Research Journal of Biological Sciences 4 (11): 1190-1192, 2009
ISSN: 1815-8846


# The Evaluation of the Effect of Tannin of Oak Leave on *in vitro* Rumen Fermentation of Soybean Meal

T. Mohammadabadi, M. Chaji, M. Eslami and M. Bojarpour
Department of Animal Science, Ramin Agriculture and Natural Resource University, Molasani, Ahwaz, Iran

**Abstract:** This study was conducted to evaluate the effect of oak leave tannin (OT, 25 and 45 g $kg^{-1}$ DM) on *in vitro* gas production and fermentative parameters of Soybean Meal (SB). Treatments were SB treated with 25 and 45 g $kg^{-1}$ oak leave tannin (OT1 and OT2, respectively). The results of experiment showed that oak leave caused to reduce the fermentable fraction (b) and gas production rate Constant (c) of SB ($p<0.05$) and the lowest (b) and (c) was for SB treated by 45 g $kg^{-1}$ oak leave tannin (104.5 mL, 0.04 mL $h^{-1}$, respectively). Soybean meal treated with OT2 had the lowest the Organic Matter Digestibility (OMD) and Metabolizable Energy (ME) (153.3 g $kg^{-1}$ OM, 13.6 MJ $kg^{-1}$ DM, respectively). Concentration of ammonia-N ($NH_3$-N) and Short Chain Fatty Acid (SCFA) decreased ($p<0.05$), when SB treated with OT and the highest $NH_3$-N and SCFA concentrations was for untreated soybean meal (36.6 mg $dL^{-1}$ and 0.88 μmol). Therefore, it may be that *in vitro* rumen fermentation and nutritive value of soybean meal are influenced by tannin content of oak leave.

**Key words:** Oak tannin, gas production, soybean meal, digestibility, concentration

## INTRODUCTION

Approximately 3 million ha of forest are covered by various oak species, in the north-west of Iran (Fatahi, 1995) that leaves used as important source of forage. Soybean meal is used as supplemental protein in dairy rations and is classified as highly degradable. It is reported that oak is containing the high level of tannin and tannin used as chemical additives for protecting and decreasing ruminal degradation of protein sources such as soybean meal because bind with protein (El-Waziry *et al*., 2007). At normal pH of the rumen, protein remains bound to the tannin but at the low pH of abomasum, the protein is released, so protein becomes available for digestion in the small intestine (El-Waziry *et al*., 2005).

The *in vitro* gas production is more efficient than the in sacco method in evaluating the effects of tannins (El-Waziry *et al*., 2007). Effects of tannin from different sources in reducing ruminal gas production and ammonia levels have been reported but there is not enough information on the effect of oak tannins on rumen fermentation of soybean meal.

Therefore, the aim of this study was to estimation the effect of oak leave tannins (OT, 25-45 g $kg^{-1}$ DM, respectively) on rumen degradation of soybean meal by *in vitro* gas production.

## MATERIALS AND METHODS

*In vitro* Gas Production (GP) technique was conducted according to the Menke and Steingass (1988). Rumen fluid was supplied from two fistulated sheep were fed a 40:60 concentrate: forage (3 kg concentrate: 2 kg alfalfa hay and 4.5 kg corn silage) in prior to the morning meal, homogenized in a laboratory blender filtered through three layers of cheese-cloth and purged with $CO_2$. The well mixed and $CO_2$ flushed rumen fluid was added to the buffered rumen fluid solution (1:2 v/v), which was maintained in a water bath at 39°C. Experimental samples were Untreated SB (USB), SB treated with 25 and 45 g $kg^{-1}$ oak leave tannin (SB + OT1 and SB + OT2, respectively). Tannin content of leave were determined by using Folin-Ciocalteu reagent in calorimetric method (Makkar and Singh, 1991). About 200 mg experimental sample (1.0 mm screen) incubated with 35 mL buffered rumen fluid under continuous $CO_2$ reflux in 100 mL calibrated glass syringes for 2, 4, 6, 8, 10, 12, 16, 24, 48, 72 and 96 h in a water bath maintained at 39°C. Samples were incubated in triplicate together with three syringes containing only incubation medium (blank).

