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Manipulation of *in vitro* ruminal fermentation and digestibility by dried rumen digesta

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ABSTRACT

This experiment was to investigate the effect of dried rumen digesta (DRD) levels on rumen kinetic gas production, rumen ammonia-nitrogen (NH_3-N) and digestibility of nutrients by using *in vitro* gas production techniques. The experimental design was a completely randomized design (CRD) and the dietary treatments were DRD supplementation at 0, 4, 8, 12, 16 and 20 mg with 0.5 g of roughage and concentrate ratio at 70:30. The gas production was recorded at 0, 0.5, 1, 2, 4, 6, 8, 12, 18, 24, 48, 72 and 96 h of incubation and was used for calculated of kinetics gas. The results revealed that crude protein, NDF and ADF in DRD were 19.4%, 42.4% and 20.9% of DM, respectively. The intercept value (a) for the different treatments representing gas production from soluble fractions, gas production from the insoluble fraction (b), potential extent of gas production ($a+b$) were not significantly different among treatments ($P>0.05$). However, gas production rate (c) and cumulative gas production (96 h of incubation) were significantly different between the supplemented groups and control ($P<0.05$) and were highest when supplementation of DRD at 8 mg and the values were 0.09 ml/h and 132.5 ml/0.5 g substrate, respectively ($P<0.05$). NH_3-N concentrations were linearly increased when increasing levels of DRD and were highest with DRD supplementation at 20 mg (25.3 mg%). Moreover, *in vitro* dry matter digestibility (IVDMD) and *in vitro* organic matter digestibility (IVOMD) were highest when 8 mg of DRD was supplemented and the mean values were 63.8% and 65.2% DM, respectively. Based on this experiment, it could be concluded that supplementation of DRD at 8 mg could improve rumen fermentation, *in vitro* kinetics gas, NH_3-N concentration and digestibility.

1. Introduction

The use of soybean meal (SBM) as a protein source in ruminant feeding is well established. However, high price of SBM in some parts of the world, and fluctuation in SBM production, have raised interest in alternative local protein sources for animal feeding (Cherdthong et al., 2011). Locally available feed resources for ruminant production in the tropics such as crop residues, agro-industrial by-products and animal west are becoming increasingly important because of rising costs and limited supplies of conventional feedstuffs (Hermansen, 2003; Wanapat, 2009).

Rumen digesta is the one of by-products of slaughterhouse, it is consisted of fermented and non-fermented of dietary feeds, rumen microorganisms and the end products of microbes metabolic activities such as microbial proteins, amino acids, vitamins and volatile fatty acids. In addition, rumen digesta contains no anti-physiological factors (Okpanachi et al., 2010). The crude protein (CP) in dried rumen digesta (DRD) ranged from 9% to 20% of DM and could be a good source of protein in

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animal feed if it is properly processed and harnessed (Adeniji and Balogun, 2002; Agbabiaka et al., 2011a,b; Esonu et al., 2006). DRD was usually discarded as waste and allowed to rot hence posing a high environmental pollution and disposal problems (Agbabiaka et al., 2011a). In Thailand, it is estimated that the total by-product of DRD in 2011 was 41,000 t of dry matter from 1.2 million of ruminant animals in abattoir (FAO, 2012). Thus, the use of DRD not only serves as animal feed ingredient, but also reduces disposal and environmental pollution problem.

Since DRD is rich in protein as well as rumen micro-organisms and some minerals, supplementation into diet of animals could increase the efficiency of by-products utilization in the feeding systems. Several studies have been succeeded by supplementation of DRD in diets of Nile Tilapia (Agbabiaka et al., 2011a), African catfish (Agbabiaka et al., 2011b), broiler chicken (Esonu et al., 2006), layers (Odunsi, 2003), and rabbits (Dairo et al., 2005; Okpanachi et al., 2010). However, no investigations have been performed on supplementation of DRD in concentrate diets of ruminants fed on rice straw. Therefore, the objective of this experiment was to investigate the effect of DRD levels on rumen kinetics gas production, *in vitro* rumen fermentation and digestibility of nutrients by using *in vitro* gas production technique.

