AGRICULTURAL BIOLOGY,
2022, V. 57, Iss. 6, pp. 1025-1054
[SEL'SKOKHOZYAISTVENNAYA BIOLOGIYA]

ISSN 2412-0324 (English ed. Online)
ISSN 0131-6397 (Russian ed. Print)
ISSN 2313-4836 (Russian ed. Online)

Reviews, challenges

UDC 636.087:591.132.7:631.95

doi: 10.15389/agrobiology.2022.6.1025eng
doi: 10.15389/agrobiology.2022.6.1025rus

RUMEN METHANE PRODUCTION AND ITS REDUCTION USING NUTRITIONAL FACTORS
(review)

N.V. BOGOLYUBOVA✉, A.A. ZELENCHENKOVA, N.S. KOLESNIK, P.D. LAHONIN

Ernst Federal Research Center for Animal Husbandry, 60, pos. Dubrovitsy, Podolsk District, Moscow Province, 142132 Russia, e-mail 652202@mail.ru (✉ corresponding author), aly4383@mail.ru, kominisiko@mail.ru, lakhonin.99@mail.ru
ORCID:
Bogolyubova N.V. orcid.org /0000-0002-0520-7022
Zelenchenkova A.A orcid.org/0000-0001-8862-3648
Kolesnik N.S. orcid.org/0000-0002-4267-5300
Lahonin P.D. orcid.org/0000-0002-7354-0337
The authors declare no conflict of interests
Acknowledgements:
Supported financially by the Ministry of Science and Higher Education of the Russian Federation (FGGN-2022-0009).
Received October 5, 2022

Abstract

Methane is a powerful greenhouse gas with a higher global warming potential than carbon dioxide. Agriculture, especially animal husbandry, is considered the largest sector of anthropogenic methane production. Of farm animals, ruminants are the main producers of methane. Its world production and emissions are increasing due to abundant population of ruminants. The hydrogenotrophic scenario of methanogenesis from hydrogen and carbon dioxide, carried out by ruminal archaea, prevails. Over the past 50 years, numerous research papers have substantially improved our understanding of rumen fermentation and methanogenesis to develope strategies for assessing and reducing methan emission (K.A. Beauchemin et al., 2020). One of the proposed strategies is dietary intervention, i.e. improved dietes and the use of nutritional factors that affect the ruminal microbiota. The quality, feed preparation, the ratio of concentrated and roughage feeds affect methane emissions. Some feeds may increase propionate production or decrease acetate production by reducing the level of ruminal hydrogen converted to methane. Another strategy is the use of modifiers, the feed additives that directly or indirectly inhibit methanogenesis, and biocontrol manipulation using defaunization agents, bacteriocins, bacteriophages, and immunization aimed at reducing the counts of methanogens. The strategy may be also based on genetically or technologically improved productivity performance. With higher productivity, the relative methane emission per unit of meat or dairy product is reduced (M. Islam et al., 2019). Fat additives, organic acids, probiotics, ionophores, phytogenics can serve as strategies to reduce methane formation in ruminants (M. Wanapat et al., 2021; R.D. Marques et al., 2021; S.H. Kim et al., 2020). Feeding manipulation is a simplistic and pragmatic approach to improve animal productivity with a reduced CH_4 emission (M.D. Najmul et al., 2018). In the review, along with a description of methanogenesis, we also summaraized modern research data on the influence of various alimentary factors (i.e., special diets, phytogenic saponins, tannins, flavonoids and essential oils) on CH_4 emission. The type of diet, the quality of bulky and concentrated feeds, their chemical composition, ratio, pre-feeding preparation affect methane emission in ruminants. However, a promising approach to mitigate methane emissions is adding a small amount of grain to roughage and feeding high quality forages with less fiber and higher levels of soluble carbohydrates. Phytogenics made from various botanical parts of plants is a cheap and environmentally friendly agents to reduce greenhouse gas emissions. Phytogenics also positively affect animal resistance. There are few studies on the in vitro efficacy of flavonoids and other secondary plant metabolites as agents for reducing methane emissions. The data obtained are variable and depend on the type of herbal preparations, their characteristics and the diet fed to the animals. Further in vivo studies should establish the optimal dosages of phytogenics that provide a positive effect. The combination of various phytogenics seems to be relevant and promising. An integrated approach should provide high fragmentation activity, effective digestion and assimilation of feed nutrients.

Keywords: ruminants, greenhouse gases, methanogenesis, diet quality, diet composition, phytogenics, saponins, tannins, flavonoids, essential oils

The impact of greenhouse gas (GHG) emissions on climate change has become a global and publicly discussed environmental and health problem both in the world and in Russia. Agriculture is one of the largest sources of greenhouse gases, and wise use of the potential of the industry can limit the rate of global warming to 2 °C by the end of the century [1].

The efforts of the global community to prevent climate change are predominantly focused on reducing carbon dioxide (CO_2) emissions. At the same time, methane (CH_4), nitrous oxide (N_2O) and other CO_2-free greenhouse gases emitted during the production of crop and livestock products also contribute to global warming. Methane (CH_4) is one of the three major greenhouse gases (GHGs), in addition to carbon dioxide (CO_2) and nitrous oxide (N_2O), with a global warming potential 28 times greater than that of carbon dioxide (CO_2) [1-3]. Agriculture, due to the increase in land use and the reduction of CO_2 absorption spaces (forests, organic soils), is involved in increasing the production of carbon dioxide. Livestock and especially ruminants are the largest source of direct emissions; synthetic fertilizers also contribute heavily to direct emissions; livestock and fish farms account for 31% of greenhouse gases [4]. The livestock sector accounts for approximately 18% of global anthropogenic GHG emissions.

Among livestock, ruminants produce about 81% of greenhouse gases [5] due to massive methanogenesis by rumen microbes, which produce 90% of the total CH_4 emitted by ruminants [6]. Ruminants emit about 115 million tons of CH_4 annually, which is formed as a result of fermentation carried out in the rumen by a complex of bacteria, archaea, protozoa and fungi [7]. Globally, CH_4 emissions from dairy and beef production account for 30% and 35% of livestock emissions, respectively. Buffaloes and small ruminants contribute less, accounting for 8.7% and 6.7% of industry emissions, respectively [8]. Cows and other ruminants hold the record for methane emissions. In their multi-chambered stomachs, bacteria help digest food by synthesizing methane as a by-product. It is released into the atmosphere through belching, although a small part of it is also produced in the intestines. The digestive system of other farm animals differs from that of ruminants. Chickens and pigs emit less greenhouse gases, but their amount is many times greater than that produced by plants of nuts or peas. Fish reared in fresh water also serve as a source of greenhouse gases: excrement and unused food are deposited on the bottom of ponds, where there is almost no oxygen, that is, conditions ideal for the appearance of methane are created.

The harmful effect of methane on the state of the atmosphere is confirmed by the fact that with a conventionally accepted global warming potential (GWP) of carbon dioxide equal to 1, for methane GWP = 21, the half-life of methane is 11 years, and the duration of stay in the Earth atmosphere exceeds 100 years. It follows that methane as a greenhouse gas is no less dangerous than carbon dioxide.

Ruminants can produce between 250 and 500 liters of methane per day, and the contribution of cattle to global warming, which may occur in the next 50-100 years, is estimated at just under 2%. While emissions per unit of livestock production have decreased, global emissions have risen due to an increase in animal populations [8]. By 2050, total CH_4 emissions from ruminants are expected to increase significantly due to increased demand for milk and meat, given the growing world population [9]. This determines the importance of the problem of reducing CH_4 emissions in animal husbandry and the attention to environmental issues in general on the part of government structures [10].

It is possible to reduce the formation of methane in the digestive system of animals through the use of various feed additives, antibiotics and vaccines, as well as through the inclusion of high-quality roughage in the diets of cattle. In addition, a reduction in the volume and intensity of emissions can be achieved through the use of modern methods to increase the productivity of animals. This strategy is very attractive as it increases farm profits at the same time [11].

Methane emissions from animals вdepends on the amount of feed consumed, the type of carbohydrates in the diet, the methods of preparing feed for feeding, feed additives of various nature that regulate the state of microbial processes. Management of these processes can reduce methane formation in the rumen of ruminants and, as a result, methane emissions into the atmosphere. The study of biochemical, microbiological and genetic aspects of methane formation in the rumen of ruminants is necessary for the use of nutritional factors to reduce CH_4 emissions in animal husbandry.

In recent years, the results of a huge number of studies have been published that have improved understanding of the complex processes of rumen fermentation and methanogenesis in ruminants, as well as ideas about means and methods for reducing methane production in ruminants [12].

The purpose of this review is to summarize current data on the effect of alimentary factors, in particular, the structure and composition of diets, phytogenics of various nature (saponins, tannins, flavonoids, and essential oils), on the formation of methane in ruminants.

Mechanisms of methane formation in ruminants. The microbial ecosystem of the rumen is very stable and optimized due to the natural selection of microorganisms, but not completely efficient. One reason for this is the loss of energy due to methane emissions [13]. For the host animal, the formation of CH_4 means a loss of 2 to 12% of the total energy intake that could be available for growth or production [14].

Carbohydrates are the main source of energy for ruminants. In the rumen, polysaccharides (mainly cellulose, hemicellulose and starch) are hydrolyzed to glucose and other hexoses and pentoses. Further, monosaccharides are metabolized into volatile fatty acids (VFA) and CO_2. Metabolic hydrogen is released during the conversion of monosaccharides to VFAs, restoring intracellular cofactors, and cofactors must be reoxidized to continue fermentation. This is largely due to hydrogenase activity and the formation of dihydrogen (H_2, that is, molecular hydrogen). Dihydrogen does not accumulate in the rumen because it is transferred from the fermenting consortium of bacteria, protozoa, and fungi to methanogenic archaea, which use H_2 to reduce CO_2 and other one-carbon compounds to CH_4 [15, 16].

Methanogens can be divided into three groups depending on the substrate used: methane derivatives: methylotrophic, hydrogenotrophic, and acetate (acetoclastic) [17, 18]. H_2 and CO_2 serve as the main substrates of methanogens, and hydrogenotrophic methanogenesis is considered to be the predominant route of CH_4 formation in the rumen [19]. There is a wide variety of methanogenic archaea in shape (cocci, spirilla, rods of various shapes), mobility (mobile and immobile) and other properties, but the general physiological characteristics of methanogens are the need for anaerobiosis and the use of energy, the formation of which is associated with methane bosynthesis, as its only source [20]. According to a meta-analysis of global data, 90% of rumen methanogens belong to the genera [21] *Methanobrevibacter* (63.2% of the methanogen population), *Methanomicrobium* (7.7% of the methanogen population), *Methanosphaera* (9.8%), “rumen cluster C”, currently called *Thermoplasma* (7.4%), and *Methanobacterium* (1.2%). The production of methane from H_2 and CO_2 lowers the partial pressure of H_2, thus allowing the fermentation to continue. Without H_2 removal, H_2 accumulation will inhibit further reoxidation of reduced cofactors which, in turn, will consequently inhibit VFA production [16, 22]. In addition, the functional group of methanogens also uses formate (up to 18% of the total amount of methane in the rumen), acetate, methanol, methylamines (mono-, di- and trimethylamine) and alcohol, but due to the biological characteristics of these microorganisms, this plays a small role in formation of this gas [23]. For example, *Methanosphaera stadtmanae* produces methane only through the reduction of methanol with the participation of hydrogen, having one of the most stringent energy exchanges of all methanogenic archaea [21].

The formation of methane consumes the maximum amount of hydrogen in the rumen. A smaller part of it is used for the production of propionate. A strong positive relationship between hydrogen and propionate concentrations indicates that elevated H_2 levels in the rumen may activate reactions that involve hydrogen in propionate production [24]. Propionate (an alternative hydrogen scavenger for CH_4) is the main glucose precursor in ruminants, so it is desirable to increase its levels in animals with a high demand for glucogen precursors [25]. Reductive acetogenesis (formation of acetate from CO_2 and H_2) is also desirable as a process for incorporating hydrogen into metabolism, since acetate serves as an energy source and building block in the synthesis of long chain fatty acids. However, reductive acetogenesis is thermodynamically inferior to methanogenesis in the normal rumen, but can be a useful hydrogen sink to enhance methanogenesis-inhibited rumen fermentation. Theoretically, the redirection of hydrogen from methanogenesis to fermentation end products that can be taken up and used by the host animal, as well as to the synthesis of microbial biomass, not only helps to reduce CH_4 emissions, but also has the potential to increase the productivity of the animal. However, this potential has not been consistently realized so far [26].

The rumen microbiota plays an important role in the production of biogenic methane. Information on how the hereditary factors of the host influence on the variability of the rumen microbiota and their combined effect on methane emissions are limited. Q. Zhang et al. [27], using a Bayesian model, in a sample of 750 dairy cows, the joint contribution of the host genotype and microbiota to the host's methane emission was estimated. The study showed that host genotype and microbiota accounted for 24% and 7% of variations in host methanogenesis activity, respectively. In addition, it appeared that certain host genes were significantly associated with the composition of the rumen microbiota [27].

Strategies to reduce methane emissions. According to various authors, methane emissions from dairy cows range from 151 to 497 g/day [28]. This value depends on climatic conditions [29], genotype [30], type of productivity, age [31], as well as the quality and composition of the diet [32, 33] and the provision of food needs of animals [34, 35]. Thus, lactating cows produce more CH_4 (354 g/day) than dry cows (269 g/day) and heifers (223 g/day). Dairy sheep emite 8.4 kg of CH_4 per animal per year. Holstein cows produce more CH_4 (299 g/day) than crossbreeds (264 g/day). Methane emissions from heifers grazing on fertilized pastures are higher (223 g/day) than their counterparts on uncultivated pastures (179 g/day). In beef cattle, average CH_4 emissions range from 161 to 323 g/day. Adult beef cows produce 240-396 g CH_4 daily, Suffolk sheep 22-25 g daily. Annual CH_4 emissions per bison are 72 kg [28]. In a 10-year follow-up in New Zealand, S.J. Rowe et al. [36] noted that sheep with low CH_4 emission had higher wool shearing, were leaner, and differed from their high CH_4 counterparts in fatty acid muscle tissue profile [36].

