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THE RELATIONSHIP BETWEEN DIETARY PROXIMATE ANALYSIS AND GREENHOUSE GAS EMISSIONS DETERMINED USING IN VITRO METHODOLOGY

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Abstract

The enteric production of greenhouse gas (GHG) is determined in part from the fermentation of dietary carbohydrates. The current work looked at 147 rations from lactating dairy herds across New Zealand spanning a period of 12 months which were incubated in a closed *in vitro* gas production system to determine the relationships between proximate analysis measurements and methane production. Rations were collected from lactating commercial herds and included a range from 0% pasture through to 100% pasture in the diet.

In a ruminant diet additivity of the proximate analysis of raw materials is assumed when designing rations, due to the ease of formulation. However, in the dynamic fermentation environment within the rumen, additivity of dietary components is not expected to be linear. For this reason, correlations of methane with fermentation characteristics were determined.

The current study found a weak relationship between methane production from the *in vitro* system and proximate analysis. The correlation coefficients were positive for protein, starch, and total non-fibre carbohydrates, being 0.02, 0.22 and 0.29 respectively, and negative for ash, acid detergent fibre and neutral detergent fibre, being -0.07, -0.29 and -0.48 respectively.

The coefficients were stronger for parameters determined during fermentation. Correlation coefficients were positive for apparent dry matter digestibility (ADMD) and true dry matter digestibility (determined as ADMD less microbial biomass produced) being 0.78 and 0.66 respectively.

Based on this study is concluded that *in vitro* fermentation parameters provide a better predictor of methane production than proximate analysis.

Introduction

There is currently a very large focus on greenhouse gas (GHG) from agriculture in New Zealand. Agriculture is said to contribute 48% of New Zealand's GHG profile, and a large part of this is from enteric methane production from the rumen (Ministry for the Environment, 2020). Research has focussed on identifying and understanding any nutritional influences on methane production.

There is a suspected tension between urinary nitrogen excretion and methane production of feeds, referred to as "pollution swapping". In a pasture type system, urinary nitrogen excretion is a function of dietary protein (Clark et al., 2010) and dietary protein in New Zealand is commonly in excess of animal requirements (Wilkinson and Waldron, 2017). An implication that then follows is that formulating for a lower protein diet will reduce urinary N excretion.

As diets are formulated for lower N content, other chemical parameters will necessarily increase. Because enteric methane is in part related to dietary carbohydrate fractions, it is conceivable that increasing carbohydrate fractions may result in higher methane production.

This study looked at a large data set of gas production data, determined by *in vitro* fermentation, from a broad range of complete rations taken from commercial New Zealand dairy farms. The data was examined *a posteriori* to test whether lower protein diets produced more methane. Furthermore, it examines some key proximate values for any correlation to methane production.

Methodology

Lactating dairy herd rations (n=147) were harvested from across New Zealand from commercial dairy farms during a 12-month period (June 2018 – May 2019).

The rations were subjected to a 48-hour *in vitro* fermentation in a closed gas production system. To account for weekly variation in door fluid, a known standard ration with known fermentation characteristics was used as part of each weekly fermentation to provide a weekly correction factor that was applied to metrics determined.

Donor rumen fluid was taken from a lactating dairy cow fed a pasture-based ration. Dried samples of feeds (0.5 g) ground to a 2 mm size were incubated at 39ºC using a rumen-buffered inoculum for 48h (Mould et al, 2005). Rumen fluid to buffer ratio was 20:80.

During the incubation period, gas production was measured continuously using an automated pressure transducer system (IFM, Alltech).

VFA concentrations were measured by GC (Erwin et al., 1961) on samples taken at 48h of incubation. The stoichiometry of (Wolin, 1960) was used to estimate methane production based on VFA production.

Selected proximate analysis values and selected fermentation endpoints (listed in table 1) were examined for their correlation with calculated methane production.

Results

	Average	Minimum	Maximum
Crude protein (%DM)	21.86	14.37	30.85
Starch (%DM)	7.55	0.01	31.53
Total non-fibre carbohydrates (%DM)	20.09	1.77	42.75
Ash (%DM)	9.62	4.73	42.75
NDF (%DM)	20.01	1.77	42.78
ADF (%DM)	26.5	16.56	35.66
ADMD (%)	65	45	78
TDMD (%)	79	61	89

Table1. Average, minimum and maximum values of selected proximate analysis results and selected fermentation results.

