.* (2004)^[33] reported that enzyme supplementation beneficially affects feed efficiency in fattening rabbits.

Despite the potential of methane-reducing feed additives, their effectiveness has not been thoroughly investigated in intensive rabbit farming. Instead, the focus has been on improving productive traits, including reducing mortality through gut health, and enhancing growth, carcass, and meat quality in rabbits. A limited number of studies on feed additives have shown some positive outcomes. Marounek *et al.* (1997) examined various antibiotics for methanogenesis and fermentation in vitro using rabbit caecal contents. While ionophores typically decrease CH₄ in the rumen by reducing H₂ production, their study found that ionophore antibiotics (maduramicin, monensin, lasalocid, and salinomycin; commonly used as anticoccidials) stimulated caecal methanogenesis in rabbits. It has been suggested that these ionophores may inhibit H₂-dependent acetate formation, leading to increased H₂ availability for methanogens (Marounek *et al.*, 1997). In a related study, Piattoni *et al.* (1998)^[181] investigated the effects of monensin and bromoethansulfonic acid on rabbits.

In fasted rabbits, bromoethansulfonic acid addition (1 mM) reduced CH₄ by 14% without altering the fermentation pattern, while monensin decreased CH₄ by 51%, accompanied by a 29% reduction in total volatile fatty acids (VFA) production, primarily affecting butyrate (37%) and acetate (33%). However, bromoethansulfonic acid (20 mM) depressed CH₄ markedly by 93% in the unfasted rabbits, while monensin increased CH₄ by 56% and led to a decreased total level of VFA of 16%, mainly butyrate (34%). As the addition of monensin did not decrease acetogenic or reductive acetogenic bacteria, the increase in CH₄ was suggested to be due to a depression of autotrophic activity, with more H₂ being available for methanogenesis. In nonruminant animals, the use of plant secondary metabolites in mitigating methanogenesis found an application in the study of Chen *et al.* (2014). Pogány Simonová *et al.* (2015)^[184] found that *Enterococcus faecium* CCM7420 strain administration improved rabbit growth performance, possibly affecting gut function and nutrient uptake.

7.2 Mitigating of greenhouse gas emission through proper manure management in intensive rabbit farming

Montes *et al.* (2013)^[163] outline various approaches to decrease N₂O and CH₄ emissions during storage. When properly managed, composting can effectively mitigate CH₄ emissions from solid manure storage. Samer (2015)^[192] noted that while adding straw to solid manure reduces CH₄ emissions, it may sometimes increase N₂O emissions. Rivera and Chará (2021)^[192] suggest that the most effective methods for reducing N₂O emissions from manure deposits include: (i) maintaining anaerobic conditions in deposits (e.g., by ensuring they are tightly packed and covered); (ii) transitioning from deep-bed systems to liquid manure systems (although this requires more water); and (iii) incorporating straw to break down ammonium. In livestock systems where animals are confined continuously, periodically, or year-round, proper manure management, including handling, storage, and application, is crucial for mitigation.

To reduce greenhouse gas emissions from animal manure, the following actions are necessary: minimizing air exposure and

composting manure reserves to reduce methane emissions, and using urease inhibiting agents to slow down or prevent the conversion of urea (found in animal urine and manure) to nitrous oxide (Chadwick *et al.* 2011). The composition of animal diets influenced greenhouse gas emissions from manure, with animals fed a 0.25% SB diet producing significantly lower emissions ($p < 0.05$): 30% less ammonia (NH₃), 25% less methane (CH₄), 9% less nitrous oxide (N₂O), and 8% less carbon dioxide (CO₂) compared to the control group. Biagini *et al.* (2021) found that a feed additive derived from a water-soluble biopolymer (SB) obtained from urban gardening waste effectively reduced ammonia and greenhouse gas emissions from rabbit manure.

The Min *et al.* (2020) reported that enhancing animal diets and incorporating feed additives can significantly reduce methane emissions from enteric fermentation and manure storage. Research indicates that the production and emission of CH₄ and N₂O from manure are influenced by factors like feed digestibility, animal type, handling methods, and environmental conditions like sunlight, humidity, precipitation, and wind (Gupta *et al.* 2007; Brown *et al.* 2002; Gaur *et al.* 1984)^[30]. Petersen *et al.* (2013)^[93] note that due to the biological nature of GHG emissions and the unique properties of manure, emissions can be controlled through appropriate handling, treatment, and storage practices. The study suggests that promoting sustainable rabbit farming practices, including proper manure management and utilization, can significantly reduce environmental impact.