The development of strategies to reduce the release of methane in the body of ruminants during the fermentation process is of scientific and practical interest. The proposed approaches fall into several categories. For example, there are strategies that affect methanogenesis through nutritional factors. In particular, some feeds increase the production of propionic acid or reduce the production of acetate, reducing the concentration of H_2, which can potentially serve as a source of methane. Feeding strategies also use so-called modifiers that change processes in the rumen, the substances that directly or indirectly inhibit methanogenesis or provide biological control (defaunation, bacteriocins, bacteriophages, and immunization) of the rumen biota, aimed at reducing the content of methanogens. Increasing animal productivity at the genetic level and by optimizing housing conditions for better use of nutrients in the body, which increases feeding efficiency and reduces gas emissions per unit of product (meat or milk). If the annual milk yield remains constant but comes from fewer cows, overall CH_4 emissions will be reduced.

A number of proposed strategies to reduce methane production in ruminants have been reviewed previously, including many that have been revised [2, 37, 38]. Reviews on methods for measuring methane emissions and their application [39-41], including in dairy cattle [42, 43] are of particular interest, as well as the study of methanogens and their role in methanogenesis [44].

Changing feeding patterns is a simplistic and pragmatic approach that can lead to higher animal performance and lower CH_4 emissions [4]. Changing diets is the most common example of such a strategy. Among the ways to control methanogenesis using nutritional factors, two main categories can be distinguished - improving the quality of food and changing the amount consumed per feeding, as well as the use of feed additives that either directly inhibit methanogens or change metabolic pathways, leading to a decrease in the production of substrate for methane synthesis.

F e e d q u a l i t y. Considerable attention is paid to the study of the effect of feed quality and diet structure on methane production in ruminants (Table 1). The rate of methane production in the rumen depends on the composition of the diet, the type of carbohydrates (cellulose or starch), proteins and lipids, which make the biggest impact on methanogenesis [21, 35], as well as physiological factors, e.g., the time of digestion in the rumen.

Feed quality is known to affect CH_4 production in the rumen [32, 45]. High-quality feed (e.g., young plants) can reduce CH_4 emission by altering the metabolic pathway, as this feed contains more easily fermentable carbohydrates and less neutral detergent fiber (NDF), which improves digestibility and increases the rate of passage of the feed through the gastrointestinal tract [46]. Feeding corn silage was reported to linearly decrease CH_4 output (21.7; 23.0; 21.0 and 20.1 g/kg DM) and CH_4 emissions as a share of total energy intake (6.3, 6.7, 6.3 and 6.0%) when using plants of later stages of maturity [47]. However, other authors have not noted differences in methane emissions when changing the stage of maturation of the grass used for haymaking [48]. Methane release during fermentation differs between grazing ruminants on natural and artificial pastures [49, 50] and also depends on the quality of grass stand [46]. Different feed types can also contribute differently to CH_4 emissions due to differences in chemical composition [51]. So, when replacing a fibrous concentrate with a starchy concentrate, methane production decreased by 22%, and when using the so-called protected starch, by 17%. Methane production was lower for legumes than for cereals (by 28%) and for silage compared to hay (by 20%) (51, 52). Legume feeds have lower CH_4 yields due to the presence of condensed tannins, low fiber content, high dry matter intake, and high transit rate [53]. Increasing consumption of alfalfa as a concentrate replacement can significantly reduce CH_4 emissions [54].

1. Methane emission in animals, as Influenced by feed quality and diets

Factor	Animsals, *n*	Method for methane measurement	Effect on methane peoduction	Reference
Feed quality (high, medium and low)	12 heifers (6 of Holstein breed and 6 of Charolais × Simmental breed, 12 months, BW 310 kg)	Sulfur hexafluoride (SF_6) tracer gas	The amount of methane per 1 kg of digested organic matter was highest on low-quality diets	[32]
Animal age, content of concentrates	45 heifers aged 9, 12 and 15 months, rations with different levels of concentrates (30, 40, 50%)	Sulfur hexafluoride (SF_6) tracer gas	Heifers aged 9, 12 and 15 months with an average weight of 267.7; 342.1 and 418.6 kg produced 105.2; 137.4 and 209.4 g CH_4/day. Average ratio of CH_4 to gross energy consumption 0.054; 0.064; 0.0667. With an increase in the level of concentrates, the release of methane decreased	[31]
Diet composition	40 Continental × British bulls (6 months, BW 252 kg)	Sulfur hexafluoride (SF_6) tracer gas	Methane production per day with high roughage or bulky feed (83.5% silage:11.5% grain) was 42% higher than high grain diets (41.8% silage:41.7% grain)	[150]
Feed quality (poor quality hay + protein supplements: .CF at 0.29% BW or DS at 0.41% of BW)	23 crossbred British bulls (BW 344 kg)	Open cycle gas quantification chamber (GreenFee emission monitoring system GEM; C-Lock Inc., Rapid City, SD)	Animals treated with .CF had higher CH_4 emissions (211 g/day) than those who received DS (197 g/day). With protein supplements, the emissions were higher than in the control (175 g/day). Methane emissions as a percentage of GE consumption were the lowest when animals consumed DS (7.66%), intermediate when .CF was consumed (8.46%), and the highest in control (10.53%)	[33]
Diet composition, animal genotype, age	Rumen contents of crossbred cows Limousin × Swiss (meat) and Limousin × Holstein (milk-meat)	in vitro	The first factor is diet: flaxseed reduced methane yield (by 6.5%), total gas production (by 3.6%), and methane/total gas ratio (by 2.7%) The second factor is the genotype: a lower methane output (by 15%) was noted in the Limousin × Swiss crossbreed cows compared to the Limousin × Holstein crossbreed cows. The third factor is age. In meat animals, methane emissions increased with age; in dairy and meat animals, the highest values were in young and old animals.	[30]
Feed quality	16 lambs received a diet of ryegrass (before flowering and at a late flowering phase)	Sulfur hexafluoride (SF_6) tracer gas	No difference observed	[48]

Continued Tabel 1

Feed quality and diet composition	9 heifers (BW 329 kg) zebu Brahman received one of three diets: hay GQ, hay LQ and low quality hay + molasses + urea (LQ + A)	Sulfur hexafluoride (SF_6) tracer gas	Methane emission (g/day) was the same in the LQ (110.4) and LQ + A (125.8) groups and lower than in the GQ group (181.5). The values of CH_4/kg of DM consumed were maximum in the LQ (31.0) and LQ + A (29.8) groups and lower in the GQ diet (23.0) (30% reduction in emissions compared to the LQ group)	{45]
Feed quality	Natural grassland and sorghum, natural grassland and alfalfa	Sulfur hexafluoride (SF_6) tracer gas	Methane emissions were lower in cows grazing on sorghum than those grazing on natural grass pastures; in cows on natural pastures and fed with alfalfa hay, methane emission was the same. Poor quality diets increase methane output	{49]
Feed quality and consumption	56 lactating dairy Holstein-Friesian cows, ration of grass silage, corn silage and compound feed (70:10:20). Animals are divided into 2 groups, with high (day 96 of lactation) and low (day 217 of lactation) consumption of DM	Open circuit gas quantification chamber	The total amount of methane released per day did not differ between the groups. Relative methane emissions (12.8±0.56 g/kg of milk) were lower (by 12%) with high feed intake and higher milk yield. Methane emissions increased as grass quality deteriorated, regardless of consumption level	[35]
Forage quality (cultivated and natural pastures)	11 Swiss lactating cows	Sulfur hexafluoride (SF_6) tracer gas	When grazing cows on cultivated pastures, methane emissions per unit of both consumed and digested organic matter were lower than when grazing on natural pastures	[50]
Feed quality	12 crossbred (Hu × Han) dry ewes (aged 3 years, BW 32 kg) received corn stover, alfalfa and concentrates (60:0:40, 60:15:25, or 60:30:10)	Respiratory chamber (open circuit)	Increasing the share of alfalfa in the diet reduced methane emission per day, including in relation to the consumed DM and OM	[54]
Feed quality	Different grazing systems consisted in changing the density of animals per ha (1 cow/ha and 2.5 cows/ha) that alters the quality of the grass stand	Sulfur hexafluoride (SF_6) tracer gas	Methane emissions per unit GE consumption (4.6%) was low for grazing animals	[46]

Continued Table 1

Consumption level	290-302 beef cattle (420 kg) and 1105-1251 beef cattle (430 kg), ad libitum/limited feeding	Automatic sampling systems	In CH_4 emissions from the feedlot, one peak was observed during the day with ad libitum feeding, and several peaks with limited feeding. Total emissions did not change	[34]
Diet nutritional value and climatic conditions	30 Simmental dry cows and Gelbfi beef cows (663 kg BW)	Sulfur hexafluoride (SF_6) tracer gas	The use of protein supplements in low-protein diets and prolonged exposure to cold reduced CH_4 emissions	[29]
Diet composition	8 lactating Holstein-Friesian cows, silage based rations or silage + hay	Respiratory chamber (open circuit)	Cows fed a diet based on silage and hay had higher daily methane emissions. There were no differences in methane emissions per 1 kg DM consumed or per 1 kg of milk	[52]
Diet composition	16 lactating cows	Respiratory chamber	Adding treated oilseeds as a source of fatty acids reduced methane production by an average of 13%	[62]
Consumption level and quality of ryegrass silage	56 Holstein-Friesian lactating cows	Respiratory chamber (open circuit)	Improving the quality of grass silage by harvesting feed at an earlier stage of plant growth significantly reduces intestinal CH_4 emissions regardless of DMC	[35]

N o t e. BW — bodyweight, DM — dry matter, CF — cotton flour, DS — dry stillage, GQ — good quality, LQ — low quality, A — additives, GE — gross energy, OM — organic matter, DMC — dry matter consumption.

Feed handling and storage also affect CH_4 emissions [55, 56]. For example, milling or granulating can reduce CH_4 emissions per kg of dry matter ingested, as their smaller particle size accelerates their degradation in the rumen. Methanogenesis is generally lower with ensiled feed [52] (presumably, because ensiled feed is already partially fermented during ensiling). Another study [35] showed that intestinal CH_4 emissions from dairy cows at different levels of feed intake depended on the nutritional value and chemical composition of grass silage. Feed based on young plants with less fiber and increased soluble carbohydrates has improved quality, and the addition of a small amount of grain to the forage also gives a favorable result.

The formation of methane in the rumen of ruminants also depends on the amount and composition of concentrates in the diet [54]. With fewer cell walls and easily fermentable carbohydrates (starch and sugar), concentrates promote propionic acid production, reducing CH_4 release [55]. It was noted that the reduction of CH_4 emissions occurred at the addition of concentrates to diets in amounts of 80 and 90%, while no effect was observed at their proportion equal to 35 or 60% [57]. Increasing the proportion of concentrates in the diet of ruminants is not a good strategy for reducing methane production, as diets high in concentrates are low in structural fiber and will compromise rumen function in the long term, leading to subacute or acute acidosis. Probably, it is necessary to select the optimal ratios of roughage and concentrates in the structure of the diet.

The composition of concentrates also influences rumen gas formation, as different ingredients have different carbohydrate compositions. Among non-structural components, sugar is more methanogenic than starch. All carbohydrate fractions contribute to the formation of CH_4, of which starch is the least (probably due to the formation of VFAs with a predominance of propionate). A large amount of starch in the diet reduces intestinal energy loss compared to diets dominated by roughage [58]. Starch fermentation promotes propionate production in the rumen by creating an alternative H_2 sink [59], lower rumen pH, inhibiting methanogen growth, reducing protozoa in the rumen, and limiting interspecific H_2 transfer between methanogens and protozoa. In addition, feeding starch, which can avoid rumen fermentation, potentially provides energy to host animals while ruminal methanogenesis is inhibited. Up to 30% of corn starch may not be fermented in the rumen and digested in the small intestine [60]. Data on the effect of protected starch on the reduction of methane emissions is still very limited, which requires further study of the problem. Sugar, on the other hand, is rapidly and completely degraded in the rumen, increasing butyrate production at the expense of propionate, thereby making sugar concentrates more methanogenic than starch [61]. Sugars increase butyric acid production at higher H_2 partial pressure and higher rumen pH, as confirmed by I.K. Hindrichsen and M. Kreuzer [61] who reported a 40% increase in CH_4 production with sucrose (compared to starch) at high rumen pH, while methane production decreased at low pH.

Replenishing protein deficiency with protein supplements [29] and adding processed oilseeds (as a source of fatty acids) [62] to diets can significantly reduce CH_4 emissions.

Thus, the type of diet of ruminants, the quality of bulky and concentrated feeds and their chemical composition, the ratio of roughage and concentrated feeds, and the preliminary preparation of feeds affect methane emissions into the atmosphere. A promising approach to reduce methane emissions is to add a small amount of grain to forage and feed high quality feed, feed with less fiber and a higher content of soluble carbohydrates.

Feed additives affecting methane production. *Fat supplements*

[63]. The mechanism of suppression of methanogenesis by fat is induced by reducing the fermentation of organic substances, the digestibility of fiber, as well as by direct inhibition of methanogens in the rumen [64]. Data on the methane production in ruminants when fed fat supplements are quite contradictory. For example, the additional inclusion of linseed oil in the diet of cattle contributed to an increase in the species diversity of the rumen microbiota, the number of bacteria of the phylum *Bacteroidetes* (64.2%), as well as a significant increase in representatives of the rumen archaea domain involved in methanogenesis [65].