After 96 h of incubation, the medium of each syringe used for determination ammonia-N ($NH_3$-N) concentration using distillation method (Kjeltec 2300 Autoanalyzer, Foss Tecator AB, Hoganas, Sweden). Cumulative gas production data were fitted to the exponential equation:

**Corresponding Author:** T. Mohammadabadi, Department of Animal Science, Ramin Agriculture and Natural Resource University, Molasani, Ahwaz, Iran

$$Y = b\,(1\text{-}e^{-ct})$$

Where:

b = Gas production from the fermentable fraction (mL)
c = Gas production rate constant (mL $h^{-1}$)
t = Incubation time (h)
Y = Gas produced at time t

The values of Organic Matter Digestibility (OMD) and Metabolisable Energy (ME) of samples were calculated by the equation of Menke and Steingass (1988), OMD (g $kg^{-1}$ OM) = 148.8 + 8.89 GP + 4.5 CP + 0. 651 XA and ME (MJ $kg^{-1}$ DM) = 2.20 + 0.136 GP + 0.057 CP + 0.0029 $CP^2$. Short Chain Fatty Acids (SCFA) were determined by the equation reported by (Getachew *et al*., 1999). SCFA (Fmol $L^{-1}$) = 0.0239 GP B 0.0601. CP and XA were crude protein and ash and GP was the net gas production after 24 h incubation.

Data of *in vitro* gas production, ME, OMD, $NH_3$-N and SCFA were subjected to analysis as a completely randomized design using the General Linear Model (GLM). Duncan's multiple range test was used to compare treatment means at ($p<0.05$).

## RESULTS AND DISCUSSION

Tannin content of oak leave was 39 g $100^{-1}$ DM, respectively. Oak leave tannin decreased *in vitro* gas production parameters, ME, OMD, SCFA (Table 1) and $NH_3$-N (Fig. 1) of soybean meal ($p<0.05$). Soybean meal treated with oak 45 g $kg^{-1}$ DM leave had the lowest b and c fractions. This result reported by El-Waziry *et al*. (2007) for soybean meal treated by querbracho tannin.

Tabacco *et al*. (2006) reported that tannins significantly depressed gas production, probably hampering rumen microorganisms. Also the result of Bento *et al*. (2005) showed reduction in gas production by mimosa tannin may be associated with binding of it to microorganisms or their enzymes but McMahon *et al*. (2000) concluded that tannins do not simply inhibit cellulose digestion but the inhibitory effects of tannins involved the bacterial cells themselves.

The estimated ME, OMD and SCFA were decreased when SB was treated by oak leave and this decrease was maximum for SB treated by 45 g $kg^{-1}$ DM. Reducing these parameters reported by Mohammadabadi *et al*. (2010) for sunflower meal treated with tannic acid. Also Tabacco *et al*. (2006) explained that OM digestibility decreased by about 5.1% with tannin. Reduction of volatile fatty acid reported by EL-Waziry *et al*. (2005). The decrease in OMD and ME was probably due to decreased rumen degradability and formation of complexes between tannins and dietary proteins and carbohydrates, as well as reducing rumen microbial proteolytic and cellulolytic enzyme activities, general fermentative activities (Muhammed *et al*., 1994).

Table 1: Gas production parameters of soybean meal treated with tannin of oak leave

| Treatments | b (mL) | c (mL $h^{-1}$) | OMD (g $kg^{-1}$ OM) | ME (MJ $kg^{-1}$ DM) | SCFA (μmol $L^{-1}$) |
|---|---|---|---|---|---|
| USB | 142.8$^a$ | 0.090$^a$ | 180.60$^a$ | 26.60$^a$ | 0.88$^a$ |
| SB+OT1 | 128.2$^b$ | 0.070$^b$ | 167.30$^b$ | 19.50$^b$ | 0.64$^b$ |
| SB+OT2 | 104.5$^c$ | 0.040$^c$ | 153.30$^c$ | 13.60$^c$ | 0.49$^c$ |
| SEM | 0.5 | 0.001 | 0.14 | 0.33 | 0.02 |

USB: Untreated SB; SB + OT1: SB treated with 25 g $kg^{-1}$ oak leave tannin; SB + OT2: SB treated with 45 g $kg^{-1}$ oak leave tannin b: gas production from fermentable fraction; c: gas production rate constant; OMD: Organic Matter Digestibility; ME: Metabolizable Energy; SCFA: Short Chain Fatty Acid; SEM: Standard Error of Mean, means within each column with different letters are significantly different ($p<0.05$)

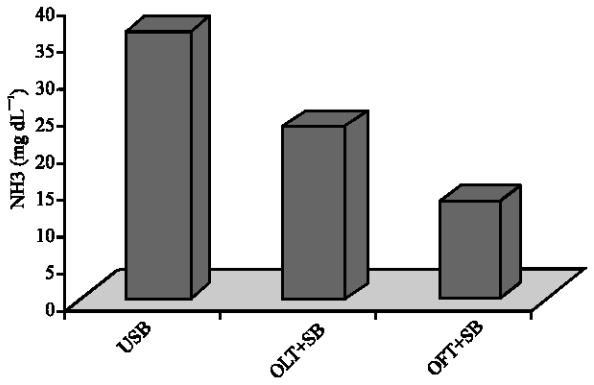


Fig. 1: The effect of tannin of oak leave on *in vitro* $NH_3$-N concentration of soybean meal

Tannin caused to altered microbial profiles in the fermentation, reduced microbial numbers and or enzyme production from the microbes (Apajalahti *et al*., 2004) and reduced microbial degradation of carbohydrates (Muhammed *et al*., 1994).

