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Original Research Article

# Dynamic model development of enteric methane emission from goats based on energy balance measured in indirect open circuit respiration calorimeter

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#### ABSTRACT

A dynamic model of methane  $(CH_4)$  emission in goats was proposed and parameterized from energy balance experimental data. The model focused on dry matter intake and fat content of the diet as explanatory variables for  $CH_4$  emission. Experimental and literature data were used to develop the model. Then, data (n=123) from five energy balance experiments were used to evaluate the model. The model was adequate to represent energy in milk, heat production and  $CH_4$  emissions. Residual analysis showed that most of the prediction errors were due to unexplained variations with small mean and slope bias (around zero with exception of  $CH_4$ ; <6%). The model tends to over-predict energy in  $CH_4$  at higher energy intake and, energy in milk and heat production at lower energy intake. Random bias was greater than 90%, signifying than more than 90% of the error was non-systematic indicating the mechanism in the model are properly represented. The model is a first step towards a mechanistic description of nutrient use by goats and, useful as a research tool for investigating energy partition in dairy goat systems. The model described in this study should be considered for preparation of enteric  $CH_4$  emissions inventories for goats.

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## 1. Introduction

Agreement was reached at the 2015 United Nation Climate Change Conference in Paris to keep global warming "well below 2°" (United Nations Framework Convention on Climate Change, 2015). Current emissions are predicted to increase global temperatures by 1.5 °C within 15 years and by 2 °C within 35 years (Howarth, 2015). As well as reduced CO<sub>2</sub> emissions, substantial reductions in methane (CH<sub>4</sub>) will be needed to achieve the target (United Nations Environmental Program and Word Meteorological Organization, 2011). Enteric CH<sub>4</sub> from livestock contributes approximately 38.6% of total agricultural emissions (FAO, 2010). Although a major portion of the CH<sub>4</sub> emission arises from cattle (73.8%) and buffalo (11.3%) in 2010, the world goat population of about 1.01 billion (FAOSTAT, 2018) produces around 4.61 million tons of enteric CH<sub>4</sub> representing 4.9% of total CH<sub>4</sub> emissions from livestock (Patra, 2014). Furthermore, CH<sub>4</sub> emissions from goats are expected to grow in the future due to enhanced growth of goat population and growing demands of milk and meat (Haenlein, 2001). As the world population continues to grow during this century, agriculture must focus on production efficiency to provide an adequate

food supply, and milk and dairy foods are important source of calories as well as protein and micronutrients (Knapp et al., 2014). Development of CH<sub>4</sub> emissions prediction models is, therefore, required to precisely estimate CH<sub>4</sub> emission from goats.

Within external influences, CH<sub>4</sub> emissions are strongly related to feed intake (Moorby et al., 2015) and dietary lipids (Beauchemin et al., 2008; Knapp et al., 2014), and both may help to quantify and mitigate CH<sub>4</sub> emissions. A number of statistical models for predicting CH<sub>4</sub> emissions from goats have been developed based on dietary composition and nutrient intake from 42 publications (Patra and Lalhriatpuii, 2016). There is not model to predicting CH<sub>4</sub> emissions in goats developed from energy transfers. The goat requires energy for self-organization, motion, harvesting food, maintenance, growth and/or milk production. Jørgensen (2015) indicated that this is important to distinguish between two forms of energy: energy that can-do work and energy that cannot do work, but is lost as heat to the environment at the temperature of the environment. Some heat (maintenance and CH<sub>4</sub> emissions) is lost during every energy transformation, and their quantification was some of the main objectives of present study.

The aim of this study was to develop, represent and assess a dynamic mathematical model for dairy goats based on flow of the energy intake though the body, quantifying  $CH_4$  emissions and total amount of milk produced.

#### 2. Materials and methods

#### 2.1. Ethics statement

The experimental procedures carried out were approved by the Committee on Animal Use and Care at the Universitat Politèctica de Valencia (Spain). Animals were cared for by trained personnel and managed in accordance with the Spanish guidelines for experimental animal protection (Royal Decree No. 1201 2005) and the European Convention for the Protection of Vertebrates used for Experimental and other Scientific Purposes (European Directive 86/609).

#### 2.2. Experimental data

To develop the model, experimental energy balance data was obtained with 8 goats. Eight Murciano-Granadina dairy goats at mid lactation (16 weeks), with similar body weight (BW;  $47 \pm 3.9 \text{ kg}$ ) were selected to determine energy balance and gas exchange. Goats were fed once per day and, each goat was offered 2.5 kg of fresh feed per day, comprising 1.0 kg forage and 1.5 kg of a standard concentrate. The concentrate consisted in 2 commercial compound feed; one with 2% of fat and the other with 5% of fat. Nutrient requirements of the goats were obtained using the recommended values of (AFRC 1993; FEDNA 2009)

 Table 1

 Descriptive statistics of the variables in the database used to develop the methane model in goats.