Organic acids. It is likely that organic acids stimulate the production of propionic acid in the rumen by acting as hydrogen scavengers, thereby reducing the amount of CH_4 [66]. Malate, acrylate, oxaloacetate, and fumarate are carbohydrate fermentation intermediates that are converted to propionate or used in anabolism to synthesize amino acids or other molecules. They can react with hydrogen, which reduces the amount of hydrogen available to form methane [21]. Organic acid supplements have mainly been tested for effects on methane synthesis in vitro with conflicting results. The use of organic acids in diets to reduce gas formation in vivo requires further study. In addition, the use of organic acids may be limited by the risk of rumen acidification causing acidosis in animals.

Ionophores. Ionophores, which can change the movement of cations (in particular, calcium, potassium, sodium) through cell membranes, are classified as antibiotics and are synthesized by soil microorganisms. Among the inophores, monensin and lasalocide are most commonly used to reduce methane emissions. The mechanism of their influence on methanogenesis is associated with the impact on the number of protozoa and bacteria in the rumen. Ionophores act as antimicrobial agents that can disrupt the concentration gradient of calcium, potassium, hydrogen, and sodium ions across certain microbial membranes, initiating an inefficient ion cycle and providing a competitive advantage for some microorganisms at the expense of others. These compounds preferentially inhibit the growth of Gram-positive bacteria that produce lactate, acetate, butyrate, formate, and hydrogen as end products, thereby reducing the availability of hydrogen for methanogens [67]. Although ionophores can reduce methane production, they also appear to impair dry matter intake in both dairy cows and beef steers (68). It has also been shown that the effect of ionophores weakens over time due to the adaptation of protozoa and the development of resistance in succinate- and propionate-producing bacteria [21]. The temporary effect of ionophores and increasing public pressure to reduce the use of antimicrobial feed additives in agricultural production limit the long-term solution to CH_4 emissions with inonophores.

Probiotics. The effect of probiotics on the formation of gases in the rumen may be based, firstly, on an increase in the number of bacteria due to the separation of degraded carbohydrates between microbial cells and fermented products, and secondly, on a shift in the processes of hydrogen utilization from methanogenesis to reductive acetogenesis. Homoacetogenic bacteria produce acetate from CO_2 and H_2 and play an important role in the recycling of enzymatic hydrogen in the colon in monogastrics. For example, co-feeding of moringa extract and a live culture of yeast (*Saccharomyces cerevisiae*) in in vitro experiments performed with goat ruminal contents reduced methane production [69].

S.H. Kim et al. [70] indicate that most probiotics reduce CH^4 production by affecting the activity of ruminal microorganisms without adversely affecting animals. In addition, probiotics enhance rumen fermentation [70]. Other studies have shown that the effect of probiotics on gas exchange depends on their composition. Thus, in vitro results in ruminants showed that conventional and encapsulated probiotics from the group of lactic acid bacteria reduced the production of methane by 6.1 and 33.1%, respectively, compared with the control. In addition, the authors noted an increase in total gas formation by 15.7 and 23.3% when using the same probiotics [71]. In the work of G. Guo et al. [72] lactic acid bacteria contributed not only to the reduction of CH_4 emission per unit of VFA yield, but also improved the quality of fermentation and digestibility of silage fiber. A decrease in the formation of methane in the rumen of dairy cows was noted when using a mixture of propionic acid and lactobacilli in the diet with a high content of starch and fiber in the diets [73]. However, the mechanism of inhibition of methane synthesis by lactic acid bacteria has not yet been fully studied, therefore, in the future, additional study of their effect on microorganisms is necessary. In a study on Holstein heifers, the use of the denitrifying ruminal bacterium Paenibacillus 79R4 (79R4) in the diet contributed to a decrease in the formation of methane in the rumen with intramuscular injection of nitrate and a decrease in nitrite toxicity (a decrease in the concentration of methemoglobin in blood plasma was noted) [74]. Feed additives containing *B. licheniformis* were effective in reducing methane emissions in sheep in vivo, with concomitant improvements in energy and protein utilization [75].

Summing up, we note that studies on the effectiveness of the use of probiotics to reduce the emission of methane and other gases are controversial, and in vivo experiments are few. Due to the availability and wide use of probiotics in animal husbandry, it is of interest to study their effectiveness and find the best products and their complexes to reduce methane formation.

Phytogenics. The term phytogenic feed additives or phytogenics was introduced in the 1980s by Delacon Biotechnik GmbH (Austria) and combines a wide group of natural substances obtained from herbs, spices and their extracts, for example, essential oils, saponins, tannins, flavonoids. Such supplements contain many active ingredients. In addition to improving the palatability and, as a result, increasing the attractiveness of the feed, they increase the enzymatic activity in the gastrointestinal tract of animals, the absorption of nutrients, exhibit antioxidant properties, improve the condition of the gastric mucosa and reproductive function [76].

We would like to dwell on this part of our review in more detail. Table 2 presents the results of in vivo studies on the use of saponins, tannins, flavonoids and essential oils to reduce methane emissions in ruminants.

S e c o n d a r y p r o d u c t s o f p h y t o b i o c e n o s e s. Plant secondary metabolites have long been considered toxic to animals and have been referred to as anti-nutritional factors [77, 78]. However, in the past few decades, interest in these components in animal nutrition has been growing due to their effect on parasite control, rumen fermentation, and methane synthesis [79].

Saponins and tannins. Recently, the potential impact of plant secondary metabolites (PSMs) in reducing methane production has been recognized. The effect of suppressing the release of this gas due to PSMs is associated mainly with the antimicrobial properties of PSMs [80]. Plants produce many secondary compounds, among which much attention has been given to condensed tannins [81, 82] and saponins [83]. The three main plant compounds effective in reducing methane emissions in vitro are condensed tannins, saponins, and essential oils [84].

Tannins are naturally occurring polyphenolic biomolecules found in the bark, wood, fruits, leaves, flowers, and roots of most plant species. Tannins are a subclass of plant polyphenols [78]. Several studies have evaluated the relationship between tannin-rich diets and CH_4 formation in ruminants both in vivo and in vitro [62, 85-87]. Tannins, depending on the chemical structure, can be divided into hydrolysable and condensed tannins [88, 89]. It should be noted that condensed tannins have been more studied with respect to their effect on methane production than hydrolysable tannins. Tannins have the ability to reduce methane synthesis in the rumen directly or indirectly by inhibiting the growth of methanogens or protozoan populations, respectively [78], which has been confirmed in in vitro studies [90, 91].

2. In vivo experiments to study the effect of saponins, tannins, flavonoids and essential oils on methane emission in ruminants

Factor	Animal species, breed	Diet	Effect on methane production	References
Condensed tannins from *Lotus pedunculatus*	Sheep aged 3-4 years and Friesian cows in the final stages of lactation	Pasture based on ryegrass, then alfalfa and then *Lotus pedunculatus*	Reducing methane emissions by the amount of CDM	[96]
Tannin extract (hydrolyzable tannins; *Castanea sativa* wood extract) and saponins (sarsaponin; *Yucca schidigera* extract)	Lambs	Hay: concentrates (1:1) and additionally wheat starch; tannins were added (1 and 2 g/kg DM or 2 and 30 mg/kg DM)	Methane emission increased at low tannin dose compared to control without additives	[119]
Concentrated tannin fodder (*Sericea lespedeza)*	24 female angora goats (BW 41.5 kg)	Pasture with *Sericea lespedeza* and cane fescue	Reducing methane emissions by 30% (g/day) and 50% (g/kg CDM)	[97]
Acacia mearnsii extract	Sheep (75 g fodder DM per kg metabolic body weight)	Partial replacement of ryegrass (*Lolium perenne*) with red clover (*Trifolium pratense*) or alfalfa (*Medicago sativa*) with the addition of 0 or 41 g of *Acacia mearnsii* extract containing 0.615 g/g KT per 1 kg DM	Reducing methane emissions by 15, 13 and 11% (kJ/MJ GE)	[98]
Condensed tannins from the plant *Lespedeza striata*	24 1 year old Boer × Spanish (7/8 Boer) crossbreeds goat kids	Sudanese sorghum + 33; 67 and 100 g tannins	Reducing the absolute emission of methane by 32.8, 47.3 and 58.4%	[99]
Foliage of two tannin-rich shrub legumes *Calliandra calothyrsus*	6 Swiss White Hill lambs	Replacement of 1/3 or 2/3 high quality herbaceous legume forage with shrub legume *Calliandra calothyrsus*	Reducing methane emissions by 24% per day per unit of feed and energy	[100]
Tannins extracted from the bark of black locust (*Acacia mearns*i, KT 60.3%)	60 lactating cows	Pasture with ryegrass, crushed triticale grain (5 kg/day), tannin (163 and 326 g/day with a decrease to 244 g/day by day 17)	Reduction of methane emissions by 14 and 29% in accordance with the dose (about 10 and 22% of CDM)	[102]

Continued Table 2

Pulp of pumpkin seeds (*Terminalia chebula*), garlic (*Allium sativum*) and their mixture	16 sheep (age 22 months, BW 29.96±1.69 kg)	Feed:concentrate (50:50) + phytonutrients (1% dietary DM, alone or mixed)	Reducing methane emissions up to 24% of digested DM and consumed DM	[103]
Yucca schidigera (YS) extract	Sheep for fattening	Grass silage:concentrate (70:30) + 120 mg YS extract/kg DM	Reducing methane emissions per body weight	[104]
Acacia mearnsii tannin extract	12 Holstein dairy cows	Pasture grass millet + 6 kg concentrates + 120 g tannin extract	Reducing methane emissions by 32%	[146]
Chestnut tannins or chestnut tannins + querbajo tannins	75 crossbred steers (BW 292±4.1 kg)	Alfalfa:barley (50:50) + chestnut tannins (0.25% DM) or chestnut tannins (0.125 or 0.75% DM) + querbacho tannins (0.125 or 0.75% DM)	No reduction in methane emissions was observed	[80]
Tea saponins alone and in combination with soybean oil	32 Huzhou weaned lambs (age 50 days, BW 14.2±1.38 kg)	60% Chinese wild rye (*Aneurolepidium chinese* Kitagawa) and 40% concentrate blend + saponins 3 g/day (or saponins 3 g/day + soybean oil 3% CDM)	Daily methane production decreased by 27.7% and 18.9% respectively	[117]
Tea saponins	12 Hu sheep (aged 7 months, BW 21.5±1.80 kg)	600 g/kg Chinese wild rye and 400 g/kg concentrate mix, 3 g/d tea saponins	Reduced CH_4 production in the rumen, effect similar to that of defaunization	[118]
Condensed tannin of tannic acacia species	Bapedi sheep (aged 1 year, BW 25±1.6 kg)	80% grass hay and 20% concentrates, refined condensed tannin (0, 30, 40, 50 g/kg DM)	Reducing methane emissions by 51-60%	[81]
Condensed tannins and saponins, obtained from *Enterolobium cyclocarpum* (Jacq.) Griseb. crushed pods mixed with foliage of *Gliricidia sepium* (Jacq.) Steud.	4 crossbred heifers (*Bos taurus* × *Bos indicus*) (aged 12 months, BW 218±18 kg)	79.9% hay of *Brachiaria brizantha* (Hochst. ex A. Rich.) and 20.1% balanced mix based on soybean meal, bran, cane molasses and minerals, 15, 30 and 45% DM based on dry and crushed leaves of *G. sepium* and pods of *E. cyclocarpum* in equal proportions	Methane emissions decreased by 0.16 times (calculated on DCP)	[120]
Dried leaves of *Leucaena leucocephala* (DLL)	4 crossbred heifers (*Bos taurus* × *Bos indicus*) (BW 310±9.6 kg)	Hay and concentrates + dried leaves of *Leucaena leucocephala* (DLL) (0, 12, 24 and 36% DM)	Reducing the formation of methane (on average by 25% (per 1 kg of DP)	[121]
Levcaena *Leucaena leucocephala* (Lam.) De Wit Cunningham variety, fresh	8 Lucerne heifers (aged 19±3 months, BW 218±18 kg)	100% chickweed *Cynodon plectostachyus* K. Schum) and 76% chickweed with 24% levcaena *Leucaena leucocephala* (Lam.) De Wit Cunningham variety fresh	No increase observed in methane emissions with an increase in productivity, which reduced methane emissions by 1 kg of production	[122]

Flour from the pods of *Samanea saman*	4 crossbred heifers (*Bos taurus* × *B. indicus*) (BW 261.5±1.29 kg)	Ground green grass feed, soy flour, wheat bran and sugar cane molasses, minerals with the addition of ground *S. saman* pods were 0, 10, 20 and 30% DM	At a dosage of ground S. saman pods of 30% DM, methane emissions per animal decreased by 50.9% (in absolute units) and by 56.9% (calculated per 1 kg CDM)	[123]
Tannins from tropical legumes *Desmanthus leptophyllus* and *D. bicornutus*	14 Droughtmaster bulls (aged 12 months, BW 296±5 kg)	Rhodes grass hay (*Chloris gayana*) with fresh desmanthus (0, 15, 31 and 22% DM)	Linear reduction of methane emissions without reduction of DM consumption	[82]
Mulberry leaf extracts and resveratrol from *Polygonum cuspidatum*	10 crossbred first-lamb ewes (Dorper × Han, BW 60.0±1.73 kg)	Basal diet without additives, supplemented with flavonoids from mulberry leaves (2 g/day per sheep) and supplemented with resveratrol (0.25 g/day per sheep)	Reducing the formation of CH_4 by 10.64% per 1 kg of CDM	[147]
Blend of essential oils containing coriander seed oil, eugenol and geranyl acetate	4 Holstein cows (BW 603±70 kg, day 296 of lactation) and 4 Belgian blue beef heifers (BW 484±111 kg)	Dairy cattle — grass silage (460 g/kg DM), corn silage (370 g/kg DM) and soybean meal (50 g/kg DM), concentrates, 0.2 g/day of a mixture of essential oils (120 g/kg DM); beef cattle — corn silage ad libitum and supplementary feeding with concentrates, 0.2 g/day of essential oils	After 6 weeks of supplementation in dairy cattle, CH_4 emissions decreased (g/day) by 15%, re-calculated for DM consumed by 14% ($p = 0.07$), in beef cattle, these indicators tended to increase by 10 and 11 % and decreased by 20% when calculated based on body weight	[142]
Coriander seed oil blend, geranyl acetate and eugenol	149 early lactating Holstein-Friesian cows	Grass feed, whole grain wheat, corn silage, 1 g of a mixture of essential oils with drinking water	Decreased methane production from 438 to 411 g/day	[143]
Essential oil blend (0.17 g/kg DM), lauric acid (65 g/kg DM), essential oil blend with lauric acid	8 cows (BW 610±59 kg)	Feed mix of 40% corn silage, 30% grass silage and 30% concentrates	The reduction in methane emissions was more pronounced when using a mixture of additives	[144]
A mixture of phytogenic supplements from dried and crushed leaves of *Populus deltoides* and *Eucalyptus citriodora* (50:50 by weight)	12 lactating Murra buffaloes (*Bubalus bubalis*) (BW 510.50±32.12 kg) at an early stage of lactation	Chopped young sorghum plants, wheat straw and mixtures of concentrates with phytogenic additives (15 g/kg DM)	Reducing the concentration of methane in exhaled air by 37.3%	[148]

N o t e. BW — bodyweight, DM — dry matter, CDM — consumed dry matter, GE — gross energy, DCP — digested crude protein, DP — digestible protein.