Methane Emission in Rabbits: Selection, Genetic Characteristics and Breeding Opportunities

Selecting animals emitting lower methane (CH₄) is an effective method to reduce CH₄ emissions, as genetic progress is cumulative over generations (Manzanilla-Pech, *et al.* 2021)^[144]. Combining data from different countries can expedite accurate genetic parameters for CH₄ traits and build a future genomic reference population, as few countries actively record CH₄. The traditional selection approach for ruminants has been based on methane emissions expressed as a proportion of feed intake (MY), which was previously perceived to be free from association with feed intake or body weight (Herd *et al.*, 2014; Donoghue *et al.*, 2016)^[98, 20]. However, this approach has been disputed due to its unpredictable response to other economic traits in beef production (Pickering *et al.*, 2015). A negative phenotypic correlation between MY and feed intake has been observed across both concentrate and forage-based diets under ad libitum feeding conditions using the GreenFeed emissions monitoring system (Bird-Gardiner *et al.*, 2017; Renand *et al.*, 2019)^[25, 188]. Breeding rabbits specifically for reduced methane production has part of rabbit breeding objectives has not been explored, possibly due to challenges in quantifying methane output from their hindgut fermentation. The target of the breeding programs in rabbits were carcass and growth characteristics, to improve efficient meat production, improved reproduction and animal health (Elfadil, *et al.* 2023; Ouyed, *et al.* 2008)^[61, 173]. Unknowingly productive performance with special reference to residual feed intake has been improved and this has implication for ceecal methane release (Fathi, *et al.* 2023).

Selection programs use various methodologies to estimate selection responses (König and Swalve. 2009)^[121]. One approach involves regressing breeding values on generations, based on genetic parameters and the model used. Other methodologies use a control population, either unselected or diversely selected, to estimate selection responses. In most

rabbit breeding programs, sire lines are selected based on post-weaning daily weight gain or weight at the end of the fattening period. This is because heavier market weight is crucial for profitability in rabbit farms, as it allows for selling more kilograms of rabbit and increasing profit (Abel-Azizi, 1998). This has potential reduction in ceecal emission. Nevertheless, such efforts would be based on genetic improvement programs that consider phenotypic and genetic correlations of traits related to gas emissions (Lynch and Walsh, 1998) [138]. Accurate breeding value (BV) estimation requires evaluating animals using production records adjusted for non-genetic factors (Lush 1994). Various ruminant species have shown a wide range of heritability and genetic correlations for CH₄-related traits (Donoghue *et al.*, 2020; Manzanilla-Pech *et al.*, 2010; Pinares-Piães *et al.*, 2013) [182]. Donoghue *et al.* (2020) [20] studied methane and carbon dioxide output rates in Angus cattle, finding h² values of 0.53 and 0.31 for CPR and MPR, respectively. The higher h² of 0.53 for CPR indicates a stronger genetic basis and potential for genetic response. When h² exceeds 0.30, selection based on individual performance allows for greater genetic improvement (Falconer and Mackays, 1996) [67]. For traits with low heritability, crossbreeding is recommended to select genetically superior animals. Crossbreeding is preferred for advancing low heritability traits as it capitalizes on heterosis but does not enhance highly heritable traits. Understanding the relationships between CH₄ emissions and other economically important production traits is crucial for developing effective selection strategies for low CH₄ yields. Heritability, a key aspect of quantitative biology, positively influences selection (Sood *et al.* 2020) [204]. The phenotypic or genetic correlations between CH₄ and other traits may impact selective breeding for low emissions. Recent studies have begun to explore these associations (Lassen and Lovendahl, 2016; Zetouni *et al.*, 2018) [2]. In animal breeding, selecting genetically superior individuals based on their phenotypic values and those of their relatives is the most economical approach. Most economically important traits are complex and controlled by multiple genes. Accurate CH₄ measurement is essential for determining useful genetic parameters like heritability. Inaccurate metrics in genetic evaluation for low CH₄ emissions can increase error variance, leading to incorrect genetic parameters. A significant selection response for methane traits is more likely when there is substantial additive genetic variance among individuals, which is the raw material for repeatability and heritability. In such cases, genetic selection for reduced CH₄ production could be feasible (Pinares-Piães *et al.* 2013) [182]. However, genetic evaluation for low CH₄ may be ineffective due to the various methods of CH₄ determination influenced by study objectives (de Haas *et al.* 2017).

Some studies target the amount of CH₄ that cows release; hence, the CH₄ production pattern may be represented in grams per day (Garnsworthy *et al.* 2012) [83]. Methane genetic evaluation is more likely to experience high environmental or non-additive genetic variation. Hence a lack of additive genetic variance is to blame for the low heritability of fitness characteristics in animal populations for any targeted traits. Breeding for reduction in methane output the ability to promote or eliminate genotypes of interest depends on the breeder's understanding of correlations between CH₄ and its proxies (Breider *et al.* 2019) [29]. The genotypic correlation establishes a greater need for selection for low CH₄ emissions. This is regardless of whether or not the association amongst the traits in question is of a phenotypic, genotypic, or environmental form. In order to find factors associated

with lowering gas emissions that may be employed as indirect selection, such as RFI, the study of correlation is therefore essential for breeding low emissions (Bezerra *et al.* 2013) [23].