Variable <sup>a</sup>	Lactating Dairy Goat fed Mixed Diet						
	(n = 8)	(n = 8)					
	Mean	Min.	Max.	SD			
Diet Composition							
Forage to concentrate ratio40/6	0						
DM percentage	93.3	92.9	93.7	0.57			
CP (% DM)	17.9	17.6	18.3	0.46			
EE (% DM)	2.3	1.8	2.8	0.72			
NDF (% DM)	38.5	34.6	42.5	5.59			
Ash (% DM)	8.1	6.9	10.7	0.99			
Starch (% DM)	15.0	12.4	17.7	3.76			
GE (MJ/kg DM)	17	16	17	0.2			
Energy balance (kJ/kg of BW <sup>0.75</sup> )							
Intake							
DMI, kg/d	2.0	1.8	2.1	0.19			
GEI	1926	1703	2106	238.4			
Energy waste							
Methane	85	68	101	17.8			
Fecal	630	537	706	125.4			
Urinary	74	53	85	32.5			
Energy can not do work							
Heat production	584	539	623	54.8			
Energy can do work							
MEI	1137	1039	1236	167.8			
Reserves	131	18	284	102.9			
Energy in milk	423	321	500	136.3			
Goat characteristics							
Body weight (kg)	47	44	51	3.9			
Milk yield (kg/d)	2.3	1.8	2.5	1.4			

<sup>&</sup>lt;sup>a</sup> DM = dry matter; CP = crude protein of diet; EE = ether extract; NDF = neutral detergent fiber; GE = gross energy; DMI = dry matter intake; GEI = gross energy intake; MEI = metabolizable energy intake.

for goats in lactation. Water was freely available at all times. The mean gross energy (GE) of the two diets on the dry matter basis (DM) was 17 MJ kg $^{-1}$ , crude protein (CP) 17.9%, and neutral detergent fiber (NDF) 38.5% (see Table 1 for details). All goats were housed in a building in which the environment was controlled by a HOBO device (HOBO probe, Onset Data Loggers, Cape Cod, MA, USA) at thermo-neutrality; 20-23 °C.

The goats were kept in individual pens for a period of 15 d to adapt to their experimental diets. Then goats were moved to individual metabolism cages for other 10 d of adaptation. Then, feed intake, total fecal and urine output, and milk were recorded daily for each goat over a 5 d period for energy balance. Each goat was milked at  $0800 \, h$  with a portable milking machine (Flaco, model DL-170, J. Delgado S.A., Ciudad Real, Spain). Representative samples of diet, feces, urine and milk were collected daily, stored at  $-20 \, ^{\circ}$ C, and pooled for energy analysis.

After collecting data for energy balance, gaseous emissions from each goat were then measured for a period of 24 h by housing them in individual metabolism cages fitted with the respirometry units, 2 animals per day. The indirect calorimetry system based on a mobile open-circuit respiration system for measuring real time gaseous exchange in small ruminants (sheep and goats) was described by Fernández et al. (2015).

Metabolizable energy intake was the difference between GE intake and energy losses in feces, urine and CH<sub>4</sub> (with an energy equivalent value of  $39.54 \, \text{kJ L}^{-1}$ ; Brouwer 1965). Brouwer (1965) developed the equations for calculation of heat production (HP) based on gas exchange and nitrogen excretion in urine. The HP was determined from measurements of  $O_2$  consumption,  $CO_2$  and  $CH_4$  production, and urine nitrogen ( $N_{\text{urine}}$ ):

HP (kJ) = 
$$16.18 \times O_2 + 5.02 \times CO_2 - 2.17 \times CH_4 - 5.99 \times N_{urine}$$

where gases were expressed in L  $d^{-1}$  and  $N_{urine}$  in g  $d^{-1}$ .

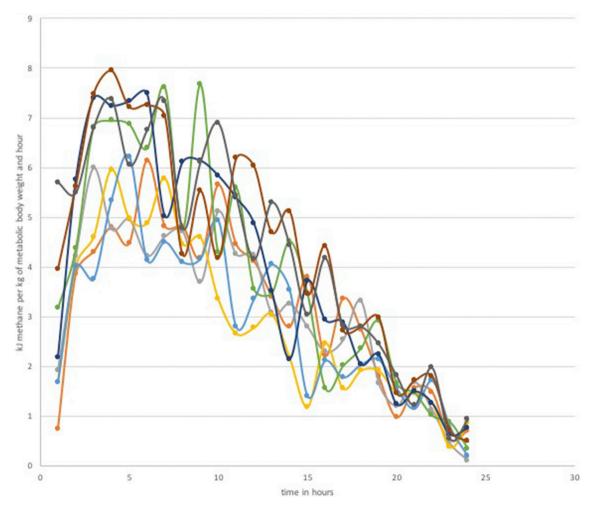


Fig. 1. Hourly evolution pattern of energy lost in CH<sub>4</sub> from the 8 goats used to develop the model.

Retained energy was determined as the difference between metabolizable energy intake and the energy retained in milk plus the energy that cannot do work (HP). Fig. 1 represent the hourly pattern for CH<sub>4</sub> production and the 8 goats. Goats were fed once per day at the beginning, and approximately after 24 h only some left over were observed.

Next,the chemical analysis was briefly described following. Feed and feces were dried in a forced air oven at 55 °C for 48 h and then grounded to pass 1 mm screen. So, dry matter intake (DMI) was obtained. Urine and milk were dried by lyophilization. Chemical analyses were conducted according to methods of AOAC (2000) for dry matter (no. 934.01), ash (no. 942.05), ether extract (EE) (no. 920.39) and CP (no. 968.06). GE content was determined in an adiabatic bomb calorimeter (Gallenkamp Autobomb; Loughborough, UK). NDF concentration of diets was determined using filter bags and a fiber analyzer (A220; ANKOM Technologies, Fairport, NY, USA) following AOAC (2000) official methods (no. 973.18). Nitrogen were analyzed by Dumas principle (TruSpec CN; LECO Corporation, St. Joseph, MI, USA).

## 2.2.1. Model description

The goat model was set up to simulate indoor facilities in which animals are grouped in lots by their production potential; special attention was paid over dry matter intake and fat content of the diets. Each element of the model is specified by initial conditions. The initial conditions derived from actual measurements and fractional rates derived mainly from the experimental and empirical information. To evaluate the goat model, information from different studies from literature was used. The model consisted of a dynamic system of differential equations, coded in R (R Core Team, 2016).

