



# **Methane Emission Factor in Italian Mediterranean Buffalo According to Production Management**

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## **Authors' contributions**

*This work was carried out in collaboration among all authors. Author NC designed the study, wrote the protocol, writing the article. Author LA done the literature searches, performed the statistical analysis. Author GA managed the analysis, analysing the data. Author RP designing the study, writing the article, author GG managed the analysis. Author FS designed the study, performed the statistical analysis, wrote the protocol, managed the analysis and written the article. All authors read and approved the final manuscript.*

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## **ABSTRACT**

The environmental impact of greenhouse gases caused by livestock farms plays a fundamental role due to the implications and environmental consequences that livestock practices entail, affecting the stability of the entire ecosystem connected to them, especially as a consequence of the growing demand for products of animal origin.

The aim of this work was to quantify the CH<sub>4</sub> emissions factor in lactating buffaloes by comparing four different types of livestock management: family, conventional, organic and sustainable.

To determine the enteric CH<sub>4</sub> emissions from buffalo, information about animal production and farm management was analyzed, and the CH<sub>4</sub> emission factor was calculated using the IPCC Tier 2 model.

ANOVA was conducted to evaluate significant differences between the farms; Pearson's correlation was used to evaluate the relationship between parameters.

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In a conventional farm, the CH<sub>4</sub> emission factor for buffalo was 27.69 kg CH<sub>4</sub>/head/yr compared to 22.77 and 21.61 kg CH<sub>4</sub>/head/yr respectively for organic and family-run. These data may also depend on the higher protein and fiber content in the administered *unifeed*. Furthermore, the ratio of enteric emissions factor of CH<sub>4</sub> / gross energy intake ratio reflected these data (12.04 vs 10.93 vs 10.16 vs 10.65 for conventional, organic, sustainable, and family-run farms, respectively).

**Keywords:** CH<sub>4</sub> emission factor; buffalo cows; managements; organic; conventional.

## 1. INTRODUCTION

Due to a growing concern about climate change, greenhouse gas (GHG) emissions have become one of the major issues in all industrial sectors [1]. Agricultural activity accounts for around 60% and 50% of global anthropogenic emissions of nitrous oxide (N<sub>2</sub>O) and methane (CH<sub>4</sub>), respectively, and the livestock sector has been recognized as an important contributor to the emissions of greenhouse gases [2].

More than 70% of CH<sub>4</sub> is generated by anthropogenic activities, including animal husbandry (27%; enteric fermentation in livestock, manure management), paddy rice cultivation (26%), petroleum sources (26%), waste management (12%) and biomass combustion (9%) [3].

Enteric fermentation of ruminants is the largest source of CH<sub>4</sub> emissions in the livestock sector [4]; it is therefore necessary to accurately estimate the enteric production of methane in these species to develop a national greenhouse gas inventory and establish mitigation strategies for greenhouse gas emissions from primary livestock production.

Concerns about global warming have increased the pressure to promote environmentally sustainable livestock production. Therefore, research focused on assessing greenhouse gas emissions from the livestock sector is essential to develop more sustainable practices.

The methodologies suggested by the IPCC guidelines [5] for the estimation of the enteric methane production by cattle are Tier 1 (default values), Tier 2 (including diet and energy intake considerations), and Tier 3 (country-specific methodologies and parameter estimates). Tier 1, the least accurate approach, provides tabular fixed values. The Tier 2 methodology is commonly used to quantify enteric CH<sub>4</sub> emissions from cattle and estimate methane emissions from enteric fermentation of individual cattle by calculating a CH<sub>4</sub> emission factor (MEF,

Kg CH<sub>4</sub>/head/year). Which is the product of a CH<sub>4</sub> conversion factor (MCF: percentage of gross energy [GE] in feed converted into CH<sub>4</sub>) and daily GE intake (MJ/head/day).

According to Xue et al. [6] studied the effects of methane (CH<sub>4</sub>) emissions as a function of manure management in large ruminants in China, and concluded that the country of breeding strongly influences the results. Therefore, it is essential to carry out these studies on animals reared in a particular country to provide the latter with all the information necessary to proceed with the appropriate development of policies and mitigation strategies to reduce the production of this greenhouse gas.