7.4 Improving Improve Residual Feed Intake to Enhance Feed Efficiency in Rabbits: A Key to Mitigating Methane Emissions.

Environmental sustainability is significantly influenced by feed efficiency (Cesari *et al.* 2018) [33]. Compared to traditional methods, residual expressions of enteric emissions offer a more equitable evaluation of an animal's potential to produce methane (Smith *et al.* 2021) [201]. These assessments incorporate factors such as dry matter intake (DMI), growth, feed efficiency, and carcass output in relation to enteric emissions. In recent years, considerable research has been dedicated to understanding and reducing rumen methane production (Van Nevel and Demeyer, 1996) [220]. The primary goal is to boost feed efficiency, as methane generation significantly reduces feed energy. Additionally, methane inhibition is considered crucial for addressing global warming and preserving the ozone layer. Despite substantial improvements in animal production efficiency over the past four decades, feed remains the most significant expense. Minimizing nutrient losses associated with animal production is essential for reducing environmental impact (Gidenne *et al.* 2017) [202].

Improving feed efficiency (FE) is crucial for enhancing production efficiency and reducing greenhouse gas emissions. Selective breeding for improved FE has been recognized as a strategy to decrease CH₄ emissions and combat global warming (Beauchemin, *et al.* 2011) [18]. By breeding rabbits for better feed utilization, CH₄ output can be reduced through increased productivity linked to production efficiency. The feed conversion ratio (FCR), a measure of feed efficiency, is vital for assessing the financial and environmental performance of farming systems (Gidenne *et al.* 2017) [202]. To effectively select for FE that leads to lower CH₄ emissions, it is important to accurately identify true FE variations across different experimental conditions (Fischer *et al.* 2018). The concept of residual intake is a well-established method for quantifying individual differences in feed efficiency. Residuals from a linear regression of intake represent comparable biological processes that use or release energy and can serve as a proxy for residual feed intake. Huhtanen *et al.* (2021) noted that genetics plays a role in FE as a production trait.

Over the last 15 years, rabbit farm FCR has seen a 10% improvement, accompanied by a 10% reduction in nitrogen and phosphorus outputs. FCR is influenced by factors such as health, feeding, stock management, and genetics (Gidenne *et al.* 2017) [202]. The study suggests that FCR could potentially be reduced to 3.0 in the coming decade. Various elements affect FCR, including breeding management, animal genetic potential, feed quality, feeding strategy, animal losses, slaughter age, and housing conditions. The importance of feed efficiency is not unexpected, given that breeding programs have historically overlooked this aspect. Only recently has attention shifted towards addressing the issue of low CH₄ emissions in ruminants (Guinguina *et al.*, 2021b) [92]. Enhancing feed efficiency in rabbits has the potential to increase producer profitability while simultaneously reducing the environmental impact of rabbit production. Feed efficiency, often expressed as residual feed intake (RFI), is a critical indicator for evaluating the performance and profitability of farming systems. A correlation exists between RFI in ruminant species and nutrient utilization, making it

valuable for selecting for feed efficiency (FE) (Huhtanen *et al.* 2021).

RFI is a measure of FE that offers superior statistical properties compared to conventional FE measurements based on growth factor ratios. Dekkers and Gilbert (2010) [49] identified RFI as one of the most widely used attributes for studying feed efficiency in ruminants. Martin *et al.* (2021) observed that RFI as a predictor of FE is becoming increasingly important in selecting for low CH₄ emissions. This has driven a shift towards sustainable production efficiency, as it is not necessary to derive efficiency as a ratio between production and feed intake (Huhtanen *et al.* 2021). RFI serves as an ideal proxy for use in genetic evaluation for low emissions, as it measures efficiency variations among individuals (Kenny *et al.*, 2018) [117].

Research by Olijhoek *et al.* (2018) [171] suggests that breeding can enhance feed efficiency (FE) in dairy cows, potentially reducing methane (CH₄) emissions per unit of milk produced. Gill *et al.* (2010) [88] proposed that regular measurement of feed conversion efficiency in ruminants could accelerate genetic improvement and decrease greenhouse gas (GHG) emissions per unit of animal product. According to Steinfeld *et al.* (2006) [206], increasing cow productivity is the most straightforward and feasible approach to minimize CH₄ emissions from dairy cows. Studies have shown that higher feed intake and milk production correlate with lower CH₄ emissions per unit of milk (Garnsworthy, 2004) [83].