The model contains six pools (kJ kg of BW $^{-0.75}$ ) represented by capital letter and with a box, and the inputs and outputs to and from the pools are the flows (kJ kg of BW $^{-0.75}$  h $^{-1}$ ) and, are represented by arrows and denominated by the abbreviation F (Fig. 1 and Table 2). The amount of energy gained minus the amount of energy lost to the environment will tell us how much energy is accumulated. Therefore, the pool or accumulation will change with time depending on the magnitude of the fluxes, and the change is described by a differential equation of the form:  $dPOOL/dt = F_{in} - F_{out}$ . The model was programmed in R software. A fourth order Runge-Kutta method with an integration step size of 0.05 h was used for numerical integration, and the model was run until 24 h was achieved for each level of DM intake.

A model was developed assuming mass action (F = k x POOL; being k the rate constant) and saturating flux (i.e. Michaelis-Menten [ $F = Mx/(1 + (K_m/POOL))$ ); where Mx is the maximal energy rate and K the affinity constant]. Table 2 describes all stocks, fluxes and symbols used to develop the model.

Diagrammatic representation of the model is shown in Fig. 2. Description of pools and the associated differential equations follow below and abbreviations are summarized in Table 2.

**Table 2** Pools, fluxes and symbols used in the model.

Label	Description
Energy pools (kJ kg of BW <sup>-0.75</sup> )	
FA	Feed available
D	Digestive tract
RM	Rumen
M	Metabolism
R	Reservoir
MLK	Milk
Energy fluxes (kJ kg of $BW^{-0.75} h^{-1}$ )	
F <sub>FA D</sub>	Gross energy intake to digestive tract
F <sub>D feces</sub>	Waste of fecal energy
F <sub>D M</sub>	Digestible energy flux to metabolism
F <sub>D RM</sub>	Energy taken by rumen microbial population
F <sub>M urine</sub>	Waste of energy in urine
$F_{M_R}$	Metabolizable energy flux to reservoir
F <sub>M_heat</sub>	Heat energy that cannot do work
F <sub>M MIK</sub>	Flux of metabolizable energy to milk
F <sub>RM CH4</sub>	Waste of energy from methane
Fractional rates (h <sup>-1</sup> )	
k <sub>i</sub>	fractional rate of F <sub>FA D</sub>
k <sub>d</sub>	fractional rate of F <sub>D M</sub>
$k_{ij}$	fractional rate of F <sub>M urine</sub>
$k_r$	fractional rate of F <sub>M R</sub>
k <sub>h</sub>	fractional rate of F <sub>M heat</sub>
k <sub>m</sub>	fractional rate of F <sub>M MLK</sub>
Reference constants	
R_EE (%)	Minimum fat inclusion in mixed diets
n	Power exponent
Mx	Maximum energy rate
K	Affinity constant in Michaelis Menten equation
Inputs	
BW (kg)	Input value of body weight
GE (kJ/gDM)	Diet input value of gross energy
EEd (%)	Diet input value of fat

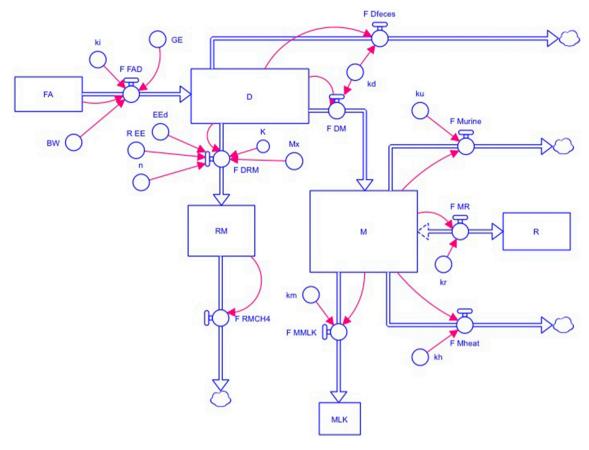


Fig. 2. Diagrammatic representation of the mathematical model (using Stella software). See Table 2 for legend.

Intake pool, FA (g x  $d^{-1}$ ). The intake pool included the dry matter intake, it is the food available. The initial amount of DMI was 2.0 kg  $d^{-1}$  on average, determined experimentally, and the average BW of the eight goats was 47 kg. This pool had one outputs. The output was the conversion from dry matter intake to gross energy intake per metabolic body weight. The energy content of the diet (GE) was 17 kJ g DM $^{-1}$  and the metabolic BW was the kg of BW $^{-0.75}$ . The rate constant ki ( $h^{-1}$ ) was one of the parameters to estimate and the initial value observed was 0.20. Therefore, the flux from pool E to pool D was the hourly gross energy intake ( $F_{E,D}$ ) from outside to digestive tract.

Intake pool, FA (g x  $d^{-1}$ ). Differential equation:

$$dFA = -F_{E\_D}$$
  
Outputs:

 $F_{FA\_D} = (ki \ x \ E \ x \ GE) \ x \ BW^{-0.75}$ 

The E pool size was expressed by the integral equation:

$$E = \int_{0}^{24} \frac{dFA}{dt} + iE$$

Representing the quantity of dry matter accumulated during initial time (0) and final time (24), being iFA the initial pool size (2.0 kg DMI  $d^{-1}$ ).