There are several possible hypotheses explaining the mechanisms of action of tannins on the reduction of methane formation in the animal body [89]. One of them suggests a direct effect of condensed tannins on the methanogenic archaea of the rumen due to the binding of protein adhesin or parts of the cell membrane, which disrupts the formation of the methanogen-protozoal complex, reduces the interspecies transfer of hydrogen, and inhibits the growth of methanogen. The high molecular weight and polyphenolic nature of tannins lead to the formation of complexes with microbial enzymes or cell walls. The activity shown can cause inhibition of cellulolytic or proteolytic bacteria or methanogens [92]. The mechanism of action of tannins strictly depends on their chemical structure, as well as on the type of bacteria [78]. Another possible explanation is indirect inhibition by reducing the availability of nutrients (carbohydrates, amino acids) for rumen microorganisms and the formation of tannin-protein complexes in the rumen [93], which reduces feed digestibility and disrupts the structure of the rumen microbiota. The latter theory suggests that the condensed tannins themselves act as hydrogen scavengers, reducing its availability for carbon dioxide reduction to methane [89]. Condensed tannins have been found to be more nutrient-binding than hydrolyzed tannins, mainly due to their higher degree of polymerization, making them more difficult to degrade in the rumen [91]. In another work, the same authors note that, on the contrary, hydrolyzed tannins had a greater ability to precipitate protein, which is associated with increased biological activity and a higher ability to suppress the formation of methane compared to condensed tannins [91].

An in vivo study in fistula sheep examined the direct inhibition of certain Gram-positive specialized ruminal fibrolytic bacteria (*Fibrobacter succinogenes*, *Ruminococcus albus*, *Ruminococcus flavefaciens*, *Butyrivrio proteoclasticus*) by condensed tannins [94]. G.C. Waghorn and S.L. Woodward [95] report that condensed lotus tannins reduce methane production on a dry matter basis by about 15% in sheep and dairy cows. A similar effect has been noted in other studies [96]. Feeding goats with the perennial *Lespedeza cuneata*, which contains condensed tannins, showed a 57% reduction in methane production per kg of dry matter ingested compared to that observed in goats fed a mixture of *Digitaria ischaemum* and *Festuca arundinacea* [97]. It has been found that methanogenesis is reduced by 13% in sheep eating *Acacia mearnsii* with a tannin content of 41 g/kg dry matter [98]. A decrease in methanogenesis with the use of tannins from the plant *Lespedeza striata* in the diet of goats was noted by G. Animut et al. [99]. Tannin-containing *Callinada calothyrsus* and *Fleminga macrophylla* reduced methane production in lambs by 24% [100], but condensed tannin extract from *Schinopsis quebrachocolorado* [62] and tannin-containing sorghum silage [101] fed to cattle did not suppress methanogenesis. A decrease in methanogenesis in in vivo experiments on cows and sheep using tannin from various sources has been noted in a number of studies [102-104].

Saponins are natural detergents chemically defined as high molecular weight glycosides in which sugars are linked to a triterpene or steroidal aglycone moiety. As secondary plant metabolites, saponins have the ability to modulate rumen fermentation while reducing methane production and ammonia concentration [105]. Saponins mainly affect the population of protozoa [106-108], disrupting their cell membrane integrity [109, 110]. The symbiosis of protozoa with methanogenic bacteria in the rumen is well known and it has been suggested that selective suppression of protozoa may be a promising approach to reduce methane production. Plants rich in saponins have the potential to increase microbial protein flux from the rumen, increase feed efficiency, and reduce methanogenesis.

R. Wallace et al. [111] suggested that saponins can destroy protozoan cells by forming complexes with sterols on the membrane surface, which are then destroyed and disintegrated. In addition, some saponins affect various types of membrane proteins, such as Ca^{2+} channel proteins and Na^+/K^+ ATPase [112]. E. Ramos-Morales et al. [113] suggest that the effect of saponins on protozoa is temporary beause the bacteria can break down saponins into sapogenins, compounds that cannot affect protozoa.

Saponins have been shown to inhibit protozoa in vitro and also limit the availability of hydrogen for methanogenesis [114]. An in vitro study showed that liquid extracts of *Yucca schidigera* and *Quillaja saponaria* added in amounts from 2 to 6 ml/l of rumen fluid, reduce the number of protozoa in the rumen and can potentially change the ammonia content, propionate concentration and the ratio of acetate to propionate. In the same study, the effect of *Y. schidigera* was manifested in a decrease in the rate and formation and volume of methane, depending on the dose, by 42 and 32%, respectively, while in *Q. saponaria*, the effect of inhibition of methanogenesis was not manifested [115].

In an in vivo study, dietary extract of *Y. schidigera* (120 mg) reduced methane production in fattening sheep (104). In a study by L. Holtshausen et al. [116], cows received whole *Y. schidigera* plant powder (10 g/kg dry matter) or whole *Q. saponaria* plant powder (10 g/kg dry matter), both powders containing saponin. The authors stated that previous in vitro studies have shown a reduction in methane production at higher doses of saponins (15 g/kg DM or more), but these values were avoided in vivo to minimize the impact on feed digestibility [114]. Under natural conditions, no effect of the herbal supplement was found, and the authors concluded that the decrease in in vitro methane content was probably due to a decrease in digestibility and fermentation of the feed (116). Tea saponins, alone or in combination with fat supplements, have been shown to reduce methane emissions in sheep in vivo [117, 118].

Combinations of tannins and saponins in ruminant diets have proven to be effective in terms of methane emissions [119, 120]. In a number of in vivo experiments on ruminants, a decrease in methane emission in animals was also noted when saponins and tannins were included in the diet [121-123].

Flavonoids and essential oils. Flavonoids are C_6-C_3-C_6 polyphenols found in seeds and vegetables that exhibit anti-inflammatory, antioxidant, and antimicrobial properties [124]. Flavonoids are highly biologically active, reducing or preventing cellular damage caused by free radicals [125]. Flavonoids act on gram-positive microorganisms by inhibiting the functions of the cytoplasmic membrane, inhibiting the synthesis of the bacterial cell wall or nucleic acids. Flavonoids included in the diet of ruminants have been shown to increase productivity by increasing the production of propionate compared to acetate [126]. The influence of various flavonoids (flavones, myricetin, naringin, catechin, rutin, quercetin and kaempferol) at a concentration of 4.5% of the DM on the microbial activity of the rumen in vitro was evaluated [127]. The results showed that all flavonoids, except for naringin and quercetin, reduced the ability of the microbiota to degrade dry matter. Gas production decreased under the influence of flavone, myricetin and kaempferol, while naringin, rutin and quercetin markedly increased its production. Flavonoids significantly suppressed methane production. The total concentration of VFAs decreased in the presence of flavone, myricetin and kaempferol. All flavonoids, except for naringin and quercetin, significantly reduced the activity of carboxymethyl cellulase, cellulase, xylanase, and β-glucosidase, purine content, and microbial protein synthesis. Under the influence of flavones, myricetin, catechin, rutin and kaempferol, the microbial population of the rumen was reduced. The growth of the population of protozoa and methanogens was suppressed by naringin and quercetin. The results of this study showed that naringin and quercetin at 4.5% DM are potentially suitable for suppressing methane production without any negative effect on microbial fermentation in the rumen.

A commercial citrus extract of a mixture of flavonoids reduced methane production, the abundance of hydrogenotrophic methanogenic archaea, while increasing propionate concentration and population of *Megasphaera elsdenii* in vitro [128]. In Holstein cows, when an extract of alfalfa flavonoids (60 mg/kg of body weight) was added to the diet, the ratio of valeric acid and the total amount of VFAs in the rumen increased, the composition of milk and the digestibility of nutrients improved, and there was a trend towards an increase in the ruminal population of *Butyrivibrio fibrisolvens* [129]. In an in vitro experiment, the flavonoid luteolin-7-glucoside was found to reduce methane [130]. Based on available data, flavonoids have the ability to reduce methane emissions, but further in vivo studies are needed.

In vitro experiments have examined the effect of the combination of garlic powder and bitter orange extract on methane production, rumen fermentation, and feed digestibility in various diet structures (ratios of roughage grasses to concentrates) [131]. The results showed a strong suppression of methane production in all variants. For a diet consisting only of grass, the effectiveness of the additive was 44.0%, when the diet was supplemented with concentrates in a ratio of 20:80, it was 69.2%. The use of flavonoids significantly increased the concentration of ammonia nitrogen and lowered pH, while the digestibility of organic matter and fiber did not decrease. When using these nutritional factors, regardless of diets, there was a change in rumen fermentation with less acetate and more propionate and butyrate, with an increase in total VFAs.

There is a known method for reducing the concentration of methane in the rumen of ruminants through the use of medicinal plants — wormwood herb (10.0 g/kg DM diet), elecampane rhizomes and roots (6.0 g/kg DM diet) [132]. It has also been proposed to orally administer a food composition containing flavanones from a citrus plant. The authors used compositions with different combinations of components: neohesperidin, poncirin, and naringin [133].

Essential oils are volatile plant-derived secondary metabolites with very strong antimicrobial properties that inhibit the growth and viability of most microorganisms in the rumen [134]. The mechanism of action of essential oils varies depending on their type [135]. All essential oils contain chemical components (terpenoids, phenols and phenols) and functional groups that have strong antimicrobial properties. Due to their lipophilic nature, essential oils have a high affinity for microbial cell membranes [136]. When essential oil is used, methanogenesis in the rumen is reduced, especially due to the reduction of microbial populations. Nevertheless, the mechanisms of the influence of essential oils on the processes of fermentation in the rumen of ruminants require more in-depth study.

The effect of plant secondary metabolites on methane production has been studied in vitro [137]. Nine concentrations of the following metabolites were compared: 8-hydroxyquinoline, α-terpineol, camphor, bornyl acetate, α-pinene, thymoquinone, and thymol. All compounds can alter rumen fermentation and reduce CH_4 production. The minimum concentrations that reduce the production of CH_4 were as follows: 8-hydroxyquinoline 8 mg/l, thymoquinone 120 mg/l, thymol 240 mg/l, α-terpineol + camphor + bornylacetate + α-pinene 480 mg/l. The authors attribute these effects to changes in the structure of the rumen bacterial community [137]. As shown by ion semiconductor sequencing, the influence of secondary plant metabolites was most pronounced in the predominance of the relative abundance of the families *Lachnospiraceae*, *Succinivibrionaceae*, *Prevotellaceae*, unclassified *Clostridiales* and *Ruminococcaceae*. CH_4 production correlated negatively with the relative abundance of *Succinivibrionaceae* and positively with the relative abundance of *Ruminococcaceae*.

In other in vitro experiments, the effect of *Macleaya cordata* extract at six concentrations (0.01, 0.11, 0.21, 0.31, 0.41 and 0.51%) was studied when incubated for 12 and 24 h for methane formation [138]. Methane emission decreased depending on the dose of *Macleaya* extract after 3, 6, 9, and 12 h of incubation, but increased after 24 h. The addition of 0.11% *M. cordata* extract effectively reduced methane production without affecting in vitro digestion of DM.

Research by D. Petri et al. [139] showed in vitro that a substrate containing a mixture of medicinal plants (wormwood, chamomile, fumitory and mallow) had a strong antioxidant capacity in the rumen content and had the potential to reduce methane production. Thymol at a dose of 200 mg/l, when incubated in the cicatricial contents for 24 h, contributed to a decrease in methane formation, which the authors attribute to changes in the quantitative composition of bacteria, archaea, and protozoa [140].

The use of a mixture of essential oils, bioflavonoids and tannins in animal diets significantly reduced the total gas emission, which was noted for methane in an in vitro experiment after 16, 20 and 24 h of incubation. In addition, a decrease in the concentration of acetic acid and an increase in the concentration of propionic acid were observed in the rumen after 16 and 24 h. The group of animals receiving the mixture showed an increase in milk yield and DM consumption while maintaining the milk quality [141].