In recent years, several countries have incorporated feed efficiency, also known as residual feed intake (RFI), into their breeding objectives (Manzanilla-Pech *et al.*, 2022; González-Recio, 2020) [144, 14]. Current research indicates a positive correlation between RFI and methane emissions (Difford *et al.*, 2020; Li *et al.*, 2021; López-Paredes *et al.*, 2020) [52, 133]. Consequently, indirect selection for lower-emitting animals using RFI could be a near-term strategy to achieve desired reductions in methane output (Olijhoek, *et al.* 2020) [171]. Efforts to decrease methane emissions and improve feed efficiency in ruminants have been pursued simultaneously (Archer *et al.*, 2017) [11]. The genetic improvement approach for long-term, cumulative gains in feed efficiency could potentially be integrated into strategies for reducing methane production. Researchers are exploring the possibility of incorporating emission reduction strategies into breeding programs, with a focus on enhancing feed efficiency in ruminant species. Arthur *et al.* (2001) found that selecting animals for efficiency (low RFI) resulted in offspring that were slightly leaner with lower intake, but performed similarly to those selected for high RFI.

RFI has a moderate heritability ranging from 0.26 to 0.43, with repeatability across diets varying from 0.33 to 0.67. Heritability estimates for various feed efficiency metrics or RFI in sheep are limited but range from 0.11 to 0.49 (Cammack *et al.*, 2005; Fogarty *et al.*, 2006; Paganoni *et al.*, 2017; Tortoreau *et al.*, 2020) [76, 176, 212]. A meta-analysis of 39 published studies estimated the heritability of RFI in cattle at 0.33 ± 0.01 (range of 0.07–0.62) (Berry and Crowley 2013) [22]. The only published heritability estimates for New Zealand's maternal effect on CH₄ are early estimates reported by Johnson *et al.* (2018) [113]. Heritability of CH₄ output per unit of feed intake for beef and dairy sheep has been low (0.13–0.35).

7.5 Crossbreeding as a Tool for Climate-Friendly Rabbit Farming: Reducing Methane Emissions through Genetic Improvement.

Several studies have reported positive effects of crossbreeding in productive traits in different animal species, beef cattle (Barani *et al.* 2024; Assan *et al.* 2024; Wientjes, *et al.* 2020) [13, 222], dairy cattle (Kathambi, *et al.* 2025; Meseret, *et al.* 2022; Mrode *et al.* 2021) [15, 116, 153, 164], pig (Fabbri *et al.* 2024; Duenk *et al.* 2021; Popova *et al.* 2020) [64, 58, 185], and chickens (Dzungwe *et al.* 2024; Fikerneh *et al.* 2023; Bassey *et al.* 2022) [60, 75, 17]. For rabbits crossbreeding has shown its impact growth rate and production efficiency (Masara, *et al.* 2024; Kumar, *et al.* 2023; Cherwon, *et al.* 2020; Assan, 2017) [154, 151, 126, 13], however even if the methane was not measured efficient growth of animal is directly linked to methane release reduction from animals. This connection has been established in relation to decreasing enteric gas emissions in ruminants. Enhanced growth and feed efficiency in crossbred rabbits are key factors in methane reduction. Feed efficiency, evaluated through feed conversion ratio (FCR), is essential for assessing rabbit farming system performance (Maertens and Gidenne, 2016) [141]. Crossbreeding offers a solution to develop breeds that excel in nutrient utilization efficiency, reduce environmental impact, and surpass current meat production and reproductive standards. Nutrient utilization efficiency is known to reduce environmental impact through low GHG emissions.

Crossbreeding is a traditional method of improving production efficiency in rabbits by mating individuals with different genetic backgrounds. It aims to produce offspring with desirable traits, but the best breeds for improving production traits and methane reduction are not fully explored. Crossbreeding is an animal breeding strategy that influences breed composition, enhancing animal performance and potentially altering enteric ruminal and possibly ceecal gas emissions. The extensive research on crossbreeding in rabbit production suggests promising tool that in addition to improve efficiency of production can also be reducing rabbit ceecal methane emissions (Assan *et al.* 2024; Palka *et al.* 2023; Derewicka, *et al.* 2021) [13, 178, 51]. Rabbit crossing has gone as far as triple crossing programs which means that efficiency of production is likely to be improved, however no work has been attached to quantification of ceecal methane in these ventures. In principle we may suggest the success story of rabbit crossbreeding might be indirectly address methane production. Crossbreeding systems can be employed in rabbits to enhance production through the utilization of complementary breed effects, heterosis and the ability to compare the performance of different breeds and their crosses (Brahmantiyo, *et al.* 2021; Boiko, *et al.* 2020; Gharib, *et al.* 2008) [28, 26, 86].