Digestive Tract pool, D ( $kJ \times kg$   $BW^{-0.75}$   $day^{-1}$ ). The digestive tract pool includes one input and three outputs. The input was the flux of energy from energy intake to digestive tract ( $F_{FA\_D}$ ) described previously. The outputs are the waste of energy from the digestive tract to feces ( $F_{D\_feces}$ ), that is the opposite to the energy digestibility. Other output is the digestibility; obtained experimentally and defined as  $F_{D\_M} = kd \times D$ , being the fractional rate kd = 0.67. The last output from this pool was energy used for rumen fermentation ( $F_{D\_RM}$ ). As Robinson et al. (2016) mentioned, within external influences,  $CH_4$  emissions are strongly related to feed intake and dietary lipids. Intake was considered in the first pool and dietary lipids was considered in

the rumen pool. So, R\_EE represent the ether extract or fat content of the diet. Due that our model was based goats fed mixed diets and allocated indoor, a reference value of fat was 1.8%. EEd represented the actual fat content of the mixed diet. Besides, the flux  $F_{D\_RM}$  included a saturation function, due that after place the daily meal into the feeder we observed a rapid rate of CH<sub>4</sub> production with a maximum value of 8 kJ kg BW<sup>-0.75</sup> h<sup>-1</sup> (Fig. 1). This higher value was accompanied with a K value of 55 kJ kg BW<sup>-0.75</sup>.

Digestive Tract Pool, D (kJ x kg  $BW^{-0.75}$  day<sup>-1</sup>). Differential equation:

$$\begin{split} dD = F_{FA\_D} - F_{D\_M} - F_{D\_feces} - F_{D\_RM} \\ Inputs: \end{split}$$

$$F_{FA\_D} = (ki \ x \ E \ x \ GEd) \ x \ BW^{-0.75}$$
  
Outputs:

$$\begin{aligned} F_{D\_M} &= kd \ x \ D \\ F_{D\_feces} &= (1 - kd) \ x \ D \\ F_{D\_RM} &= \ Mx \ x \frac{\left(\frac{R\_EE}{EEd}\right)^n}{\frac{D}{(K+D)}} \end{aligned}$$

Where n was the exponent of the EE ratio with an observed value of 0.30. In the Michaelis-Menten equation, Mx was the maximum energy waste in form of CH<sub>4</sub> per hour and K the affinity constant.

The digestive pool size was expressed by the integral equation:

$$D = \int_{0}^{24} \frac{dD}{dt} + iD$$

Representing the quantity of digestible energy accumulated during initial time (0) and final time (24 h), being iD the initial pool size (that we assumed equal to 0).

Rumen Pool, RM ( $kJ \times kg \ BW^{-0.75} \ day^{-1}$ ). The digestive tract pool includes one input and one outputs. The input  $F_{D\_RM}$  was defined previously and the output was the energy from the rumen that was emitted to the environment in the form of  $CH_4$  ( $F_{RM\_CH4}$ ).

Rumen Pool, RM (kJ x kg  $BW^{-0.75}$  day<sup>-1</sup>). Differential equation:

$$\begin{aligned} dRM = F_{D\_RM} - F_{RM\_CH4} \\ Inputs: \end{aligned}$$

$$F_{D\_RM} = Mx x \frac{\left(\frac{R\_EF}{EEd}\right)^n}{\frac{D}{(K+D)}}$$

Outputs:

 $F_{RM CH4} = RM$ 

The rumen pool size was expressed by the integral equation:

$$RM = \int_{0}^{24} \frac{dRM}{dt} + iRM$$

Representing the quantity of rumen energy from initial time (0) and final time (24 h), being iRM the initial pool size (0). *Metabolism pool*,  $M(kJ \times kg \ BW^{-0.75} \ day^{-1})$ . The metabolism pool represents the metabolizable energy intake. The metabolism pool includes one inputs and four outputs. The fractional rates of the inputs and outputs were obtained experimentally from the trial described above. The input was the flux of digestible energy defined beyond  $(F_{D_m})$ . The outputs comprise the energy that cannot do work  $(F_{M_n})$  and was lost to the environment  $(kh = 0.51 \ h^{-1})$ , the waste of energy lost in urine  $(F_{M_n})$  with an observed fractional rate  $ku = 0.065 \ h^{-1}$ , the flux of energy conducted to milk production  $(F_{M_n})$ , with a fractional rate  $km = 0.34 \ h^{-1}$ ) and the reservoir energy  $(F_{M_n})$  with a  $kr = 0.115 \ h^{-1}$ .

Metabolism pool, M (kJ x kg  $BW^{-0.75}$  day<sup>-1</sup>). Differential equation:

$$\begin{split} dM = & F_{D\_M} - F_{M\_urine} - F_{M\_R} - F_{M\_heat} - F_{M\_MLK} \\ Inputs: \end{split}$$

$$F_{D\_M} = kd x D$$
  
Outputs:

$$F_{M \text{ urine}} = k_u x M$$

$$F_{MR} = k_r x M$$

$$F_{M\_heat} = k_h x M$$

$$F_{M\_MLK} = k_m x M$$

The metabolism pool size was expressed by the integral equation:

$$M = \int_{0}^{24} \frac{dM}{dt} + iM$$

Representing the quantity of metabolizable energy, being iM the initial pool size (0).

Reservoir pool,  $R(kJx kg BW^{-0.75} day^{-1})$ . The reservoir pool includes one input. Due that the model was developed for dairy goats in mid or late lactation, we have assumed only one direction flux, from M to R  $(F_{M_R})$ .

Reservoir pool, R (kJ x kg  $BW^{-0.75}$  day<sup>-1</sup>).

Differential equation:

$$dR = F_{M\_R}$$
Inputs:

$$F_{M R} = k_r x M$$

The reservoir pool size was expressed by the integral equation:

$$R = \int_{0}^{24} \frac{dR}{dt} + iR$$

Representing the quantity of energy accumulated during the day, being iR the initial pool size (0).

Milk pool, MLK (kJ x kg  $BW^{-0.75}$  day<sup>-1</sup>). The milk pool includes one input and represent the energy use by the mammary gland for milk production.