In general, it should be emphasized that the number of studies on the effect of flavonoids and other secondary plant metabolites on methane production in vivo is very limited. In addition to the examples above, the effectiveness of the methane synthesis suppression in the cattle rumen with dietary mixture of essential oils are reported [142-146]. Other in vivo experiments have examined sheep and buffalo gassing during fermentation as influendes by a mixture of phytogenic supplements in diets [147, 148]. Thus, the number of in vivo studies on the use of flavonoids and other secondary plant metabolites to reduce methane emissions is very limited. The results obtained are variable and depend on the type of metabolite, its characteristics and the diet of the animals. In addition to continuing research to assess the potential of phytogenics for animal husbandry practice, long-term observations are needed in connection with the possible adaptation of ruminal microorganisms to a bioactive metabolite, as well as identifying differences between its effects in vitro and in vivo. A promising way to reduce methane emissions into the atmosphere is the integrated use of various phytogenics in animal diets.

Summing up, we note several important, in our opinion, aspects. Although the efforts of geneticists, breeders, and animal nutrition specialists have significantly reduced methane emissions per unit of livestock production, the growing demand for food requires further reduction in both the intensity of emissions per unit of production and absolute emissions per animal. However, the available evidence on the effectiveness of various strategies to reduce methane emissions in ruminant species is conflicting [149]. Modern methods make it possible to more accurately assess the formation of greenhouse gases in the animal body, but remain expensive and technically complex, so their application is mainly limited to scientific research. The development of biomarkers for methane production is at a relatively early stage and should become a priority in the future. It also requires additional study of probiotics, phytogenics, other feed factors and their complexes as potential means of reducing methane emissions, taking into account the structure of diets, dosage, animal species and other factors. In addition, it is important to understand that researchers do not yet have sufficient information about the impact of strategies to reduce methane emissions into the atmosphere on productivity, animal health, the state of the antioxidant and hormonal systems, and the structure of the rumen microbiome.

In summary, over the past 50 years, a significant amount of research has been carried out that has deepened the understanding of the complex processes of fermentation and methanogenesis in the rumen in ruminants and made it possible to gain an understanding of the means by which methane production can be reduced. However, sustainable strategies for dealing with the problem have not yet been adopted. As the results of studies show, the use of feed factors of various nature (ionophores, probiotics, plant secondary metabolites) can serve as a cheap and environmentally friendly strategy to reduce methane formation in ruminants with a positive effect on animal tolerance. A combination of various phytogenics seems to be an actual and promising approach. In numerous in vitro studies, the effectiveness of reducing methane emissions depends on many factors. Therefore, an integrated approach is needed to reduce gas formation in ruminants while maintaining the state of enzymatic processes, digestibility and assimilation of nutrients in feed rations.

REFERENCES

1. Pachauri R.K., Allen M.R., Barros V.R., Broome J., Cramer W., Christ R., Church J.A., Clarke L., Dahe Q., Dasgupta P., Dubash N.K., Edenhofer O., Elgizouli I., Field C.B., Forster P., Friedlingstein P., Fuglestvedt J., Gomez-Echeverri L., Hallegatte S., Hegerl G., Howden M., Jiang K., Jimenez Cisneroz B., Kattsov V., Lee H., Mach K.J., Marotzke J., Mastrandrea M.D., Meyer L., Minx J., Mulugetta Y., O'Brien K., Oppenheimer M., Pereira J.J., Pichs-Madruga R., Plattner G.-K., Pцrtner H.-O., Power S.B., Preston B., Ravindranath N.H., Reisinger A., Riahi K., Rusticucci M., Scholes R., Seyboth K., Sokona Y., Stavins R., Stocker T.F., Tschakert P., van Vuuren D., van Ypserle J.-P. *Climate change 2014: synthesis report. Contribution of working groups i, ii and iii to the fifth assessment report of the intergovernmental panel on climate change* /R. Pachauri, L. Meyer (eds). Geneva, Switzerland, IPCC, 2014.
2. Islam M., Lee S.S. Advanced estimation and mitigation strategies: a cumulative approach to enteric methane abatement from ruminants. *Journal of Animal Science and Technology*, 2019, 61(3): 122-137 (doi: 10.5187/jast.2019.61.3.122).
3. Skytt T., Nielsen S.N., Jonsson, B.G. Global warming potential and absolute global temperature change potential from carbon dioxide and methane fluxes as indicators of regional sustainability — a case study of Jämtland, Sweden. *Ecological Indicators*, 2020, 110: 105831 (doi: 10.1016/j.ecolind.2019.105831).
4. Haque M.N. Dietary manipulation: a sustainable way to mitigate methane emissions from ruminants. *Journal of Animal Science and Technology*, 2018, 60(1): 15 (doi: 10.1186/s40781-018-0175-7).
5. Hristov A.N., Oh J., Lee C., Meinen R., Montes F., Ott T., Firkins J., Rotz A., Dell C., Adesogan A., Yang W., Tricarico J., Kebreab E., Waghorn G., Dijkstra J., Oosting S. *Mitigation of greenhouse gas emissions in livestock production. A review of options for non-CO_2 emissions. FAO Animal Production and Health Paper No.177.* P.J. Gerber, B. Henderson, H.P.S. Makkar (eds.). FAO, Rome, 2013.
6. McAllister T.A., Meale S.J., Valle E., Guan L.L., Zhou M., Kelly W.J., Henderson G., Attwood G.T., Janssen P.H. Ruminant nutrition symposium: use of genomics and transcriptomics to identify strategies to lower ruminal methanogenesis. *Journal of Animal Science*, 2015, 93(4): 1431-1449 (doi: 10.2527/jas.2014-8329).
7. Sandoval-Pelcastre A.A., Ramírez-Mella M., Rodríguez-Ávila N.L., Candelaria-Martínez B. Árboles y arbustos tropicales con potencial para disminuir la producciyn de metano en ruminates. *Tropical and Subtropical Agroecosystems*, 2020, 23(33): 1-16.
8. Opio C., Gerber P., Mottet A., Falcucci A., Tempio G., MacLeod M., Vellinga T., Henderson B., Steinfeld H. *Greenhouse gas emissions from ruminant supply chains — a global life cycle assessment.* FAO, Rome, 2013.
9. Gerber P.J., Steinfeld H., Henderson B., Mottet A., Opio C., Dijkman J., Falcucci A., Tempio G. *Tackling climate change through livestock — a global assessment of emissions and mitigation opportunities.* FAO, Rome, 2013.
10. *Ukaz Prezidenta RF ot 4 noyabrya 2020 g. № 666 «O sokrashchenii vybrosov parnikovykh gazov»* [Decree of the President of the Russian Federation of November 4, 2020 No. 666 «On the reduction of greenhouse gas emissions»]. Available: https://www.garant.ru/products/ipo/prime/doc/74756623. Accessed: 22.08.2022 (in Russ.).
11. Petrunina I.V., Gorbunova N.A. *Pishchevye sistemy*, 2022, 5(3): 202-211 (doi: 10.21323/2618-9771-2022-5-3-202-211) (in Russ.).

12. Beauchemin K.A., Ungerfeld E.M., Eckard R.J., Wang M. Review: Fifty years of research on rumen methanogenesis: lessons learned and future challenges for mitigation. *Animal*, 2020, 14(S1): 2-6 (doi: 10.1017/S1751731119003100).
13. Calabrò S. Plant secondary metabolites. In: *Rumen microbiology: from evolution to revolution*. A.K. Puniya, R. Singh, D.N. Kamra (eds.). Springer, New Delhi, India, 2015: 153-189 (doi: 10.1007/978-81-322-2401-3_11).
14. Rooke J.A., Wallace R.J., Duthie C.A., McKain N., de Souza S.M., Hyslop J.J., Ross D.W., Waterhouse T., Roehe R. Hydrogen and methane emissions from beef cattle and their rumen microbial community vary with diet, time after feeding and genotype. *British Journal of Nutrition*, 2014, 112(3): 398-407 (doi: 10.1017/S0007114514000932).
15. Cammack K.M., Austin K.J., Lamberson W.R., Conant G.C., Cunningham H.C. Tiny but mighty: the role of the rumen microbes in livestock production. *Journal of Animal Science*, 2018, 96(2): 752-770 (doi: 10.1093/jas/skx053).
16. Stewart R.D., Auffret M.D., Warr A., Wiser A.H., Press M.O., Langford K.W., Liachko I., Snelling T.J., Dewhurst R.J., Walker A.W., Roehe R., Watson M. Assembly of 913 microbial genomes from metagenomic sequencing of the cow rumen. *Nature Communications*, 2018, 9: 870 (doi: 10.1038/s41467-018-03317-6).
17. De la Fuente G., Yacez-Ruiz D.R., Seradj A.R., Balcells J., Belanche A. Methanogenesis in animals with foregut and hindgut fermentation: a review. *Animal Production Science*, 2019, 59(12): 2109-2122 (doi: 10.1071/AN17701).
18. Poulsen M., Schwab C., Borg Jensen B., Engberg R.M., Spang A., Canibe N., Нщjberg O., Milinovich G., Fragner L., Schleper C., Weckwerth W., Lund P., Schramm A., Urich T. Methylotrophic methanogenic thermoplasmata implicated in reduced methane emissions from bovine rumen. *Nature Communications*, 2013, 4(1): 1428 (doi: 10.1038/ncomms2432).
19. Solden L.M., Naas A.E., Roux S., Daly R.A., Collins W.B., Nicora C.D., Purvine S.O., Hoyt D.W., Schückel J., Jørgensen B., Willats W., Spalinger D.E., Firkins J.L., Lipton M.S., Sullivan M.B., Pope P.B., Wrighton K.C. Interspecies cross-feeding orchestrates carbon degradation in the rumen ecosystem. *Nature Microbiology*, 2018, 3(11): 1274-1284 (doi: 10.1038/s41564-018-0225-4).
20. Wolin M., Millert L.C., Stewart S. Microbe-microbe interactions. In: *The rumen microbial ecosystem*. P.N. Hobson, S. Stewart (eds.). Springer, Dordrecht, 1997: 467—491 (doi: 10.1007/978-94-009-1453-7_11).
21. Patra A., Park T., Kim M., Yu Z. Rumen methanogens and mitigation of methane emission by anti-methanogenic compounds and substances. *Journal of Animal Science and Biotechnology*, 2017, 8(1): 13 (doi: 10.1186/s40104-017-0145-9).
22. Van Lingen H.J., Plugge C.M., Fadel J.G., Kebreab E., Bannink A., Dijkstra J. Thermodynamic driving force of hydrogen on rumen microbial metabolism: a theoretical investigation. *PLoS ONE*, 2016, 11(10): e0161362 (doi: 10.1371/journal.pone.0161362).
23. Huws S.A., Creevey C.J., Oyama L.B., Mizrahi I., Denman S.E., Popova M., Mucoz-Tamayo R., Forano E., Waters S.M., Hess M., Tapio I., Smidt H., Krizsan S.J., Yácez-Ruiz D.R., Belanche A., Guan L., Gruninger R.J., McAllister T.A., Newbold C.J., Roehe R., Dewhurst R.J., Snelling T.J., Watson M., Suen G., Hart E.H., Kingston-Smith A.H., Scollan N.D., do Prado R.M., Pilau E.J., Mantovani H.C., Attwood G.T., Edwards J.E., McEwan N.R., Morrisson S., Mayorga O.L., Elliott C., Morgavi D.P. Addressing global ruminant agricultural challenges through understanding the rumen microbiome: past, present, and future. *Frontiers in Microbiology*, 2018, 9: 2161 (doi: 10.3389/fmicb.2018.02161).
24. Wang M., Wang R., Xie T.Y., Janssen P.H., Sun X.Z., Beauchemin K.A., Tan Z.L., Gao M. Shifts in rumen fermentation and microbiota are associated with dissolved ruminal hydrogen concentrations in lactating dairy cows fed different types of carbohydrates. *Journal of Nutrition*, 2016, 146(9): 1714-1721 (doi: 10.3945/jn.116.232462).
25. Cantalapiedra-Hijar G., Abo-Ismail M., Carstens G.E., Guan L.L., Hegarty R., Kenny D.A., McGee M., Plastow G., Relling A., Ortigues-Marty I. Biological determinants of between-animal variation in feed efficiency of growing beef cattle: Review. *Animal*, 2018, 12(s2): s321-s335 (doi: 10.1017/S1751731118001489).
26. Ungerfeld E.M. Inhibition of rumen methanogenesis and ruminant productivity: a meta-analysis. *Frontiers in Veterinary Science*, 2018, 5: 113 (doi: 10.3389/fvets.2018.00113).
27. Zhang Q., Difford G., Sahana G., Lovendahl P., Lassen J., Lund M.S., Guldbrandtsen B., Janss L. Bayesian modeling reveals host genetics associated with rumen microbiota jointly influence methane emission in dairy cows. *The ISME Journal*, 2020, 14(8): 2019-2033 (doi: 10.1038/s41396-020-0663-x).
28. Broucek J. Production of methane emissions from ruminant husbandry: a review. *Journal of Environmental Protection*, 2014, 5(15): 1482-1493 (doi: 10.4236/jep.2014.515141).
29. Bernier J.N., Undi M., Plaizier J.C., Wittenberg K.M., Donohoe G.R., Ominski K.H. Impact of prolonged cold exposure on dry matter intake and enteric methane emissions of beef cows overwintered on low-quality forage diets with and without supplemented wheat and corn dried distillers' grain with solubles. *Canadian Journal of Animal Science*, 2012, 92(4): 493-500 (doi:

