METHAGENE

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Ranking of cows for methane yield is determined by the host animal

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To study the host animal's control on methane (CH_4) emission, 100 cows were ranked according to CH_4/DMI emission using respiratory chambers. Five low- and five high-emitters were selected and subjected to different diets in three 35-d periods. High grass, low grass and red clover based-diets differing in forage to concentrate ratio were fed. Intake of DM tended to be lower for the low- compared with high-emitter cows. Both groups had similar milk yield. Digestibility of OM and NDF was lower for low- compared with high-emitters. Low-emitters tended ($P = 0.08$) to have lower CH_4/DMI than high-emitters (20.9 vs 22.5 g/kg). Rumen DM fill, measured by rumen evacuation, did not differ between diets or groups. Rumen molar Ac:Pr tended to be lower for low- than high-emitters (3.09 vs 3.39; $P = 0.09$). The results suggest that CH_4/DMI ranking of cows is determined by the host animal irrespective of the diet fed.

Nuclear Magnetic Resonance to identify rumen metabolites as proxies for methane emissions

Riccardo Bica, Dusan Uhrin, Javier Palarea-Albaladejo, Richard Dewhurst

This study investigated the use of nuclear magnetic resonance (NMR) to measure rumen metabolites and explore variation related to diet type and methane emissions. The ultimate objective of this work is to develop a simple proxy for methane emissions that is robust across diet types and methane pathways (hydrogenotrophic and methylotrophic). We had 224 rumen fluid samples obtained from 4 different experiments (12 different diets) conducted between 2013 and 2017. Methane measurements were obtained over 2-day periods in respiration chambers. Rumen samples were obtained using a naso-gastric tube at the end of the chamber measurement period and (for 14 samples) by buccal swabbing. Samples were analysed for volatile fatty acids by HPLC and were also analysed using 1H NMR spectroscopy at 600 MHz.

Methane emissions ranged from 9.7 to 31.2 g/kg DM intake (mean = 19.60; s.d. = 4.70). Acetate/propionate ratio (HPLC) ranged from 0.92 to 7.85 mol/mol (mean = 3.42; s.d. = 1.29). There was good agreement between acetate/propionate ratios measuring using HPLC and NMR, with both having a good ability to predict methane emissions (g/kg DM intake). A principal component analysis confirmed that concentrate diets were more associated with propionate whereas the mixed and forage diets were characterised by higher levels of acetate and butyrate.

A predictive model of CH_4 emissions using a partial least square approach provided a cross-validated R^2 of 0.59 when using only metabolites as predictors, and R^2 of 0.72 when including more variables (DM intake, animal weight, diet ME concentration). Other less prominent metabolites were evident in the NMR spectra and these explained a small, but significant, proportion of the variation. Further work is exploring the nature and origin of these peaks. 1H NMR spectra from buccal swabs had very different profiles to those of rumen fluid samples and there appears to be little prospect of using this as a simplified way to obtain samples for metabolite analysis.

Breeding goals and the use of selection index theory to reduce methane emissions

I.S. Breider, Y. de Haas, O. González-Recio, P.C. Garnsworthy, E. Wall

A potential route to reduce methane (CH_4) emissions from dairy cattle is to include CH_4 into breeding goals. To do so, we need to understand the genetic control of CH_4 and its relationships with production and fitness traits. As estimates of such genetic parameters become known, selection index theory can explore how to incorporate CH_4 into breeding goals. This study analyses three scenarios adding CH_4 to national breeding goals (UK, Spain & Netherlands) with; 1) no restriction on CH_4 emissions (i.e., current breeding goal); 2) no genetic change in CH_4, and 3) an economic value assigned to CH_4. Preliminary results show that current breeding goals lead to increased CH_4 emissions. Restricting CH_4 genetic change to zero decreases the economic gain with 3 to 14%. Assigning an economic value to CH_4 still increases CH_4, but 11 to 15% less than without restrictions, whilst the overall economic gain decreases with 1 to 4%.