Milk pool, MLK (kJ x kg  $BW^{-0.75}$  day<sup>-1</sup>).

Differential equation:

$$dMLK = F_{M\_MLK}$$
Inputs:

 $F_{M\_MLK} = k_m x M$ 

The milk pool size was expressed by the integral equation:

$$MLK = \int_{0}^{24} \frac{dMLK}{dt} + iMLK$$

Representing the quantity of energy accumulated during the day in the mammary gland for milk production, being iMLK the initial pool size (0).

#### 2.2.2. Parameter estimation

The dynamic model was implemented in R software (2016) and the function ode of the deSolve (solving differential equations) package (Soetaert et al., 2010) for numerical solution of initial first order problems was used. The solution was achieved using the Isoda integration method with absolute and relative error tolerance of  $10^{-6}$ . The root means square error

(RMSPE) measures the magnitude of the difference between the output from the model and the experimental data. The DEoptim function of the package DEoptim (performing the Differential Evolution algorithms) searches for minima of the objective function between lower and upper bounds on each parameter to be optimized. The algorithm evaluates different combinations of the parameters in the provides ranges, comparing them by the RMSE. Bootstrapping is a statistical method for assigning measures of accuracy, such as confident interval, to the parameter estimates. Nonparametric bootstrap resampling was run and 1000 resampling was performed and the confidence interval was determined (Efron, 1979).

## 2.3. Evaluation of the mathematical model

#### 2.3.1. Lactation trial

Using residues of the crop and processing industries to feed livestock has the advantage to obviating the need for costly waste management programs. Data from five energy balance experiments (two unpublished) conducted at the Universitat Politècnica de Valencia were used to validate the model (López et al., 2014; Criscioni and Fernández, 2016; Ibáñez et al., 2016). These trials evaluated the response of lactating goats in terms of intake, energy and N balance, apparent total tract digestibility and milk performance, when cereal were replaced with horticulture byproducts. Fibrous by-products are cheaper than cereal grains and is an interesting alternative, recycling and reusing to fed dairy goats. The trial of López et al. (2014) studied the effect of replacing corn grain with citrus pulp, Criscioni and Fernández (2016) replaced oats with rice bran, Ibáñez et al. (2016) replaced barley grain with soy hulls and orange pulp, and the other two unpublished studies replaced barley with lemon pulp, and cereals with beet pulp, respectively. All studies had shown the possibility to substitute the cereal grain with fibrous byproducts successfully. And to maintain isoenergetic diets, those diets with fibrous byproduct had fat added. The trials encompassed a total of 123 multiparous Murciano-Granadina goats in mid lactation. Intake was *ad libitum* with diets offered at 110% of consumption on the preceding few days. Half the daily ration was offered at 08:00 and half at 16:00 h, respectively. Goats had free access to water. A summary of the data used in the model evaluation is given in Table 5.

## 2.3.2. Model evaluation

The adequacy of the model developed was assessed by residual analysis. The observed values of energy in milk (MLK), daily HP and CH<sub>4</sub>, emission was compared with model predictions. An assessment of the error of the predicted relative to the observed values was made by calculation of the root mean squared prediction error (RMSPE). The prediction error was assessed by calculating the mean square prediction error (MSPE). The MSPE was decomposed (Bibby and Toutenburg, 1977) into error in central tendency (ECT), error due to regression (ER), and error due to disturbances (ED). Root MSPE was used as a measure of accuracy of prediction (RMSPE). All calculations were performed by R software (2016).

# 3. Results

# 3.1. Model development

The model had four parameters and the parameters estimation of the model used observations of 8 energy balance goats; energy intake and output of CH<sub>4</sub>, fecal, urinary and milk. The initial and final values of optimized parameters, obtained by RMSPE, with their standard deviation (SD) and variation coefficient (CV) are shown in Table 3. The parameters ki and n had a CV around 0.12 and K and Mx around 0.06. The results of the nonparametric bootstrap are shown in Table 4. After 1000

**Table 3**Initial and final parameters estimation and standard deviation of optimized model parameters.

Parameters <sup>a</sup>	Initial Values	Final Values	SD	CV
ki	0.20	0.17	0.021	0.11
n	0.30	0.25	0.035	0.13
K	55.0	59.0	2.83	0.05
Mx	8.0	8.8	0.57	0.07

<sup>&</sup>lt;sup>a</sup> Parameters abreviation is given in Table 2; SD = standard deviation; CV = variation coefficient.

**Table 4**Methane model with the estimates obtained by bootstrap analysis in the current analysis.

Parameters	Original sample	Mean	Bootstrap <sup>a</sup>	
			Lower limit	Upper limit
ki	0.1694	0.1694	0.1692	0.1697
n	0.2523	0.3569	0.1294	0.5696
K	59.08	59.09	58.84	59.228
Mx	8.829	9.224	8.401	10.043

<sup>&</sup>lt;sup>a</sup> Parameter fit.

**Table 5**Descriptive statistics of the variables in the database used to validate the methane model in goats.

Variable <sup>a</sup>	Lactating Dairy Goat fed Mixed Diet $ (n = 123) $				
	Mean	Min.	Max.	SD	
Diet Composition					
Forage to concentrate ratio40/60					
DM percentage	90.4	87.5	93.2	0.87	
CP (% DM)	16.3	13.2	16.5	2.17	
EE (% DM)	2.4	1.6	5.3	0.99	
NDF (% DM)	34.1	21.0	58.9	7.93	
Ash (% DM)	8.1	6.9	10.7	0.99	
Starch (% DM)	25.6	1.5	41.6	9.47	
GE (MJ/kg DM)	17	16	18	0.5	
Energy balance (kJ/kg of BW <sup>0.75</sup> )					
Intake					
DMI, kg/d	1.752	1.285	2.352	0.1998	
GEI	1746	1424	2140	140.2	
Energy waste					
Methane	97	66	116	12.6	
Fecal	491	300	794	124.6	
Urinary	46	25	116	12.9	
Energy can not do work					
Heat production	623	570	692	31.2	
Energy can do work					
MEI	1113	907	1307	86.2	
Reserves	69	-101	210	70.15	
Energy in milk	422	392	456	14.4	
Goat characteristics					
Body weight (kg)	44.4	33.0	60.5	4.74	
Milk yield (kg/d)	1.825	1.408	2.349	0.21	

