DOI: 10.12731/2658-6649-2023-15-2-163-177
UDC 636.084

Original article | Animal Husbandry

EFFECT OF DIETARY SUPPLEMENTATION WITH FOSSIL SHELL FLOUR ON ENTERIC METHANE OUTPUT AND POSITION-DEPENDENT VARIATIONS IN DOHNE-MERINO WETHERS

O.O. Ikusika, C.T. Mpendulo

This study aimed to investigate the influence of fossil shell flour (FSF) supplementation levels on Dohne-Merino wethers' position on enteric methane output. Twenty-four Dohne-Merino wethers (20.0±1.50 kg B.W.) were randomly assigned for 84 days to either of four dietary treatments: basal or basal diet supplemented with 2%,4% or 6%FSF on a dry matter basis. The enteric methane output was measured using a portable Laser Methane Detector (LMD) machine during feeding, standing, and resting activities. The highest volume of enteric methane was obtained from wethers supplemented with 4%, followed by 6%, 0%, and 2% FSF. Higher enteric methane emission was observed for resting wethers than those feeding and standing (P < 0.05). Including fossil shell flour in Dohne-merino wethers' diets at 4% and 6% increases enteric methane output (P<0.05). Dohne-merino sheep emit more enteric methane when resting than when feeding or standing idle.

Keywords: *fossil shell flour; enteric methane; animal position; Dohne-merino ram*

For citation. *Ikusika O.O., Mpendulo C.T. Effect of Dietary Supplementation with Fossil Shell Flour on Enteric Methane Output and Position-Dependent Variations in Dohne-Merino Wethers. Siberian Journal of Life Sciences and Agriculture, 2023, vol. 15, no. 2, pp. 163-177. DOI: 10.12731/2658-6649-2023-15-2-163-177*

Introduction

In the last two decades, there has been great concern about global warming due to a rise in the volume of many atmospheric gasses, leading to increased atmospheric temperature [1]. These gases include methane, carbon dioxide, and nitrous oxide often called greenhouse gases. It has been projected that greenhouse effects in the next century will bring about the distribution of new

deserts in the world and change the range of pest that affects plants, which may threaten the existence of animals and human health [2]. The emission of greenhouse gases from the animal production sector and their effects on climate variability is a major concern worldwide [3]. About 98% of CH_4 output accrues to the agricultural sector are from the livestock sector [4]. Ogino et al. [5] observed that enteric methane constituted about 50% to 60% of GHG emitted in ruminant production at the farm scale. The CH_4 emissions from the animal production sector are estimated to be 2.2 billion tons of carbon dioxide equivalent, accounting for 35% of the global anthropogenic methane emissions [6]. However, in sub-Saharan Africa, it is projected to rise [7]. Enteric methane is a natural by-product of the fermentation processes in the large intestine of ruminant animals and is released into the atmospheric environment through breathing [8]. According to [9] nitrous oxide and methane have higher global warming potentials than carbon dioxide. While nitrous oxide has a global warming potential range of 296 to 310 times higher than CO_2, it has been reported that CH_4 is about 25 times more effective in trapping heat in the atmosphere [10, 11]. Depending on the feed intake and rumen activity, the rate of enteric methane production varies with individual animals [12]. Besides its negative effect on global warming, methane accounts for a significant amount of animals' energy loss during grazing or browsing. Sallaku et al.[13 and [1] both reported that energy loss due to methane emission could be as high as 12% of gross energy (G.E.) intake, thereby reducing livestock productivity and the economic efficiency of ruminant production. Therefore, mitigating the emission of CH_4 in ruminants without altering animal production performance is a desirable approach to reducing global greenhouse gases emissions and improving feed conversion efficiency.

Sheep have the largest population among the small stock globally [14] hence their contribution to GHG is high [1]. The Dohne-Merino has been one of the fastest breeds of sheep, spreading across many continents [15, 16] accounting for over 36 % of the GHG emissions from livestock species. The by-product of microbial fermentation of feeds in the rumen of sheep is methane. Sallaku et al. [13] reported that the amount of enteric CH_4 emitted in sheep is influenced by breed and purposes of the animal, quality and type of forage, diet composition, feed intake and digestibility. Also, activities of the animals, such as resting, standing, or feeding, as well as feed additives, have been reported to affect the volume of methane output in other livestock [17]. The animals' position and activities have been reported to affect the amount of methane generated. Roessler et al. [18] reported that when a goat is lying,

it generates more enteric methane than standing. Likewise, [19] observed that when goats are ruminating or resting after a long journey, higher methane is generated than when the animal is standing after feeding.

