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Evaluation effect of *Zataria multiflora* water extract on degradibility of soybean meal with gas product technique

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ABSTRACT

The aim of the present study was conducted to survey effect of adding different levels (0 and 0.3 ml/30ml buffered rumen fluid) of zataria multiflora water extract (ZMWE) on soybean meal (SBM) degradability were studied by in vitro gas producing techniques. Gas production test with mixtures of filtered rumen liquid of three Taleshi native male cattle rumen in times of 2, 4, 6, 8, 12, 24, 48, 72 and 96 hours were performed. The results showed that gas volume at 24 h incubation (for 200 mg dry samples), were 56.38 and 53.72 ml/200 mg DM for soybean meal, and zataria multiflora water extract (0.3 ml/30 ml buffered rumen fluid) respectively. Gas volume at 48 h incubation (for 200 mg dry samples), were 62.43 and 59.45 ml/200 mg DM for soybean meal, and zataria multiflora water extract (0.3 ml/30 ml buffered rumen fluid) respectively.

Keywords: Incubation; Zataria Multiflora; Soybean Meal; Gas Production Technique; Taleshi Native Male cattle; Rumen.

Abbreviations: ZMWE, Zataria Multiflora Water Extract; SBM, Soybean Meal; ZM, Zataria Multiflora.

INTRODUCTION

Modification of rumen microbial fermentation to decrease methane and ammonia nitrogen production using feed additives, such as antibiotics, has proved to be a useful strategy to improve production efficiency in dairy cattle [13, 2]. The public concern over the routine use of antibiotics and growth promoters in livestock production has increased recently because of the risk of the antibiotic residues presence in milk and meat and its effect on human health [19]. These led to its prohibition in the European Union in 2006 in animal feeding. Accordingly, there is greater interest in using plants and plant extracts as alternatives to feed antibiotics to manipulate ruminal fermentation, improve feed efficiency and animal productivity [3, 4 and 19]. Many plants produce secondary metabolites such as phenolic compounds, essential oils, and sarsaponins [3, 4 and 19]. That affect was microbial activity [19]. The *in vitro* gas production technique developed by Menke et al [14] is a very useful tool for the rapid screening of feeds to

assess their potential as energy sources for ruminant animals, Blummel and Becker [1], assuming that the volume of gas produced reflect the end result of the fermentation of the substrate to short chain fatty acids (SCFA), microbial biomass and the neutralization of the SCFA [23, 21 and 22]. This technique has been used by Blummel and Orskov [15] to determine gas production at several incubation times and values obtained could describe the pattern of fermentation of feed by using the model of [11, 21]. In addition, the application of models permits the fermentation kinetics of the soluble and readily degradable fraction of the feed and the more slowly degradable fraction to be described, [7, 21, 23 and 22]. The rumen has been well recognized as an essential fermentation that is capable of preparing end-products particularly volatile fatty acids and microbial protein synthesis as major energy and protein for the ruminant host, hence, the more efficient the rumen is, the optimum the fermentation end products are being synthesized [20]. In recent years, there have been increasing interests, researches conducted as well as reviews in relation to rumen studies, rumen ecology and rumen manipulation [20, 8, 12, 6, 9, 5 and 10]. The objective of this study were to evaluate effects of zataria multiflora water extract (0.3 ml/30ml buffered rumen fluid) on degradability, of soybean meal (SBM) using *in vitro* gas production technique.

MATERIALS AND METHODS

2.1. Zataria multiflora and soybean meal (SBM) samples

Soybean meal samples were obtained from commercial sources in Iran. During summer season Zataria multiflora samples were collected from different parts of Esfahan province. Next, there were drying for one week, and homogeneous mixture were papered for nutritive chemical analyzes. For determination of (zataria multiflora extract) effects, we added zataria multiflora water extract with tow doses (0 and 0.3 mL: 200 mg sample) into gas test syringes. All samples were then ground in a laboratory mill through a 1 mm screen.