Comparison of the GreenFeed technique with the SF6 technique in methane measurement on grazing cows

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In order to develop sustainable mitigation strategies, accurate and efficient measurement techniques for enteric CH_4 production are necessary. The sulphur hexafluoride (SF_6) tracer technique is an established and labour intensive method (Johnson et al., 1994). An alternative method may be the GreenFeed (GF) system (C-Lock Technology Inc., Rapid City, SD, Hammond et al., 2015). It has been shown that ruminal fermentation traits are linked to CH_4 emission (Moss et al. 2000). The objective of the present study was to compare and to evaluate the two measurement methods, SF_6 and GF, with grazing cows. The experiment was carried out with 20 multiparous Holstein Friesian dairy cows. The cows grazed as a single herd. The SF_6 technique and the GF system were applied in parallel over a 6-d period and the GF measurements were further conducted to complete a period of 11 d. Within this 6-d period, rumen fluid samples were taken twice from each cow and the individual herbage DM intake was estimated (Mayes et al., 1986). CH_4 emission data obtained with the GF were averaged from day 1 to day 6 (P1), to day 8 (P2) and to day 11 (P3) and compared with daily CH_4 emission data determined with the SF_6 technique, which were averaged across the 6-d sampling period. For the statistical analysis these averaged CH_4 emission data of P1, P2 and P3 were used. Differences between the data obtained with the SF_6 technique and the GF technique were analysed using the Mixed Model Procedure of SYSTAT 13. Lin's concordance correlation coefficient (CCC) was calculated using the R package 'epiR'. Pearson correlation coefficients between CH_4 yield, using the GF (P1, P2, P3) and the SF_6 technique, and each measured rumen fluid variable were assessed (SYSTAT 13). In general, the values derived from the SF_6 technique were higher than those obtained from the GF ($P < 0.01$). Regarding CH_4 production, the agreement (CCC) between the two methods applied was weak in each period (0.14 - 0.17). Using GF, there was a significant and moderate correlation between CH_4 production and dry matter intake (DMI) in P1 ($r = 0.51$, $P = 0.03$). In P1, it was observed that the molar percentage of acetate was correlated positively ($r = 0.54$, $P = 0.02$) with CH_4 yield measured by the GF. There was a negative correlation with the molar percentage of propionate ($r = - 0.60$, $P < 0.01$). When the duration of the GF measurements was extended from 6 to 11 days the correlations increased slightly. We could not find any significant relationships between the SF6 technique and GF CH_4 data as well as the measured characteristics. High and variable background concentrations of SF_6 are likely the main reason. However, in order to obtain more reliable data of enteric CH_4 for a given situation, in view of slightly increasing correlation due to the number of visits and their potentially more even distribution over 24 h, a measurement period over more than 6 days is recommended for studies with GF.

Extending method comparison theory to methane missions in animal science

G.F. Difford, Y. de Haas, J. Lassen and P. Løvendahl

With technological advances, new instrumentation and methods are continuously being developed and so too are statistical methods for comparing them. In the case where the reference method is time consuming, costly or limited in its application, it is of interest to the researcher to evaluate to what extent a new method agrees with the existing reference method. This involves decomposing the measurements into different sources of (dis)agreement, e.g. linear association, accuracy and precision. When extending the application to different fields of animal science, researches may place higher emphasis on differing components of agreement. For instance acute nutritional trials with limited numbers of replications may place higher emphasis on accuracy and precision whilst large scale genetic evaluations may forego precision in order to obtain records on vast numbers of animals. Here we discuss existing methods of methane emissions in ruminants and discuss possible pitfalls in their analysis

Greenhouse gases emissions in sheep, relationship with liveweight, feed intake and residual feed intake: first results

D. Francois and D. Marcon

A batch of 56 young Romane breed rams have been GHG (CH_4 and CO_2) phenotyped under 2 Sheep GreenFeed units during May and June 2017 months. They were previously phenotyped for feed intake and residual feed intake under a low energy concentrates diet. Then there were fed a total mixed ration and during these two months they emitted 52 ą 11 g/day CH_4 and 1568 ą 194 g/day CO_2. Phenotypic correlation were low with liveweight (0.10 for emitted CH_4 and 0.16 for emitted CO_2), very low with daily feed intake (- 0.01 for emitted CH_4 and 0.01 for emitted CO_2), and negative for residual feed intake (-0.18 for emitted CH_4 and -0.27 for emitted CO_2) suggesting that the most feed efficient rams are not those who emit less.