There was no correlation evident between the dietary crude protein content and the amount of methane produced *in vitro* (r=0.02, not significant) (Figure 1)

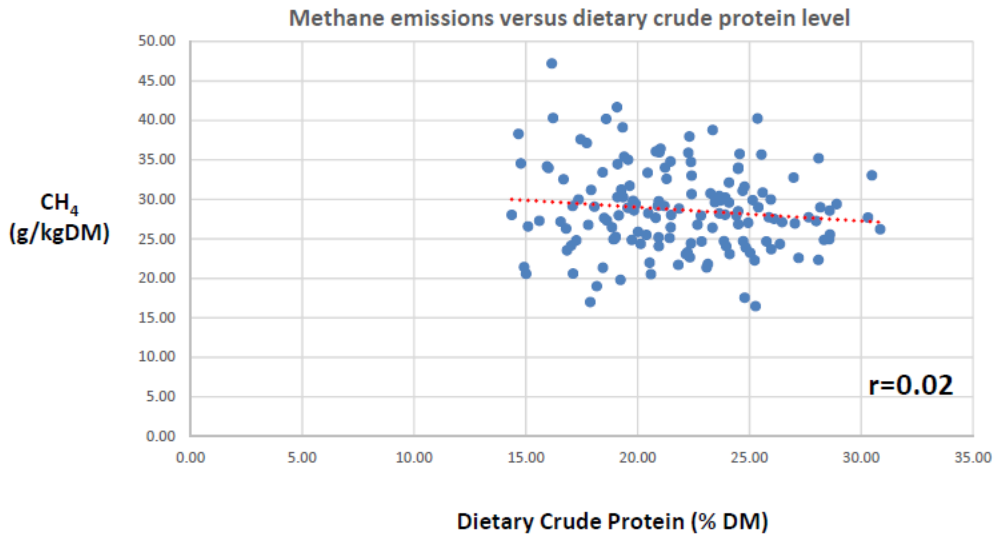

Figure 1: Methane emissions (g/kgDM) versus dietary crude protein (%DM)

The correlation coefficients for starch, and total non-fibre carbohydrates were 0.22 (p<0.01) and 0.29 (p<0.01) respectively, and negative for ash, acid detergent fibre and neutral detergent fibre, being -0.07 (NS), -0.29 (p<0.01) and -0.48(p<0.01) respectively.

The correlation coefficients were stronger for parameters determined during fermentation. Correlation coefficients were positive for apparent dry matter digestibility (ADMD) and true dry matter digestibility (determined as ADMD less microbial biomass produced) being 0.78 (P<0.01) and 0.66 (p<0.01) respectively.

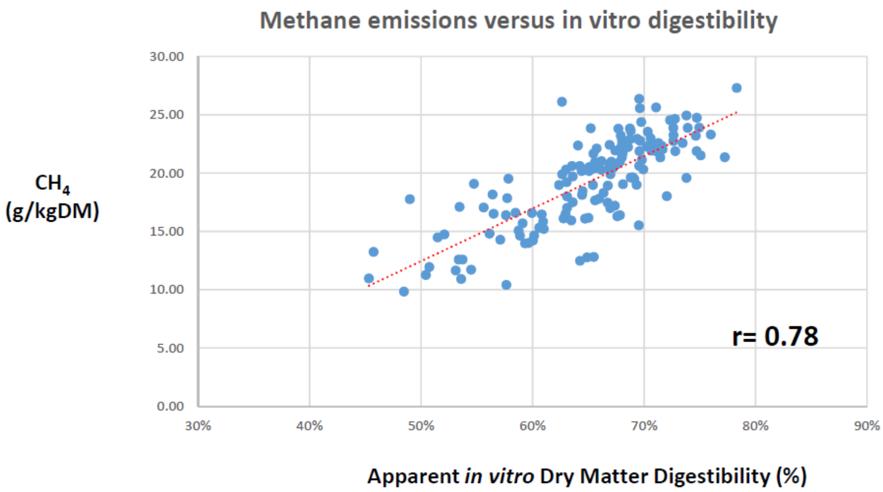

Figure 2: Methane emissions (g/kgDM) versus apparent in vitro digestibility (%)

Discussion

It has been previously proposed that lowering the dietary protein levels and subsequently lowering the urinary nitrogen excretion can lead to a reduction in nitrate leaching however it also may raise enteric methane production (Gregorini et al., 2010, Gerber et al., 2013). Results from the current study show that there is only a very weak correlation between dietary crude protein and other individual proximate analysis values and methane production. This is likely because the rumen environment is a dynamic place and interactions between different raw materials are not linear. The methane production will be contingent on which raw materials are introduced into the diet with reformulation. For example, high neutral detergent fibre (NDF) materials reduce fermentation overall, and the subsequent gas production and raw material combinations that result in higher propionate production result in less methane overall.

While proximate analysis of raw materials can be summed additively, the effect in a rumen is not considered to be additive. A recent comprehensive study on the prediction of enteric methane production using the dairy CH_4 database from GLOBAL NETWORK project concluded that information on dry matter intake (DMI) is a key factor to predict enteric methane production (Niu et al., 2018). A strong correlation between apparent dry matter digestibility and methane production as found in the current study is in line with these previous findings. It is proposed that fermentation parameters are expected to be better predictors of methane production in complete rations.

However further work is necessary to determine the effect of pasture in the final ration on dry matter digestibility and methane production.

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