<sup>&</sup>lt;sup>a</sup> DM = dry matter; CP = crude protein of diet; EE = ether extract; NDF = neutral detergent fiber; GE = gross energy; DMI = dry matter intake; GEI = gross energy intake; MEI = metabolizable energy intake.

resampling, the best parameters obtained by RMSPE were into the bootstrap lower and upper limit. The bootstrap means parameters were used for model evaluation (see Table 4 and Fig. 3).

#### 3.2. Model evaluation

The RMSPE can be used as a measure of model adequacy. The model prediction errors were reasonable for all predicted outputs; < 5% of the observed values (Table 6). Values 4.94% in heat production indicated that some variation that remains to be explained. The slope bias was close to zero for milk and HP, and less precision was found for energy in CH<sub>4</sub> (3.40%). Mean bias represents the accuracy of the model being around zero for HP, however some disturbances were found for milk (1.07%) and CH<sub>4</sub> energy (6.0%). Random bias was greater than 90%, signifying than more than 90% of the error was non-systematic indicating the mechanism in the model are properly represented.

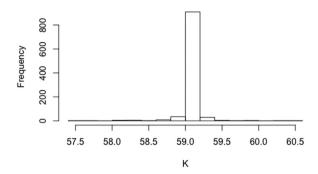
Plots of observed versus predicted values in feces, urine and milk are shown in Fig. 4. The model under predicted milk energy and HP, and over predicted energy in  $CH_4$ . The standardized residual error was 13.5, 30.86 and 2.75 for milk energy, HP and  $CH_4$ , respectively. Analyses of residuals are shown in Fig. 5. This method involves regressing residuals against predicted values, as residuals are not correlated with predictions and the slope of residuals regressed on prediction must be zero if the model is unbiased. Slope of residuals versus predicted were positive for energy in  $CH_4$ , indicating that the model overpredicted the amount of energy emitted as  $CH_4$  as predicted amount increased. The slope was negative for energy in milk and  $CH_4$  indicating that the model underpredicted as predicted energy increased.

## 4. Discussion

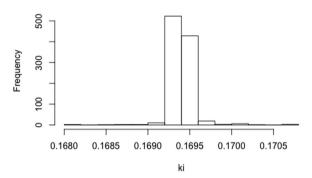
The main objective of the model was to describe the partition of the energy that can-do work (available to product milk) from the energy that cannot do work, and it is lost to the environment in form of CH<sub>4</sub> and heat.

The pioneer study of Blaxter and Clapperton (1965) used more than 2500 determinations of the 24 h production of CH<sub>4</sub> by indirect calorimetry, although the linear model included energy digestibility and level of feeding, no goats were studied. INRA (2017) proposed estimated equation of energy losses in CH<sub>4</sub> based on organic matter intake, BW, level of feeding and concentrate level of diet. The inputs of the model developed in this study were the BW, DMI and EE of the diet. Patra and Lalhriatpuii (2016) in goats demonstrated that intake of nutrients was stronger determinant of methane production than nutrient composition, they found a strong relationship between CH<sub>4</sub> production and DMI or energy intake (R<sup>2</sup> ranged from 0.75 to 0.85). Different studies also reported that feed intake (DM or energy) was the key explanatory variable for prediction

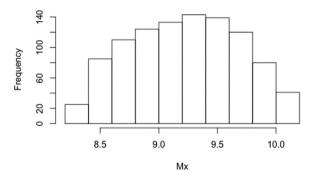
## Bootstrap results; run number=1000



# Bootstrap results; run number=1000



# Bootstrap results; run number=1000



# Bootstrap results; run number=1000

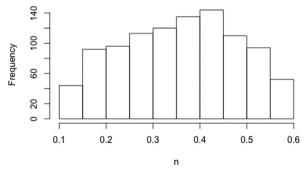


Fig. 3. Nonparametric Bootstrap for each parameter; ki, n, K and Mx.

**Table 6**Model evaluation: prediction errors and decomposition associate with prediction of the outputs.

Variable <sup>a</sup>	Observed	Predicted	RMSPE <sup>b</sup>	Mean bias, %	Slope bias, %	Random bias, %
Milk energy	421.5	422.9	3.19	1.07	0.83	98.10
Heat production	623.3	622.0	4.94	0.20	0.70	99.10
Methane energy	96.53	95.83	2.99	6.00	3.40	90.60

a Variable abbreviation is given in Table 2.

<sup>&</sup>lt;sup>b</sup> RMSPE = root mean square prediction error as a percentage of observed mean.