The breeding of dairy buffaloes (*Bubalus bubalis*) is traditional in the swampy areas of the central and southern plains of Italy. For zootechnical purposes, the buffalo species is today considered to have a dual aptitude, although milk production is more important than meat production; in fact, the buffalo species has produced 123 million tons of milk and 4.2 million tons of meat worldwide.

According to FAO estimates [7], the world livestock sector is responsible for 14.5% of total anthropogenic emissions. Considering the main greenhouse gases, at a global level, livestock farms would be responsible for the emission of about 9% of carbon dioxide, 37% of methane, and 65% of nitrous oxide. In particular, global anthropogenic emissions amount to about 9 million tons of CO<sub>2</sub>eq per year.

74% of the world's emissions of livestock origin are caused by cattle. This is mainly due to the abundance of dairy cattle, but also to a large amount of methane and nitrous oxide emitted by beef cattle compared to other animals. According to recent estimates reported by ANSA, sheep contribute 9% of world emissions, followed by buffaloes (7%), pigs (5%), and goats (4%).

In Italy, agriculture is responsible each year for about 7% of total national emissions and about

3.2% of emissions are attributable to livestock farming, as reported by [8,9]; in Italy, regardless of the type of farming, more or less intensive, the differences in terms of methane emissions between cattle and buffaloes are considerable. This data could be justified considering the number of animals raised certainly to the advantage of bovine animals.

The aim of this work was to quantify CH<sub>4</sub> emissions factor in lactating buffaloes by comparing four different types of livestock management: family, conventional, organic and sustainable.

In Italy, to date, CH<sub>4</sub> emissions have been estimated using country-specific emission factors for cattle only; while for other livestock species, the default emission factors [5] are used.

Therefore, in this experimental work, methane in dairy buffaloes was calculated following the Tier 2 method by [5].

## 2. MATERIALS AND METHODS

### 2.1 Experimental Design

The study was conducted in Southern Italy, in four different farms, and involving 30 Italian Mediterranean lactating Buffaloes in each farm, at the same stage of lactation, average production in the previous lactation, and parity.

The quantification of CH<sub>4</sub> emission factor in lactating buffaloes was obtained by comparing four different types of livestock management: family, conventional, organic, and sustainable.

The farms were selected according to the management system:

1. Family-run system (FRS): A small-sized farm, where the ration was composed by polyphyte hay and integrated concentrate; it is spread over about 6 hectares and raises a total of 80 heads, of which about 38 were in production at the time of the experimental test. The food ration administered was composed of polyphyte hay, flour-ground maize, and integrated compound feed.
2. Conventional system (CS): Has a total extension of 260 Ha. Approximately 1000 animals are present in the breeding, and at the time of the experimental test there

were about 310 lactating animals, divided into production groups. The total mixed ration was composed of corn silage, medical hay and polyphyte hay, feed (corn flour, soybean, protein concentrate, wheat bran and supplements of mineral and vitamin).

3. Organic system (OS): It extends for about 200 Ha, and raises about 630 animals. At the time of the experimental test, there were about 209 lactating buffaloes, divided into production groups. The ration administered consisted of corn silage, alfalfa hay and alfalfa wrap, straw and integrated compound feed.
4. Sustainable system (with a forage production sustainable for the environment) (SS): It has a total extension of about 20 Ha. Around 300 heads are present on the farm, and at the time of the experimental test about 100 of them were in lactation, divided into three production groups. It produces "Nobile" mozzarella (a cheese characterized by milk from free grazing animals, which feed freely, without forcing). The ration administered consisted of an integrated compound feed and polyphyte hay.

From these buffaloes, samples of total mixed ration and milk were taken, on which the *unifeed* chemical-nutritional characteristics and the chemical composition of the milk were calculated respectively.

### 2.2 Chemical-nutritional Characteristics and Nitrogen Balance in Different Farms

The *unifeed* samples were collected directly by the manager as soon as discharged by the mixer wagon, for three consecutive days, and analyzed in duplicate.

The feces and milk samples were collected directly in the rectal ampoule, in all 30 animals per farm, and on the same days as the sampling of *unifeed* samples. Moreover, in this case, the analyses were carried out in duplicate.

The chemical-nutritional analysis was carried out with near-infrared spectroscopy (NIR) and reported in Table 1.

Immediately after drying, grinding was done with a 1.1 mm grinding grid.