In recent times, reducing emissions of greenhouse gases from livestock production is attracting the use of supplements or feed additives in manipulating the rumen community. Thota et al. [20] reported that the mean enteric CH_4 emissions (l/day) were significantly lower in sheep fed with probiotics supplemented diet than in sheep fed without probiotics supplementation and reduced by 21.9 per cent as compared to the non-supplemented diets. Similarly, [21] reported that allicin supplementation effectively lowered daily CH_4 emissions in sheep by reducing the population of ruminal protozoans and methanogens. The use of inorganic feed additives to mitigate greenhouse gases is either toxic to the animal or exhibits only transient effects on methanogens [2]. Using natural products as additives to mitigate the emission of greenhouse gas in livestock benefits the livestock, the environment and consumers of the animal.

The most recently sought for use as a feed additive is fossil shell flour (FSF), among the common natural products used as feed additives. Ikusika et al. [22] observed that FSF as a feed additive benefits sheep production in terms of growth performance, feed preference, and wool quality. A little information is available on the impact of FSF in sheep diets on methane gas production. Fossil shell flour is a naturally occurring, silicon-rich sedimentary rock made up of fossilized remains of millions of diatoms, a type of hard-shelled plant algae originally deposited millions of years ago in the earth from dried-up seas and lakes [22, 23]. They are readily available, cost-effective, healthy, and eco-friendly for animals and humans. Because of the antimicrobial activities of FSF, as reported by [24] it would reduce methane production by militating methanogenesis microbes. Against this background, this study investigates the impact of varying FSF supplementation on enteric methane production at a different animal position in Dohne-Merino wethers. It was hypothesized that including fossil shell flour into the diet of Dohne-merino wethers would decrease the enteric methane production.

Methodology

Ethical approval

Ethical clearance was obtained from the University of Fort Hare Animal Ethics and Use committee before the commencement of the feeding trials (approval number: MPE041IKU01).

Study site description

The experiment was conducted at the honeydale farm, University of Fort Hare, Research farm, Alice, South Africa. It lies at a longitude of 26º 50' E, and latitude of 32°46' S. The annual rainfall is between 480-490 mm, and a temperature range between 24.6 º C and 11.1 º C (average is 17.8 º C) at an altitude of 535 meters above sea level.

Animal, experimental design and management

Twenty-four five-month-old Dohne-Merino wethers weighing 20 ± 1.5kg on average were selected and bought from a commercial farm in Mitford village, Tarkastad, Eastern Cape province, South Africa. The wethers were randomly allotted into four treatments (n = 6). They were individually housed (1.5 m × 1.5 m) in a well-ventilated roofed animal building with a concrete floor and exposed to the same environmental condition. The experiment lasted 105 days, excluding 14 days of the adaptation period. The wethers had access to sufficiently clean and fresh water ad libitum daily. Each wether was ear-tagged and labelled for identification on a diet basis.

Experimental Diets

The diets for the wethers consisted of concentrate and hay at a 40:60 ratio. The basal diet was made up of maize (8%), sunflower oil cake (10 %), molasses (5%), wheat offal (15%), limestone (1.5%), salt 0.3%, sheep mineral-vitamin premix (0.2%), 30 % teff and 30 % Lucerne. The ingredients were purchased from Monti Feeds (pty) Ltd, East London, South Africa. All ingredients were thoroughly milled and mixed evenly. The feed was formulated to meet the used sheep's nutritional (energy and protein) requirements [25]. The four dietary groups were: basal diet (0%); basal diet +2% FSF; basal diet +4 % FSF and basal diet + 6% FSF. The wethers were fed at 8:00h and 15:00h at 4 % of the body weight (on a dry matter basis). The food-grade Fossil shell flour was purchased from Eco-Earth (Pty) Ltd, Port Elizabeth, South Africa, which produces this product under a license by the Department of Agriculture, Forestry and Fisheries of South Africa.

Proximate analysis of the experimental diets

Dry matter, crude protein, crude fibre, ether extract and total ash of samples were analyzed in triplicates using the standard procedure described in [26]. The proximate composition of the experimental diet is presented in Table1.

Table 1.