2.2. Procedure of Zataria multiflora extracts preparation

The zataria multiflora water extract were prepared according to Patra et al [16]; Sallam et al [19] with some modifications. The zataria multiflora materials were dried at 50°C and ground in mills to pass a 1 mm sieve and 100 g placed in 1000 ml of distilled water solvent. The flasks of all the solvents were stoppered and agitated with a magnetic stirrer for 24 h at room temperature. Then the solutions were centrifuged at 3000 g for 10 min. The residue was re-extracted with 500 ml of methanol for 24 h stirring at room temperature and centrifuged again at 3000 g for 10 min. The zataria multiflora water extract were combined. Distilled water was evaporated from the solution at approximately 85°C using a rotary-evaporator [19].

2.3 Treatments and experimental design

The tow doses (0, 0.3 ml/30 ml buffered rumen fluid) zataria multiflora water extract were added to the soybean meal samples.

2.4. *In vitro* gas production

Fermentation of soybean meal samples were carried out with rumen fluid was obtained from three fistulated Taleshi native male cattle fed. The samples were incubated in the rumen fluid in calibrated glass syringes following the procedures of Menke and Steingass [14] as follows. 200 mg dry weight of the sample was weighed in triplicate into calibrated glass syringes of 100 ml in the absence and presence of doses (0.3 ml/30 ml buffered rumen fluid) zataria multiflora water extract. The syringes were pre-warmed at 39°C before injecting 30 ml rumen fluid-buffer mixture into each syringe followed by incubation in a water bath at 39°C. The syringes were gently shaken 30 min after the start of incubation and every hour for the first 10 h of incubation.

Gas production was measured as the volume of gas in the calibrated syringes and was recorded before incubation 2, 4, 6, 8, 12, 24, 48, 72 and 96 hours after incubation. All samples were incubated in triplicate with three syringes containing only rumen fluid-buffer mixture (blank).

RESULTS AND DISCUSSION

3.1. *In vitro* gas production

Gas production volumes (ml/200 mg DM) for soybean meal and zataria multiflora water extract (0.3 ml/30ml buffered rumen fluid) at different incubation times shown were in Figure1 and 2.

Gas production volumes (ml/200 mg DM) for soybean meal and zataria multiflora water extract (0.3 ml/30ml buffered rumen fluid) at different incubation times shown were in Table 1.

The results showed that gas volume at 8 h incubation (for 200 mg dry samples), were 37.81 and 35.32 ml/200 mg DM for soybean meal and zataria multiflora water extract (0.3 ml/30ml buffered rumen fluid) respectively. Gas volume at 12 h incubation (for 200 mg dry samples), were 42.23 and 42.12 ml/200 mg DM for soybean meal and zataria multiflora water extract (0.3 ml/30 ml buffered rumen fluid) respectively. Gas volume at 24 h incubation (for 200 mg dry samples), were 56.38 and 53.72 ml/200 mg DM for soybean meal and zataria multiflora water extract (0.3 ml/30ml buffered rumen fluid) respectively. Gas volume at 48 h incubation (for 200 mg dry samples), were 62.43 and 59.45 ml/200 mg DM for soybean meal and zataria multiflora water extract (0.3 ml/30ml buffered rumen fluid) respectively. Salamat azar et al., [18] estimation effect of tree doses *thyme methanolic extract* (0, 0.15 and 0.3 ml/30 ml buffered rumen fluid) on degradability kinetics, of sunflower meal and report gas volume at 48 h incubation (for 200 mg dry samples), soluble fraction (a), insoluble but fermentable fraction (b), potential gas production (a + b) and rate constant of gas production (c) of sunflower meal were 44.99, 3.60, 49.32, 52.92 ml/200 mg DM and 0.135 ml/h, gas volume at 48 h incubation (for 200 mg dry samples), soluble fraction (a), insoluble but fermentable fraction (b), potential gas production (a+ b) and rate constant of gas production (c) of *thyme methanolic extract* (0.15 ml/30 ml buffered rumen fluid) were 29.91, 0.53, 36.25, 36.79 ml/200 mg DM and 0.049 ml/h, respectively. Rezaei et al., [17] evaluation effect of tree doses *fennel methanolic extract* (0, 0.5 and 1 ml/30 ml buffered rumen fluid) on degradability, of soybean meal and report gas volume at 12 h incubation (for 200 mg dry samples), soluble fraction (a), insoluble but fermentable fraction (b), potential gas production (a + b) and rate constant of gas production (c) of soybean meal were 51.620, 1.767, 70.880, 72.647 ml/200 mg DM and 0.100 ml/h, gas volume at 12 h incubation (for 200 mg dry samples), soluble fraction (a), insoluble but fermentable fraction (b), potential gas production (a + b) and rate constant of gas production (c) of *fennel methanolic extract* (0.5 ml/30 ml buffered rumen fluid) were 54.970, 4.302, 70.919, 75.221 ml/ 200 mg DM and 0.088 ml/h, respectively. Gas volume at 24 and 48 h incubation (for 200 mg dry samples), of soybean meal were 65.370 and 71.240 ml/200 mg DM, while for *fennel methanolic extract* (0.5 ml/30ml buffered rumen fluid) were 65.470 and 71.883 ml/200 mg DM, respectively.

