

ISSN 1819-1878

Asian Journal of
Animal
Sciences

The Effect of Different Levels of Molasses as Silage Additives on Fermentation Quality of Foxtail Millet (*Setaria italica*) Silage

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Abstract: The aim of this study was to examine the effect of different levels of molasses on fermentation quality and nutritive value of foxtail millet silage. Silage of foxtail millet is tested in micro-silos, treatments included control (no additive), 2.5, 5 and 7.5% molasses. Silages were assessed using both method of appearance evaluation and method of DM, pH, CP, NDF, ADF, TVFA, WSC, aerobic stability and DM degradation. Each of treatments was determined after 60 days. Treated silages had better appearance quality than the control silages in Flieg's method. All of the silages had good and very good degrees in the method based on DM, pH. The treated silages had lower pH compared with the control ($p < 0.05$). The control group had lower DM content than treated silages ($p < 0.05$). The concentration of ADF, NDF and WSC in the treated silage decreased with increasing level of addition molasses ($p < 0.05$). The amount of CP, DDM, DE, ME, TDN and TVFA in the treated silages increased with increasing level of addition molasses ($p < 0.05$). Application of 7.5% molasses as an additive to foxtail millet resulted in improved degradation dry matter and aerobic stability ($p < 0.05$).

Key words: Silage, foxtail millet, molasses, degradability, aerobic stability

INTRODUCTION

The summer annual grasses are of considerable importance to livestock producers. They have the advantages of making rapid growth, giving good forage yield and, if cut at the proper stage of growth, providing good forage quality. The major summer annual grasses are the sorghums, sudangrasses and millets (pearl millet, browntop millet, foxtail millet and Japanese millet). German millet and Hungarian millet are types of foxtail millet (Baltensperger, 2001). Foxtail millet is some of the best species for drought conditions that they have rapid rate of growth, high ability of adaptation with tropical region, high percentage of protein. They are very leafy and multi-stemmed, allowing them to regrow after grazing or cutting. They can produce as much tonnage as corn with about 60% as much rainfall as corn needs. Summer annual grasses can be used for grazing, hay, silage, or greenchop. Millet should be harvested at the mid-dough stage for ensiling (Hamilton *et al.*, 2005).

In the research had done by Hamilton *et al.* (2005) the process of vacuum ensilage of '*Setaria sphacelata* (cv. Nandi) (33% DM; 7% soluble carbohydrates) was studied. The silage was well preserved in a chemical sense (pH 4.5; lactic acid 1.7%, volatile acids 1.2% DM; volatile bases 9.8 % total N) but, because of the structural rigidity of the harvested grass, air could not be completely excluded even from the polythene-covered vacuum stack. The temperature reached 43°C in the first week of storage and considerable surface wastage occurred. The silage (DM digestibility 42%, voluntary DM intake 81 g kg⁻¹0.75) was of poorer quality than the grass harvested (DM digestibility

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54%, voluntary DM intake 84 g kg^{-1} 0.75). The poor nutritive value of the silage is attributed to the nature and composition of the material ensiled rather than to any defect in the ensilage process itself and may be a feature of most silage made from tropical grasses.

The reason for these finding is that without additives, setaria can be turned into acceptable silage with pH of 4.07 but the pH of the silage was reduced with the addition of 4% molasses and it's quality was improved (Aminah *et al.*, 2001).The ensilage of two harvests of *Setaria sphacelata* (Nandi) and *Chloris gayana* (C.P.I. 16144) with and without added sucrose has been examined in laboratory silos. Without added sucrose, second harvest *S. sphacelata* and both harvests of *C. gayana* produced acetic acid silage. With added sucrose, silages were initial acidification to pH c. 3.8, rapidly (Catchpoole, 2003).

Moisture content at the time of harvest (too wet or too dry) may also be a problem if the proper maturity is not matched to the area. A good fermentation process is not only dependent on the type and quality of the forage crop, but also on the harvesting and ensiling technique. Attention to details such as speed of harvesting, moisture content, length of chop, silage distribution and compaction can greatly influence the fermentation process and storage losses. Molasses is commonly used to provide readily available energy for lactic acid fermentation and it has positive effects on pH and lactic acid levels (Haigh, 1988; McDonald *et al.*, 1991; Snyman and Joubert, 1996). When major bacterial population is lactic acid bacteria, fermentation products are mainly lactic acid and acetic acid and ethanol at low level, this type of silage increases dry matter intake, dry matter and organic matter digestibility and thus increase animal performance in ruminants. It was reported that molasses stimulates silage fermentation but it is not able to prevent proteolysis enough due to slow reduction in pH with molasses addition (Baytok *et al.*, 2005).