Genetic improvement is primarily achieved crossbreeding, which allows for additive progress and complementarity between breeds or strains. This method exploits the effect of heterosis at each generation, but requires a complex scheme involving maintenance and selection of pure stocks and multiplication and diffusion of crossbred females. The tangible outcome of improved production efficacy and performance is the reduction of CH₄ yield, resulting in a favorable carbon footprint (Huhtanen *et al.*, 2021). In tropical climates, crossbreeding presents a viable strategy for enhancing performance and lowering gut emissions (Maciel *et al.*, 2019) [139]. Successful crossbreeding relies on breed compatibility, while complementarity refers to the competitive advantage of one crossbreed over another in performance (Ragab, *et al.* 2016) [187]. Well-designed crossbreeding systems can increase animal productivity by up to 21%, thereby reducing the carbon footprint of beef

production (Mokolobate *et al.*, 2014) ^[161]. Cross-induced growth improves efficiency in terms of product measure, as it should theoretically allocate more feed nutrients to productivity. Consequently, these animals should produce less polluted excrement and lower CH₄ emissions per unit basis (Fraaser *et al.* (2014)

Cross-bred cows generally exhibit higher mature weight and maturing rate, leading to efficient feed utilization and reduced CH₄ emissions. Hybridizing adaptive local breeds with foreign breeds shows promise in lowering CH₄ emissions in beef cattle (Donoghue *et al.*, 2016; Hayes *et al.*, 2016) ^[56]. For example, Zebu bos indicus demonstrates remarkable resilience and adaptability to tropical temperatures, resulting in crossbreds with higher growth rates due to effective feed utilization and lower CH₄ emissions. Crossbreeding genetically improves animal performance, which can be directly influenced by both the quality and quantity of the diet provided to crossbreds (Luchyn, *et al.* 2023) ^[134]. Effective systematic crossing has been shown to enhance animal performance and may offer a productive approach to minimizing GHG emissions. Crossing will lead to a reduction in the total number of animals that need to be reared by increasing output per animal as a result, will increase productivity, hence the carbon foot principle-reduction approach. Science-genetics through crossing is an intriguing means of mitigation through its influence on improved production efficiency.

Maciel *et al.* (2019) ^[139], in their conclusion, noted that crossbreeding could be a possibility to enhance production efficiency while decreasing CH₄ per ADG under tropical temperature conditions. This will result in a lower release of methane per kg of meat produced. Barwick *et al.* (2019) ^[16] noted that the expenses in feed-factored livestock breeding program indices will affect the potential to minimize GHG emissions in beef cattle. If a breeding objective's feed expenditure is high, multiple-trait selection will lower emission levels simultaneously, maximizing economic efficiency. Whenever it is low, more substantial growth will be recommended, which at first could end up resulting in an undesirable rise in GHG emissions. In theory, crossbreeding rabbits will reduce the amount of methane produced by the caecum. The development of synthetic lines in developing countries is the most efficient strategy, resulting from crossing local populations or breeds with selected European strains (Abd El-Aziz, *et al.* 2002) ^[4]. These lines can be specialized maternal or paternal lines or multi-purpose lines. This approach benefits from complementarity between local populations and selected strains, and allows for half of the heterosis (Assan, 2024) ^[13]. Currently, marker-assisted selection (MAS) is not widely used in rabbit selection programs in developing countries (Abu *et al.* 2008). is in order

7.6 Mitigation through Microbiome Manipulation to minimize methane production in rabbits

The digestive system type, feed quantity, and cecal microbial population influence methane emissions in rabbits (Misiukiewicz *et al.* 2021; Baca-González, *et al.* 2020; Clauss *et al.*, 2020) ^[158, 41, 14]. Studies on reducing methane production in ruminants through microbiome manipulation have shown promise. These approaches encompass using probiotics as feed additives, eliminating protozoa via rumen defaunation, modifying cellulolytic and protozoan activity, developing vaccines to limit methanogenesis, and utilizing chemicals to directly suppress methane formation (Tavendale *et al.* 2005; Subhrat *et al.* 2016; Wedlock *et al.* 2013) ^[208]. The applicability of these methods to rabbits remains