Multiple comparisons of methods for measuring methane emissions by dairy cows

Phil Garnsworthy, Gareth Difford, Matt Bell

Many methods are available for measuring methane emissions by individual animals, each with its own particular features that can influence results. Little is known of the equivalence or lack thereof between methods. In WG2 we collated a database of results from 23 studies comparing at least two methods applied to the same individual cows (n= 8 - 44). Methods included chambers, SF_6, LMD and sniffers (Guardian, Gasmet and Greenfeed). Concordance coefficients comparing chambers with alternative methods ranged from 0.30 to 0.79; coefficients comparing alternative methods ranged from 0.14 to 0.79. For each dataset, a model was fitted to predict methane from live weight, milk yield and lactation week. Models were validated against an independent chamber dataset, and mean prediction errors ranged from 15 to 45% (mean 25%) of observed methane. We conclude that although different methods measure different aspects of methane emissions, there is a significant underlying agreement between methods.

Effect of selected medicinal herbs from central Europe on methane production from in vitro ruminal fermentations

Lubomira Gresakova, Zora Varadyova, Svetlana Kisidayova

The objective of this *in vitro* ruminal fermentation study was to compare methane production of twenty selected traditional medicinal herbs. The selection was based on their antiparasitic, anti-inflammatory, carminative, antioxidant and immunological properties. The following dry medicinal herbs were used: roots of dandelion, calamus, marshmallow, butterbur and elecampane; flowers of marigold, mallow and chamomile; leaves of ribwort plantain and rosemary; seeds of fennel and whole overground herbs of St. John's-wort, yarrow, nettle, chicory, goldenrod, fumitory, hyssop, lemon balm and wormwood. Qualitative phytochemical screening of herbs revealed the presence of medically active compounds (tannins, flavonoids and glycosides). Methane production varied among the herb treatments ($P < 0.001$), however, pH and SCFA in fermentations was not significantly influenced. The results point to the promising beneficial effects of some medicinal herbs without adverse effect on rumen fermentation.

Relations between imputation and population parameters on the prediction accuracy of methane production

Jody Leigh Edmunds, Filippo Biscarini and Pablo Orozco terWengel

As a result of high-throughput genotyping and commercialisation of SNP chip arrays, imputation, a statistical method for the prediction of missing data, has become a useful tool in livestock genomics. A workflow was successfully developed utilising various statistical packages (e.g. R, PLINK, Zanardi and Beagle) to estimate the accuracy imputation of missing genotypes in heterogeneous livestock populations, focusing on the impact on the prediction accuracy. The data comprised of both genotypic and phenotypic datasets and was subject to various scenarios including, population size and structure and percentage of missing data, all of which had an effect on imputation accuracy. Specific scenarios were selected for analysis with phenotypic data on rumen methane emissions, identifying relations between imputation and population parameters on the prediction accuracy of methane production. Conclusions drawn strengthen predictions of anthropogenic greenhouse gas emissions thus, assisting their mitigation and sustainable livestock breeding.

Breeding goals to reduce methane in beef cattle

J. Lopez-Paredes, O. González-Recio, and R. Alenda

Reduction of green-house gas emissions in agriculture and livestock is gaining attention and beef cattle population should consider it in the breeding goal. In this study, methane emissions (CH_4) were estimated in Blonde d´Aquitaine population using gross energy requirements approximation expressed in kilograms of CH_4 per slaughtered calf per year. Emissions were calculated for the suckler cow and the calf in feedlot process representing 69 and 31% of CH_4, respectively. Genetic correlation were estimated for cow CH_4 (h^2=0.18) and calf CH_4 (h^2=0.42) and eleven traits. Economic values for cow and calf CH_4 were estimated in three scenario: 1) non-considering CH_4. 2) Inclusion a tax per CH_4 kg. 3) Establishing a quota of CH_4 per farm and year. Three selection indexes were developed including CH_4 of cow and calf and age at first calving, calving interval, calving ease, weaning weight, carcass weight gain, carcass conformation score, culled cow carcass weight, and culled cow carcass conformation score for the three scenarios. Economic weights for CH_4 were higher under the Scenario 2. Functional traits (calving interval, age at first calving, calving ease) enhanced their importance in the Scenario 2 and whereas production traits (growth) showed to be more relevant in the Scenario 3, in comparison to Scenario 1.