The objective of this study was to determine the effect of different levels of molasses as silage additives on quality fermentation, ruminal digestibility and aerobic stability of foxtail millet silage.

MATERIALS AND METHODS

This study was conducted in Gorgan University of Agriculture Sciences and Natural Resources, autumn 2006. Whole foxtail millet was harvested by a one row forage harvester at dough stage of kernel maturity. Four different silages were prepared from chopped forage. Silage treatments included control (no additive), 2.5, 5 and 7.5% molasses. Ensiling were performed by stamping as much of chopped plant material into the barrels as possible. By this action most of the air was excluded. After ensiling each barrel was sealed off tightly with a lid. The barrels were then stored for 60 days in a dark room with a temperature ranging between 20-25°C. All of the silos for each treatment were opened after 60 days of ensiling. The DM content of fresh forage and silage samples was determined by drying (70 to 75 g) in a forced-air oven at 60°C for 48 h. After drying, samples were ground. Ten gram samples of silage from each treatment were diluted 100 mL sterile deionized water and blended for 2 min. Silage pH determined immediately. Acid detergent fiber and Neutral detergent fiber was analyzed by using sulfite and amylase (Van Soest *et al.*, 1991). Total nitrogen was determined after total combustion (LECO Corporation, CNS 2000 Analyzer; St. Joseph, MI) and CP was calculated by multiplying total nitrogen by 6.25. Starch was analyzed by using the method described by Poore *et al.* (1993). Fresh millet silage (25 g) was added to a dilution bottle that contained 225 mL of sterile quarter-strength Ringer's solution (Oxoid BR54; Unipath, Basingstoke, UK) and homogenized in a blender for 1 min. After blending, water extracts were filtered through Whatman 54 filter paper (Whatman Inc., Clifton, NJ), acidified with 50% sulfuric acid and frozen before analysis of water-soluble carbohydrates (WSC) (Nelson, 1944). The pH of silage extracts was determined within 20 min of homogenization. After 60 days of ensiling, when silos were opened, silage was mixed well and a 2 kg sample was returned to

its respective silo. No physical packing of the silage took place. A thermometer was placed in the geometric center of each silage mass and temperatures were recorded every 3 h. A double layer of cheesecloth was placed on the top of each silo to prevent contamination but allowed for penetration of air because silos were incubated between 21 and 22°C. Aerobic stability was defined as the number of hours before a 2°C increase in temperature of the silage mass relative to ambient temperature (Mills and Kung, 2002). The temperature and pH of silages were recorded for 144 h, each 2 h once. For the determination of *in situ* ruminal DM, dry millet silage samples were ground to pass through a 2 mm screen and 0.5 g of silage was weighed into Dacron bags (5×10 cm, 53- μm pore size; Ankom, Fairport, NY). Duplicate bags were placed in the rumen of a fistulated sheep fed a diet that was 82.4% alfalfa hay/grass hay, 8.8% soybean meal and 8.8% dry rolled barely. Bags were incubated for 0, 4, 8, 16, 24, 48 and 72 h. After removal from the sheep, bags were washed with water until effluent was clear and then were dried at 55°C for 48 h. Dried bags were weighed and DM disappearance was determined. The chemical composition of Foxtail millet and silages was analyzed in a completely randomized design by the general linear models procedure of SAS/STAT (1996). Factors were ADF%, NDF%, CP%, WSC%, TVFA (Mmol kg⁻¹), DDM%, DE (Mmol kg⁻¹), ME (Mmol kg⁻¹), TDN%. Means were separated by Duncan test at 0.05 probability level (Duncan, 1955). The kinetics of ruminal DM disappearance *in situ* was estimated by the Naway software.

RESULTS

Appearance Evaluation

In this study, each of the factors including like to smell, color and structure (appearance characters) based on Flieg's score has given a number (Horiguchi and Takahashi, 2007). Treated foxtail millet silages with the additives had better appearance than Control group. All of levels of molasses in this study produced good silage.