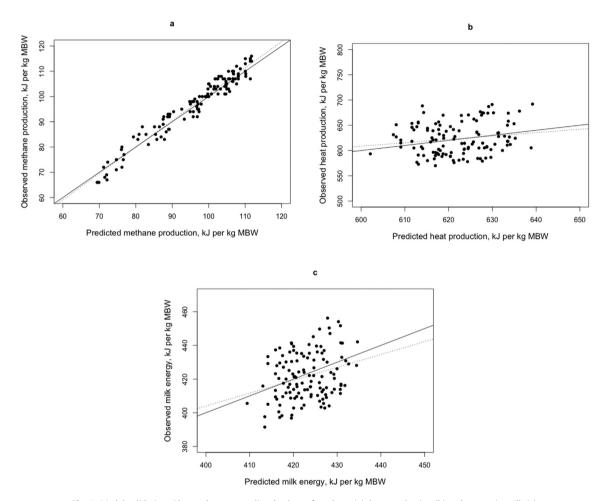


Fig. 4. Model validation: Observed versus predicted values of methane (a), heat production (b) and energy in milk (c).

equations of CH<sub>4</sub> emission in cattle with R<sup>2</sup> from 0.68 to 0.85 (Mills et al., 2003; Yan et al., 2009; Ramin and Huhtanen, 2013). Grainger and Beauchemin (2011) reported that increasing the level of starch and lipids, plus decreasing NDF and acid detergent fiber in diets, reduced the CH<sub>4</sub> production. However, Patra and Lalhriatpuii (2016) did not include the concentration of EE in their model, and Ellis et al. (2007) included DMI and EE intake in their equations and the prediction of CH<sub>4</sub> in cattle was improved. Although some studies excluded EE concentration as an explanatory variable, EE in diets inhibits the growth and activity of methanogens, lowering CH<sub>4</sub> production in the rumen (Grainger and Beauchemin, 2011). During the last 30 years, dairy goat feeding systems in Spain have passed from grazing to have the animals confined on the barn, feeding with mixed diets rich in concentrates, that was partially replaced with fiber-byproduct and the amount of EE in diets was increased (Daza et al., 2004). The composition of a diet can shifts the microbial population in the rumen and consequence the production of volatile fatty acids. When the dynamic model from the present study was run with inputs of 44 kg of BW, 2 kg of DM intake, 17 MJ of GE kg DM<sup>-1</sup> and 3.2% EE, the CH<sub>4</sub> production was 97 kJ kg BW<sup>-0.75</sup> d<sup>-1</sup> (14.8 g CH<sub>4</sub> kg DMI<sup>-1</sup>). This value decrease to 76 kJ kg BW<sup>-0.75</sup> d<sup>-1</sup> (11.7 g CH<sub>4</sub> kg DMI<sup>-1</sup>), when the amount of EE was 5.6%. Similar variation was found in the experimental study of Bava et al. (2001) comparing forage with non-forage diet in Saanen goats at mid lactation. They found values of CH<sub>4</sub> production of 131 and 115 kJ kg BW<sup>-0.75</sup> d<sup>-1</sup> for diets with 3.2 and 5.6% of EE, respectively. The differences in our

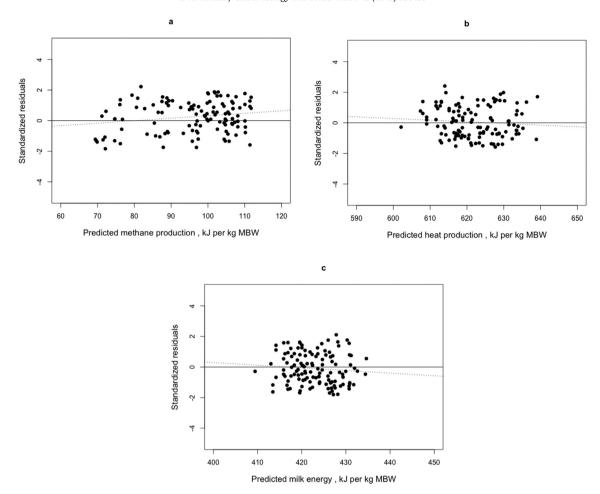


Fig. 5. Model validation: Residuals plot of methane (a), heat production (b) and energy in milk (c).

simulation were of 21 kJ kg BW $^{-0.75}$  d $^{-1}$ , and 16 kJ kg BW $^{-0.75}$  d $^{-1}$  for Bava et al. (2001) study. The differences and trends detected between the simulation and the experimental study of Bava et al. (2001) was close, although in the Bava et al. (2001) study the breed was Saanen with higher BW (55 kg), greater intake (2.8 kg DM $^{-1}$ , on average), greater GE (19 MJ kg $^{-1}$ ) and milk production. In the study of Aguilera et al. (1990) with Granadina goat at mid and late lactation the CH<sub>4</sub> production was 111 and 97 kJ kg BW $^{-0.75}$  d $^{-1}$ , respectively. Similar values to observed in our simulation with 3.2% of EE, although the differences here were due to the stage of lactation because diet consisted on pelleted alfalfa and barley during the whole lactation and, no information about the level of fat was available. Comparing 3.2 and 5.6% EE, the model reduced CH<sub>4</sub> by 9% for each percent increased in fat diet (the reduction was 5% for Bava et al., 2001). Patra (2013) found lower reduction (4%) in dairy lactating cows than in the dynamic model developed in dairy goats. INRA (2017) included three meta-analyses of data of the literature, globally pooling 205 treatments, and have proposed an equation based in lipids contents to calculate CH<sub>4</sub> mitigations. The EE ranged from 2 to 10% and the reduction per each percent in EE was 5% (Giger-Reverding et al., 2003), 8% (Moate et al., 2011) and 10% (Grainger and Beauchemin, 2011).