Proximate analysis of the experimental diets

Items	**Percentage (%)**
Maize	8
Sunflower oil cake	10
Molasses	5
Wheat bran	15
Limestone	1.6
Sheep premix	0.2
Salt	0.3
Grinded leucine hay (alfalfa)	30
Grinded teff hay	30
Chemical composition	
Dry matter (% as fed)	95.5
Organic matter	85.22
Energy ME	24.67
Crude Protein	14.56
Ash	10.33
Ether extract	1.7
Crude Fibre	22.60

Mineral analyses

The mineral composition of the dietary FSF used is shown in Table 2. In determining the FSF's mineral content, 5.0 g of the sample was weighed in triplicate and burnt at 550 º C in a muffle furnace for 5.5 hours. The residues were cooled in a desiccator before dissolving in 100 ml of deionized water. Suitable salts of the elements were used to make their standards. The standard mineral solutions were injected into the atomic absorption spectrophotometer (Jenway, FPSP 210 model 6305, United Kingdom), and concentration was obtained. These standards determined Mg, Zn, Fe, Cd, Ca, Al, Mn, and B in an unknown feed sample. Na and K's concentrations were determined using a flame photometer (Jenway Models PFP7 and PFP7/C, Cole-Parmer, United Kingdom).

Table 2.

Mineral composition of Fossil shell flour (FSF)

Items	**Quantity**
DM %	93
Ca	0.40
% CaO (calculated from %Ca)	0.55

End of table

Mg	0.21
%MgO (calculated from %Mg)	0.34
K%	0.16
Cu (mg/kg)	30
Na (mg/kg)	923
Zn(mg/kg)	118
Fe(mg/kg)	7944
Mn(mg/kg)	69
P (as P2O5)	0.037
Sulfate Sulfur (S)%	0.062
Aluminum (Al) %	0.065
Vanadium (V) %	0.00438
Boron (B) %	0.0023

Measurement of methane production

The measurement of methane was done using a laser methane detector LMD (Crowcon Detection Instruments Ltd., Oxford shire, United Kingdom). Measurements were carried out weekly from the trial's inception during three different wethers' activities, including resting, feeding, and standing. Also, during the last 7 days of the experiment, methane output was measured daily for the same three activities of the wethers. Methane gas column density was measured by directing the hand-held LMD machine targeting (visible HeNe) at wethers' nostrils for 25s per wethers at a distance of 2 m. The 2-m space was considered safe to prevent the disturbance of the animal's activity, as described by [27] and [19]. The effect of methane in the atmosphere from the measured results was discounted using the offset function of the LMD. All measurements were taken at approximately the same time of day (1000h-1100h). Three measurements were taken from individual wethers during each activity. Methane eructed was determined per activity using standard respiratory coefficients per activity, then translated to an equivalent emission per day. Methane production was also evaluated in relation to dry matter intake (DMI). A laser methane detector (LMD) measures methane emission in ppm-m, which is not equivalent to g/kg/d. Therefore, to know how much methane is being produced per wether, methane was determined on a DMI basis.

Methane eructed during activity MTV = MMD × TVr / 106ml, [27]

Where: MTV is the enteric methane in breath in ml during ruminating; MMD is the enteric methane detected by LMD converted from ppm-m to ml.

TVr is the tidal volume during different activities

Tidal volume (feeding) = 3100 ml, tidal volume (standing) = 3800 ml for dairy animals. These were then converted using livestock units to represent sheep, where 0.5 LU cow is equivalent to 0.1 livestock unit (L.U.) sheep [28] for sub-Saharan Africa.

The TVr for sheep were, therefore: TVr feeding = 620 ml, TVr standing = 760 ml, TVr ruminating = 760 ml

Methane eructed per activity per day MTTA = MTV × RTA [27]

MTTA is the amount of enteric methane produced during an activity (rumination, feeding, just standing).

Methane eructed per day M.D. = MTTA x (T.D. × RTA) ml/day [27] Where: M.D. is daily enteric methane

T.D. is daytime in seconds

RTA is the total time spent on an activity

RTA standing = 1440, RTA feeding = 2880, and RTA ruminating = 7200 By substitution and use of specific density conversion factor, daily enteric methane in grams (MDG) is:

MDG (g/day) = MD x 0.00066715 (CH4 density in g/ml) [27]

Methane (l/day) = 0.0305 DMI(g/day) – 4.441 [29] M (kg/head/day) = DMI x 0.0188 + 0.00158 [30].

