

Evaluation of Biscuit by-Product Supplemented with *Saccharomyces Cerevisiae* using *in Vitro* Gas Production Technique

M. Besharati, A. Karimi, and Z. Nemati

Abstract— An *in vitro* gas production technique was used in this study to elucidate the effect of active live yeast on gas production. The treatment contained 0, 2.5, 5 and 7.5 g yeast *Saccharomyces cerevisiae* (Sc) per kg of biscuit by-product based on DM, respectively. 2 h after incubation time the treatment with Sc 7.5 g/kg DM had the highest gas production ($P<0.05$). At the early incubation times (4 and 6 h), the treatments 1 and 2 (treatment with Sc, 0 and 2.5 g/kg DM, respectively) had the highest gas production volume among treatments, but up to 8 h the gas production volume in treatment 4 (treatments with Sc, 7 g/kg DM) was the lowest gas production volume ($p<0.05$). It was concluded that *in vitro* gas production value of biscuit by-product was improved with addition of yeast *Saccharomyces cerevisiae* at 2.5 g/kg DM, but *Saccharomyces cerevisiae* at 5 and 7.5 g/kg DM decreased gas production volume.

Index Terms— Biscuit by-Product, *in Vitro* Gas Production, Probiotic, *Saccharomyces Cerevisiae*.

I. INTRODUCTION

IN Middle East, animals suffer from under feeding and malnutrition in winter due to the shortage of locally produced feeds which are not sufficient to cover the nutritional requirements of animals. A major constraint to increasing livestock productivity in developing countries is the scarcity and fluctuating quantity and quality of the year-round supply of conventional feeds. These countries experience serious shortages in animal feeds of the conventional type. In order to meet the projected high demand of livestock products and to fulfill the future hopes of feeding the millions and safeguarding their food security, the better utilization of non-conventional feed resources which do not compete with human food is imperative. There is also a need to identify and introduce new and lesser known food and feed crops. An important class of non-conventional feeds is by-product feedstuffs which are obtained during harvesting or processing of a commodity in which human food or fibre is

derived. The amount of by-product feedstuffs generally increases as the human population increases and economies grow [1,2].

Several factors have lead to increase interest in by-product feedstuffs, such as pollution abatement and regulations, increasing costs of waste disposal and changes in perception of the value of by-product feedstuffs as economical feed alternatives [1].

Probiotics present an attractive alternative to the use of chemical and hormonal promoters in the livestock growth production industry. The preparations contain have been used for production safe by micro-organisms many years and thus are that in food generally accepted as both the farmer and the final consumer. *Saccharomyces cerevisiae* supplementation in ruminant diets can be increased DMI, production performance, cellulose degradation, and nutrient digestibility [3]. The gas measuring technique has been widely used for evaluation of nutritive value of feeds. Gas measurement provides a useful data on digestion kinetics of both soluble and insoluble fractions of feedstuffs [4]. In the gas method, kinetics of fermentation can be studied on a single sample and therefore a relatively small amount of sample is required or a larger number of samples can be evaluated at time. Besharati et al. [5] showed that probiotic can improve the *in vitro* gas production. The purpose of this study was to study effect of adding different levels of *Saccharomyces cerevisiae* on *in vitro* gas production of biscuit by-product.

II. MATERIALS AND METHODS

Samples of rumen fluid were collected from two fistulated sheep fed twice daily a diet containing forage (400 g/kg) plus concentrate (600 g/kg) after morning feeding, strained through four layers of cheesecloth. Gas production was measured by Fedorak and Hruby [6] method. Approximately 300 mg of dried and ground (2mm) biscuit by-product sample was weighted and placed into serum bottles. The treatment contained 0, 2.5, 5 and 7.5 g yeast *Saccharomyces cerevisiae* (Sc) per kg of biscuit by-product based on DM, respectively. Buffered rumen fluid with McDougal buffer (20ml, ratio of buffer to rumen fluid was 2:1) was pipetted into each serum bottle [7]. The gas production was recorded after 2, 4, 6, 8, 12, 16, 24, 36, and 48 h of incubation. Total gas values were corrected for the blank incubation, and reported gas values are expressed in ml per 1 g of DM.