Heritability of methane emission measured on commercial dairy farms

Marcin Pszczola

We estimated the heritability of the methane emissions from dairy cows using the Fourier Transform Infrared Spectroscopy during milking in an automated milking system by implementing the random regression method. The methane measurements were taken on 485 Polish Holstein-Friesian cows at two commercial farms located in western Poland. The overall daily estimated methane emission was 279 g/d. Average heritability was 0.27 (average SE 0.09). We have shown that the estimated methane emission is a heritable trait and that the heritability level changes over the course of lactation. Genetic correlations between particular between the neighboring DIM was high and decreased with the increasing distance between DIM, reaching zero at the biggest distances. The observed changes and low genetic correlations between distant DIM suggest that it may be important to consider the period, in which methane phenotypes are collected.

Individual methane emission measured with GreenFeed systems

Gilles Renand

Individual methane emission was measured with GreenFeed systems during 8 to 12 weeks on three groups of growing Charolais cattle while they were tested for feed intake and growth. Two groups of 2-year old heifers were ad libitum fed a roughage diet: 250 heifers received herb silage and 74 heifers natural meadow hay complemented with 1 kg rapeseed meal. One group of 81 one year old young bulls received a pelleted diet mainly composed of dehydrated alfalfa hay, wheat middling, dehydrated beat pulp and wheat bran. Mean performance were: 534, 508, 498 kg mean live weight (MLW); 8.75, 7.89 9.29 kg/d dry matter intake (DMI); 932, 360, 1765 g/d daily gain (ADG) and 206, 206, 154 g/d daily methane production (MPR). Feed Efficiency (FEff) and Methane Yield (MPY = MPR/DMI) were respectively 109, 46, 192 g/kg (ADG/DMI) and 24, 26, 17 g/kg (MPR/DMI). Correlations between methane emission and feed efficiency traits were calculated among each experimental populations.

Genetic analysis of CH_4 concentrations in the breath of dairy cows measured by Laser Methane Detector

Diana Sorg, Sarah Mühlbach, Frank Rosner, Jelena Kecman, Hermann H. Swalve

Methane profiles were taken with the portable Laser Methane Detector (LMD; Tokyo Gas Engineering Solutions, Tokyo, Japan) in the breath of 622 dairy cows on three commercial farms in Germany. Each profile was reduced to a point measurement by separating the respiratory from the eructation values. Heritability for different CH_4 phenotypes was 0.05-0.23, depending on the statistic model. Genetic correlation between these phenotypes was 0.71-0.95. A high correlation between the phenotypes was to be expected since they were all derived from the same profile and contained overlapping subsets of data points. It seems possible to rank cows according to their breath CH_4 concentration with the LMD, despite the lower accuracy compared to other methods. One advantage of the LMD is its mobility and adaptability under commercial conditions, which makes this method an interesting candidate for further research on new phenotypes for genetic selection on lower methane emissions.

Co-recording with the Laser Methane Detector: method comparisons towards joint genetic evaluations

Diana Sorg

Short-term breath concentration methods for CH_4 are cheap and high throughput, with the trade-off of a loss of precision and accuracy as compared to the gold standard respiration chamber. The LMD was used to co-record CH_4 on dairy cattle populations in Germany, Denmark and Poland together with the GreenFeed, NDIR and FTIR spectroscopy methods, respectively. LMD breath profiles had a repeated measures correlation (r_p) with the GreenFeed flux measures of CH_4 in g/d of 0.66 (±0.23). The LMD measures had a good r_p (0.60 and 0.57) with CH_4 g/d from NDIR and FTIR spectroscopy techniques. Since the r_p is the upper threshold for genetic correlations, in principal the LMD can be used as a flexible tool to record CH_4 as well as improve genetic connectedness between research herds employing other methods.

Agreement between methane measurements in dairy cows obtained from two non-invasive infrared methods

Mateusz Sypniewski, Tomasz Strabel, Adam Cieslak, Małgorzata Szumacher-Strabel, Marcin Pszczola

We compared methane concentrations measured on Polish-Holstein Friesian cows kept in commercial conditions using Fourier transform infra-red spectroscopy (FTIR) and nondispersive infrared (NDIR) installed in automatic milking system. Measurements were carried out for 5 days. Differences between daily means of gas concentration were not significant. Repeatability of gas concentrations averaged per milking using FTIR was higher than of NDIR. Coefficient of individual agreement for concentrations averaged per milking equalled 0.87 for CH_4 and 0.78 for CO_2, concordance correlation coefficient were 0.4 for both gases, and repeatability correlation was 0.85 for CH_4 and 0.84 for CO_2. Pearson's correlation for uncorrected phenotypes was 0.75 for both CH_4 and CO_2.

