

BIOPARAMETRICS LTD

Maize Link Project

Gas Production

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BACKGROUND

Bioparametrics Ltd. provides commercial evaluation of forages and feeds using in-vitro gas production techniques IVGPT developed at Edinburgh University over the past twenty years

There is a recent upsurge in interest in USA and Canada reported in Feedstuffs Dec (2010) with the realization that rumen management requires measurements on the rates of fermentation of carbohydrate and protein fractions in the rumen. This makes it possible to control rumen pH flux by dietary means with digestion kinetics measured in minutes rather than hours using IVGPT.

There is an expectation that a better understanding of gas production data may reduce the need for in-vivo measurements requiring surgically modified animals. The Gas production technique used in this study used rumen liquor donated from euthanized and slaughtered sheep.

METHOD

Bioparametrics standard carbohydrate fermentation SOP (2002) has evolved to include a protein evaluation system to improve the accuracy of measurement of carbohydrate fermentation Palmer (2006).

Five syringes are used for each sample. Carbohydrate fermentation is done in duplicate and protein fermentation is done in triplicate with readings taken over 72 hours.

1. The cumulative CHO gas production from fermentation is the net gas remaining after corrections are applied to the observed gas volumes using the standardization procedures set out in Bioparametric SOP (2004). Sample blanks (3 for nitrogen and 3 for carbohydrate) are fitted with a two pool model. The first pool is a measure of fermentable material; the second is the measure of fermentable microbial matter. The first pool is subtracted from gas volumes.
2. Gas volumes are adjusted so that standard A sample parameters are the same from every gas run.
3. Protein gas volumes and Carbohydrate gas volumes (net of fermented protein gas volumes) are fitted to estimate fermentation rates in both quickly and slowly fermenting pools plus lag times for slowly fermenting protein, starch and NDF pools.

Stoichiometry is the key to interpreting gas data this requires coefficients expressing release of ml of gas from fermenting mg of substrate dry matter. Values were found for rates, lag times and volumes by fermenting pure substrates published by Jessop and Herrero. These values correspond with results from the literature on IVGP.

RESULTS

Appendix 1 Tables 1-4

Standard output from commercial evaluation reports submitted in PDF format is tabulated here. This shows the pool sizes rates and lag times derived from stoichiometric fitting of gas data. Proxy measures from NIR have been substituted for small items e.g. sugar and volatiles not in the Eurofins data.

Wet Chemistry is used to determine pool sizes but it is the lag time and rate of fermentation which contributes most to the energy release during the residence time in the gut. The single variable defined as fermentability of carbohydrate explains most of the variability in ME.

Appendix 2

Dry Matter (carbohydrate + protein) net gas volumes at hours and protein gas volumes at hours are tabulated here.

Appendix 3

Sparse Data equivalent to Dacron bag measures is tabulated here for direct estimation of Orskov and Macdonald parameters.

DISCUSSION

Appendix 3 shows that direct estimates of in-vitro **a**, **b**, **c** values from gas data do not match in-vivo **a**, **b**, **c** values. Gas production alone can reveal what is happening during the first six hours following ingestion of a meal. The **a** values in this study are roughly one tenth of the **b** values.

The in-vivo measures are closer to parity between **a** and **b** values with **a** roughly 10 percentage points less in **a** than **b**.

	in-vitro				in -vivo			
	aDM	bDM	cDM	a+bDM	Dma	Dmb	Dmc	Dma+b
min	5.10	55.71	0.039	61.54428	25.30	35.60	0.009	76.3
max	9.10	86.75	0.08	94.46	54.10	65.00	0.04	100.10
mean	7.10	70.46	0.06	77.56	41.63	47.50	0.02	89.13

An alternative model is needed to predict **a**, **b**, **c** rate of loss of the **b** fraction in-vivo and this can be derived from appendix 1 data.

Looking at in-vitro DM parameters **aDM** values range from (5.1-9.1) these numbers are very small by comparison to in-vivo Dma (25.3-54.10). This is because the **aDM** value at time zero consists of ash, oil, and un-fermentable materials, everything else ferments over time. Consequently the **bDM** value from the sparse data in-vitro is large. The fermentation rate **c** is also large because the **b** fraction fermenting from time zero contains the **a** fraction of DMA and would be better described as (a + b)DM.

A small **c** value for in-vivo prediction can be predicted by regressing on losses from a pool containing only slow starch and NDF with the first non zero time point after all quickly fermenting **a** material

has vanished. Applying this technique we can derive a different set of gas production a, b, c values more closely resembling in-vivo a, b, c values.

Solubility must not be confused with fermentability as a means of disappearance because the rate of fermentation is a lot less than the rate of solubilization which is certainly not infinite. Soluble fractions have a measurable rate of fermentation apparent when data is reported at frequent intervals.

Recognition of the differences between methods which have a similar purpose is required.

In-vivo measures losses from a micro-porous bag. The results tell us nothing about what was lost or how, where and when it was utilized. Use of that data requires assumptions about solubility and passage.

Gas production tells us how fast materials are fermented in gas tight syringes. How or where substrates are utilized also depends on assumptions concerning solubility and passage. Predictions of ME from gas production have a wider range than that predicted from digestibility data.

CONCLUSIONS

1. A quickly vanishing dry matter fraction can be identified from gas production data tabulated in appendix 1. as sugar + quick starch + OQCHO + oil + ash + fermentation acids.
2. A slowly vanishing dry matter fraction composed of slow starch and fermentable NDF at a measurable fractional rate.
3. The slowly vanishing fraction goes at a fractional rate of 0.01 to 0.04 much less than the fermentation rates observed in gas production
4. Nearly 80% of the variability in ME can be explained by the fermentability of carbohydrates
5. Varietal difference in fermentation characteristics may be more important than variation in starch content of forage maize.
6. A new model set of c rate parameters remains to be calculated

REFERENCES

Bioparametrics Ltd, 2004 Carbohydrate digestion SOP

Bioparametrics Ltd, 2004 Standardization of Rumen Liquor SOP

Feedstuffs 13 December 2010 Gas fermentation a promising tool

Herrero, M., Jessop, N.S., 1996. Relationship between in vitro gas production and neutral detergent fibre disappearance in three tropical grasses. *Animal Science* 62 (Abstract), 682.

Palmer MJ 2006 Development of the in vitro gas production technique to estimate protein degradation in the rumen (University of Edinburgh)