10.4141/cjas2012-040).
30. Wang S., Pisarcikova J., Kreuzer M., Schwarm A. Utility of an in vitro test with rumen fluid from slaughtered cattle for capturing variation in methane emission potential between cattle types and with age. *Canadian Journal of Animal Science*, 2017, 98(1): 61-72 (doi: 10.1139/cjas-2016-0238).
31. Dong L., Li B., Diao Q. Effects of dietary forage proportion on feed intake, growth performance, nutrient digestibility, and enteric methane emissions of Holstein heifers at various growth stages. *Animals*, 2019, 9(10): 725 (doi: 10.3390/ani9100725).
32. Boadi D.A., Wittenberg K.M. Methane production from dairy and beef heifers fed forages differing in nutrient density using the Sulphur hexafluoride (sf6) tracer gas technique. *Canadian Journal of Animal Science*, 2002, 82(2): 201-206 (doi: 10.4141/A01-017).
33. Shreck A.L., Zeltwanger J.M., Bailey E.A., Jennings J.S., Meyer B.E., Cole N.A. Effects of protein supplementation to steers consuming low-quality forages on greenhouse gas emissions. *Journal of Animal Science*, 2021, 99(7): skab147 (doi: 10.1093/jas/skab147).
34. Li Z., Liao W., Yang Y., Gao Z., Ma W., Wang D., Cai Z. CH_4 and N_2O emissions from China's beef feedlots with ad libitum and restricted feeding in fall and spring seasons. *Environmental Research*, 2015, 138: 391-400 (doi: 10.1016/j.envres.2015.02.0).
35. Warner D., Bannink A., Hatew B., Van Laar H., Dijkstra J. Effects of grass silage quality and level of feed intake on enteric methane production in lactating dairy cows. *Journal of Animal Science*, 2017, 95(8): 3687-3699 (doi: 10.2527/jas.2017.1459).
36. Rowe S.J., Hickey S.M., Jonker A., Hess M.K., Janssen P., Johnson T., Bryson B., Knowler K., Pinares-Patino C., Bain W., Elmes S., Young E., Wing J., Waller E., Pickering N., McEwan J.C. Selection for divergent methane yield in New Zealand sheep — a ten-year perspective. *Proc. of the 23rd Conf. of the association for the advancement of animal breeding and genetics (AAABG)*. Armidale, New South Wales, Australia, 2019: 306-309.
37. Korotkiy V.P., Zaytsev V.V., Buryakov N.P., Kuchin A.V., Ryzhov V.A., Turubanov A.I. *Sposob snizheniya metanogeneza u krupnogo rogatogo skota. C1 2777053 (RF), A 61 K 38/00. OOO Nauchno-tekhnicheskiy tsentr «Khiminvest» (RF), № 2021137457. Zayav. 16.12.2021. Opubl. 01.08.2022* [Method for reducing methanogenesis in cattle. C1 2777053 (RF), A 61 K 38/00. LLC Scientific and Technical Center Khiminvest (RF), № 2021137457. Appl. 12.16.2021. Publ. 08.01.2022] (in Russ.).
38. De Mulder T., Peiren N., Vandaele L., Ruttink T., De Campeneere S., Van de Wiele T., Goossens K. Impact of breed on the rumen microbial community composition and methane emission of Holstein Friesian and Belgian Blue heifers. *Livestock Science*, 2018, 207: 38-44 (doi: 10.1016/j.livsci.2017.11.009).
39. Hristov A.N., Kebreab E., Niu M., Oh J., Bannink A., Bayat A.R., Boland T.M., Brito A.F., Casper D.P., Crompton L.A., Dijkstra J., Eugène M., Garnsworthy P.C., Haque N., Hellwing A.L.F., Huhtanen P., Kreuzer M., Kuhla B., Lund P., Madsen J., Martin C., Moate P.J., Muetzel S., Mucoz C., Peiren N., Powell J.M., Reynolds C.K., Schwarm A., Shingfield K.J., Storlien T.M., Weisbjerg M.R., Yácez-Ruiz D.R., Yu Z. Symposium review: uncertainties in enteric methane inventories, measurement techniques, and prediction models. *Journal of Dairy Science*, 2018, 101(7): 6655-6674 (doi: 10.3168/jds.2017-13536).
40. Huhtanen P., Cabezas-Garcia E.H., Utsumi S., Zimmerman S. Comparison of methods to determine methane emissions from dairy cows in farm conditions. *Journal of Dairy Science*, 2015, 98(5): 3394-3409 (doi: 10.3168/jds.2014-9118)
41. Kumar S., Choudhury P.K., Carro M.D., Griffith G.W., Dagar S.S., Puniya M., Calabro S., Ravella S.R., Dhewa T., Upadhyay R.C., Sirohi S.K., Kundu S.S., Wanapat M., Puniya A.K. New aspects and strategies for methane mitigation from ruminants. *Applied Microbiology and Biotechnology*, 2014, 98(1): 31-44 (doi: 10.1007/s00253-013-5365-0).
42. Knapp J.R., Laur G.L., Vadas P.A., Weiss W.P., Tricarico J.M. Invited review: Enteric methane in dairy cattle production: Quantifying the opportunities and impact of reducing emissions. *Journal of Dairy Science*, 2014, 97(6): 3231-3261 (doi: 10.3168/jds.2013-7234).
43. Negussie E., de Haas Y., Dehareng F., Dewhurst R.J., Dijkstra J., Gengler N., Morgavi D.P., Soyeurt H., van Gastelen S., Yan T., Biscarini F. Invited review: Large-scale indirect measurements for enteric methane emissions in dairy cattle: a review of proxies and their potential for use in management and breeding decisions. *Journal of Dairy Science*, 2017, 100(4): 2433-2453 (doi: 10.3168/jds.2016-12030).
44. Tapio I., Snelling T.J., Strozzi F., Wallace R.J. The ruminal microbiome associated with methane emissions from ruminant livestock. *Journal of Animal Science and Biotechnology*, 2017, 8(1): 7 (doi: 10.1186/s40104-017-0141-0).
45. Montenegro J., Barrantes E., DiLorenzo N. Methane emissions by beef cattle consuming hay of varying quality in the dry forest ecosystem of Costa Rica. *Livestock Science*, 2016, 193: 45-50 (doi:10.1016/j.livsci.2016.09.0).
46. Chiavegato M.B., Rowntree J.E., Carmichael D., Powers W.J. Enteric methane from lactating beef cows managed with high- and low-input grazing systems. *Journal of Animal Science*, 2015, 93(3): 1365-1375 (doi: 10.2527/jas.2014-8128).

47. Hatew B. *Low emission feed: opportunities to mitigate enteric methane production of dairy cows.* Wageningen University, 2015.
48. Molano G., Clark H. The effect of level of intake and forage quality on methane production by sheep. *Australian Journal of Experimental Agriculture*, 2008, 48(2): 219 (doi: 10.1071/ea07253).
49. Gere J.I., Bualy R.A., Perini A.L., Arias R.D., Ortega F.M., Wulff A.E., Berra G. Methane emission factors for beef cows in Argentina: effect of diet quality. *New Zealand Journal of Agricultural Research*, 2021, 64(2): 260-268 (doi: 10.1080/00288233.2019.1621).
50. Alvarado-Bolovich V., Medrano J., Haro J., Castro-Montoya J., Dickhoefer U., Gómez C. Enteric methane emissions from lactating dairy cows grazing cultivated and native pastures in the high Andes of Peru. *Livestock Science*, 2021, 243: 104385 (doi: 10.1016/j.livsci.2020.1043).
51. Benchaar C., Pomar C., Chiquette J. Evaluation of dietary strategies to reduce methane production in ruminants: a modelling approach. *Canadian Journal of Animal Science*, 2001, 81(4): 563-574 (doi: 10.4141/A00-119).
52. Gislon G., Colombini S., Borreani G., Crovetto G.M., Sandrucci A., Galassi G., Rapetti L. Milk production, methane emissions, nitrogen, and energy balance of cows fed diets based on different forage systems. *Journal of Dairy Science*, 2020, 103(9): 8048-8061 (doi: 10.3168/jds.2019-18134).
53. Beauchemin K.A., Kreuzer M., O'Mara F., McAllister T.A. Nutritional management for enteric methane abatement: a review. *Australian Journal of Experimental Agriculture*, 2008, 48(2): 21-27 (doi: 10.1071/EA07199).
54. Wang C., Zhang C., Yan T., Chang S., Zhu W., Wanapat M., Hou F. Increasing roughage quality by using alfalfa hay as a substitute for concentrate mitigates CH_4 emissions and urinary N and ammonia excretion from dry ewes. *Journal of Animal Physiology and Animal Nutrition*, 2019, 104(1): 22-31 (doi: 10.1111/jpn.13223).
55. Martin C., Morgavi D.P., Doreau M. Methane mitigation in ruminants: from microbe to the farm scale. *Animal*, 2010, 4(3): 351-365 (doi: 10.1017/S1751731109990620).
56. Albores-Moreno S., Alayón-Gamboa J.A., Ayala-Burgos A.J., Solorio-Sánchez F.J., Aguilar-Pérez C.F., Olivera-Castillo L., Ku-Vera J.C. Effects of feeding ground pods of Enterolobium cyclocarpum Jacq. Griseb on dry matter intake, rumen fermentation, and enteric methane production by Pelibuey sheep fed tropical grass. *Tropical Animal Health and Production*, 2017, 49(4): 857-866 (doi: 10.1007/s11250-017-1275-y).
57. Lovett D., Lovell S., Stack L., Callan J., Finlay M., Conolly J., O'Mara F.P. Effect of forage/concentrate ratio and dietary coconut oil level on methane output and performance of finishing beef heifers. *Livestock Production Science*, 2003, 84(2): 135-146 (doi: 10.1016/j.livprodsci.2003.09.010).
58. Beauchemin K.A., McAllister T.A., McGinn S.M. Dietary mitigation of enteric methane from cattle. Perspectives in Agriculture. *Veterinary Science, Nutrition and Natural Resources*, 2009, 4(035): 1-18 (doi: 10.1079/PAVSNNR20094035).
59. Murphy M.R., Baldwin R.L., Koong L.J. Estimation of stoichiometric parameters for rumen fermentation of roughage and concentrate diets. *Journal of Animal Science*, 1982, 55(2): 411-421 (doi: 10.2527/jas1982.552411x).
60. Orskov E.R. Starch digestion and utilization in ruminants. *Journal of Animal Science*, 1986, 63(5): 1624-1633 (doi: 10.2527/jas1986.6351624x).
61. Hindrichsen I.K., Kreuzer M. High methanogenic potential of sucrose compared with starch at high ruminal ph. *Journal of Animal Physiology and Animal Nutrition*, 2009, 93(1): 61-65 (doi: 10.1111/j.1439-0396.2007.00779.x).
62. Beauchemin K.A., McGinn S.M., Benchaar C., Holtshausen L. Crushed sunflower, flax, or canola seeds in lactating dairy cow diets: effects on methane production, rumen fermentation, and milk production. *Journal of Dairy Science*, 2009, 92(5): 2118-2127 (doi: 10.3168/jds.2008-1903).
63. Llonch P., Haskel M.J., Dewhurs R.J., Turner S.P. Current available strategies to mitigate greenhouse gas emissions in livestock systems: An animal welfare perspective. *Animal*, 2017, 11(2): 274-284 (doi: 10.1017/S1751731116001440).
64. Patra A.K. The effect of dietary fats on methane emissions, and its other effects on digestibility, rumen fermentation and lactation performance in cattle: a meta-analysis. *Livestock Science*, 2013, 155(2-3): 244-254 (doi: 10.1016/j.livsci.2013.05.023).
65. Sheyda E.V., Lebedev S.V., Miroshnikov S.A., Duskaev G.K., Ryazanov V.A., Grechkina V.V., Rakhmatullin Sh.G. *Zhivotnovodstvo i kormoproizvodstvo*, 2021, 104(2): 84-95 (doi: 10.33284/2658-3135-104-2-84) (in Russ.).
66. Wanapat M., Viennasay B., Matra M., Totakul P., Phesatcha B., Ampapon T., Wanapat S. Supplementation of fruit peel pellet containing phytonutrients to manipulate rumen pH, fermentation efficiency, nutrient digestibility and microbial protein synthesis. *Journal of the Science of Food and Agriculture*, 2021, 101(11): 4543-4550 (doi: 10.1002/jsfa.11096).
67. Marques R.D., Cooke R.F. Effects of ionophores on ruminal function of beef cattle. *Animals*, 2021, 11(10): 2871 (doi: 10.3390/ani11102871).
68. Appuhamy J.R., Strathe A.B., Jayasundara S., Wagner-Riddle C., Dijkstra J., France J., Kebreab E. Anti-methanogenic effects of monensin in dairy and beef cattle: a meta-analysis. *Journal*

