

## Methane, energy balance and feed efficiency MIR predictions evaluation on dairy cow's population

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Methane (CH<sub>4</sub>) is one of the major greenhouse gas (GHG) emissions. Along with other sources it is naturally produced during the rumen fermentation process of ruminants. Different factors could have an impact on CH<sub>4</sub> quantity emitted i.e. feed, feeding system, herd management or genetic selection for efficient animals. The study aim was to compare milk MIR CH<sub>4</sub> predictions relevance obtained with different equations based on same reference dataset but including or not fix effects (1,203 data). The CH<sub>4</sub> models were built on SF<sub>6</sub> tracer gas method and respiratory chambers measurements with PLS, CPPLS and GLMNET methods and considered the stage of lactation. The model performances have been statistical validated in yearly dairy data production. Pearson correlation analysis has been made with milk MIR indicators. For energy efficiency were found positive correlation with GLMNET model. Negative correlations were found between all CH<sub>4</sub> models and blood NEFA, C18-1Cis9, Energy-Detect, KetoMIR, INSAT and positive correlation showed SCFA and MCFA and SAT. The GLMNET models showed positive correlation with EB NEL and ME. The correlations fit well with the usual metabolic effects of extreme body fat mobilization e.g. an increased concentration of NEFA, ketone bodies, long chained unsaturated fatty acids, a decreased concentration of MCFA and an extreme negative EB. The effect can be explained by reduced feed intake and rumen activity in these situations. The limits of applicability must still be defined in order to ensure the relevance of the predictions obtained, therefore in ReMissionDairy project the GLMNET model predictions are used in a production report for field consultants and advisors in order to reduce the CH<sub>4</sub> emission and the CH<sub>4</sub> per kg milk. To better understand the CH<sub>4</sub> results team work with filed experts and also climatic environmental experts is necessary.

### Abstract

**Keywords:** MIR spectra, ECM, CH<sub>4</sub>, energy balance, feed efficiency, energy efficiency

## Introduction

Mid infrared spectroscopy (MIR) is using the infrared light from the electromagnetic spectrum which shows specific absorption patterns when sent through a milk sample caused by frequency dependent interactions with the chemical bonds of the chemical milk components. With the help of milk MIR spectra, a wealth of information can be obtained by establishing relationships with reference methods. Since 2012 researchers in the MIR spectroscopy are working on creating predictions models based on milk MIR spectral data and dairy cow phenotypes like standard milk components, new milk MIR components like ketone bodies (Grelet C. *et al.*, 2016), fatty acids (Grelet C. *et al.*, 2014), minerals (Soyeurt H. *et al.*, 2009) and MIR based blood components (BHB, NEFA, Glucose, IBF1, Insuline, Calcium) (Dale L. *et al.* 2019) and traits like energy balance (NEL and ME) (Dale L. *et al.*, 2019). The current research focus is the detection of indirect quantities, such as: methane (CH<sub>4</sub>) emissions (Dehareng *et al.*, 2012), furthermore Vanlierde (2019) present a set of CH<sub>4</sub> models based on spectral data and SF<sub>6</sub> and climatic chamber measurements. CH<sub>4</sub> is one of the major greenhouse gas (GHG) emissions. Along with other sources it is naturally produced during the rumen fermentation process of ruminants. Different factors could have an impact on CH<sub>4</sub> quantity emitted i.e. feed, feeding system, herd management or genetic selection for efficient animals. The eMissionCow project focused on the prediction of emission and efficiency related phenotypes and their potential applications, whereas the ReMissionDairy project is testing the estimated values for feed efficiency, energy balance and methane emissions on different pilot farms in 4 federal states of Germany. The ReMissionDairy projects aim is to develop feeding strategies to increase efficiency and to decrease emissions of CH<sub>4</sub>. The study aim was to compare milk MIR CH<sub>4</sub> predictions relevance obtained with different equations and compare energy balance, energy efficiency and different CH<sub>4</sub> equations based on MIR spectra predictions. The objective of this study was to evaluate different CH<sub>4</sub> equation based on SF<sub>6</sub> tracer gas method and respiratory chambers in order to improve the quality of the predictions obtained and choose the best model.

