

## ***In vitro* Gas Production Profiles in Some Concentrate Ingredients**

A. Taghizadeh, H. Janmohamadi, G.A. Moghadam and J. Shodja

Department of Animal Science, Faculty of Agriculture, University of Tabriz, Iran

**Abstract:** The *in vitro* gas production technique was used for determine of fermentation characteristics of barley grain and corn grain. Triplicate samples were used for measuring gas production. The data were fitted by equation of  $P = A(1-e^{-ct})$  that P is gas production at time t, A is gas production of soluble and insoluble fraction, c is rate of gas production and t is time of incubation. The gas production was measured at 2, 4, 6, 8, 12, 16, 24, 36 and 48 h. The gas production of corn grain at 2 h was lower than barley grain ( $p < 0.05$ ), that can be resulted from slow degradation of starch in corn grain compared to barley grain. However, the sum of gas production of soluble and insoluble (a+b) in corn grain was significantly more than barley grain. The rate of gas production of barley grain was higher than the corn grain. The pattern of fermentation for corn grain differed from that in the barley grain.

**Key words:** Barley grain, corn grain, gas production

### INTRODUCTION

Nowadays most available information on the degradation behavior of feeds in ruminants is based on in situ disappearance in the rumen, which measure the appearance of nutrients. The replacement of method for in situ method that can be measured soluble fraction fermentation characteristics is *in vitro* gas production system. This method offer much information on the degradation washable and non-washable but degradable of starch, protein and cell walls (Chai *et al.*, 2003). Chai *et al.* (2003) reported high correlation between gas production and starch degradability. Tagizadeh (2004a, b) reported high relationship for gas production and in situ techniques at 24 h incubation for barley grain, corn grain and molasses. Blummel and Ørskov (1993) showed that dry matter disappearance can be predicted by gas production technique. The current study was carried out to determination of pattern fermentation of barley grain and corn grain using *in vitro* gas production technique.

### MATERIALS AND METHODS

**Chemical analysis:** All samples (barley grain, corn grain) were ground through a 1 mm screen in a wiley mill (model 4, Arthur H. Thomas Co, Philadelphia, PA). Neutral detergent fiber and ADF were measured according to the method of Van Soest *et al.* (1991). Determination of N were conducted using the Kjeldahl method in an automated Kjelfoss apparatus (Foos Electric,

Copenhagen, Denmark). Dry matter was determined by drying the whole samples in a forced air oven at 55°C until a constant weight was achieved.

***In vitro* gas production:** Two male sheep (38±1.5 kg) were used as donors of ruminal fluid for the preparation of inoculums. The sheep were fed a diet comprising (as fed) 72 g kg<sup>-1</sup> CP and 1.9 Mcal kg<sup>-1</sup> ME (containing 550 g kg<sup>-1</sup> alfalfa hay, 400 g kg<sup>-1</sup> barley grain, 50 g kg<sup>-1</sup> wheat bran and 20 g kg<sup>-1</sup> limestone). Equal volumes of ruminal fluid from each sheep collected 2 h after the morning feeding were combined and strained through four layers of cheesecloth and mixed with buffer prewarmed to 39°C (2 buffer : 1 rumen liquor). The inoculum was dispensed (20 mL) per vial in to substrate 35 mL serum vial (containing of 300 mg per vial) which had been warmed to 39°C and flushed with oxygen free CO<sub>2</sub>. The vials were sealed immediately after loading and were affixed to a rotary shaker platform (lab-line instruments Inc Melors dark, USA) set at (120 rpm) housed in a incubator. Vials for each time point, as well as blanks (containing no substrate), were prepared in triplicate. Gas production was measured in each vial after 2, 4, 6, 8, 12, 16, 24, 36, 48, 72 and 96 h of incubation using a water displacement apparatus (Fedorak and Hrudey, 1983).

**Calculation and statistical analyses:** Gas production was calculated as mL g<sup>-1</sup> sample DM and gas production values over time for each sample were fitted to a one-component McDonald model:  $P = A(1-e^{-ct})$  that P is the volume of gas production (mL g<sup>-1</sup> DM) in time of

Table 1: The chemical composition of test feeds (%DM)

Feed	Item				
	DM%	CP%	NDF%	ADF%	Hemicellulose <sup>1</sup>
Corn grain	82.56	11.86	10.0	6.24	3.76
Barley grain	93.08	13.61	13.2	6.70	6.30

<sup>1</sup> - Hemicellulose = NDF-ADF

Table 2: The gas production of test feeds (mL g<sup>-1</sup> DM)

Feed	Incubation time												
	2	4	6	8	12	16	24	36	48	72	96	A	C
CG	17.6 <sup>b</sup>	45.4 <sup>b</sup>	66.8 <sup>b</sup>	88.3 <sup>b</sup>	126.8 <sup>b</sup>	169.9 <sup>b</sup>	223.9 <sup>b</sup>	268.1	297.8	313.6	320.8	326.5 <sup>a</sup>	0.04 <sup>b</sup>
BG	31.8 <sup>a</sup>	72.0 <sup>a</sup>	106.7 <sup>a</sup>	141.5 <sup>a</sup>	191.7 <sup>a</sup>	228.0 <sup>a</sup>	260.9 <sup>a</sup>	281.8	291.9	295.5	297.4	296.9 <sup>b</sup>	0.09 <sup>a</sup>
SEM	2.763	2.930	3.763	5.248	7.225	8.710	10.057	12.306	12.277	11.410	11.092	10.00	0.002

incubation, A is gas production of soluble and insoluble fraction, c rate of gas production of insoluble fraction and t is time. Parameters A and c was estimated by an iterative least square method using a nonlinear regression procedure of the statistical analysis system (SAS, 1990).