**Table 1. Chemical composition of diets and chemical-nutritional characteristics of milk (% on dry matter) in different farms**

Farm		FRS	CS	OS	SS
Chemical-nutritional characteristics		$\mu \pm \sigma$	$\mu \pm \sigma$	$\mu \pm \sigma$	$\mu \pm \sigma$
Dry matter (DM)	%	68.85 A $\pm$ 4.56	57.71 B $\pm$ 2.24	57.21 B $\pm$ 2.87	58.95 B $\pm$ 3.02
DM intake	Kg/day	19.41 B $\pm$ 0.95	22.40 A $\pm$ 1.29	21.37 A $\pm$ 1.02	11.34 B $\pm$ 0.90
Crude protein	%	15.02 A $\pm$ 1.82	16.26 A $\pm$ 1.67	11.94 B $\pm$ 1.52	14.61 A $\pm$ 1.85
Crude lipids	%	4.50 B $\pm$ 0.95	6.03 A $\pm$ 0.80	5.54 A $\pm$ 0.73	3.95 B $\pm$ 0.62
Crude fibre	%	17.80 B $\pm$ 1.73	20.72 A $\pm$ 1.85	20.69 A $\pm$ 1.74	17.61 B $\pm$ 1.53
Ash	%	7.20 A $\pm$ 1.2	3.38 B $\pm$ 1.18	8.46 A $\pm$ 1.09	5.51 B $\pm$ 1.12
NSC	g <sup>-kg DM</sup>	38.31 A $\pm$ 3.03	34.84 B $\pm$ 3.21	31.61 B $\pm$ 2.98	32.29 B $\pm$ 3.13
Starch	%	25.33 B $\pm$ 2.25	24.66 B $\pm$ 2.61	25.51 B $\pm$ 3.02	29.61 A $\pm$ 2.87
Milk production	Kg	9 A $\pm$ 0.97	10 A $\pm$ 1.65	9 A $\pm$ 1.25	8 B $\pm$ 1.30
Fat	%	6.78 B $\pm$ 0.99	8.73 A $\pm$ 1.04	8.17 A $\pm$ 1.00	9.15 A $\pm$ 0.98
Density	°SH	1.0353 $\pm$ 0.015	1.0383 $\pm$ 0.011	1.0375 $\pm$ 0.009	1.0352 $\pm$ 0.0.1
Lactose	%	5.15 B $\pm$ 0.35	5.56 A $\pm$ 0.33	5.41 A $\pm$ 0.21	5.25 B $\pm$ 0.28
RDM	%	10.75 b $\pm$ 1.02	11.55 a $\pm$ 0.61	11.22 a $\pm$ 0.70	11.13 a $\pm$ 0.74
Protein	%	2.94 B $\pm$ 0.18	4.31 A $\pm$ 0.15	4.02 A $\pm$ 0.24	3.45 B $\pm$ 0.41
Freezing point	°C	-0.654 B $\pm$ 0.09	-0.711 A $\pm$ 0.10	-0.685 B $\pm$ 0.10	-0.691 B $\pm$ 0.1
Salts	%	0.82 B $\pm$ 0.06	0.99 A $\pm$ 0.05	0.95 A $\pm$ 0.03	0.96 A $\pm$ 0.07
pH		6.7 $\pm$ 0.05	6.6 $\pm$ 0.13	6.72 $\pm$ 0.12	6.65 $\pm$ 0.10
FCM	kg	9.55 $\pm$ 0.07	10.69 $\pm$ 0.01	9.49 $\pm$ 0.04	8.51 $\pm$ 0.02
DM intake/kg of milk	kg/kg of milk	1.30 A $\pm$ 0.15	1.35 A $\pm$ 0.21	1.43 A $\pm$ 0.18	0.86 A $\pm$ 0.11

FRS = family-run system; CS = Conventional system; OS = Organic system; SS = Sustainable; RDM = residual dry matter; FCM = fat corrected milk; A, B:  $P < 0.01$ ; a, b:  $P < 0.05$

Before making such determinations, the sample was dried in a forced ventilation oven at 65 °C for 48 h until the weight was stable

The NIR analysis was performed in the wavelength range between 1100 and 2500 nm, with a reading resolution of about 2 nm, for a total number of 700 reading points for each scan. Before the analysis, the instrument was subjected daily to the revision and calibration of the optical groups (called SST, System Suitability Test), to optimize the readings.