## Material and methods

From the work at the Walloon Agricultural Center and University of Liege, Gembloux Agro Bio-Tech, in cooperation with OptiMIR/EMR partners, the Methagene Group and the European project GplusE, a CH<sub>4</sub> equation with SF<sub>6</sub> tracer gas method and respiratory chambers measurements was created. The spectral data set was first standardized by applying the OptiMIR/EMR method (Grelet *et al.*, 2015) and pre-processed by Savitzky-Golay first derivative to remove the offset differences between samples for baseline correction, before performing Legendre polynomial modelling. To identify the main variables that were positively or negatively associated with CH<sub>4</sub> emission, the data was submitted to polynomial regression in combination with lasso parameter optimization as implemented in the “glmnet” R package.

The Legendre polynomial data based on DIM for the 212 OptiMIR wavenumbers of spectral data were used as input variables. The global spectrometric equations for energy balance calculated by the two evaluation systems net energy lactation (NEL) (GfE, 2001) and metabolizable energy (ME) (Susenbeth, 2018) is the result of a collaboration between the German optiKuh project consortium and the “German Association for Quality and Performance Testing e.V.” (DLQ). optiKuh is a collaborative project, of 12 research farms from different German states such as Baden Württemberg, North Rhine-Westphalia, Bavaria, Schleswig-Holstein, Rhineland-Palatinate, Lower Saxony and Mecklenburg-Western Pomerania, funded by the German Federal Ministry of Food and Agriculture. Between 2014 and 2017 a dairy cow feeding experiment with weekly collection of milk samples and daily individual feeding data provided reference values of both energy balances (NEL, ME). The local MROs and associated milk laboratories, organized in DLQ, provided milk recording results and standardised as

Table 1. Milk MIR models for CH<sub>4</sub> emission and energy status.

Milk Biomarker	Unit	#LV	$\phi$	SD	SEC	R <sub>1c</sub>	SECV	R <sub>1cv</sub>	RPD	Use
CH <sub>4</sub> Emission	[g/d]	12	1266	97.00	50.00	0.73	54.00	0.69	1.80	- <sup>1</sup>
Energy Balance – NEL*	[MJ/d]	12	2.47	17.29	8.27	0.75	8.27	0.75	2.00	0 <sup>2</sup>
Standardized							7.53	0.84	2.50	
Not Standardized							8.08	0.76	2.00	
Energy Balance – ME**	[MJ/d]	12	0.08	23.54	8.99	0.85	8.94	0.85	2.58	0
Standardized							8.42	0.89	3.05	
Not Standardized							9.06	0.84	2.48	
Energy Efficiency – NEL	[MJ/kg]	8	2.47	17.29	0.27	0.89	0.27	0.89	3.00	+ <sup>3</sup>
Standardized							0.28	0.90	3.10	
Not Standardized							0.29	0.88	2.93	
Energy Efficiency - ME	[MJ/kg]	8	7.54	1.48	0.42	0.91	0.42	0.91	3.26	+
Standardized							0.42	0.91	3.30	
Not Standardized							0.46	0.89	3.05	
Feed Efficiency	[ECM/kg]	10	1.54	0.27	0.09	0.90	0.08	0.89	3.03	0
Standardized							0.08	0.90	3.13	
Not Standardized							0.08	0.88	2.97	

<sup>1</sup>Use “-” means RPD between 0-2 a very poor model class with application as allowing to compare groups of cows and distinguish between high or low values.

<sup>2</sup>Use “0” means RPD between 2-3 a poor model class with application as rough screening.

<sup>3</sup>Use “+” means RPD between 3-5 a fair model class with application as screening.