Table 1. Gas production volumes (ml/200 mg DM) for soybean meal and zataria multiflora water extract (0.3 ml/30ml buffered rumen fluid) at different incubation times.

	Incubation times								
Treatments	2	4	6	8	12	24	48	72	96
soybean meal	9.31	20.4	27.61	37.81	42.23	56.38	62.43	64.18	64.59
$ZMWE_{0.3}$	8.44	18.236	26.20	35.32	42.12	53.72	59.45	61.27	61.27

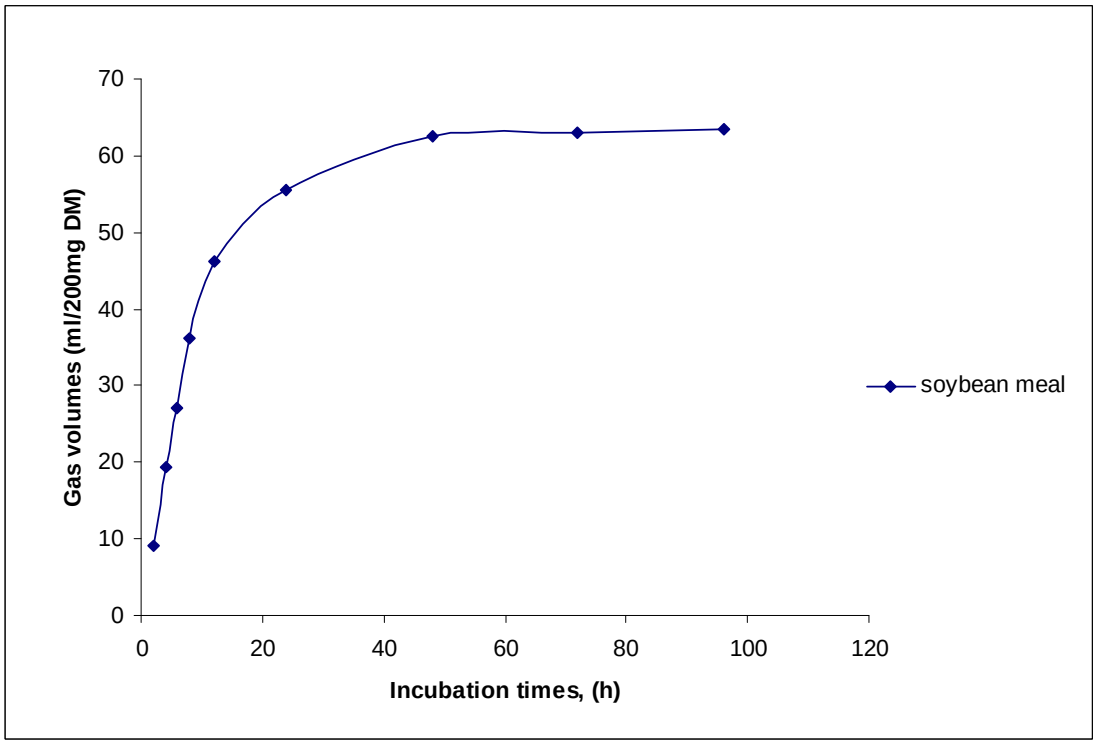

Figure1. Gas production volumes (ml/200 mg DM) for soybean meal at different incubation times.