For calibrations of the *unifeed* chemical composition, the calibration curves contained in a database formed by the registrations obtained from previous studies were used and updated.

For the quantification of the nitrogen eliminated with the urines, we considered a value equal to about 30% on average during the lactation, so it is a calculated and not a determined value.

### 2.3 Milk Analysis

The study was conducted in four different farms, and on 30 Italian Mediterranean lactating Buffalo for each farm, at different stage of lactation and a different parity, as the representativeness of the average conditions of the livestock.

Milk yield was calculated by recording the consecutive milking production on the same day, and milk samples collected and analyzed.

Nutrient composition on milk was determined, on 50 ml samples, using a Milk-Lab PRO (MilkLab – United Kingdom), which is based on mid-infrared spectroscopy. For pH determinations, a portable pH-meter Mettler Toledo was used.

We also proceeded to calculate the fat correct milk for the energy content (FCM, kg), using the formula of [10], where fat and protein (g · kg<sup>-1</sup>) represent the respective contents in the analyzed milk:

$$\{[(\text{fat} - 40 + \text{protein} - 31) \times 0.01155] + 1\} \times \text{milk yield}$$

The parameters analyzed were temperature, density, freezing point, pH, and fat content, residual dry matter (RDM), proteins, lactose, salts, and FCM (fat corrected milk), and are reported in Table 1.

### 2.4 Estimation of the Emission Factor of Methane

The following parameters were calculated as suggested by the IPCC guidelines:

#### 2.4.1 Net energy required by the animal for maintenance (MJ/d)

$$NE_m = C_{fi} \times (\text{weight})^{0.75}$$

Where:

C<sub>fi</sub>: a coefficient varying according to animal category (0.322 for non-lactating buffalo and 0.335 for lactating buffalo)

Weight: animal metabolic body weight, in Kg

#### 2.4.2 Net energy for lactation (MJ/d)

$$NE_l = \text{Kg of milk per day} \times (1.47 + 0.40 \times \text{Fat})$$

#### 2.4.3 Net energy for pregnancy (MJ/d)

$$NE_p = C_{\text{pregnancy}} \times NE_m$$

Where:

C<sub>pregnancy</sub>: pregnancy coefficient = 0.10.

#### 2.4.4 Ratio of net energy available in a diet for maintenance to digestible energy consumed = REM

$$\text{REM} = 1.123 - (4.092 \times 10^{-3} \times \text{DE}) + [1.126 \times 10^{-5} \times (\text{DE})^2] - (25.4/\text{DE})$$

Where:

DE: digestible energy expressed as a percentage of gross energy.

According to some studies, the equation to estimate the gross energy intake (GEI) is the following:

#### 2.4.5 Gross energy intake for buffalo (MJ/head/yr)

$$\text{GEI} = [(\text{NE}_m + \text{NE}_l + \text{NE}_p) / \text{REM}] / (\% \text{DE}/100)$$

The emission factor should be calculated using the following equation:

#### 2.4.6 Emission factor, kg CH<sub>4</sub>/head/yr

$$\text{EF} = (\text{GEI} \times Y_m \times 365 \text{ d/yr}) / (55.65 \text{ MJ/kg CH}_4)$$

Where:

EF: emission factor, Kg CH<sub>4</sub>/head/yr.

GEI: gross energy intake, MJ/head/d.

Y<sub>m</sub>: methane conversion rate, namely the fraction of gross energy contained in feed

converted to methane. The Ym value is 6% in buffalo cows.

## 2.5 Statistical Analysis

ANOVA was carried out to evaluate significant differences between the farms. Pearson's correlation was used to evaluate the relationship between the parameters.

All statistical methods of data evaluation were done using [11].

## 3. RESULTS

Data related to dry and organic matter intake and chemical-nutritional characteristics of milk are given in Table 1.

The dry matter administered, as a function of live weight, was found to be similar for the conventional and organic farm (0.022 and 0.021 kg/kg LW, respectively), being lower in the family company (0.019 kg/kg LW) and significantly lower in sustainable conduction (0.011 kg/kg LW).

While for the conventional and organic farm the values obtained are in line with those reported in the literature [12], a sustainable farm is significantly different.