\*NEL balance (GfE 2001); \*\* ME balance (Susenbeth, 2018)

well as non-standardised MIR spectral data from FOSS and Bentley FTIR analysers. With financial support from DLQ the weekly milking and MIR spectral data were combined with energy balance data to establish the MIR calibration equation of the two energy balances. Following the same Methodology and using the same optiKuh feeding experiment data source energy efficiency reference values and MIR spectra were combined within the eMissionCow project. The energy efficiency is defined here as the quotient of the energy intake and the amount of ECM per cow and day. Global models for energy efficiency NEL and ME and feed efficiency with a cross-validation trial from all research farms with around 1,511 animals were then created. In order to evaluate the effect of non-optimized feeding regimes on animal health, veterinary diagnosis and treatment data were linked from the LKV BW dairy database. This type of data is routinely recorded at LKV BW starting with the GMON project in 2011 for around 1,000 farms. Combined with milk recording data each milk sample from these farms can be associated with a health status.

The CH<sub>4</sub> calibration model was performed with 10-fold cross validation on a subset of 1,203 samples. This model is now also available to DLQ through the eMissionCow project as a new consortium member. The CH<sub>4</sub> model is expanded to include measurements on Simmental cows. This increased the variability of the data and the robustness of the equation (Table 1). With a RPD<sub>cv</sub> of 1.8 it can be used for distinction of high and low values. Nearly 26,000 energy balances on NEL basis and nearly 29,000 energy balances on ME basis of the 12 experimental farms have been used for the new equations. It can be emphasized that there is a difference of 0.5 in RPD (Table 1.) between standardized and non-standardized devices. But for a better variability and a better robustness of the models, we have combined the standardized and non-standardized spectra and this model will be used for validation in commercial herds. It has to be pointed out that for the non-standardized spectral data only a maximum of 1 % relative deviation between the official fat content provided by the laboratories and the fat content derived from the RobustMilk MIR equation was accepted.

## Results and discussions

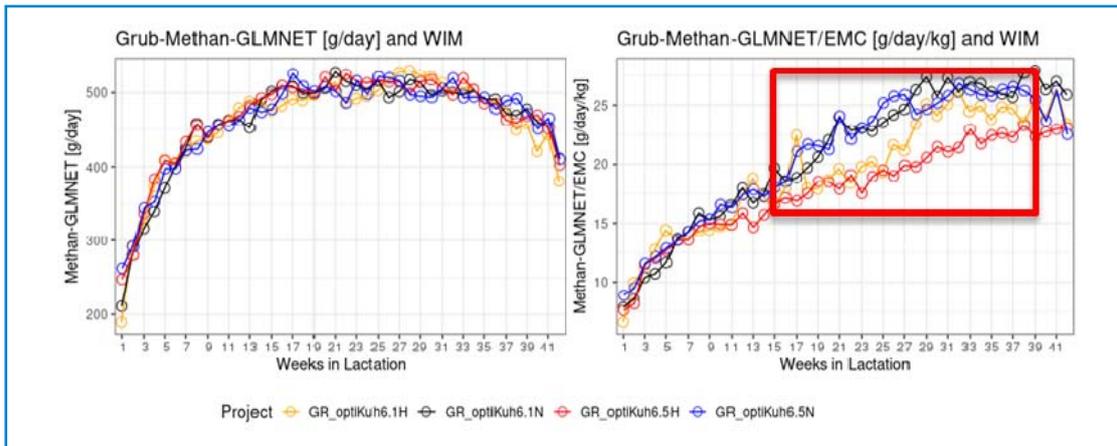


Figure 1. Absolute and relative (per kg ECM) calculated CH<sub>4</sub> emission over weeks in lactation in different feeding regimes

