



Influence of inoculum source in a gas production method

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Abstract

Gas production techniques are used in many laboratories to study fermentation kinetics of ruminant feeds, and the major source of variation is often the inoculum. Fifteen substrates (two legume hays, two tropical grass hays, one fresh tropical grass, five temperate grasses, soybean meal, maize grain, maize silage, wheat straw, sugarcane bagasse) were used to measure fermentation gas release with a semi automated system, and a sigmoidal model was fitted to gas production data from rumen fluid collected from eight fistulated sheep and two cows. Comparisons were made between cattle and sheep inocula and between inocula prepared using different proportions (v/v) of rumen liquid and solid phases (1:0, 0.75:0.25, 0.67:0.33 and 0.5:0.5). There were no differences between estimates of asymptotic gas production, and organic matter digestibility, with the different species inocula, but rates of fermentation were higher with rumen fluid inocula from cattle versus sheep. Rumen contents with no solid phase produced more gas, whereas a 1:1 ratio of liquid:solid increased digestibility.

Abbreviations: A, the asymptotic gas production of the France model; ADF, acid-detergent fibre; CP, crude protein; DEG, degradability; DM, dry matter; DMD, DM degradability; GP, gas production; G48, gas production after 48 h of incubation; G96, gas production after 96 h of incubation; LAG, lag time; NDF, neutral detergent fibre; OM, organic matter; OMD, OM degradability; REL1, ratio between G48 and G96; REL2, ratio between G96 and A; RL_{cattle}, cattle rumen fluid; RL_{sheep}, sheep rumen fluid; SCFA, short chain fat acids

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However, the rate of gas production was not affected by the proportion of solid phase in the rumen inoculum.

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1. Introduction

Feed evaluation for ruminants is often based on an estimate of rumen digestibility, even though such measurements alone cannot predict how ruminants will utilise feed nutrients. However, *in vivo* digestibility depends on a range of factors, including potential digestibility, rate of digestion, fractional rumen outflow and efficiency of microbial production. Thus a number of methodologies to estimate potential digestibility and/or rate of digestion (e.g., Tilley and Terry, 1963; Ørskov and McDonald, 1979) have been developed. *In vitro* gas production systems (e.g., Wilkins, 1974; Menke et al., 1979; Pell and Schofield, 1993; Theodorou et al., 1994; Mauricio et al., 1999) provide an estimate of dry matter (DM) and/or organic matter (OM) digestion and are an indicator of the end products produced, either directly as gas production (GP) or indirectly as short chain fat acids. The popularity of *in vitro* gas systems to evaluate ruminant feeds has largely been due to high analytical capacity and low cost.

However, the major source of variation of this, and other, *in vitro* techniques is with the inoculum used and its method of preparation. Considerable animal diurnal variation in the quality of rumen fluid inoculum, prepared identically, is known to exist both within and among donor animals. Several factors influence GP, such as microbial activity (Jessop and Herrero, 1996) that could be affected by collection frequency (Nagadi et al., 1999), time of collection (Menke and Steingass, 1988) and the final dilution with buffer (Pell and Schofield, 1993; Rymer et al., 1999).

In addition, the method of inoculum preparation may influence results (Rymer et al., 1999). Microbial populations differ between the solid and liquid phases of rumen contents with the majority of the cellulolytic bacteria attached to feed particles within the solid phase and the opportunistic, or non-specialised, rumen microflora being in the liquid phase. Ideally, the composition of microorganisms in the inocula should be representative, in terms of quantity and quality, of that found in the rumen of the donor animal. The contribution of the solid phase associated microorganisms to degradation is therefore vital, especially when forages with high cell wall contents are examined.

A further aspect is whether the species of donor animal used has a bearing on the quality of the inoculum. The use of small ruminants, especially sheep, is widely accepted in both digestibility and metabolism studies, such as ruminal *in situ* techniques, partly because of ease of animal handling, reduced animal maintenance costs and lower feed requirements versus larger ruminants such as cattle. Indeed digestibility values obtained in sheep fed at energy maintenance are accepted as the worldwide standard for most ruminant feeding systems (Cone et al., 1996, 2002).

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