The intake of DM per kg of milk was 1.30 kg/kg milk for the family business, 1.35 kg/kg milk for the conventional farm, 1.43 kg/kg milk for the organic farm and 0.86 kg/kg for a sustainable system. From the analysis of the data it is clear that, while for family-run, conventional and organic farms the differences do not appear significant, sustainable farm differs significantly ( $p < 0.01$ ) from the other three.

The nitrogen balance, in different farms, are reported in Table 2.

The amount of nitrogen secreted in milk and excreted in feces and urine as a percentage of nitrogen consumption was 108% for buffalo breed in the organic farm, 104% for that breed in family-run, 86% for the breed in conventional and 98% for the breed in sustainable one.

There were significant differences ( $p < 0.05$ ) on the quantity of nitrogen excreted by feces and milk, and the differences appear to be evident in organic and sustainable farms, which excreted less nitrogen through milk than conventional and

family-run systems; regarding the nitrogen excreted through feces, the difference occurs between farms. Regarding the nitrogen excreted through feces, in farms where a food ration that satisfies less maintenance and production needs (family-run and sustainable), the difference appears more evident; indeed, in this managements system there is major excretion of nitrogen.

The efficiency of the use of the nitrogen of ration was found to be equal to 48.02, 30.79, 23.58, and 44.04 respectively for the family, conventional, organic, and sustainable farm.

Dry matter intake (kg) per fat corrected milk produced (kg DM/kg FCM) was equal to 1.30 in case of the family-run farm, 1.35 for the conventional, 1.43 for the organic, and 0.86 for the sustainable farm.

Data on Gross Energy Intake (GEI, in MJ/head/yr) and Emission Factor (EF, in Kg CH<sub>4</sub>/head/yr) for buffalo cows were calculated for each animal in each farm, and are reported in Table 3.

Another important point emerging from the analysis of the experimental data is that, although the energy requirement for maintenance is the most important aspect to estimate the CH<sub>4</sub> emission factor, as shown in Fig. 1, this required energy is not influenced by production, while the daily energy requirement rises according to the amount of milk produced. Hence, the fraction of energy required for maintenance is also reduced. Consequently, it appears clear that farm management strongly influences the amount of produced CH<sub>4</sub>.

## 4. DISCUSSION

Rearing system ensuring fewer requirements of nitrogen of maintenance and production, in relation to fibre and NSC content (17.61 and 32.29%, respectively) and high starch content (29.61%), excrete more nitrogen through faeces compared to the other three systems.

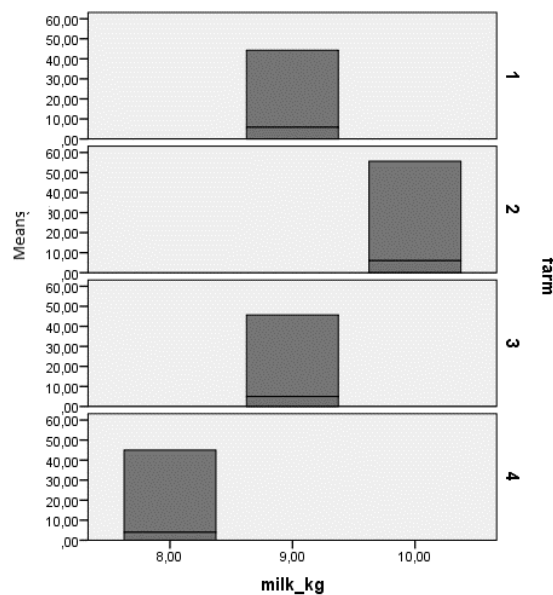
In the study conducted to evaluate the utilization of the residual feed intake (RFI) as a feed efficiency selection tool and its relationship with methane emissions, [13] reported that the selection of more efficient buffalo heifers has multiple benefits, such as decreased feed intake and less emission of methane.