of Dairy Science, 2013, 96(8): 5161-5173 (doi: 10.3168/jds.2012-5923).
69. Pedraza-Hernández J., Elghandour M.M.M.Y., Khusro A., Camacho-Diaz L.M., Vallejo L.H., Barbabosa-Pliego A., Salem A.Z.M. Mitigation of ruminal biogases production from goats using Moringa oleifera extract and live yeast culture for a cleaner agriculture environment. *Journal of Cleaner Production*, 2019, 234: 779-786 (doi: 10.1016/j.jclepro.2019.06).
70. Kim S.H., Mamuad L.L., Islam M., Lee S.S. Reductive acetogens isolated from ruminants and their effect on in vitro methane mitigation and milk performance in Holstein cows. *Journal of Animal Science and Technology*, 2020, 62(1): 1-13 (doi: 10.5187/jast.2020.62.1.1).
71. Abdelbagi M., Ridwan R., Fidriyanto R., Rohmatussolihat, Nahrowi, Jayanegara A. Effects of probiotics and encapsulated probiotics on enteric methane emission and nutrient digestibility in vitro. *IOP Conference Series: Earth and Environmental Science*. IOP Publishing, 2021, 788: 012050 (doi: 10.1088/1755-1315/788/1/012050).
72. Guo G., Shen C., Liu Q., Zhang S.L., Shao T., Wang C., Wang Y., Xu Q., Huo W. The effect of lactic acid bacteria inoculums on in vitro rumen fermentation, methane production, ruminal cellulolytic bacteria populations and cellulase activities of corn stover silage. *Journal of Integrative Agriculture*, 2020, 19(3): 838-847 (doi: 10.1016/S2095-3119(19)62707-3).
73. Jeyanathan J., Martin C., Eugène M., Ferlay A., Popova M., Morgavi D.P. Bacterial direct-fed microbials fail to reduce methane emissions in primiparous lactating dairy cows. *Journal of Animal Science and Biotechnology*, 2019, 10: 41 (doi: 10.1186/s40104-019-0342-9).
74. Latham E.A., Pinchak W.E., Trachsel J., Allen H.K., Callaway T.R., Nisbet D.J., Anderson R.C. Paenibacillus 79R4, a potential rumen probiotic to enhance nitrite detoxification and methane mitigation in nitrate-treated ruminants. *Science of The Total Environment*, 2019, 671: 324-328 (doi: 10.1016/j.scitotenv.2019).
75. Deng K.D., Xiao Y., Ma T., Tu Y., Diao Q.Y., Chen Y.H., Jiang J.J. Ruminal fermentation, nutrient metabolism, and methane emissions of sheep in response to dietary supplementation with *Bacillus licheniformis*. *Animal Feed Science and Technology*, 2018, 241: 38-44 (doi: 10.1016/j.anifeedsci.2018).
76. Partnerskiy material. Fitogeniki: nastoyashchee i budushchee. *Agroinvestor, 30 aprelya 2019*. Available: https://www.agroinvestor.ru/business-pages/31677-fitogeniki-nastoyashchee-i-budushchee/. No date (in Russ.).
77. Kaur N., Agarwal A., Sabharwal M., Jaiswal N. Natural food toxins as anti-nutritional factors in plants and their reduction strategies. In: *Food chemistry*. M. Sen (ed.), Scrivener Publishing LLC, 2021: 217-248 (doi: 10.1002/9781119792130.ch8).
78. Vasta V., Daghio M., Cappucci A., Buccioni A., Serra A., Viti C., Mele M. Invited review: plant polyphenols and rumen microbiota responsible for fatty acid biohydrogenation, fiber digestion, and methane emission: experimental evidence and methodological approaches. *Journal of Dairy Science*, 2019, 102(5): 3781-3804 (doi: 10.3168/jds.2018-14985).
79. De Nardi R., Marchesini G., Li S., Khafipour E., Plaizier K.J.C., Gianesella M., Ricci R., Andrighetto I., Segato S. Metagenomic analysis of rumen microbial population in dairy heifers fed a high grain diet supplemented with dicarboxylic acids or polyphenols. *BMC Veterinary Research*, 2016, 12(1): 2074161 (doi: 10.1186/s12917-016-0653-4).
80. Aboagye I.A., Oba M., Koenig K.M., Zhao G.Y., Beauchemin K.A. Use of gallic acid and hydrolyzable tannins to reduce methane emission and nitrogen excretion in beef cattle fed a diet containing alfalfa silage. *Journal of Animal Science*, 2019, 97(5): 2230-2244 (doi: 10.1093/jas/skz101).
81. Ngámbi J.W., Selapa M.J., Brown D., Manyelo T.G. The effect of varying levels of purified condensed tannins on performance, blood profile, meat quality and methane emission in male Bapedi sheep fed grass hay and pellet-based diet. *Tropical Animal Health and Production*, 2022, 54(5): 263 (doi: 10.1007/s11250-022-03268-7).
82. Suybeng B., Charmley E., Gardiner C.P., Malau-Aduli B.S., Malau-Aduli A.E.O. Supplementing Northern Australian beef cattle with desmanthus tropical legume reduces in-vivo methane emissions. *Animals (Basel)*, 2020, 10(11): 2097 (doi: 10.3390/ani10112097).
83. Ku-Vera J.C., Jiménez-Ocampo R., Valencia-Salazar S.S., Montoya-Flores M.D., Molina-Botero I.C., Arango J., Gómez-Bravo C.A., Aguilar-Pérez C.F., Solorio-Sánchez F.J. Role of secondary plant metabolites on enteric methane mitigation in ruminants. *Frontiers in Veterinary Science*, 2020, 7: 584 (doi: 10.3389/fvets.2020.00584).
84. Min B.R., Pinchak W.E., Hume M.E., Anderson R.C. Effects of condensed tannins supplementation on animal performance, phylogenetic microbial changes, and in vitro methane emissions in steers grazing winter wheat. *Animals (Basel)*, 2021, 11(8): 2391 (doi: 10.3390/ani11082391).
85. Jayanegara A., Goel G., Makkar H.P.S., Becker K. Reduction in methane emissions from ruminants by plant secondary metabolites: effects of polyphenols and saponins. In: *Sustainable improvement of animal production and health*. N.E. Odongo, M. Garcia, G.J. Viljoen (eds). FAO, Rome, Italy, 2010: 151-157.
86. Jayanegara A., Leiber F., Kreuzer M. Meta-analysis of the relationship between dietary tannin level and methane formation in ruminants from in vivo and in vitro experiments. *Journal of Animal Physiology and Animal Nutrition*, 2012, 96(3): 365-375 (doi: 10.1111/j.1439-0396.2011.01172.x).
87. Min B.R., Solaiman S. Comparative aspects of plant tannins on digestive physiology, nutrition

and microbial changes in sheep and goats: a review. *Journal of Animal Physiology and Animal Nutrition*, 2018, 102(5): 1181-1193 (doi: 10.1111/jpn.12938).
88. Goel C., Makkar H.P.S., Becker K. Methane mitigation from ruminants using tannins and saponins. *Tropical Animal Health and Production*, 2011, 44(4): 729-739 (doi: 10.1007/s11250-011-9966-2).
89. Naumann H.D., Tedeschi L.O., Zeller W.E., Huntley N.F. The role of condensed tannins in ruminant animal production: advances, limitations and future directions. *Revista Brasileira de Zootecnia*, 2017, 46: 929-949 (doi: 10.1590/s1806-92902017001200009).
90. Bhatta R., Uyeno Y., Tajima K., Takenaka A., Yabumoto Y., Nonaka I., Enishi O., Kurihara M. Difference in the nature of tannins on in vitro ruminal methane and volatile fatty acid production and on methanogenic archaea and protozoal populations. *Journal of Dairy Science*, 2009, 92(11): 5512-5522 (doi: 10.3168/jds.2008-1441).
91. Jayanegara A., Goel G., Makkar H.P.S., Becker K. Divergence between purified hydrolysable and condensed tannin effects on methane emission, rumen fermentation and microbial population in vitro. *Animal Feed Science and Technology*, 2015, 209: 60-68 (doi: 10.1016/j.anifeedsci.2015.08.002).
92. Mannelli F., Daghio M., Alves S.P., Bessa R.J., Minieri S., Giovannetti L., Conte G., Mele M., Messini A., Rapaccini S., Viti C., Buccioni A. Effects of chestnut tannin extract, vescalagin and gallic acid on the dimethyl acetals profile and microbial community composition in rumen liquor: an in vitro study. *Microorganisms*, 2019, 7(7): 202 (doi: 10.3390/microorganisms707020).
93. Mueller-Harvey I. Unravelling the conundrum of tannins in animal nutrition and health. *Journal of the Science of Food and Agriculture*, 2006, 86(13): 2010-2037 (doi: 10.1002/jsfa.2577).
94. Costa M., Alves S.P., Cabo B., Guerreiro O., Stilwell G., Dentinho M.T., Bessa R.J. Modulation of in vitro rumen biohydrogenation by *Cistus ladanifer* tannins compared with other tannin sources. *Journal of the Science of Food and Agriculture*, 2017, 97(2): 629-635 (doi: 10.1002/jsfa.7777).
95. Waghorn G.C., Woodward S.L. Ruminant contributions to methane and global warming — a New Zealand perspective. In: *Climate change and managed ecosystems*. J.S. Bhatti, R. Lal, M.J. Apps, M.A. Price (eds.). Taylor and Francis, Boca Raton, 2006: 233-261.
96. Woodward S.L., Waghorn G.C., Ulyatt M.J., Lassey K.R. Early indication that feeding lotus will reduce methane emission from ruminants. *Proceedings of New Zealand Society of Animal Production*, 2001, 61: 23-26.
97. Puchala R., Min B.R., Goetsch A.L., Sahlu T. The effect of a condensed tannin-containing forage on methane emission by goats. *Journal of Animal Science*, 2005, 83(1): 182-186 (doi: 10.2527/2005.831182x).
98. Carulla J.E., Kreuzer M., Machmüller A., Hess H.D. Supplementation of Acacia mearnsii tannins decreases methanogenesis and urinary nitrogen in forage-fed sheep. *Australian Journal of Agricultural Research*, 2005, 56(9): 961-970 (doi: 10.1071/AR05022).
99. Animut G., Puchala R., Goetsch A.L., Patra A.K., Sahlu T., Varel V.H., Wells J. Methane emission by goats consuming diets with different levels of condensed tannins from Lespedeza. *Animal Feed Science and Technology*, 2008, 144: 212-227 (doi: 10.1016/j.anifeedsci.2007.10.014).
100. Tiemann T.T., Lascano C.E., Wettstein H.R., Mayer A.C., Kreuzer M., Hess H.D. Effect of the tropical tannin-rich shrub legumes Calliandra calothyrsus and Flemingia macrophylla on methane emission and nitrogen and energy balance in growing lambs. *Animal*, 2008, 2(5): 790-799 (doi: 10.1017/S1751731108001791).
101. De Oliveira S.G., Berchielli T.T., Pedreira M.D.S., Primavesi O., Frighetto R., Lima M.A. Effect of tannin levels in sorghum silage and concentrate supplementation on apparent digestibility and methane emission in beef cattle. *Animal Feed Science and Technology*, 2007, 135(3-4): 236-248 (doi: 10.1016/j.anifeedsci.2006.07.012).
102. Grainger C., Clarke T., Auldist M.J., Beauchemin K.A., McGinn S.M., Waghorn G.C. Potential use of Acacia mearnsii condensed tannins to reduce methane emissions and nitrogen excretion from grazing dairy cows. *Canadian Journal of Animal Science*, 2009, 89(2): 241-251 (doi: 10.4141/CJAS08110).
103. Patra A.K., Kamra D.N., Bhar R., Kumar R., Aggarwal N. Effect of *Terminalia chebula* and *Allium sativum* on in vivo methane emission by sheep. *Journal of Animal Physiology and Animal Nutrition*, 2011, 95(2): 187-191 (doi: 10.1111/j.1439-0396.2010.01039.x).
104. Santoso B., Mwenya B., Sar C., Gamo Y., Kobayashi T., Morikawa R., Kimura K., Mizukoshi H., Takahashi J. Effects of supplementing galacto-oligosaccharides, Yucca schidigera or nisin on rumen methanogenesis, nitrogen and energy metabolism in sheep. *Livestock Production Science*, 2004, 91(3): 209-217 (doi: 10.1016/j.livprodsci.2004.08.004).
105. Kozłowska M., Cieślak A., Jyżwik A., El-Sherbiny M., Stochmal A., Oleszek W., Kowalczyk M., Filipiak W., Szumacher-Strabel M. The effect of total and individual alfalfa saponins on rumen methane production. *Journal of the Science of Food and Agriculture*, 2019, 100(5): 1922-1930 (doi: 10.1002/jsfa.10204).
106. Kang J., Zeng B., Tang S., Wang M., Han X., Zhou C., Yan Q., He Z., Liu J., Tan Z. Effects of *Momordica charantia* saponins on in vitro ruminal fermentation and microbial population. *Asian-Australasian Journal of Animal Sciences*, 2016, 29(4): 500-508 (doi: 10.5713/ajas.15.0402).

