



In vitro ruminal dry matter degradability, microbial efficiency, short chain fatty acids, carbohydrate and protein fractionation of tropical grass-multipurpose tree species diets



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ARTICLE INFO

Article history:

Received 10 May 2013

Received in revised form

18 November 2013

Accepted 19 November 2013

Keywords:

Forage

Partitioning factor

Protein value

Small ruminant production

ABSTRACT

An in vitro experiment was carried out using the Hohenheim gas production technique to evaluate 24-h gas production, apparently and truly degraded dry matter (DM), partitioning factor (PF), short chain fatty acids, crude protein (CP) and carbohydrate (CHO) fractionation of grass and multipurpose tree species (MPTS) foliage diets. Four grasses and three MPTS were used to formulate 12 diets of equal mixtures (0.5:0.5 on DM basis) of each grass with each MPTS. In vitro gas production was terminated after 24 h for each diet. True DM degradability was measured from incubated samples and combined with gas volume to estimate PF. Diets had greater ($P < 0.001$) CP (102–183 g/kg DM) content than sole grasses (66–131 g/kg DM) and lower ($P < 0.001$) concentrations of fibre fractions. Contrary to in vitro apparently degraded DM, in vitro truly degraded DM coefficient was greater ($P < 0.001$) in diets (0.63–0.77) than in sole grasses (0.48–0.68). The PF was on average higher in diets than in sole grasses. The proportion of potentially degradable CP fractions (A₁, B₁, B₂ and B₃, based on the Cornell Net Carbohydrate and Protein System) in the diets ranged from 971 to 989 g/kg CP. Crude protein fractions, A and B₂ were greater in diets but B₁ and B₃ fractions were less in diets than in sole grasses. A similar trend was also observed in the CHO fractions. Results showed that the nutritive value of the four grasses was improved when MPTS leaves were incorporated into the diet and this could ensure higher productivity of the animals.

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1. Introduction

Grasses mixed with foliage of multipurpose tree species (MPTS) in varying proportions constitute a major portion of small ruminants' feeding both in extensive and semi-intensive systems in tropical and sub-tropical regions

including India (Devendra, 1990; Topps, 1992). Biomass of grazing resources (e.g., common lands, forest lands and roadside lands) primarily consist of grasses and foliage from MPTS. Grasses are usually low in quality and quantity particularly in summer and unable to meet the maintenance requirement of ruminants. The MPTS foliages which are rich in crude protein (CP), vitamin and minerals (Aganga and Mesho, 2008; Mtui et al., 2008) are usually used to supplement poor quality roughages (dry grasses, straws, stovers, etc.) in periods of scarcity to improve their nutritive value (Aganga, 2003). A level of 300–500 g/kg of shrub and tree leaves on dry matter (DM) basis in diets of ruminants has been recommended for improved nutrient utilisation (Devendra, 1990; Singh, 2004).

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Morus alba, *Grewia optiva* and *Leucaena leucocephala* are well adapted to hot and arid environments, persistent in almost all type of soils and constitute a substantial part of small ruminant feeding system in many countries (Trivedi, 2010).

In the present study, leaves of *L. leucocephala*, *M. alba* and *G. optiva* were evaluated with four grasses (*Pennisetum purpureum*, *Panicum maximum*, *Cenchrus ciliaris* and *Syzygium nervosum*) for their supplementary effect when used in 0.5:0.5 proportions on in vitro gas production, apparent and true substrate DM degradabilities, partitioning factor (PF), microbial mass and efficiency, and on CP and carbohydrate (CHO) fractions.

2. Materials and methods

2.1. Experimental site

The experiment was carried out at the Institute of Animal Science, University of Bonn, Germany. Samples of grasses and tree leaves were collected from nurseries maintained at farmers' demonstration blocks and fields of the Grassland and Silvopastoral Division of Indian Grassland and Fodder Research Institute, Jhansi, India.