## RESULTS AND DISCUSSION

The chemical compositions are shown on Table 1. The gas production data are shown on Table 2. The gas production of corn grain at 2 h of incubation is lower than the barley grain that can be resulted from slow degradability of corn starch (Getachew *et al.*, 1998, 2002). The difference between fermentation characteristics specially the gas production and rate of gas production at first 24 h of incubation is related to soluble carbohydrate (starch, sugar and) (Menke and Steingass, 1988). The gas production of barley grain at first 24 h is more than the corn grain (p<0.05), that showed soluble carbohydrate in barley grain is higher than the other feed. The gas production at 96 h in corn grain is numerically more than the barley grain that can be resulted from low NDF in corn grain (Table 1).

The gas yield after 24 h is resulted from fermentation of structural carbohydrate (Menke and Steingass, 1988; Getachew *et al.*, 1998). High gas production show more metabolisable energy (Datt and Sinigh, 1995) and the potential gas production in both of cereal grain in this experiment certificate this hypotheses. Lanzas *et al.* (2006) reported the gas production of barley grain and corn grain at 48 h of incubation, 280 and 330 mL g<sup>-1</sup> DM, respectively. Getachew *et al.* (2002) found that gas production of barley grain and corn grain at 24 h of incubation, 343.5 and 373.5, respectively. The rate of gas production in barley grain is more than the corn grain (Table 2).

This finding can be predicted due to slow degradability of starch in corn grain proportion to barley

grain resulting of differences in physical and chemical characteristics. The rate of gas production in both of cereal grain in our experiment is different from other reports (Getachew *et al.*, 2002; Lanzas *et al.*, 2006). The differences of gas production and rate of gas production in test feeds of this study compared to other reports, can be resulted of differences in soluble carbohydrate, structural carbohydrate, microbial rumen inoculums, time of sampling of rumen fluid, type of donor animal and gas production recording assay.

## REFERENCES

- Blummel, J.M.W. and E. Orskov, 1993. Composition of *in vitro* gas production and nylon bag degradability of roughages in predicting food intake in cattle. *Anim. Feed Sci. Technol.*, 40: 109-119.
- Chai, W.Z., A.Z. Van Golder and J.W. Cone, 2003. Relationship between gas production and starch degradation in feed samples. *Anim. Feed Sci. Technol.*, 114: 195-204.
- Datt, C. and G. Singh, 1995. Effect of protein supplementation on *in vitro* digestibility and gas production of wheat straw. *Indian J. Dairy Sci.*, 48: 357-361.
- Fedorak, P.M. and S.E. Hrudey, 1983. A Simple apparatus for measuring gas production by methanogenic cultures in serum bottles. *Environ. Technol. Lett.*, 4: 425-435.
- Getachew, G., M. Blummel, H.P.S. Makkar and K. Becher, 1998. *In vitro* gas measuring technique for assessment of nutritional quality of feeds: A Rev. *Anim. Feed Sci. Technol.*, 72: 261-281.
- Getachew, G., G.M. Crovetto, M. Fondevila, U. Krishna Moorthy, B. Singh, M. Spanghero, H. Steingass, P.H. Robinson and M.M. Kailas, 2002. Laboratory variation of 24 h *in vitro* gas production and estimated metabolizable energy values of ruminant feeds. *Anim. Feed Sci. Technol.*, 102: 169-180.

- Lanzas, C., D.G. Fox and A.N. Pell, 2006. Digestion kinetics of dried cereal grain. *Anim. Feed Sci. Technol.*, 136: 265-280.
- Menke, K.H. and H. Steingass, 1988. Estimation of the energetic feed value obtained from chemical analysis and *in vitro* gas production using rumen fluid. *Anim. Res. Dev.*, 28: 7-12.
- SAS Institute INC., 1990. SAS user's Guide: Statistics. Statistical Analysis Systems Institute Inc. Cary NC.
- Taghizadeh, A., 2004a. Differentiation of energy supplement using *in vitro* fermentation rates generated with the nylon bag and gas production technique. *Proc. Can. Soc. Anim. Sci.*, pp: 133.
- Taghizadeh, A., 2004 b. The estimation of Feed value of Energy supplement using *in vitro* Gas Production Techniques. *Proc. Can. Soc. Anim. Sci.*, pp: 134.
- Van Soest, P.J., J.B. Robertson and B.A. Levvis, 1991. Methods for dietary, neutral detergent fiber and nonstarch polysaccharides in relation to animal nutrition. *J. Anim. Sci.*, 74: 3583-3597.