Statistical analyses

The PROC MIXED procedure of Statistical Analysis Systems Institute [31] for repeated measures was used to test for the significance of inclusion level of FSF and position of wethers on methane volume. Turkey's studentized range test was used to test the significant differences between means. The statistical model used was:

Yijk= μ+ Tj + Bj +Dk + (TxBxD)jjk + eijk Where:

Yijk is methane volume μ is the overall mean

Tjs the effect of diet (i = 1,2,3,4) Bj the effect of position (i=1,2,3)

Dk is the effect of week (k, = 1,2,3,4,5,6)

(TxBxD)ijk is the interaction effect between treatment, week, and position

eijk is the error term

Results

Table 3 shows enteric methane emission from wethers fed diets with varying FSF levels during different activities. Enteric methane output was lowest in wethers fed on a diet with 0% FSF and highest in those with 6% FSF during standing, feeding, and resting ($P < 0.05$). As the FSF inclusion level increased, enteric methane output increased except for feeding and resting wethers fed on diets with 2 % and 4 % FSF ($P<0.05$). Across the diets, there were no significant differences for all the activities ($P > 0.05$). The wethers released the highest methane volume when resting and the least when feeding ($P < 0.05$).

Table 4 shows the consecutively measured methane emission, average daily feed intake, and dry matter intake in the last 7 days of the feeding trial. Both the ADFI and the DMI had a linear relationship with the amount of methane produced. Wethers fed 4 % FSF had the highest ADFI and DMI values and produced the highest ($P<0.05$) methane value. The amount of methane produced by wethers fed on a diet with 0% FSF was significantly lesser than the amount generated by wethers on 4% and 6% FSF, but not from wethers on 2% FSF ($P < 0.05$). Wethers on a 4%FSF diet emitted more methane than the wethers on 0% FSF and other FSF supplemented treatments ($P < 0.05$). In all the diets, wethers generated more methane (g/day) when they were resting than feeding or just standing ($P < 0.05$). In all the activities, wethers fed on a diet with 4 % produced more methane than those on 0 % FSF and other FSF supplemented diets ($P < 0.05$).

Figure 1 shows the amount of methane generated by the wethers on varying amounts of FSF over a period of 12 weeks. From weeks 1-3, the volume of methane produced by wethers fed on 0 %, 2%,4 %, and 6% FSF of the diets was the same ($P<0.05$). From weeks 4-6, the volume of methane emitted by wethers on 0% FSF began to be lesser than those on 2%, 4%, and 6% FSF of the diets. From weeks 7 -12, the volume of methane emitted was significantly higher in the FSF supplemented diets compared to the 0% FSF diet ($P<0.05$).

Figures 2 shows the effect of different activities of wethers on methane output at 0%, 2%,4% and 6% FSF diets. Methane output was highest ($P<0.05$) during resting and lowest during feeding at the varying inclusion levels of FSF. The methane emitted during resting was significantly different ($P<0.05$) from the volume emitted during feeding activities. In all the activities, methane output increased as the FSF inclusion levels increased up to 4% FSF levels and declined after that.

Table 3.

Enteric methane emission from Dohne-merino wethers fed on varying FSF levels during different activities in the last seven days of the trial

	Levels of FSF inclusion				
Activity	**0 %**	**2 %**	**4 %**	**6 %**	**SEM**
Standing	17.74^{a}	18.54^{a}	21.86^{a}	25.71^{a}	4.66
Feeding	15.83^{a}	22.52^{a}	13.24^{a}	19.31^{a}	4.81
Resting	38.63^{a}	42.46^{a}	42^{a}	47.50^{a}	11.25

^{abc} mean values with different superscripts across the row are significantly different ($P < 0.05$).

Table 4.

Enteric methane emission from Dohne-merino wethers fed on varying inclusion levels of FSF (grams /day)

Activity	**Levels of FSF inclusion**				**SEM**
	0 %	**2 %**	**4 %**	**6 %**	
ADFI (g)	84.69^{c}	92.86^{bc}	121.42^{a}	105.35^{b}	9.53
DMI (g)	576.29^{c}	546.11^{d}	665.76^{a}	619.84^{b}	11.84
Methane (l/day)	17.27	16.66	20.30	18.90	0.32
Methane kgDMI/year/	3,887.25	3,748.55	4569.8	4252.25	81.22
Methane (g/kg DMI)	10.65	10.27	12.52	11.65	0.218
Methane (g/day					
Standing	0.0046	0.0125	0.0198	0.023	0.000147
Feeding	0.0114	0.0017	0.0124	0.0141	0.0024
Resting	0.036	0.0511	0.301	0.0438	0.00041

^{abc} mean values with different superscripts across the row are significantly different ($P < 0.05$).