2.2. Experimental design

The experiment was arranged in a completely randomized design. Four grasses and three MPTS were used to formulate 12 diets of equal mixtures (0.5:0.5 on DM basis) of each grass with each MPTS. The dietary combinations are given below:

<i>C. ciliaris</i> – <i>L. leucocephala</i>	<i>C. ciliaris</i> – <i>G. optiva</i>	<i>C. ciliaris</i> – <i>M. alba</i>
<i>S. nervosum</i> – <i>L. leucocephala</i>	<i>S. nervosum</i> – <i>G. optiva</i>	<i>S. nervosum</i> – <i>M. alba</i>
<i>P. purpureum</i> – <i>L. leucocephala</i>	<i>P. purpureum</i> – <i>G. optiva</i>	<i>P. purpureum</i> – <i>M. alba</i>
<i>P. maximum</i> – <i>L. leucocephala</i>	<i>P. maximum</i> – <i>G. optiva</i>	<i>P. maximum</i> – <i>M. alba</i>

2.3. Sample collection

Grass and tree foliage samples were collected in mid September 2009. Grass samples were collected from two fields using a metre square quadrat while foliage was lopped from three to four trees to have a composite representative sample.

2.4. Chemical analyses

Samples of grasses and tree leaves were initially dried under shade and later oven-dried at 60 °C for 96 h. The samples were milled through 1-mm sieve and all chemical analyses were done according to the German Handbook of Agricultural Experimental and Analytical Methods (VDLUFA, 2007) and method numbers are given. The residual DM was determined by oven-drying at 105 °C (3.1). Total nitrogen (N) was estimated by Dumas combustion (4.1.2; Leco 8.1, Leco Instrument, Mönchengladbach, Germany) and CP was expressed as $N \times 6.25$, ash and

ether extract (EE) were analysed using methods 8.1 and 5.1.1, respectively. Neutral detergent fibre (NDFom; 6.5.1) determined without use of α -amylase or sodium sulphite and acid detergent fibre (ADFom; 6.5.2) were expressed exclusive residual ash. Lignin(sa) was determined by solubilisation of cellulose with sulphuric acid in the ADF residue (Van Soest et al., 1991). Hemicellulose was calculated as NDFom – ADFom. Short chain fatty acids (SCFA) concentrations were determined according to VDLUFA (2007) using a splitless injector Perkin-Elmer auto system gas chromatograph (Perkin-Elmer, Inc., Shelton, CN, USA).

2.5. In vitro gas production measurement

In vitro gas production was determined according to Menke and Steingass (1988). Rumen fluids were collected prior to feeding from three fistulated Merino sheep (≈ 70 kg body weight) fed a standard diet (600 g grass hay/600 g pelleted concentrate). Feeds were offered in two meals at 07:00 and 15:00 h at a ratio of 0.4–0.6, respectively. Equal volume of rumen fluid was collected from each sheep. Rumen fluid was strained through two layers of cheesecloth into a prewarmed, insulated flask. All laboratory handling of rumen fluid was carried out under a continuous flow of CO₂. Samples (200 ± 2 mg) were accurately weighed into 100 ml glass syringes, the pistons were lubricated with vaseline and inserted into the syringes. Syringe was the experimental unit. In vitro incubation of the samples was conducted in triplicate. Syringes were filled with 30 ml of medium consisting of 10 ml of rumen fluid and 20 ml of buffer solution as described by Menke and Steingass (1988) except that the concentration of NaHCO₃ was reduced to 33 g/l and that of (NH₄)HCO₃ increased to 6 g/l. Three blanks containing 30 ml of medium only were included. The syringes were placed in a rotor inside the incubator (39 °C) with about one rotation per min. Cumulative gas volume measurements of samples were read manually from the three replicates each at 0, 6 and 24 h. The whole process was repeated to have six analytical replicates.

2.6. In vitro apparently (ivADDM), truly degraded dry matter (ivTDDM) and microbial mass determination

In vitro apparently degraded DM was determined following the procedure of Blümmel and Lebzién (2001).

In vitro apparent degraded DM coefficient was calculated as follows:

$$\text{feed (DM) incubated} - [\text{pellet (DM)} \\ - \text{blank pellet (DM)}] / \text{feed (DM) incubated}$$

The procedure of Van Soest et al. (1991) was used to determine in vitro truly degraded DM.

The ivTDDM coefficient was calculated as follows:

$$\text{feed (DM) incubated} - \text{residue (DM) recovered} \\ \text{in the crucibles} / \text{feed (DM) incubated}$$

Microbial mass was calculated with a slight modification of the procedure of Blümmel et al. (1997b) as follows:

$$\text{pellet (DM)} - \text{blank pellet (DM)} \\ - \text{residue (DM) recovered in the crucibles}$$

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