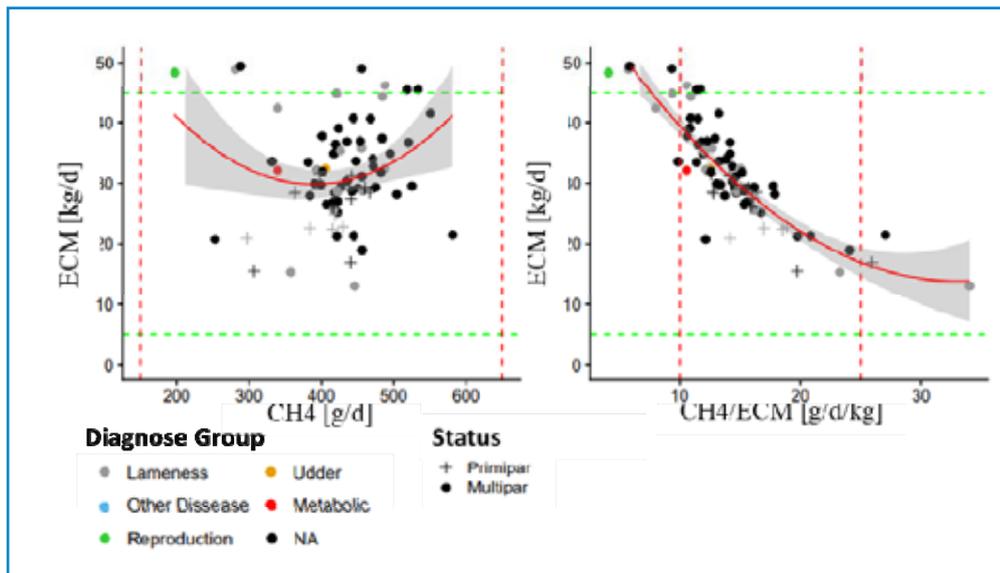


Figure 2. Absolute and relative CH<sub>4</sub> and ECM according detected health

Therefore, the equation quality is better and the RPD is higher as 2. In the case of the feed efficiency the RPD is between 3 and 5 (Table 1.), which means: the equation is satisfactory and can be used for fine grading. With the same data was modelled the feed efficiency in kg ECM/kg dry matter intake. Here the RPD is more then 3 (Table 1.). Figure 1. shows the predictions of the CH<sub>4</sub> equation with spectral data from the optiKuh project for each lactation week. It can be seen that the coloured data series are different. This is due to the four different feeding regimes, based on rations with 6.1 and 6.5 MJ NEL per kg forage DM and each with lower (N) or higher amounts of concentrates (H). The graph on the left shows the CH<sub>4</sub> emissions in grams per day, the graph on the right shows the CH<sub>4</sub> emissions relative to milk, in grams per kg ECM. The difference is much clearer visible in the relative emission. In the range from 100 to 270 lactation

days, the values of the different feeding regimes differ most clearly. The differences between the rations with lower or higher amounts of concentrates also appear very clearly here. It is very easy to see that feeding regimes with lower energy density of forage and low amounts of concentrates and a higher proportion of forage, have more CH<sub>4</sub> per day and kg of milk than feeding regimes with higher energy. In cooperation with eMissionCow and ReMissionDairy a report draft has been prepared for LKVBW test farms in order to evaluate the relevance and usability of the new predictions for herd management. The reports were designed for farmers and consultants: In the “Climate Status: CH<sub>4</sub>, feed efficiency and energy status report” can be seen the milk MIR parameters that reflect the energy status of the cow or herd. What is displayed: first two main groups of fatty acids: the de-novo group which consists of short and medium-chain fatty acids, mostly formed from the feed, and the preformed group that consist on long-chain fatty acids, mostly from the metabolization of body fat. Then there is energy balance: here is the NEL energy balance, feed efficiency, energy efficiency and energy consumption, and last CH<sub>4</sub> with absolute CH<sub>4</sub> in grams/day and relative CH<sub>4</sub> in grams/ECM kg. Also, in the report it can be seen a set of plots displaying the link between milk parameters and daily CH<sub>4</sub> emission and daily CH<sub>4</sub> emission per ECM kg (Figure 2).

In the Figure 2 it is shown the daily ECM kg over CH<sub>4</sub> emission. Compared to the left plot one can see that the milk yield has a stronger and clearly negative correlation with CH<sub>4</sub> emission per kg ECM showing that cows with higher ECM yields have a lower CH<sub>4</sub> emission per kg ECM. Figure 3. shows the link between energy efficiency and CH<sub>4</sub> emission. Again, it can be seen a better correlation with CH<sub>4</sub> emission per kg ECM. It is underlined that cows with a lower CH<sub>4</sub> emission per kg milk also have a better energy efficiency meaning less energy is used per kg ECM. The main message to be provided to the farmer and advisor is that the predictions should be kept well within borders for which reasonable thresholds have to be developed. Here is also shown higher degree regression curves with standard error borders. Furthermore, the veterinary diagnosis registered for the cows near the milk recording date were also plotted in different colours: e.g. the fertility or reproduction problems were plotted in green, the udder diseases are in orange, the metabolic distortion are red, etc. It can be seen that cows with health problems appear rather at the borders and it can