Estimation of feed conversion efficiency based on CH_4-to-CO_2 ratio revisited when 3-NOP is used as additive

Dorien Van Wesemael, Nico Peiren, Leen Vandaele, Stéphane Duval, Sam De Campeneere

When the DMI is not known, it is often suggested to use the CH_4-to-CO_2 ratio to identify the diets or the cows which have most efficient feed conversion or which produce the least CH_4. We tested if this is still correct with a feed additive like 3-NOP. In an on-farm trial with thirty highly producing Holstein Friesian cows the average CH_4-to-CO_2 ratio (g/g) for the control group (n=10) was 0.041±0.0003 and 0.030±0.0005 for the treatment group (n=20) who received 1.6 g 3-NOP/day. It was already shown that the excess of H_2 is not completely metabolized in an alternative pathway, but a part is exhaled. It is possible that part of the available CO_2 has the same fate, which would further decrease the CH_4-to-CO_2 ratio while this is no reflection of a higher feed conversion efficiency.

Therefore, caution has to be taken when data on CH_4-to-CO_2 of different trials are combined for meta-analysis of feed conversion efficiency.

Using milk Fourier-transform infrared spectra and gas chromatography-based milk fatty acid profiles to predict methane emission of dairy cows

Sanne van Gastelen, H. Mollenhorst, E.C. Antunes-Fernandes, K.A. Hettinga, G.G. van Burgsteden, J. Dijkstra, and J.L.W. Rademaker

We compared the prediction potential of gas chromatography-based milk fatty acids (MFA) and milk Fourier-transform infrared spectroscopy (FTIR) for methane (CH_4) emissions of dairy cows. Data from 9 experiments with lactating Holstein-Friesian cows with a total of 30 dietary treatments and 218 observations were used. Methane emissions were measured in climate respiration chambers. Multivariate MFA-based and FTIR-based CH_4 prediction models were developed and, subsequently, evaluated with the concordance correlation coefficient (CCC) analysis. The MFA-based CH_4 prediction models estimated CH_4 production (g/d), yield (g/kg dry matter intake), and intensity (g/kg fat- and protein-corrected milk) with a CCC of 0.72, 0.59, and 0.77, respectively. The FTIR-based CH_4 prediction models estimated CH_4 production, yield, and intensity with a CCC of 0.52, 0.40, and 0.72, respectively. These results indicate that for all CH_4 emission units, but particularly for CH_4 production and yield, the MFA-based prediction models described a greater part of the observed variation in CH_4 emission than FTIR-based prediction models.

Breeding programmes for reducing methane

Eileen Wall

This COST action has gathered key information to help inform the design of breeding programmes that incorporate methane emissions. This talk will highlight how this information could fit into breeding programmes and will quantify the importance of methane emissions (or indicators) relative to other performance traits in breeding goals (e.g., milk or meat yield, fertility), and indicate the benefit for producers when methane emissions is included in national breeding goals within EU. This will allow us to develop recommendations and suggest methods for the inclusion of methane into breeding goals.

Genetic correlations between methane production and functional and body type traits in Danish Holstein cows

Larissa Zetouni

Our aim was to investigate the genetic correlations between methane (CH_4) production and body conformation, fertility and health traits in dairy cows. Data were collected from ten commercial Holstein herds in Denmark, including 5,758 cows with records for body conformation traits; 7,390 for fertility traits; 7, 439 for health traits; and 1,397 with individual CH_4 measurements. CH_4 production was measured during milking in Automatic Milking Systems, using a sniffer approach. Correlations between CH_4 and a number of different traits were estimated. These traits were: interval between calving and first insemination, interval between first and last insemination, number of inseminations, udder diseases, other diseases, height, body depth, chest width, dairy character, top line and body condition score. Bivariate linear models were used to estimate the genetic parameters within and between CH_4 and the other traits. In general, the genetic correlations between CH_4 and the traits investigated were low. The heritability of CH_4 was 0.25, and it ranged from 0.02 to 0.07 for fertility and health traits, and from 0.17 to 0.74 for body conformation traits. Further investigations with a bigger dataset should be performed in order to establish more accurate results on how CH_4 relates with fertility, health and body conformation traits in dairy cattle, in order to take these correlations into account in a future breeding goal.