In this evaluation method, the maximum of pH for stating in very good degree silage with 30-35% DM is 4.3-4.5. In the present study, average DM was 32% and pH ranged from 3.93 to 4.88, therefore all of silages were stated in good and very good class (Table 1).

Table 1: Appearance evaluation based on dry matter and pH Flieg's method

Additive type ¹	DM (g kg ⁻¹)	pH	Judgment
Control	311.1 ^b	4.88 ^a	Good
2.5%	329.0 ^a	4.00 ^a	Very good
5%	327.5 ^a	3.93 ^a	Very good
7.5%	326.0 ^a	4.10 ^a	Very good

¹Control = Without any additive. 2.5% Molasses, 5% Molasses, 7.5% Molasses. Unlike superscript in a row differ significantly (p<0.05)

Table 2: Chemical compositions of foxtail millet forage before ensiling

Items	Fresh foxtail millet forage
Dry matter (g kg ⁻¹)	303.0
Acid detergent fibre (g kg ⁻¹)	334.1
Neutral detergent fibre (g kg ⁻¹)	547.2
Crude protein (g kg ⁻¹)	105.3
Digestible dry matter ² (g kg ⁻¹)	629.0
Digestible energy ³ (Mcal kg ⁻¹)	26.0
Metabolizable energy ⁴ (Mcal kg ⁻¹)	21.6
Total digestible nutrition ⁵ (g kg ⁻¹)	613.1

¹Control = With out any additive. 2.5% Molasses, 5% Molasses, 7.5% Molasses ²Digestible Dry Matter = 88.9-0.779 (ADF) ³Digestible Energy = 0.027+0.0427(%DDM) ⁴Metabolizable Energy = DE *0.821⁵ Total digestible nutrition = DE /0.04409 (Khalil *et al.*, 1986)

Chemical Composition

The treated silages had lower pH compared with control ($p < 0.05$) (Table 2). The control group had lower DM content than treated silages ($p < 0.05$). Content of ADF and NDF treated silage with 7.5% molasses were lower than other treatments ($p < 0.05$). The amount of CP in the treated silage with 5% molasses was more than other treatments and control group ($p < 0.05$). The WSC concentration of treated silage with 7.5% molasses were lower than other samples ($p < 0.05$). TVFA in silages content of 5 and 7.5% molasses were more than treatment of 2.5% and control group ($p < 0.05$). There are significant differences for the concentration of DDM, DE, ME, TDN in the silages and treated silages with 7.5% molasses had higher contents than the others ($p < 0.05$) (Table 3).

In situ Ruminal DM Digestibility

Coefficients and SEM of *in situ* ruminal DM digestibility is presented in Table 4. Addition of the additives to silages affected *in situ* ruminal DM digestibility of treated silages. So that, the coefficient a (That part of feed which is soluble in water) increases *in situ* ruminal DM digestion of silages with 5% molasses additive and decreases silage with 7.5% molasses. The coefficient b (That part of feed which digested by microorganisms of rumen) of silages with 7.5% molasses was the highest (0.534) and the coefficient b of silages with 5% additive was the highest (0.408). The coefficient c (The rate of degradation of part b) was the lowest in 7.5% treatment. a+b or potential degradability of dry matter in silage with 7.5% molasses was more than other treatments.

Aerobic Stability

pH value of control silage (Fig. 2) after 144 h exposure to air changed from 4.88 to 6.19 ($p < 0.05$). Temperature of control silage (Fig. 1) after 48 h exposure to air 22°C increased and after 144 h received to own peak of temperature (43.05°C). Aerobic stability of silages have showed in Fig. 3.