When SB treated with tannin of oak leave, $NH_3$-N concentration decreased that was in agreement with researches of Sliwinski *et al*. (2002). The lower ammonia concentrations by tannin were mainly due to reduce proteolysis, degradation of peptides and deamination of amino acids in the rumen (Newbold *et al*., 1990).

## CONCLUSION

In the study, the treatment of soybean meal by 25 and 45 g $kg^{-1}$ tannin of oak leave decreased degradation and gas production, OMD, ME, SCFA and $NH_3$ and the highest decrease was observed for soybean meal treated by 45 g $kg^{-1}$ tannin of oak leave.

## REFERENCES


Apajalahti, J.H.A., A. Kettunen and H. Graham, 2004. Characteristics of the gastrointestinal microbial communities, with special reference to the chicken. Worlds Poult. Sci. J., 60: 223-232.

Bento, M.H.L., H.P.S. Makkar and T. Acamovic, 2005. Effect of mimosa tannin and pectin on microbial protein synthesis and gas production during *in vitro* fermentation of 15N-labelled maize shoots. Anim. Feed Sci. Technol., 123: 365-377.

El-Waziry, A.M., M.E.A. Nasser and S.M.A. Sallam, 2005. Processing methods of soybean meal. 1-effect of roasting and tannic acid treated soybean meal on gas production and rumen fermentation *in vitro*. J. Applied Sci., 1: 313-320.

El-Waziry, A.M., M.E.A. Nasser, S.M.A. Sallam, A.L. Abdallah and I.C.S. Bueno, 2007. Processing methods of soybean meal, 2. Effect of autoclaving and Qucbraho tannin treated soybean meal on gas production and rumen fermentation *in vitro*. J. Applied Sci., 3: 17-24.

Fatahi, M., 1995. The Study of Zagros' Oak Forests and the Most Important Factors of its Destruction. Forests and Pastures Research Institute, Tehran, Iran, pp: 63.

Getachew, G., H.P.S. Makkar and K. Becker, 1999. Stoichiometric relationship between short chains fatty acid and *in vitro* gas production in presence and absence of polyethylene glycol for tannin containing browses. Proceeding of the EAAP Satellite Symposium on Gas Production: Fermentation Kinetics for Feed Evaluation and to Assess Microbial Activity, Wageningen, The Netherlands.

Makkar, H.P.S. and B. Singh, 1991. Effect of drying conditions on tannin, fibre and lignin levels in mature oak (*Quercus incana*) leaves. J. Sci. Food Agric., 54: 323-328.

McMahon, L.R., T.A. McAllister, B.P. Berg, W. Majakn and S.N. Acharya *et al*., 2000. A review of the effects of forage condensed tannins on ruminal fermentation and bloat in grazing cattle. Can. J. Plant Sci., 80: 469-485.

Menke, K.H. and H. Steingass, 1988. Estimation of the energetic feed value obtained from chemical analysis and gas production using rumen fluid. Anim. Res. Dev., 28: 7-55.

Mohammadabadi, T., M. Chaji and S. Tabatabaei, 2010. The effect of tannic acid on *in vitro* gas production and rumen fermentation of sunflower meal. J. Anim. Vet. Adv., 9: 277-280.

Muhammed, S., C.S. Stewart and T. Acamovic, 1994. Effects of tannic acid on cellulose degradation, adhesion and enzymic activity of rumen microorganisms. Proc. Soc. Nutr. Physiol., 3: 25-30.

Newbold, C.J., R.J. Wallace and N. Mckain, 1990. Effects of the ionophore tetronasin on nitrogen metabolism by ruminal microorganisms *in vitro*. J. Anim. Sci., 68: 1103-1111.

Sliwinski, B.J., C.R. Soliva, A. Machmuller and M. Kreuzer, 2002. Efficacy of plant extracts rich in secondary constituents to modify rumen fermentation. Anim. Feed Sci. Technol., 101: 101-114.

Tabacco, E., G. Borreani, G.M. Crovetto, G. Galassi, D. Colombo and L. Cavallarin, 2006. Effect of chestnut tannin on fermentation quality, proteolysis and protein rumen degradability of alfalfa silage. J. Dairy Sci., 89: 4736-4746.