The average CH<sub>4</sub> production in goats from the present model was 0.82 MJ kg DMI<sup>-1</sup>. The average value obtained by the statistical model developed by Patra and Lalhriatpuii (2016) in goats was 0.94 MJ DMI<sup>-1</sup>, similar to the value found in the present study. However, the CH<sub>4</sub> emission in cattle is higher, ranged from 1.12 to 1.49 MJ DMI<sup>-1</sup> (Ellis et al., 2007; Yan et al., 2009). It appears that CH<sub>4</sub> production is lower in goats than cattle, although different classes of animals, physiological status and feeding regimens bared not clear explanation. Tovar-Luna et al. (2010), studied the effect of dietary concentrate level on energy utilization by Alpine goats when diets had 60% or 20% of concentrate. At mid lactation, the observed CH<sub>4</sub> emissions ranged from 0.86 to 0.93 MJ DMI<sup>-1</sup> (60% of concentrate) and, from 0.68 to 0.72 MJ DMI<sup>-1</sup> (20% of concentrate). As our mixed diets had a 60% of concentrate, the CH<sub>4</sub> production from our simulation was into the range found by Tovar-Luna et al. (2010) for dietary concentrate level of 60%. However, this study did not show the EE of the diets.

The IPCC (2007) national greenhouse inventory guidelines outline methods for estimating CH<sub>4</sub> emissions from enteric fermentation. Food and Agricultural Organization (FAO, 2010) had developed some equations for estimate CH<sub>4</sub> emissions from ruminants, as well. These institutions had empirical equations and enteric CH<sub>4</sub> emissions was proportional to DMI and

thus usually normalized by expressing them on the basis of DMI, either as g CH<sub>4</sub> kg DMI<sup>-1</sup> or as a percentage of GE intake (also called Ym factor [IPCC, 2007]; the proportion of the GE intake which is lost as CH<sub>4</sub>). Kebreab et al. (2008) predicted daily emissions for lactating dairy cows that ranged from 15 to 35 g CH<sub>4</sub> kg DMI<sup>-1</sup>, while Shibata and Terada (2009) found values of 36 and 37 g CH<sub>4</sub> kg DMI<sup>-1</sup> for sheep and goats, respectively. Our value simulated for dairy goats was 14.73 g CH<sub>4</sub> kg DMI<sup>-1</sup> (assuming a value of 55.65 kJ g CH<sub>4</sub><sup>-1</sup>, Brouwer, 1965), lower than the values found by Shibata and Terada (2009). These authors based their diets on timothy and alfalfa hay and in our study, we used mixed diets with a 60% of concentrate. Using IPCC Tier 2 (2007) methodology for dairy cattle, Ym of 6.5% is suggested. With diets consisting primarily of grains, the percentage of GE intake that is converted to CH<sub>4</sub> in the rumen is typically less than 4% compared to the 6.5%, which is common for animals fed primarily forage (Beauchemin et al., 2009). Kebreab et al. (2008), using a mechanistic model (COWPOLL) for dairy cows, found a lower value for CH<sub>4</sub> emissions (5.6% of GE, on average) than IPCC (2007). Merino et al. (2001) reported that Ym ranged from 4 to 7% for dairy ewes. The Ym value in goats at mid lactation from the studies mentioned above (Bava et al., 2001; Tovar-Luna et al., 2010) ranged from 3.9 to 5%. The Ym value obtained in our goat mathematical simulation model was 5.5%, lower than the IPCC (2007) recommendation.

Methane emission from different existing model were obtained using as inputs our database (Table 5). IPCC (2007) [CH<sub>4</sub> (MJ d<sup>-1</sup>) = 0.065 x GE intake] delivered a value of 1.95 MJ d<sup>-1</sup>. FAO (2010) [CH<sub>4</sub> (MJ d<sup>-1</sup>) = (9.75–0.005 x DM digestibility, g kg<sup>-1</sup>) 100<sup>-1</sup> x GE intake] provided 1.88 MJ d<sup>-1</sup>. The linear model of Patra and Lalhriatpuii (2016) predicted 1.35 MJ d<sup>-1</sup> [CH<sub>4</sub> (MJ d<sup>-1</sup>) = (0.242 + 0.0511 x digestible energy intake] and the Mitscherlich model shown 1.29 MJ d<sup>-1</sup> [CH<sub>4</sub> (MJ d<sup>-1</sup>) = 1.721 x {1 - e<sup>(-0.0721 x ME intake)</sup>}. Table 5 shown an observed average value of 1.67 MJ d<sup>-1</sup> and our simulation predicted 1.63 MJ d<sup>-1</sup>. The models of IPCC (2007) and FAO (2010) have been suggested to prepare inventories of enteric CH<sub>4</sub> production. Both, overestimated the CH<sub>4</sub> production, whereas the Patra and Lalhriatpuii (2016) models underestimated the CH<sub>4</sub> emission. Thus, CH<sub>4</sub> production were estimated more accurately using the dynamic model developing in the current study compared with IPCC (2007) and FAO (2010).

Therefore, as Robinson et al. (2016) mentioned, within external influences, CH<sub>4</sub> emissions were strongly related to feed intake and dietary lipids, and both may help to quantify and mitigate CH<sub>4</sub> emissions. Goat energy balance data had shown us that changes in the stored energy (diet) can then be transferred along different organs, and each transfer was accompanied by energy loss. We have observed in the dynamic model that energy exchange between the goat and its environment occurs in different ways; work exchange, matter exchange and heat exchange. More studies combining chemical composition of diets and energy transfers are needed. The model described in this study should be considered for preparation of enteric CH<sub>4</sub> emissions inventories for goats.

#### 5. Conclusions

A dynamic model to predict CH<sub>4</sub> production by goats was developed and validate. The goat model was set up to simulate indoor facilities in which the goat was fed mixed rations. Body weight, dry matter intake, gross energy and fat content of the diets were useful explanatory variables to predict CH<sub>4</sub> during a day and, the dynamic model should be valuable for preparing CH<sub>4</sub> emission inventories in goats. Integration of information generated from other experiments and literature into de simulation model will contribute to a more dynamic understanding of the energy transfer and conversions in this system. The model was not set up to consider the mitigation effects of anti-methanogenic components or additives.

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