Table 3: Chemical composition foxtail millet silage after 60 day of ensiling

Items	Control	Treatment ¹		
		2.5%	5%	7.5%
Acid detergent fibre (g kg ⁻¹)	495.80 ^a	488.00 ^a	374.80 ^b	336.40 ^c
Neutral detergent fibre (g kg ⁻¹)	612.60 ^a	618.20 ^a	550.60 ^b	514.80 ^c
Crude protein (g kg ⁻¹)	108.00 ^b	105.00 ^b	118.20 ^a	91.20 ^c
Total volatile fatty acid (Mmol kg ⁻¹)	638.20 ^b	675.60 ^b	733.00 ^a	698.80 ^a
Water soluble carbohydrates (g kg ⁻¹)	11.60 ^b	15.60 ^a	11.03 ^{bc}	9.30 ^c
Digestible dry matter ² (g kg ⁻¹)	609.70 ^b	506.40 ^d	592.20 ^c	627.50 ^a
Digestible energy ³ (Mcal kg ⁻¹)	2.40 ^c	2.18 ^d	2.60 ^b	2.71 ^a
Metabolizable energy ⁴ (Mcal kg ⁻¹)	2.02 ^c	1.79 ^d	2.15 ^b	2.22 ^a
Total digestible nutrition ⁵ (g kg ⁻¹)	579.80 ^c	496.60 ^d	594.50 ^c	616.00 ^a

¹Control = Without any additive, 2.5% Molasses, 5% Molasses, 7.5% Molasses, Digestible Dry Matter = 88.9-0.779(ADF), ²Digestible Energy = 0.027+0.0427(%DDM), Metabolizable Energy = DE *0.821, Total digestible nutrition = DE / 0.04409 (Khalil *et al.*, 1986), Unlike superscript in a row differ significantly ($p < 0.05$)

Table 4: Apparent *in situ* DM digestion of foxtail millet silage

Items	Control	Treatment ¹		
		2.5%	5%	7.5%
a ²	0.071	0.074	0.111	0.060
SEM a	0.018	0.02	0.017	0.015
b ³	0.420	0.418	0.408	0.534
SEM b	0.024	0.023	0.025	0.025
c ⁴	0.060	0.062	0.055	0.039
SEM c	0.010	0.009	0.008	0.005
a+b ⁵	0.491	0.492	0.519	0.594

¹Control = Without any additive, 2.5% Molasses, 5% Molasses, 7.5% Molasses, ²a: That part of feed which is soluble in water ³b: That part of feed which digested by microorganisms of rumen ⁴c: The rate of degradation of part b ⁵a+b: Potential degradability of dry matter

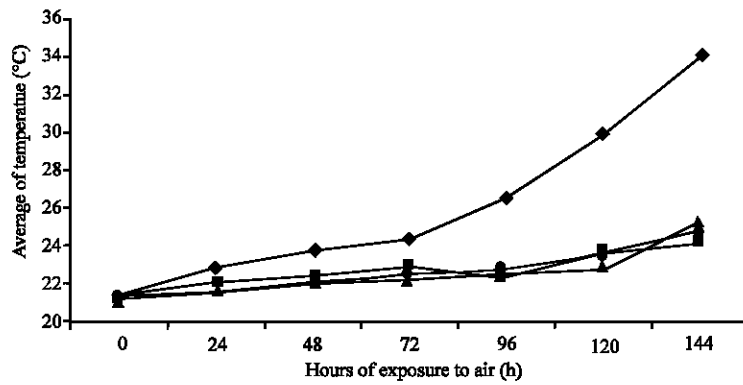


Fig. 1: Temperature changes of foxtail millet silage after exposure to air until 144 h. (◆) Control = Without any additive, (▲) 2.5% Molasses, (●) 5% Molasses, (■) 7.5% Molasses

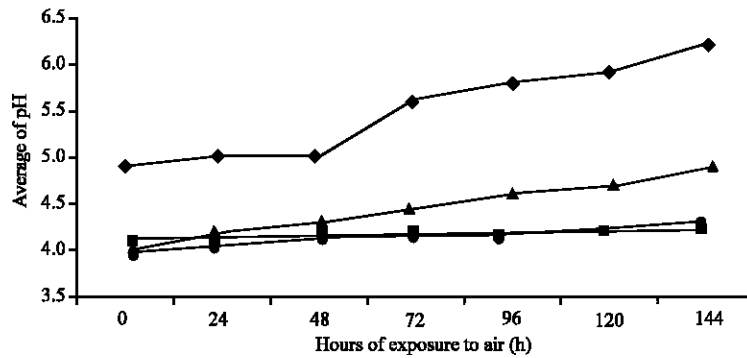


Fig. 2: pH changes of foxtail millet silage after exposure to air until 144 h. (◆) Control = Without any additive, (▲) 2.5% Molasses, (●) 5% Molasses, (■) 7.5% Molasses