The data at the different times was analyzed using completely randomized design by the GLM procedure of SAS Institute Inc [8].

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III. RESULTS AND DISCUSSION

Total gas production volume (ml/g DM) in incubation times are shown in Table 1. 2 h after incubation time the treatment with Sc 7.5 g/kg DM had the highest gas production ($P<0.05$). At the early incubation times (4 and 6 h), the treatments 1 and 2 (treatment with Sc, 0 and 2.5 g/kg DM, respectively) had the highest gas production volume among

treatments, but up to 8 h the gas production volume in treatment 4 (treatments with Sc, 7 g/kg DM) was the lowest gas production volume ($p<0.05$). The treatment 2 at the most incubation time had the highest gas production volume. It was concluded that *in vitro* gas production parameters of biscuit by-production was improved with addition of yeast *Saccharomyces cerevisiae* at 2.5 g/Kg DM level.

TABLE I
TOTAL GAS PRODUCTION VOLUME (ML/G DM) IN INCUBATION TIMES AND ESTIMATED PARAMETERS

Treatments	Incubation times (h)								
	2	4	6	8	12	16	24	36	48
Control	9.10 ^a	44.66 ^a	83.06 ^a	114.36 ^{ab}	152.70 ^a	192.83 ^a	252.33 ^a	311.44 ^{ab}	340.61 ^{ab}
Sc 2.5 g/kg DM	10.32 ^c	44.55 ^a	84.51 ^a	119.80 ^a	163.35 ^a	202.71 ^a	261.65 ^a	325.99 ^a	355.82 ^a
Sc 5 g/kg DM	14.65 ^b	25.23 ^c	66.52 ^b	101.60 ^{ab}	145.48 ^b	185.40 ^b	244.00 ^b	302.01 ^b	327.52 ^b
Sc 7.5 g/kg DM	29.75 ^a	36.89 ^b	66.63 ^b	92.61 ^c	119.84 ^c	142.66 ^c	156.98 ^c	160.04 ^c	163.44 ^c
SEM	0.86	0.94	1.95	4.53	6.89	7.09	7.38	7.03	6.34

The means within a column without common letter differ ($p<0.05$).

A significant decrease in total gas production in the yeast treatments was observed in this study (treatments 3 and 4), which was increased in previous studies [9, 10]. This may be partly associated with the decreased production of acetate in the yeast treatments, because CO₂ and H₂ are byproducts of acetate production during carbohydrate fermentation. The lower total gas values in the yeast treatment groups suggest the reduction in CH₄ was absolute and not a result of a decrease in the total gas. Lynch and Martin [11] showed that *Saccharomyces cerevisiae* reduces ruminal CH₄ production.

Saccharomyces cerevisiae (Yea-Sacc 1026; Alltech Biotechnology Center, Nicholasville, KY) increased the number of ruminal total bacteria and cellulolytic bacteria [12], increased the proportion of propionate [12, 13], and decreased lactate concentration [14]. Another *S. cerevisiae* culture (Diamond V XP; Diamond V Mills, Inc., Indianapolis, IN) stimulated the growth of the cellulolytic bacteria, *Fibrobacter succinogenes* and *Ruminococcus albus* [15, 16], and increased the proportion of propionate [16] and lactate uptake by *Selenomonas ruminantium* [17].

Beauchemin *et al.* [18] reported that the supplementation of the diet with *E. faecium* increased ($P<0.05$) the proportion of propionate and decreased ($P<0.10$) proportion of butyrate in ruminal fluid compared with the control. This may be explained by stimulating of lactic acid-utilizing bacteria, which produce propionate.

IV. CONCLUSIONS

It was concluded that *in vitro* gas production value of biscuit by-product was improved with addition of yeast *Saccharomyces cerevisiae* at 2.5 g/kg DM, but *Saccharomyces cerevisiae* at 5 and 7.5 g/kg DM decreased gas production volume.

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