**Table 2. Nitrogen balance in the stool in different farms**

Farm		FRS	CS	OS	SS
		$\mu \pm \sigma$	$\mu \pm \sigma$	$\mu \pm \sigma$	$\mu \pm \sigma$
N ingested	g/day	232.59 $\pm$ 7.46	263.32 $\pm$ 2.50	188.34 $\pm$ 4.99	229.28 $\pm$ 7.85
N eliminated with feces	g/day	111.61 A $\pm$ 0.48	81.08 B $\pm$ 1.78	85.03 B $\pm$ 4.21	100.88 B $\pm$ 0.64
N eliminated with urine	g/day	69.77 $\pm$ 2.24	79.00 $\pm$ 0.75	56.50 $\pm$ 1.50	68.78 $\pm$ 2.36
N eliminated with milk	g/day	61.13 B $\pm$ 6.75	67.52 A $\pm$ 0.63	58.04 B $\pm$ 4.69	54.11 B $\pm$ 1.75
Nitrogen efficiency	%	48.02 $\pm$ 5.24	30.79 $\pm$ 4.98	23.58 $\pm$ 4.15	44.04 $\pm$ 3.79
N total	g/day	242.51 $\pm$ 7.85	227.60 $\pm$ 3.0	202.78 $\pm$ 4.06	223.77 $\pm$ 2.29

A, B:  $P < 0.01$ **Table 3. Gross energy intake and emission factor in different farms**

Farm		FRS	CS	OS	SS
		$\mu \pm \sigma$	$\mu \pm \sigma$	$\mu \pm \sigma$	$\mu \pm \sigma$
GEI	MJ/head/yr	202.92 b $\pm$ 31.02	229.91 a $\pm$ 27.69	208.30 b $\pm$ 25.42	209.01 b $\pm$ 24.56
EF	kg CH <sub>4</sub> /head/yr	21.61 b $\pm$ 3.21	27.69 a $\pm$ 2.98	22.77 b $\pm$ 2.25	22.92 b $\pm$ 2.63
EF/GEI	%	10.65 b	12.04 a	10.93 b	10.16 b

a, b:  $P < 0.05$ **Fig. 1. Relationship between energy maintenance and milk production in Italian buffaloes**

From data relating nitrogen excretion in milk, it appears that organic and sustainable farms excrete less nitrogen than conventional ones, which is due to a lower protein content of N in the unfed administered to buffaloes, in these farms.

The intake of dry matter in the present study is similar to data reported by [12,14].

About the data on the energy balance, it is well known that in the last days of pregnancy and the first weeks of lactation ruminants are in a

negative energy balance, i.e. the nutrients that they ingest are not sufficient to meet the energy needs of the animal. This happens because the udder subtracts huge amounts of glucose, amino acids, and fatty acids for the production of milk, fat, and proteins.

The data recorded for GEI in this experiment is higher than the value reported by [15], who found the highest value of 184.95 GE intake in buffaloes. On the contrary, the methane emission factor is much lower than that calculated in this

experiment. Due to better feeding management in the other three systems comparing to conventional farm, the value for CH<sub>4</sub>, EF and GE intake were lower in these systems.

Regarding the efficiency of the use of nitrogen content in ration, this trend could be justified by the fact that the animals reared on the organic farm tend to adapt to the nitrogen deficiency by decreasing the nitrogen clearance from the kidneys, increasing the ruminal yield and decreasing the blood levels of urea, as suggested by [7].

From Fig. 1, it is evident that the energy requirement is influenced by production which also influences the methane production.

Our results represent an example of a study comparing the environmental impact of a dairy buffaloes farming system with different system managements in Italy.

## 5. CONCLUSION

This study showed that the type of corporate management in the buffalo species influences the environmental impact regarding the CH<sub>4</sub> emission factor; this result is mainly due to the diet administered to the animals.

Another important point emerging from the analysis of the experimental data is that, although the energy requirement for maintenance is the most important aspect to estimate the CH<sub>4</sub> emission factor, this required energy is not influenced by production, while the daily energy requirement rises according to the amount of milk produced. Hence, the fraction of energy required for maintenance is also reduced. Consequently, it appears clear that farm management strongly influences the amount of produced CH<sub>4</sub>. Correctly balancing the nutrients in the ration, in particular the ratio between protein and energy, to maximize the efficiency of use of both fractions at rumen level, appears essential not only to meet the nutritional needs of the animals to which the ration is intended, but also to try to have a lower impact on the environment.

It is also important to regularly check the chemical-nutritional composition of the diet and food to formulate adequate diets.

It is concluded that the Family system of rearing buffalo is emitting less methane with the highest

nitrogen efficiency and conventional system due to less DM intake per kg milk produced and lower EF/GEI, and that more research is required to achieve maximum compliance with environmental quality and standards of animal welfare.

## ETHICAL APPROVAL

Animal Ethic committee approval has been collected and preserved by the authors.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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