uncertain, and it's important to consider that the rumen and cecum may respond differently to methane reduction techniques. In herbivorous mammals, the rumen and hindgut are separate fermentation organs, with the former producing considerably more methane (Mi *et al.* 2018). Diverse methanogenic microorganisms have been identified in adult rabbit ceca. The well-documented difference between ruminants and non-ruminant herbivores (Franz *et al.*, 2010, Franz *et al.*, 2011) ^[77] illustrates this, although the reasons for higher CH₄ production in ruminants are not fully elucidated. Misiukiewicz *et al.* (2021) ^[158] proposed that improved understanding of archaeal communities in the gastrointestinal tract (GIT), including their metabolism and interactions with other microorganisms, could aid in developing new strategies to inhibit methanogens and promote acetogenesis. Acetogens are more adapted to thrive in low-substrate environments and show greater resistance to bile salts (Jezierny *et al.* 2007) ^[111], giving them an advantage in digestive tracts with lower pH and rapid passage. Examining microbial community composition and complexity in various animals is essential for understanding control mechanisms to mitigate adverse methanogenesis-related phenomena. Probiotics, which are feed supplements containing selected bacterial or yeast cultures that improve animal health by modulating gastrointestinal microflora and affecting rumen fermentation, have been evaluated in ruminants.

Protozoa provide substrate for methanogenesis and protect symbiotic archaea from oxygen toxicity, contributing 37% of rumen CH₄ emissions (Lan and Yang, 2019; Belanche, *et al.* 2014) ^[19]. In ruminants, rumen defaunation reduces organic matter digestibility, particularly acid detergent fibre (ADF) and neutral detergent fibre (NDF), subsequently decreasing food intake (Newbold *et al.* 2015; Hristove *et al.* 2013) ^[167, 102]. Cellulolytic bacteria and protozoan activity primarily influence the availability of H₂, the main substrate for methanogenesis. Probiotic bacteria are known to reduce NO₃, NO₂, and SO₄²⁻, competing with CH₄ for H₂. Rumen defaunation involves removing protozoa from the rumen ecosystem, which are significant methanogen H₂ producers. Ciliated protozoans form hydrogenosomes with methanogens on their surface and within their cell bodies. Vaccines targeting methanogenesis restrict growth by stimulating the animal's immune system to produce saliva-containing antibodies, which inhibit methanogen growth upon entering the rumen (Wedlock *et al.* 2013). A vaccine against protozoan antigens has been reported but failed to significantly reduce the ciliate population in Merino sheep, making it difficult to assess its effectiveness and requiring further research to determine its feasibility, practicality, and long-term viability (Baca-González, *et al.* 2020; Leng, 2014) ^[14]. Chemical intervention is a common approach to inhibit methanogenesis, aiming to reduce CH₄ emissions without negative effects on humans, animals, or the environment, and be cost-effective for producers while increasing output and profitability (Khampa and Wanapat, 2007) ^[118]. Ionophores are commonly used feed additives in ruminants' diets to reduce CH₄ emissions. The determination of methane resulting from the use of additives is the missing piece in the broad usage of additives in rabbits.

8. The Challenges of Addressing Low- Cecal Methane production in Rabbits: A Critical Evaluation

Cecal fermentation in rabbits, a hindgut fermentation process, generates methane, a potent greenhouse gas (Calvet *et al.* 2011). As rabbit meat production expands globally, understanding and addressing cecal methane emissions

becomes increasingly crucial. This evaluation investigates the intricacies of rabbit methane production, its environmental implications, and potential mitigation strategies, underscoring the need for targeted research and innovative approaches to minimize rabbit farming's ecological impact

8.1 Genetic Constraints on Reducing Methane Emissions in Intensive Rabbit Farming

To breed rabbits for lower methane production, effective selection procedures must first be developed, as genetic markers for methane-related traits in rabbits have yet to be identified. Moreover, it's essential to understand how genetic factors influencing methane production relate to other desirable traits. Selection for lower methane emissions could be challenging if genetically tied to other beneficial or detrimental characteristics. A key challenge is the limited understanding of methane trait variability, which is essential for informed selection decisions. With limited genetic variation, selection response is typically modest, making it challenging to achieve significant reductions in methane emissions in rabbits. Methane production is a complicated characteristic that is impacted by a variety of genetic and environmental variables, which makes breeding more challenging. A sustainable genetic improvement program should consider economic, sociological, and ecological factors. Promoters should evaluate local genetic resources, assess their interest and potential threats, and consider whether they are of interest or endangered.