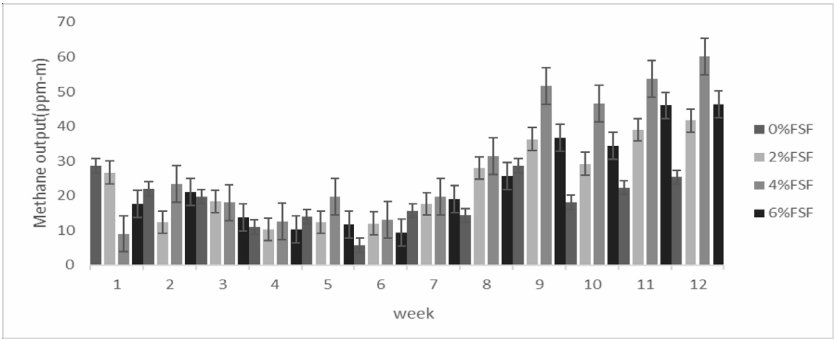

Fig. 1. Methane emission of Dohne-merino wethers at varying Fossil shell flour levels measured for 12 weeks shown as means ± standard errors. 0 % FSF, 2 %, FSF, 4 % FSF, and 6 % FSF

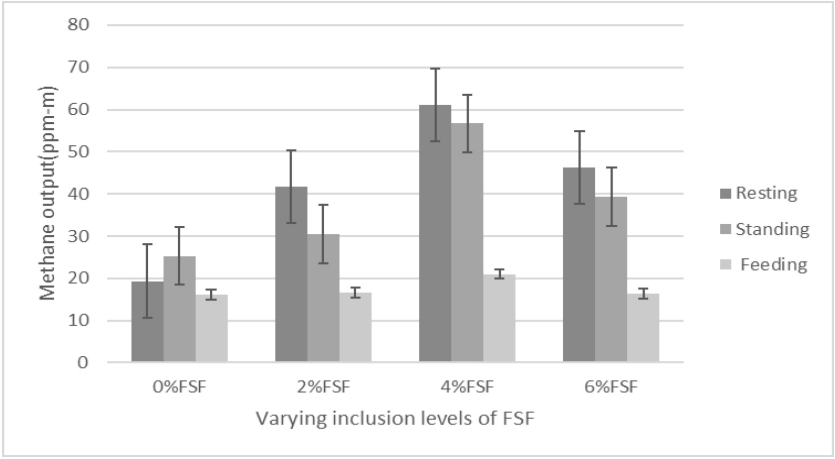

Fig. 2. Methane emission at different positions of Dohne-merino wethers fed basal diet +2%, 4% and 6% FSF shown as means ± standard errors

Discussion

The current study found that daily methane emissions (ppm-m) increased as the FSF inclusion levels increased. This was also true of both the ADFI and DMI, which increased as the FSF inclusion increased. Scholtz et al.[17] and [32] observed in their studies that when livestock consume more feed, they produce more gas than their control). Ramin and Huhtanen [33] and [34] reported that the total methane emitted by an animal is determined mainly by the DMI of the feed consumed by that animal. The results of this study align with the report from these authors. The DMI of FSF supplemented treatments was higher than the DMI of the wethers on 0 %FSF. Hence, the methane output of the supplemented diets was higher than those wethers on 0 %FSF. The reason could be because FSF increased the feed intake of the wethers, thereby increasing the DMI (g/kg) hence more feed content for fermentation. The higher methane output in wethers on FSF supplemented diets compared to those on 0 % FSF observed in this study agrees with [35] report, which considered the influence of the different amounts of FSF on in vitro gas production from West Africa Dwarf sheep. This result suggests that FSF promotes methanogen or protozoan populations. Newbold et al. [36] and[21] reported that methanogens in rumen fluid could contribute up to 25% methane emissions in sheep.