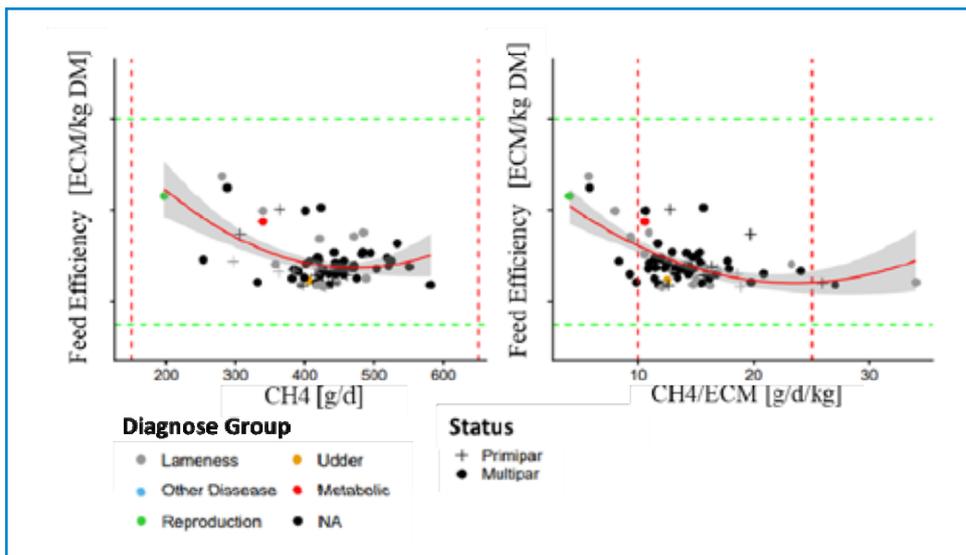


Figure 3. Absolute and relative CH<sub>4</sub> and feed efficiency according detected health.

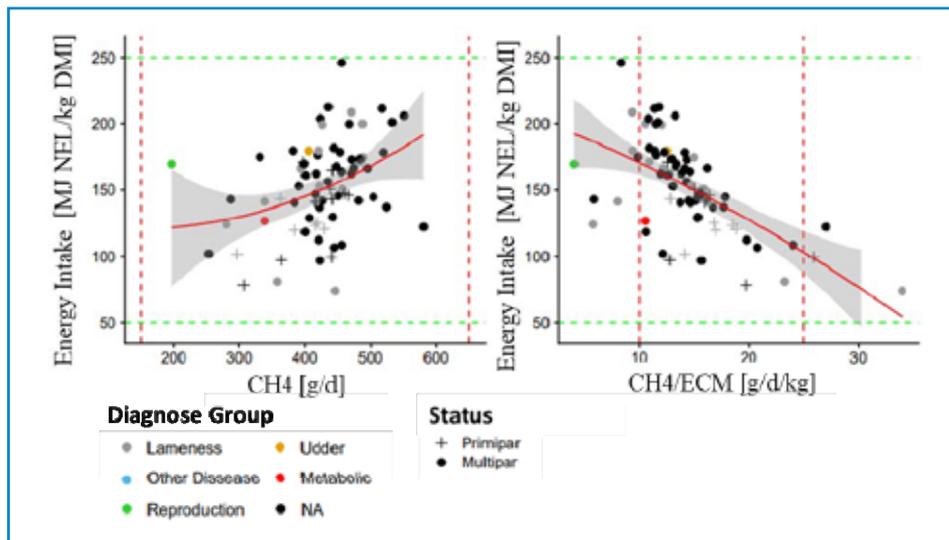


Figure 4. Absolute and relative CH<sub>4</sub> and energy intake from feed dry matter according detected health.