107. Canul-Solis J.R., Piceiro-Vazquez A.T., Chay-Canul A.J., Castillo-Sánchez L.E., Alayón-Gamboa J.A., Ayala-Burgos A.J., Aguilar-Pérez A.J., Pedraza-Beltran, Castelan-Ortega O.A., Ku-Vera J.C. Effect of the source and concentration of saponins on in vitro and ruminal methane production. *Archivos de Zootecnia*, 2019, 68(263): 362-369 (doi: 10.21071/az.v68i263.4194).
108. Wang B., Ma M.P., Diao Q.Y., Tu Y. Saponin-induced shifts in the rumen microbiome and metabolome of young cattle. *Frontiers in Microbiology*, 2019, 10: 356 (doi: 10.3389/fmicb.2019.00356).
109. Wu H., Meng Q., Zhou Z., Yu Z. Ferric citrate, nitrate, saponin and their combinations affect in vitro ruminal fermentation, production of sulphide and methane and abundance of select microbial populations. *Journal of Applied Microbiology*, 2019, 127(1): 150-158 (doi: 10.1111/jam.14286).
110. Guyader J., Eugène M., Doreau M., Morgavi D.P., Gérard C., Martin C. Tea saponin reduced methanogenesis in vitro but increased methane yield in lactating dairy cows. *Journal of Dairy Science*, 2017, 100(3): 1845-1855 (doi: 10.3168/jds.2016-11644).
111. Wallace R., McEwan N.R., McIntosh F.M., Teferedegne B., Newbold C.J. Natural products as manipulators of rumen fermentation. *Asian-Australasian Journal of Animal Sciences*, 2002, 15(10): 1458-1468 (doi: 10.5713/ajas.2002.145).
112. Chen R.J., Chung T., Li F., Lin N., Tzen J.T. Effect of sugar positions in ginsenosides and their inhibitory potency on Na^+/K^+-ATPase activity. *Acta Pharmacologica Sinica*, 2009, 30(1): 61-69 (doi: 10.1038/aps.2008.6).
113. Ramos-Morales E., Arco-Pérez A., Martín-García A.I., Yácez-Ruiz D.R., Frutos P., Hervás G. Use of stomach tubing as an alternative to rumen cannulation to study ruminal fermentation and microbiota in sheep and goats. *Animal Feed Science and Technology*, 2014, 198: 57-66 (doi: 10.1016/j.anifeedsci.2014.09.016).
114. Guo Y.Q., Liu J.X., Lu Y., Zhu W.Y., Denman S.E., McSweeney C.S. Effect of tea saponin on methanogenesis, microbial community structure and expression of mcrA gene, in cultures of rumen micro-organisms. *Letters in Applied Microbiology*, 2008, 47(5): 421-426 (doi: 10.1111/j.1472-765X.2008.02459.x).
115. Pen B., Sar C., Mwenya B., Kuwaki K., Morikawa R., Takahashi J. Effects of *Yucca schidigera* and *Quillaja saponaria* extracts on in vitro ruminal fermentation and methane emission. *Animal Feed Science and Technology*, 2006, 129: 175-186 (doi: 10.1016/j.anifeedsci.2006.11.018).
116. Holtshausen L., Chaves A.V., Beauchemin K.A., McGinn S.M., McAllister T., Odongo N.E., Cheeke P.R., Benchaar C. Feeding saponin-containing *Yucca schidigera* and *Quillaja saponaria* to decrease enteric methane production in dairy cows. *Journal of Dairy Science*, 2009, 92(6): 2809-2821 (doi: 10.3168/jds.2008-1843).
117. Mao H., Wang J., Zhou Y., Liu J. Effects of addition of tea saponins and soybean oil on methane production, fermentation and microbial population in the rumen of growing lambs. *Animal Feed Science and Technology*, 2010, 129(1-3): 56-62 (doi: 10.1016/j.livsci.2009.12.011).
118. Zhou Y.Y., Mao H.L., Jiang F., Wang J.K., Liu J.X., McSweeney C.S. Inhibition of rumen methanogenesis by tea saponins with reference to fermentation pattern and microbial communities in Hu sheep. *Animal Feed Science and Technology*, 2011, 166: 93-100 (doi: 10.1016/j.anifeedsci.2011.04.007).
119. Sliwinski B.J., Kreuzer M., Wettstein H.R., Machmuller A. Rumen fermentation and nitrogen balance of lambs fed diets containing plantextracts rich in tannins and saponins and associated emissions of nitrogen and methane. *Archieves of Animal Nutrition*, 2002, 56(6): 379-392 (doi: 10.1080/00039420215633).
120. Molina-Botero I.C., Arroyave-Jaramillo J., Valencia-Salazar S., Barahona-Rosales R., Aguilar-Pйrez C.F., Ayala Burgos A., Jacobo A., Ku-Vera J.C. Effects of tannins and saponins contained in foliage of *Gliricidia sepium* and pods of *Enterolobium cyclocarpum* on fermentation, methane emissions and rumen microbial population in crossbred heifers. *Animal Feed Science and Technology*, 2019, 251: 1-11 (doi: 10.1016/j.anifeedsci.2019.01.011).
121. Montoya-Flores M.D., Molina-Botero I.C., Arango J., Romano-Mucoz J.L., Solorio-Sánchez F.J., Aguilar-Pérez C.F., Ku-Vera J.C. Effect of dried leaves of Leucaena leucocephala on rumen fermentation, rumen microbial population, and enteric methane production in crossbred heifers. *Animals*, 2020, 10(2): 300 (doi: 10.3390/ani10020300).
122. Molina I.C., Angarita E.A., Mayorga O.L., Chará J., Barahona-Rosales R. Effect of *Leucaena leucocephala* on methane production of Lucerna heifers fed a diet based on *Cynodon plectostachyus. Livestock Science*, 2016, 185: 24-29 (doi: 10.1016/j.livsci.2016.01.009).
123. Valencia Salazar S.S., Piceiro Vázquez A.T., Molina Botero I.C., Lazos Balbuena F.J., Uuh Narváez J.J., Segura Campos M.R., Avilés L.R., Solorio Sánchez F.J., Ku Vera J.C. Potential of *Samanea saman* pod meal for enteric methane mitigation in crossbred heifers fed low-quality tropical grass. *Agricultural and Forest Meteorology*, 2018, 258: 108-116 (doi: 10.1016/j.agrformet.2017.12.262).
124. Yejun L., Su Kyoung L., Shin Ja L., Jong-Su E., Sung Sill L. Effects of Lonicera japonica extract supplementation on in vitro ruminal fermentation, methane emission, and microbial population. *Animal Science Journal*, 2019, 90(9): 1170-1176 (doi: 10.1111/asj.13259).
125. Moiseeva E.A., Kravchenko I.V., Shepeleva L.F., Bordey R.Kh. Accumulation of photosynthetic pigments and secondary metabolites in leaves of galega (*Galega orientalis* Lam.) cv. Gale depending on stand age and agrotechnologies during introduction in the Middle taiga of Western Siberia. *Sel'skokhozyaistvennaya biologiya* [*Agricultural Biology*], 2022, 57(1): 44-65

(doi: 10.15389/agrobiology.2022.1.44eng).
126. Olagaray K.E., Bradford B.J. Plant flavonoids to improve productivity of ruminants — a review. *Animal Feed Science and Technology*, 2019, 251: 21-36 (doi: 10.1016/j.anifeedsci.2019.02.004).
127. Oskoueian E., Abdullah N., Oskoueian A. Effects of flavonoids on rumen fermentation activity, methane production, and microbial population. *BioMed Research International*, 2013, 2013: ID 349129 (doi: 10.1155/2013/349129).
128. Seradj A.R., Abecia L., Crespo J., Villalba D., Fondevila M., Balcells J. The effect of Bioflavex® and its pure flavonoid components on in vitro fermentation parameters and methane production in rumen fluid from steers given high concentrate diets. *Animal Feed Science and Technology*, 2014, 197: 85-91 (doi: 10.1016/j.anifeedsci.2014.08.013).
129. Zhan J., Liu M., Su X., Zhan K., Zhang C., Zhao G. Effects of alfalfa flavonoids on the production performance, immune system, and ruminal fermentation of dairy cows. *Asian-Australasian Journal of Animal Sciences*, 2017, 30(10): 1416-1424 (doi: 10.5713/ajas.16.0579).
130. Sinz S, Kunz C., Liesegang A., Braun U., Marquardt S., Soliva C.R., Kreuzer M. In vitro bioactivity of various pure flavonoids in ruminal fermentation, with special reference to methane formation. *Czech Journal of Animal Science*, 2018, 63: 293-304 (doi: 10.17221/118/2017-CJAS)
131. Ahmed E., Fukuma N., Hanada M., Nishida T. The efficacy of plant-based bioactives supplementation to different proportion of concentrate diets on methane production and rumen fermentation characteristics in vitro. *Animals*, 2021, 11(4): 1029 (doi: 10.3390/ani11041029).
132. Nurzhanov B.S., Ryazanov V.A., Sheyda E.V., Duskaev G.K., Rakhmatullin Sh.G. *Sposob snizheniya kontsentratsii metana v rubtse zhvachnykh zhivotnykh. C1 2780832 (RF), A23K 10/30, A23K 50/10, 04.10.2022. Federal'noe gosudarstvennoe byudzhetnoe nauchnoe uchrezhdenie «Federal'nyy nauchnyy tsentr biologicheskikh sistem i agrotekhnologiy Rossiyskoy akademii nauk» (RF). № 2022106708. Zayavl. 15.03.2022. Opubl. 04.10.2022* [Method for reducing the concentration of methane in the rumen of ruminants. C1 2780832 (RF), A23K 10/30, A23K 50/10, 10.04.2022. Federal State Budgetary Scientific Institution «Federal Scientific Center for Biological Systems and Agrotechnologies of the Russian Academy of Sciences» (RF). № 2022106708. Appl. 03.15.2022. Publ. 04.10.2022] (in Russ.).
133. Bal'sel's Teres Zh., Krespo Montero F.Sh. *Sposob snizheniya metanogeneza u zhvachnykh zhivotnykh. C1 2576195 (RF). № 2014146434/13. Zayavl. 18.04.2013. Opubl. 27.02.2016* [Method for reducing methanogenesis in ruminants. C1 2576195 (RF). № 2014146434/13. Appl. 04.18.2013. Publ. 27.02.2016] (in Russ.).
134. Hu Q., Zhou M., We, S. Progress on the antimicrobial activity research of clove oil and eugenol in the food antisepsis field. *Journal of Food Science*, 2018, 83(6): 1476-1483 (doi: 10.1111/1750-3841.14180).
135. Benchaar C., Greathead H. Essential oils and opportunities to mitigate enteric methane emissions from ruminants. *Animal Feed Science and Technology*, 2011, 166-167: 338-355 (doi: 10.1016/j.anifeedsci.2011.04.024).
136. Zhou X., Zhang N., Zhang J., Gu Q., Dong C., Lin B., Zou C. Microbiome and fermentation parameters in the rumen of dairy buffalo in response to ingestion associated with a diet supplemented with cysteamine and hemp seed oil. *Journal of Animal Physiology and Animal Nutrition*, 2022, 106(3): 471-484 (doi: 10.1111/jpn.13616).
137. Joch M., Mrázek J., Skřivanová E., Čermák L., Marounek M. Effects of pure plant secondary metabolites on methane production, rumen fermentation and rumen bacteria populations in vitro. *Journal of Animal Physiology and Animal Nutrition*, 2018, 102(4): 869-881 (doi: 10.1111/jpn.12910).
138. Zeng Z., Sheng P., Zhang H., He L., Huang J., Wang D., Gui G.The effect of *Macleaya cordata* extract on in vitro ruminal fermentation and methanogenesis. *Food Science & Nutrition*, 2021, 9(8): 4561-4567 (doi: 10.1002/fsn3.2436).
139. Petrič D., Mravčáková D., Kucková K., Čobanová K., Kišidayová S., Cieslak A., Ślusarczyk S., Váradyová Z. Effect of dry medicinal plants (wormwood, chamomile, fumitory and mallow) on in vitro ruminal antioxidant capacity and fermentation patterns of sheep. *Journal of Animal Physiology and Animal Nutrition*, 2020, 104(5): 1219-1232 (doi: 10.1111/jpn.13349).
140. Yu J., Cai L., Zhang J., Yang A., Wang Y., Zhang L., Guan L.L., Qi D. Effects of thymol supplementation on goat rumen fermentation and rumen microbiota in vitro. *Microorganisms*, 2020, 8(8): 1160 (doi: 10.3390/microorganisms8081160).
141. Rossi C.A.S., Grossi S., Dell'Anno M., Compiani R., Rossi L. Effect of a blend of essential oils, bioflavonoids and tannins on in vitro methane production and in vivo production efficiency in dairy cows. *Animals*, 2022, 12(6): 728 (doi: 10.3390/ani12060728).
142. Castro-Montoya J., Peiren N., Cone J.W., Zweifel B., Fievez V., De Campeneere S. In vivo and in vitro effects of a blend of essential oils on rumen methane mitigation. *Livestock Science*, 2015, 180: 134-142 (doi: 10.1016/j.livsci.2015.08.010).
143. Hart K., Jones, H., Waddams K., Worgan H., Zweifel B., Newbold C. An essential oil blend decreases methane emissions and increases milk yield in dairy cows. *Open Journal of Animal Sciences*, 2019, 9(03): 259-267 (doi: 10.4236/ojas.2019.93022).
144. Klop G., Dijkstra J., Dieho K., Hendriks W.H., Bannink A. Enteric methane production in lactating dairy cows with continuous feeding of essential oils or rotational feeding of essential oils

and lauric acid. *Journal of Dairy Science*, 2017, 100(5): 3563-3575 (doi: 10.3168/jds.2016-12033).

145. Rakhmatullin Sh.G., Nurzhanov B.S., Duskaev G.K., Kvan O.V., Sheyda E.V. *Zhivotnovodstvo i kormoproizvodstvo*, 2021, 3(104): 94-103 (doi: 10.33284/2658-3135-104-3-94) (in Russ.).
146. Alves T.P., Dall-Orsoletta A.C., Ribeiro-Filho H.M.N. The effects of supplementing acacia mearnsii tannin extract on dairy cow dry matter intake, milk production, and methane emission in a tropical pasture. *Tropical Animal Health and Production*, 2017, 49(8): 1663-1668 (doi: 10.1007/s11250-017-1374-9).
147. Chen D., Chen X., Tu Y., Wang B., Lou C., Ma T., Diao Q. Effects of mulberry leaf flavonoid and resveratrol on methane emission and nutrient digestion in sheep. *Animal Nutrition*, 2015, 1(4): 362-367 (doi: 10.1016/j.aninu.2015.12.008).
148. Dey A., Attri K., Dahiya S.S., Paul S.S. Influence of dietary phytogenic feed additives on lactation performance, methane emissions and health status of Murrah buffaloes (*Bubalus bubalis*). *Journal of the Science of Food and Agriculture*, 2021, 101(10): 4390-4397 (doi: 10.1002/jsfa.11080).
149. Van Gastelen S., Dijkstra J., Bannink A. Are dietary strategies to mitigate enteric methane emission equally effective across dairy cattle, beef cattle, and sheep? *Journal of Dairy Science*, 2019, 102(7): 6109-6130 (doi: 10.3168/jds.2018-15785).
150. Boadi D.A., Wittenberg K.M., Scott S.L., Burton D., Buckley K., Small J.A., Ominski K.H. Effect of low and high forage diet on enteric and manure pack greenhouse gas emissions from a feedlot. *Canadian Journal of Animal Science*, 2004, 84(3): 445-453 (doi: 10.4141/a03-079).