Though the result obtained for wethers on FSF supplemented diets (between 10.27 to 12.52 g/kg D.M.) were higher compared with wethers of FSF

non-supplemented diet (10.65 g/kgDM), it is still lower than the estimation given for South Africa commercial sheep by [34]. This study also observed that more methane was emitted during the 8th and 12th week compared to the 1st to 7th week. Methane output was inconsistent in the early period of the trial. Hence, methane from wethers on 0 % FSF was higher than those on 2 %,4 %, and 6 % FSF during the 1st and 3rd week. However, wethers on FSF supplemented diets emitted far higher methane volume than 0 % FSF during the last 5 weeks of the trial. This could be because FSF has increased the palatability of the FSF supplemented diets, thereby increasing the average daily feed intake, which increases the amount of methane generated from such wethers.

The animal's position and activities during the day affect the amount of enteric methane generated at a particular time [37]. A positive relationship in methane output has been observed between lying behaviour and rumination activities in dairy cows[38]. Similarly, [19] reported a positive correlation between CH_4 output and animal activities. During the day, an animal is either eating, standing, or resting (during which they ruminate on what they have eaten), and these 3 positions were considered. Chagunda et al. [27] and [19] reported that when an animal is quiet and relaxed during rumination, methane emission is higher than when an animal is eating or standing. The result obtained from this study agreed with these authors' reports, in that the wethers emitted more methane output during resting than when standing or feeding. The explanation could be that when an animal is eating (feeding), lesser microbial activities in the rumen (reservoir of microbes) are going on compared to when the wether is resting. When wethers are eating, most activities occur in the mouth. At this stage, enzymes and very few counts of microbes contained in the saliva are involved. Also, continued dilution of the rumen during eating and peristaltic contractions for disturbance of microbial activities compared to the resting period decreases methane production. During the eating period, particles are also larger, thereby reducing microbial activity. However, when wethers are resting, regurgitation is one characteristic they exhibit. This involves bringing back from the rumen to the mouth, feeds they have previously swallowed while feeding for proper chewing, grinding, and mixing. Regurgitation breaks down, feeds into small particles, increases surface area for rapid fermentation, and releases more soluble locked-in crystalline structures, making them available. Therefore, more methane is produced due to more soluble, allowing room to increase microbial growth and population.

Conclusion

In this study, methane production from Dohne Merino wethers was relative to the animal activity, with resting producing more gas than when feeding or standing. Diets supplemented with FSF produce more methane gas than non-supplemented diets. When feeding of FSF goes beyond 5 weeks, a greater volume of methane may be generated because of an increase in average daily feed intake promoted by the continuous addition of FSF. The enteric methane production is directly proportional to ADFI and DMI of the wether.

Funding. This research received no external funding.

Disclosure of statement. The authors report no conflict of interest.

Data availability statement. Data will be available on request

Acknowledgement. The authors wish to acknowledge the Department of Livestock and Pasture Science, University of Fort, South Africa, for providing the platform and enabling environment for research

References

1. Intergovernmental Panel on Climate Change (IPCC). Press Release 8 October 2018. (accessed on 11/04/2019)
2. Moss A.R., Jouany J.P., Newbold J. Methane production by ruminants: its contribution to global warming. *Annales de Zootechnie*, 2000, vol. 49, pp. 231–253. https://hal.science/hal-00889894
3. Alemu A.W., Dijkstra J., Bannink A., France J., Kebreab E. Rumen stoichiometric models and their contribution and challenges in predicting enteric methane production. *Anim. Feed Sci.Technol.*, 2011, vol. 166–167, pp. 761–778. https://doi.org/10.1016/j.anifeedsci.2011.04.054
4. Otter L. The South African agricultural GHG inventory. Department of Agriculture, Forestry and Fisheries, 2010. South Africa.
5. Ogino A., Orito H., Shimadad K., Hirooka H. Evaluating environmental impacts of the Japanese beef cow-calf system by the life cycle assessment method. *Anim. Sci. J.,* 2007, vol. 78, pp. 424–432. https://doi.org/10.1111/j.1740-0929.2007.00457.x
6. Food and Agriculture Organization (FAO). Statistical Yearbook, 2013. Rome, Italy.
7. Tanaka K., O'Neill B.C. The Paris Agreement zero-emissions goal is not always consistent with the 1.5 °C and 2 °C temperature targets. *Nat. Clim. Change,* 2018, vol. 8, pp. 319–324. https://doi.org/10.1038/s41558-018-0097-x
8. Wiedemann S.G., Yan M.J., Henry B.K., Murphy C.M. Resource use and greenhouse gas emissions from three wool production regions in Australia.

