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Effects of Molasses and Bacterial Inoculant on Chemical Composition and Aerobic Stability of Sorghum Silage

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ABSTRACT

This experiment was conducted to determine the effects of an inoculant (lactobacillus plantarum, 10^6 g kg^{-1} fresh forage) and molasses (%5), on chemical composition and aerobic stability of sorghum (speedfeed variety) silage. The data were analyzed as Completely Randomized Design with 5 replicates for each treatment. The pH of all treated silage was below 4 and did not significantly differ ($p>0.05$). The DM content was same in all treatments ($p>0.05$). Ash content was greater in silage treated with molasses than inoculants and control ($p<0.05$). Crude Protein (CP) did not significantly differ among treatments ($p>0.05$). Ammonia-N concentration was higher in silage treated with molasses than inoculants and control ($p<0.05$). Neutral Detergent Fiber (NDF) content did not significantly differ among treatments ($p>0.05$). Silage treated with inoculant had greater disappearance Acid Detergent Fiber (ADF) compared to silages treated with molasses and control ($p<0.05$). Crude Fat (CF) was not significantly different among treatments ($p>0.05$). The pH and temperature was measured regularly each 2 h for 12 days after opening silos. Finally silage affected by additives ($p<0.05$). Silage treated with bacterial inoculant had higher pH and temperature (4/66, 26/46) compare to silage treated with molasses (4/49, 24/97) and control (4/03, 24/17) ($p<0.05$). Spoilage in forages tissues was more than in silage treated with bacterial inoculation compare to others ($p<0.05$).

Key words: Bacterial incubation, molasses, chemical composition, aerobic stability, sorghum, silage

INTRODUCTION

Irregular and inadequate supply forage is the critical constraint for profitable livestock production in developing countries (Nisa *et al*., 2008). Iran is a dry land in the world. In this country cannot cultivate forage in all seasons. There for need to plant that can use water more efficiently and struggle with harsh climate. One of important forage for making silo is sorghum. It can sustain in harsh and dry land and use water more efficiently (Sanchez *et al*., 2002). The cultivation of sorghum for silage production has become an increasingly common practice justified by its agronomic characteristics and chemical composition. Some hybrids are characterized by the presence of tannin in the grains (Oliveria *et al*., 2007). To expand the utility of sorghum as a forage crop, breeders have focused on traits likely to affect its yield and nutritive quality. Yield is a reflection of the plant's potential to accumulate high Dry Matter (DM) content in its organs and have high NDF digestibility (Ben-Ghedalia *et al*., 2001). Ensiling is the most important process to store forage. Rapid drop in pH during ensiling and increasing organic acid concentration can inhibit

the growth of undesirable microorganisms (Pahlow *et al.*, 2003). Use inappropriate method and do not have knowledge about making silo, case large amounts of organic material spoiled and converted to CO_2, NH_4 and etc. (Oliveria *et al.*, 2007). There are different objectives in using silage additives. The main target is to prevent secondary fermentation and to decrease butyric acid production. The effectiveness of additives depends on the degree of preventing such fermentation in silages (Aksu *et al.*, 2004). Use appropriate additives can increase nutritional value and have positive effect on silage quality (Weinberg *et al.*, 2002). For better preservation, the forage must have high concentration of fermentable carbohydrates, low buffering capacity, relatively low Dry Matter (DM) content (20-30%) and adequate lactic acid bacteria. Molasses has approximately 70% dry matter. In this kind of fermentation, large amount of crude protein was converted to ammonia (Kung *et al.*, 2000). Molasses have high Ash and crude protein content that may be interfering to buffering capacity. Lactobacillus plantarum raise hemofermentation and decline pH at beginning of ensiling process. Acidic environment can preserve of water soluble carbohydrate (WSC). The low level of residual (WSC) in silage provide additional advantage to aerobic stability. Since excess levels of (WSC) in silage may increase the danger of secondary undesirable fermentation and silage spoilage after its exposure to aerobic condition (Miron *et al.*, 2005). Weinberg *et al.* (1993) used LAB as silage additive for corn, sorghum and wheat and reported that some inoculated silages spoiled faster upon exposure to air. Regression analysis indicated that aerobic deterioration of inoculated silage was associated with high concentration of residual WSC and lactic acid and lack of volute fatty acid. The aim of this experiment was to determine the effects of molasses and bacterial inoculant (*Lactobacillus plantarum*) on sorghum silage.