8.2 Practical Impediments to Low-Methane Intensive Rabbit Production: A Critical Analysis

It is uncertain whether the methods established for quantifying methane production in ruminants (Knap *et al.* 2014) will be suitable for rabbits. Measuring methane in rabbits is likely to present numerous challenges, possibly due to their small size, which can make accurate measurements difficult and costly. Precise quantification of rabbit methane is essential for developing selection methods. Implementing effective rabbit selection programs demands substantial resources, infrastructure, and accurate genetic parameters related to methane production. Moreover, creating breeding programs that balance methane reduction with other rabbit reproductive and productive traits can be intricate. Rabbit breeding has intensified to produce triple crosses, potentially complicating methane assessment across different rabbit populations. Quantifying cecal methane and deriving specific values in rabbits will be challenging due to inter-breed genetic variability in performance that may affect cecal emissions. Rabbits display significant genetic diversity in size, ranging from dwarf to giant breeds (Blasco *et al.* 2014). Commercial production utilizes medium-sized breeds for reproduction due to high prolificacy and large breeds as terminal sires for rapid growth, facilitating doe management, reducing maintenance costs, and enabling high growth rates in rabbits, resulting in varying cecal fermentation.

Calvet *et al.* (2011) investigated indoor environment and gas emissions in rabbit farms using a photoacoustic multi-gas analyzer. However, they found methane measurements below 10 ppm unreliable due to cross-correlation with water vapor. As most CH₄ readings fell within this range, methane emissions were excluded from the study. There is a need to develop rabbit-specific methane emission measurements from animals and assess greenhouse gas emissions associated with their manure management. Accurate quantification of cecal methane is crucial for effective breeding of low-

methane rabbits. Methane emission measurement is vital for breeding ruminants with low enteric emissions, as it affects the breeding values used to estimate genetic parameters (Chagunda, 2013) ^[38]. It remains unknown whether the various methods developed to measure methane in ruminants will be effective for rabbit breeding. These methods include respiration chambers, GreenFeed systems, SF₆ tracer techniques, open-circuit respiration chambers, closed-circuit chambers, infrared spectroscopy, portable methane analyzers, whole-room calorimeters, automated methane measuring systems, and modeling and simulation (IEA, 2021; Hammond *et al.* 2013) ^[95].

8.3 Dietary Constraints on Cecal Methane Mitigation in Intensive Rabbit Production

The complexity of breeding for feed efficiency in rabbit production may be influenced by dietary diversity. Diet affects methane production, potentially limiting the effectiveness of breeding for reduced methane output across various diets (Manzanilla-Pech *et al.*, 2022) ^[144]. Methane production can be impacted by non-genetic factors such as diet, production system, temperature, and management practices, making breeding more complex (Sikiru *et al.* 2024). In ruminants, diet significantly affects the intensity and yield of enteric methane emissions (Tseten *et al.*, 2022) ^[216]. Methane production varies due to factors like feed quality, fiber content, concentrate-to-forage ratio, starch content, fat content, and protein content (Lileikis *et al.*, 2023). Improving ingredient combinations and nutritional management approaches can help reduce methane generation in ruminants by understanding these dietary factors (Beauchemin *et al.*, 2019) ^[18]. Methanogenesis, H₂ concentrations, and substrate availability for fermentation are also influenced by rumen composition and digestibility.

8.4 Genotype-Environment Interactions: A Key Constraint in Breeding Rabbits for Low Methane Emission

Animal breeding has been significantly impacted by the interactions between genetics and environment, which can compromise the selection response of individual traits. Environmental factors can influence genetic expression, making it challenging to achieve consistent results in methane production. In animal breeding, genotype x environment interaction (GEI) is essential for selecting genotypes that demonstrate adaptability and durability for desired traits (Rauw and Gomez-Raya, 2015; Kolmodin, *et al.* 2003). The effectiveness of genetic improvement efforts can be affected by significant variations in quantitative measures of CH₄ phenotypic plasticity, indicating the presence of GEI. To reduce CH₄ emissions while maintaining animal performance and livestock production efficiency, it is important to understand genotype by environmental components, such as diet and microbial profile, when selectively breeding ruminants. Establishing relevant genetic criteria for genetic assessment requires understanding the relationship between genetics, nutrition, and the microbiome on methane production. Genetic evaluation of CH₄ is challenging due to the increased likelihood of enteric gas emissions across settings, despite the anticipated genetic correlation. Therefore, when breeding ruminants for low methane gas levels, it is crucial to consider the interplay of genetics, nutrition, and microbial profile (Moss *et al.* 2000).

8.5 Economic and Social Limitations of Breeding Low-Methane Rabbits in Intensive Farming Systems

Implementing breeding programs for low methane production in rabbits may require substantial investments in resources and infrastructure. However, the limited market demand or incentives for low-methane rabbit products may make it difficult to justify such breeding programs. Additionally, breeding for low methane production may not align with social or cultural values, potentially hindering adoption, especially in developing countries.