J. Clean. Prod., 2016, vol. 122. pp. 121–132. https://doi.org/10.1016/j.jclepro.2016.02.025
9. Fraser M.D., Fleming H.R., Theobald V.J., Moorby J.M. Effect of breed and pasture type on methane emissions from weaned lambs offered fresh forage. *J. Agric. Sci.*, 2015, vol. 153(6), pp. 1128–1134. https://doi.org/10.1017/S0021859615000544
10. Intergovernment Panel on Climate Change (IPCC). guidelines for National Greenhouse Gas Inventories. Prepared by the National Greenhouse Gas Inventories Programme. Eds: Eggleston H.S., Buendia L., Miwa K., Ngara T., Tanabe K. Published: IGES, 2006. Tokyo, Japan.
11. Australian National Greenhouse Accounts: National Inventory Reports (ANIR). Climate Change and Energy Efficiency, Commonwealth of Australia, 2009. Canberra, ACT.
12. Chaturvedi I., Dutta T.K., Singh P.K., Sharma A., Kumar M., Rao B. Effect of Herbal Feed Additives on IVDMD, Methane and Total Gas Production Via in-vitro Study. *J. Agroecol. Nat. Resour. Manag.,* 2014, vol. 1(2), pp. 108-112.
13. Sallaku E., Vorpsi V., Jojic E., Sallaku F., Dodona E. Evaluation of methane emissions from animal farms in Shkodra district-Albania. *Res. J. Agric. Sci.,* 2011, vol. 43, pp. 484- 489.
14. Food and Agriculture Organization (FAO). Statistical Yearbook. 2017. Rome, Italy
15. McMaster, C. Birth of a breed. The Dohne Merino Story. Simon Says Advertising, 2015. Port Elizabeth, RSA.
16. Cardellino, R. 2016. The Dohne In South America. Cardellino Delta Consultants, 2016. Montevideo, Uruguay.
17. Scholtz M.M., Van Ryssen J.B.J., Meissner H.H., Laker M.C. A South African perspective on livestock production in relation to greenhouse gases and water usage. *South African Journal of Animal Science*. 2013, vol. 43(3), pp. 247-254. http://dx.doi.org/10.4314/sajas.v43i3.2
18. Roessler R., Chefor F., Schlecht E. Using a portable laser methane detector in goats to assess diurnal, diet- and position-dependent variations in enteric methane emissions. *Comput. Electron. Agric.* 2018, vol. 150, pp. 110-117. https://doi.org/10.1016/j.compag.2018.04.010
19. Washaya S., Mupangwa J., Muchenje V. Chemical composition of Lablab purpureus and Vigna unguiculata and their subsequent effects on methane production in Xhosa lop-eared goats. *S. Afr. J. Anim. Sci.*, 2018, vol. 48(3), pp. 445-458. http://dx.doi.org/10.4314/sajas.v48i3.5
20. Thota P., Mahender S.C., Ramana D.B.V. Effect of Probiotic Supplementation on Nutrient Digestibilities, Growth Performance and Enteric Methane Emissions