MATERIALS AND METHODS

Silage preparation: The (speedfeed variety) of sorghum was cultivated during summer of 2008 in North Iran_Gorgan (this experiment started at June and late until November). Whole sorghum was harvested at mid-dough stage. It had approximately 300 g kg^{-1} DM content. It was chopped (1 cm theoretical length of cut) by forage harvester (1260 Auto-max, gehl, Westbend, WI, USA). The chopped sorghum without less time mixed by three treatments: 1-molasses (5%), 2-inoculation (lactobacillus plantarum, used at rate of 106 cfu g^{-1}) and water (control). Each treatment was sprayed on chapped sorghum and completely mixed by hand. Each group of treated forage was packed in five laboratory silo (50 cm diameter ×100 cm height, 20 kg capacity).during ensiling, forage was compacted until air between particle size get off and than all silo covered by plastic and finally put door. After ended ensiling, All silage stored in dark and cold (20-25) place (Aksu *et al.*, 2006).

Chemical analysis: All silage was opened after 60 days of ensiling. Ensiled forage samples were dried in a forced air oven at 60°C for 48 h (Miron *et al.*, 2005). After drying, samples grounded to 1 mm. Residual ash assayed at 600°C for 4 h (Miron *et al.*, 2005). Crude protein and fat was determined by auto kjeldahl and soxtec that described by (Official Method 4.2.11 and 4.5.05 according to AOAC (2005). Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) contents determined by method Van Soest *et al.* (1991). The pH of each sample was determined in triplicate using approximately 25 g ensilage added to 100 mL of distilled water. After hydration for 10 min using a blender, the pH was measured using a digital pH meter (Aksu *et al.*, 2006). A subsample of the filtrate (10 mL) was combined with 0.8 mL of 650 g L^{-1} trichloroacetic acid (TCA), refrigerated on ice for 30 min and centrifuged at 16500×g for 20 min at 4°C as described by

Broderick and Kang (1980). Temperature also measured by digital thermometer. The pH and temperature was measured regularly every 2 h during 12 days.

Statistical analyses: Data were analyzed using the general linear model procedure of SAS Institute (2005). The all data for chemical composition were analyzed as a completely randomized design with five treatments and five replications per treatment. Data was analyzed using the following model:

$$\gamma_{ij} = \mu+\beta_j +\varepsilon_{ij}$$

where, γ_{ij} is the observation (parameter), μ the overall mean, β_j the effect of treatment and εij is the residual error.

Data from aerobic stability was analyzed as a Repeated Measure Design. The model is:

$$Y_{ijkl} = \mu+\alpha_i+Tl+(\alpha T)il+e_{ijkl}$$

where, γ_{ijkl} is the observation (parameter), μ the overall mean, aj the effect of treatment, Tl effect of time, (aT)il is alternate effect between time and treatment and e_{ijkl} is the residual error.

RESULTS AND DISCUSSION

Table 1 show results of chemical composition in silage that treated with five treatments. The pH in all silage was similar and below 4. Ash concentration was significantly higher in silage treated with molasses than bacterial inoculant ($p<0.05$). Molasses have high content of minerals that can increase ash. The high content of minerals in silage treaded with molasses may have resulted high Ash content in this silage. This result according with (Gofeen and KHalifa, 2007; Aksu *et al.*, 2006; Donmez *et al.*, 2003). Silage treated with molasses had higher ammonia-NH3 than other silages ($p<0.05$). Results show that proteolysis may be increased by using molasses (Guo *et al.*, 2007) Molasses has high concentration of soluble carbohydrate that can stimulate heterofermentation process in silage but could not inhibit proteolysis (Aksu *et al.*, 2006). Clostridia can grow in silage that has high soluble carbohydrate. They can degrade protein to ammonia (Ward *et al.*, 2001). Deamination process decrease in low pH (Slottner and Bertilsson, 2006). High concentration ammonia-NH3 in silage has not negative effect in ruminant (Randby, 2000).