9. Breeding Rabbits for a Greener Future: Minimizing Cecal Methane Production in Intensive Systems Through genomics and Phenomics

Over the past ten years, animal breeding across various species has been profoundly influenced by genomic technologies. Breeders extensively utilize genomic selection as a key approach to forecast trait inheritance and identify genetically superior animals. Genome science has become a central element in breeding programs for diverse animal species (Kenney *et al.*, 2023; Visser *et al.*, 2023; Ablondi *et al.*, 2022; Cortes-Hernández *et al.*, 2021; Dixti *et al.*, 2020; Baes and Schenkel, 2020; Cole *et al.*, 2020)^[223]. Research by Prez-Enciso *et al.* (2021) revealed that the anticipated advancements in genomic technology have been exceeded, facilitating widespread implementation of genomic selection. The effectiveness of breeding rabbits to decrease methane gas emissions will largely rely on the application of genomics (Hayes *et al.* 2012). This is attributed to the fact that progress in these areas has enhanced our comprehension of genetic variation in animal production traits and their influence on productivity (Esen *et al.* 2022; Fu and Yuna, 2022). Contemporary animal breeding datasets are extensive and expanding, partly due to the accessibility of high-density SNP arrays and cost-effective sequencing technologies (Cole *et al.* 2012). Pech *et al.* (2016) argue that employing genomic science to select individuals with low emissions is crucial for breeding aimed at improvement, which is difficult to achieve through alternative methods.

The creation of tools such as chips enables rapid and affordable genotyping for thousands of Single Nucleotide Polymorphisms (SNP), allowing for the introduction of 'genomic selection' (Burridge, *et al.* 2024). This facilitates more precise estimation of breeding values at younger ages and for traits that are challenging to measure in breeding animals (Visser *et al.*, 2023; Baes and Schenkel, 2020). Genomic selection is particularly advantageous in breeding programs where key objective traits are difficult to measure, such as methane-related traits and their proxies. It also enables the selection of new traits by establishing reference populations where traits like feed intake can be efficiently measured. In comparison to traditional selection based on pedigree, it is less crucial that the animals whose phenotypes are measured are directly related to the selection candidates. Genomic selection has been demonstrated to nearly double the rates of genetic gain in other ruminant species (Xu *et al.* 2020).

Kadarmideen *et al.* (2006) noted that using genome tools or markers to improve animal performance in growth, carcass, and reproduction will impact CH₄ emissions. It is now recognized that enhancing feed efficiency is more likely to reduce ruminant CH₄ release into the atmosphere. Mijena and Getiso (2021) propose that the genome is an appealing approach for selecting better-adapted livestock genetics. This could indirectly contribute to reducing CH₄ emissions by promoting production efficiency in animals that perform

optimally in specific environments. Determining genomic breeding values for genetic selection may be the most environmentally friendly method to decrease enteric CH₄ emissions from ruminants. De Haas *et al.* (2021) observed that advancements in genotyping will mitigate the challenges of capturing CH₄ at the individual cow level, and genomic prediction will enable faster adoption. However, obtaining accurate, high-throughput phenotypic data has emerged as a bottleneck in livestock genomics and related fields. Thus, relevant phenotypes are necessary to improve understanding of complex relationships and underlying biological and physiological systems, such as methane production in rabbits through expanded methane traits associated characteristics.

Phenomes represent a cutting-edge development in animal breeding and can be used alongside genomic techniques to better comprehend the genetics of methane production (Pérez-Enciso and Steibel, 2021). The development of integrated high-throughput breeding and phenomics application in breeding can benefit ruminant breeding for low methane gas levels (Waseem *et al.* 2022). Zhao *et al.* (2019) argue that the era of high-throughput automated multipurpose and reliable phenotypic technologies is becoming increasingly important as tools for accelerating genetic gain in breeding operations as highly efficient characterization capabilities rapidly advance. Pickerig *et al.* (2015), Steibel, (2023) and Houle *et al.* (2010) have all contributed to the field of phenomics, a study of phenotypic information at the organismal level. This research can help estimate genetic parameters and explain significant consequences, such as GHG emissions in animal species. High-throughput sequencing and breeding techniques, along with advanced genome-editing technologies incorporating artificial intelligence, offer new opportunities for methane reduction in animal species. The methane metrics for ruminants' vs monogastric herbivores like rabbits, however, could pose as limitations.

10. Implications

This study concludes that large-scale rabbit farming has significant environmental repercussions, particularly regarding methane emissions and climate change. To mitigate these effects, adopting sustainable practices such as manure management, dietary modifications, and breeding programs that prioritize methane reduction is essential. Regulatory frameworks and continuous research are also crucial for environmentally responsible rabbit farming. By addressing methane emissions from rabbit farming, the industry can contribute to global efforts to combat climate change while maintaining sustainable protein production. Ultimately, incorporating methane reduction strategies into rabbit breeding programs and farming practices can enhance the long-term sustainability of the industry.

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