in Deccani Ram Lambs. *J. Anim. Res.*, 2017, vol. 7(6), pp. 1009-1017. https://doi.org/10.5958/2277-940X.2017.00152.8
21. Ma T., Chen D., Tu Y., Zhang N., Si B., Deng K., Diao Q. Effect of supplementation of allicin on methanogenesis and ruminal microbial flora in Dorper crossbred ewes. *Journal of Animal Science and Biotechnology*. 2016, vol. 7(1), pp. 1-7. https://doi.org/10.1186/s40104-015-0057-5
22. Ikusika O.O., Mpendulo C.T., Zindove T.J., Okoh A.I. Fossil Shell Flour in Livestock Production: A Review. *Animals,* 2019, vol. 9(3), pp. 1–20. https://doi.org/10.3390/ani9030070
23. Koster H. Diatomite in Animal Feeds. 2013. https://www.agrisilica.co.za/en/8-diatomite-in-animal-feeds
24. Chen Y., Liu K. Preparation of granulated N-doped TiO2/diatomite and its application of visible light degradation and disinfection. *Power Tech.,* 2016, vol. 303, pp. 176-191. http://dx.doi.org/10.1016/j.powtec.2016.09.038
25. National Research Council (NRC). Nutrient requirements of small ruminants: sheep, goats, cervids, and new world camelids. 2007. Washington, D.C. USA.
26. Association of Officiating Analytical Chemists (AOAC). Official Method of Analysis Association of Officiating Analytical Chemist, 18th ed.; Association of Officiating Analytical Chemists: Washington, DC, USA, 2005.
27. Chagunda M.G.G., Ross D., Roberts D.J. On the use of a laser methane detector in dairy cows. *Comput. Electron. Agric.*, 2009, vol. 68(2), pp. 157–160. https://doi.org/10.1016/j.compag.2009.05.008
28. Chilonda P., Otte J. Indicators to monitor trends in livestock production at national, regional and international levels. *Livestock Research for Rural Development*, 2006, vol. 18(8), article 117. https://lrrd.cipav.org.co/lrrd18/8/chil18117.htm (accessed on 21/11/2019).
29. Shibata M., Terada F., Iwasaki K., Kurihara I., Nishida T. Methane production in heifers, sheep and goats consuming various hay concentrate ratio diets. *Anim. Sci. Technol.,* 1992, vol. 64, pp. 790-796. https://www.jstage.jst.go.jp/article/chikusan1924/63/12/63_12_1221/_pdf
30. Howden S.M., Reyenga P.J. Methane emissions from Australian livestock: implications of the Kyoto Protocol. *Aust. Journal of Agric. Res.,*1999, vol. 50(8), pp. 1285–1292. https://doi.org/10.1071/AR99002
31. Statistical Analysis System(SAS) version 9.1. Inst. Inc., Cary, NC, USA.
32. Tao H., Guo F., Tu Y., Si B., Xing Y., Huang D., Diao Q. Effect of weaning age on growth performance, feed efficiency, nutrient digestibility and blood-biochemical parameters in Droughtmaster crossbred beef calves. *Asian-Australas J Anim Sci.,* 2018, vol. 31(6), pp. 864–872. https://doi.org/10.5713/ajas.17.0539

33. Ramin M., Huhtanen P. Development of equations for predicting methane emissions from ruminants. *J. Dairy Sci.,* 2013, vol. 96(4), pp. 2476–2493. https://doi.org/10.3168/jds.2012-6095
34. Du Toit C.J.L., van Niekerk W.A., Meissner H.H. Direct greenhouse gas emissions of the South African small stock sectors. *South African J. Anim. Sci.*, 2013, vol. 43, pp. 340–361. https://doi.org/10.4314/sajas.v43i3.8
35. Emeruwa C.H. Growth Performance of West African Dwarf Sheep Fed Diets Supplemented with Fossil shell flour., University of Ibadan, Nigeria. 2016.
36. Newbold C.J., Lassalas B. J.P. The importance of methanogens associated with ciliate protozoa in ruminal methane production in vitro. *Lett. Appl. Microbiol.,* 1995, vol. 21(4), pp. 230–234. https://doi.org/10.1111/j.1472-765x.1995.tb01048.x
37. Chagunda M.G.G. Opportunities and challenges in using the Laser Methane Detector to monitor enteric methane emissions from ruminants. *Animal,* 2013, vol. 7, pp. 394–400. https://doi.org/10.1017/S1751731113000724
38. Grobler S.M., Scholtz M.M., van Rooyen H., Mpayipheli M., Neser F.W.C. Methane production in different breeds, grazing different pastures or fed a total mixed ration, as measured by a laser methane detector. *South African J. Anim.Sci.*, 2014, vol. 44, pp. S12– S16. http://www.scielo.org.za/pdf/sajas/v44n5/03.pdf

AUTHORS CONTRIBUTION

Ikusika Olusegun carried out conceptualization, investigation, writing, data collection and methodology, while Thando Mpendulo did the statistical analysis, supervision and validation of the research work.

DATA ABOUT THE AUTHORS

Olusegun Oyebade Ikusika

Department of Livestock and Pasture Sciences, Faculty of Science and Agriculture, University of Fort Hare
1, King Williamstown Rd, Alice, 5700, Eastern Cape, South Africa
Oikusika@ufh.ac.za

Conference Thando Mpendulo

Department of Livestock and Pasture Sciences, Faculty of Science and Agriculture, University of Fort Hare
1, King Williamstown Rd, Alice, 5700, Eastern Cape, South Africa

Поступила 25.06.2022 Received 25.06.2022
После рецензирования 07.08.2022 Revised 07.08.2022
Принята 21.11.2022 Accepted 21.11.2022