Silage treated with bacterial inoculant had higher disappearance ADF compare others ($p<0.05$). Bacterial inoculant stimulates growing lactic acid bacterial. They product acid lactic and decrease pH. Acidic condition can solve hemicelluloses. Finally it converted to simple sugar and ADF content decrease. Acidific condition also can protect sugar and preserve sugar of spoiling and decrease

Table 1: Effect of treatments on chemical composition of sorghum silages after 60 days of ensiling

Treatments	pH	(DM%)	Ash (DM%)	CP (DM%)	NH_3 (g kg^{-1})	NDF (DM%)	ADF (DM%)	CF (DM%)
Molasses	4.49±0.031	26.55±0.58^{c}	8.24±0.18^{a}	6.53±0.15^{bc}	4.21±0.07^{a}	62.54±1.05	44.0±1.09^{b}	4.55±0.11^{b}
Bacterial	4.66±0.028	27.16±0.73^{bc}	6.64±0.18^{b}	6.23±0.32^{c}	3.99±0.052^{c}	56.41±5.61	48.8±0.92^{a}	5.75±0.27^{a}
Control	4.03±0.043	27.75±0.39^{abc}	8.00±0.25^{ab}	6.20±0.22^{c}	3.99±0.035^{c}	57.00±2.73	43.0±0.7^{b}	4.71±0.041^{b}
F-value	42.64	4.08	1.52	4.43	4.43	1.71	6.21	3.4
p-value	0.0001	0.005	0.2347	0.01	0.01	0.223	0.0025	0.036

Values with different letters are significantly different

starch content (Slottner and Bertilsson, 2006). Although lactic acid bacterial make a safe condition to growing useful microorganism (Slottner and Bertilsson, 2006). The concentration of Crude Fat (CF) was affected by bacterial inoculant ($p<0.05$). After harvesting plant, enzymes in forage start to digestion cell content. At beginning silage process Lactic acid bacterial grow very fast in silage treated by bacterial inoculant. Lactic acid decline pH immediately and prevent activity of lipase enzyme. In other hand bacterial inoculant makes a cold fermentation and protect of plant components (Kung *et al*., 2000).

The result showed that pH and temperature in silage treated by bacterial inoculation significantly increase than silage treated by molasses and control ($p<0.05$). In silage treated by bacterial inoculation (*Lactobacillus platarum*) hemofermentation stimulate and product only lactic acid. It is consumed by clostridia as source of energy (Pahlow *et al*., 2003). Finally lactic acid concentration decrease, upset pH increase and condition get safe for growth undesirable microorganisms (Fig. 2). Molasses stimulate heterofermentation. This kind of fermentation products butyric acid and acetic acid they have antimicrobials activity and eliminate growing fungi, mold and yeast but increase aerobic stability in feed out phase compare bacterial inoculation that decrease aerobic stability (Fig. 1). When clostridia grow on silage degraded organic materials and product CO_2, CH_4 and heat that increase mass silage. During this process the silage tissues change and degraded (Hassant *et al*., 2007).

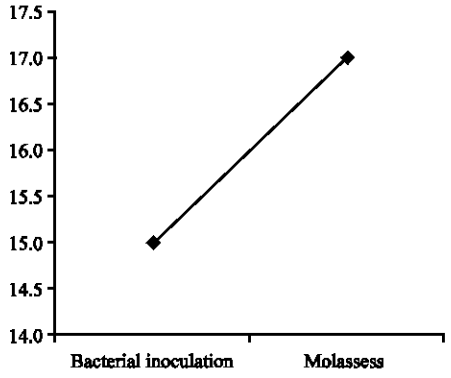

Fig. 1: Silage tissues quality in two type of silos

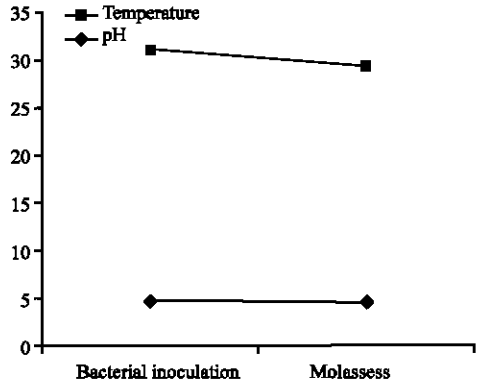

Fig. 2: Average of pH and temperature in different kinds of treatments after 12 days

CONCLUSION

Bacterial inoculation preserves forage properly and make cold fermentation that can protect of nutrient materials but could not supply silage in feed out phase. Molasses increase nutrient content and with acetic acid and butyric acid protect silage against spoilage. Molasses with carbohydrate concentration increase heterofermentation and decline pH, preserve of organic materials and increase palatability. *Lactobacillus plantarum* stimulate hemofermentation and increase lactic acid concentration finally decrease pH and maintenance organic materials. Bacterial inoculant was not successful in feed out phase.

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