2. Materials and methods

2.1. Experimental design and dietary treatments

The experimental design was a completely randomized design (CRD) and the dietary treatments were DRD supplementation at 0, 4, 8, 12, 16 and 20 mg with 0.5 g of roughage and concentrate ratio at 70:30. Rice straw was used as a roughage source. Concentrates were formulated to contain cassava chip, rice bran and molasses as an energy source while, coconut meal, palm kernel meal and urea as the protein sources. DRD was collected from local abattoir at Bann Sri-Tan, Khon Kaen Province, Thailand; and sundried for 2 days. Samples of DRD, roughage and concentrates were dried at 60 °C, then ground to pass a 1-mm sieve (Cyclotech Mill, Tecator, Sweden) and used for chemical analysis and in the *in vitro* gas test. The samples were analyzed for dry matter (DM), ash and crude protein (CP) using the procedures of AOAC (1998), neutral detergent fiber (NDF) and acid detergent fiber (ADF) according to Van Soest et al. (1991). The ingredients and chemical compositions of concentrate, rice straw and DRD used in the *in vitro* experiment are shown in Table 1.

Table 1
Ingredient and chemical composition of concentrate, rice straw and dry rumen digesta (DRD) used in the experiment.

	Concentrate	DRD	Rice straw
Ingredients, kg DM			
Cassava chips	45.9		
Brewer's gain	13.7		
Rice bran	6.7		
Coconut meal	11.8		
Palm kernel meal	13.9		
Pure sulfur	0.5		
Mineral premix	1.0		
Molasses	3.0		
Urea	3.0		
Salt	0.5		
Chemical composition			
Dry matter, %	94.2	97.4	96.0
	% of DM		
Organic matter	92.2	92.5	87.9
Crude protein*	18.0	19.4	2.1
Ash	7.8	7.5	12.1
Neutral detergent fiber	19.0	42.4	75.1
Acid detergent fiber	8.5	20.9	55.6

* Crude protein contents in dietary treatments were 6.9%, 7.0%, 7.0%, 7.1%, 7.2%, and 7.3% of DM and dietary treatments were DRD supplementation at 0, 4, 8, 12, 16 and 20 mg with 0.5 g of roughage and concentrate ratio at 70:30.

2.2. Animals and preparation of rumen inoculum

Two male, rumen-fistulated swamp buffaloes with body weight of 350 ± 50 kg were used as rumen fluid donors. Swamp buffalo rumen fluid was collected from animals fed with concentrate (14% CP and 74% TDN) at 0.5% of BW in two equal portions, at 07:00 h and 16:00 h and rice straw was fed on *ad libitum* basis. The animals were kept in individual pens and clean fresh water and mineral blocks were offered as free choice. The animals received the diets for 20 d before the rumen fluid was collected. On day 20, 1000 ml rumen liquor was obtained from each animal before the morning feeding. The rumen fluid was filtered through four layers of cheesecloth into pre-warmed thermo flasks and then transported to the laboratory.

2.3. In vitro fermentation of substrates

Samples of 0.5 g of roughage and concentrate ratio (70:30) were weighed into 50 ml serum bottles and supplementation with the respective DRD levels. For each treatment, five replications were prepared. Ruminal fluid from each animal was mixed with the artificial saliva solution of Menke and Steingass (1988) in a proportion 2:1 (ml/ml) at 39 °C under continuous flushing with CO_2 and 40 ml of rumen inocula mixture were added into each bottle under CO_2 flushing. Bottles were sealed with rubber stoppers and aluminum caps and incubated at 39 °C (96 h) for *in vitro* gas test. Thirty minutes after starting the incubation, the bottles were gently mixed and then mixed three times every 3 h. For each sampling time, five bottles containing only the rumen inocula were included within each run and the mean gas production values of these bottles were used as blank. The blank values were subtracted from each measured value to give the net gas production.

2.4. Sample and analysis

During the incubation, data of gas production was measured immediately after incubation at 0, 0.5, 1, 2, 4, 6, 8, 12, 18, 24, 48, 72 and 96 h by using a pressure transducer and a calibrated syringe. Cumulative gas production data were fitted to the model of Ørskov and McDonald (1979)