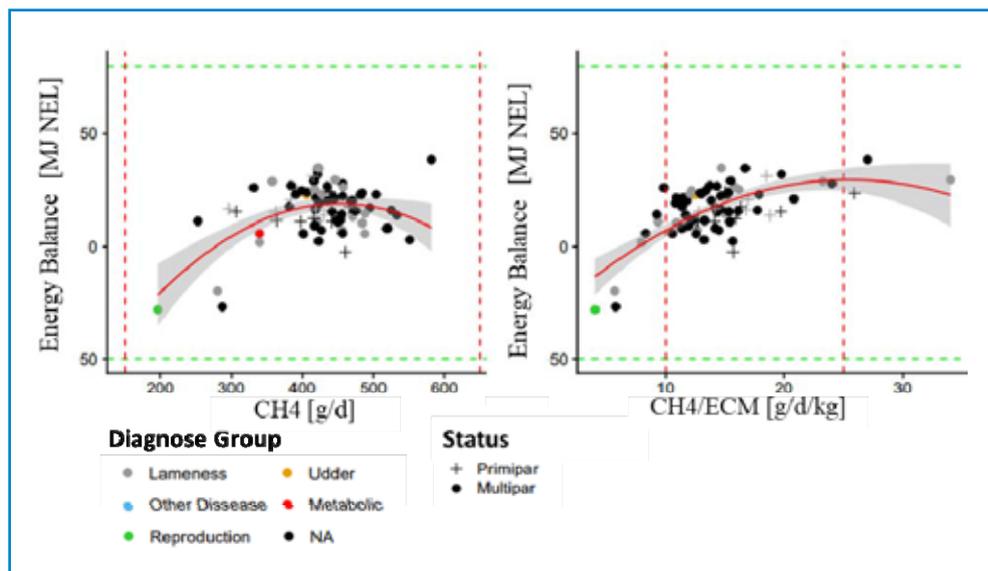


Figure 5. Absolute and relative CH<sub>4</sub> and energy balance NEL [MJ] according detected health.

tell whether the herd management goes in the right direction with regard to feeding efficiency management. This link has to be elaborated further. In the Figure 4, it can be seen the plots over absolute and relative CH<sub>4</sub> emission per day and energy intake from feed dry matter.

The plots show the energy intake per kg dry matter and the higher energy concentration in the feeding is linked with the lower CH<sub>4</sub> emission per kg ECM. The plots at the bottom show the link to energy balance again on the right side with a stronger and clearly positive correlation to CH<sub>4</sub> emission prediction per kg milk. Though negative energy balance (Fig. 5.) is linked to lower CH<sub>4</sub> emission this should be avoided

because extreme negative energy balance comes mostly with reduced feed intake. Extreme body fat mobilisation followed by health problems. This is indicated by the red versus the green bounded area. There is a correlation between diagnosis frequency in that day and the relative CH<sub>4</sub> emission per day but also to other milk component concentrations. The diagnoses observed on that testday were mainly lameness but also udder and metabolic or reproduction diseases. It has been determined that the diagnosis frequencies had positive correlation to fat and almost all fatty acids (FA) groups but not with De Novo FA. In the same time, it can be seen that CH<sub>4</sub> had a positive correlation to energy intake from feed dry matter as well as ECM and MY and also to DeNovo FA. Negative Pearson correlation to CH<sub>4</sub> was found for fat, protein, feeding efficiency, as well as to lactoferrin, natrium and magnesium, also to blood components: adiponectin, insulin, IGF, Preform FA and C17 and total C18.

The Clima Status report for farmers and advisors emphasises energy related MIR predictions. The report is designed to show the link between these predictors in order to help them understand the effects and improve the feeding management. The equations for the determination of CH<sub>4</sub>, the energy balances (NEL, ME) and the feed efficiency (NEL, ME) are statistically highly meaningful and allow a comparison between groups of cows. The future developments that could be carried out at all levels are for example reports and applications for feed and energy efficiency, because MIR-based applications for reducing emissions and optimizing feeding are good tools for reducing emissions and optimizing feeding. Another future development could be the calculation of new breeding values based on correlations between genetics and MIR indicators.

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## Conclusions

## Acknowledgments

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