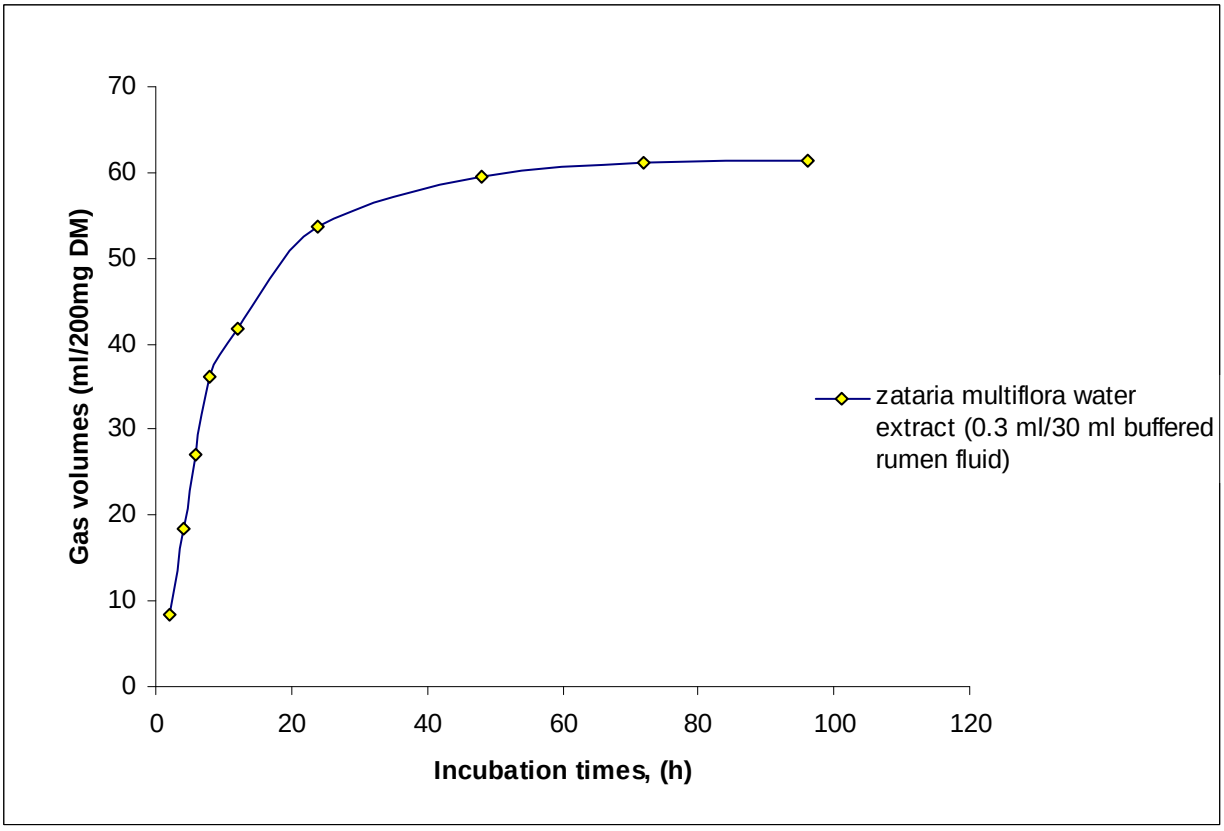

Figure2. Gas production volumes (ml/200 mg DM) for zataria multiflora water extract (0.3 ml/30ml buffered rumen fluid) at different incubation times.

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