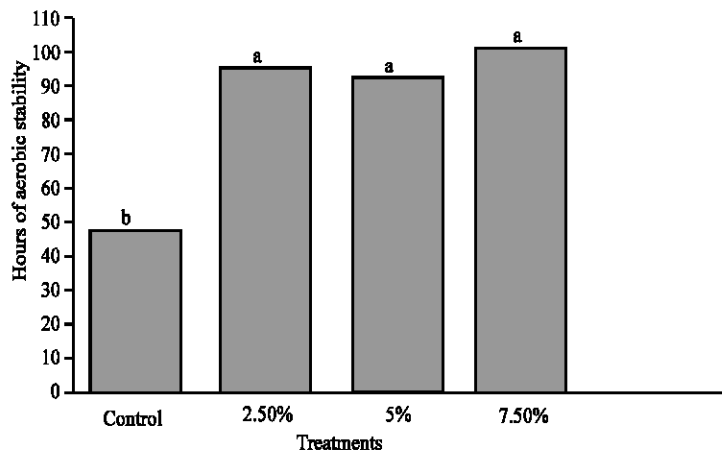


Fig. 3: The aerobic stability of foxtail millet silage. Control = Without any additive, 2.5% Molasses, 5% Molasses and 7.5% Molasses. ^{a,b}Bars with unlike letters differ ($p < 0.05$)

DISCUSSION

Appearance Evaluation

Setaria can be ensiled into reasonable silage, but the quality can be improved with the addition of molasses before ensiling (Aminah *et al.*, 2001). Applications of molasses additive improve quality of fermentation silage (McDonald *et al.*, 1991).

Chemical Composition

Molasses stimulates silage fermentation and reduce pH of silages (McDonald *et al.*, 1991; Aminah *et al.*, 2001; Baytok *et al.*, 2005). Observations about of DM showed that the control group had lower DM content. The reason for this finding is that addition molasses to silage and stimulating fermentation, increase population of lactic acid bacteria, improve quality of silage and avoid of losses dry matter (McDonald *et al.*, 1991). Addition and mixing some additives to forage before ensiling resulted to increase dry matter content of silage due to improve quality of fermentation (Harrison and Blauwikel, 1994).

In this experiment content of ADF and NDF of silages decrease with increase percentage of molasses. These data showed that molasses is a stimulant of silage and caused to increase analysis in cell wall (Baytok *et al.*, 2005). One half of hemi-cellulose analysis during of fermentation silage. Molasses is a stimulant fermentation additive because of containing sugar that utilize by microorganisms as the nutrition matter and increase their fermentation activity (McDonald *et al.*, 1991).

Molasses had 8.5% CP, On the other hand, additives containing carbohydrates result to decrease ammonia-N by stimulating fermentation and via this effect improve amount and quality of protein silage (McDonald *et al.*, 1991).

In this experiment WSC concentration of treated silage with 7.5% level of molasses were more than others. Non structural carbohydrates are water soluble mainly and ferment in silage during the fermentation. After fermentation remain very little water soluble carbohydrate which has produced by enzyme activity and hemi-cellulose hydrolysis because lactic acid bacteria don't attack to starch (McDonald *et al.*, 1991).

Results of this study detected that with increase level of molasses additive to silage, increase amount of TVFA. Additives containing of carbohydrates provide necessary energy for lactic acid bacteria to producing lactic acid. Usually in such silages, pH decrease, concentration of lactic acid increase and final quality of silages improve. Treated silages with 7.5% Molasses had higher contents of DDM, DE, ME, TDN than the other treatments, these finding is harmonic with decrease in ADF because calculation of these parameter related to ADF. Reducing ADF in the similar studies reported which it is due to effect of molasses on raising fermentation silage (McDonald *et al.*, 1991; Baytok *et al.*, 2005).

In situ Ruminal DM Digestibility

The reason of vacillations in amount of the coefficient a may be related to the different degree of the silage fermentation and production more volatile fatty acids. Usually feeds which have lower coefficient a, they have higher coefficient b. The coefficient c was low in treatment 7.5% molasses because the coefficient a was low, it can reduce rate of growth of rumen microorganisms and the coefficient b was high which can caused to slow and monotonous analysis feeds into the rumen. It institute fixed situation in the rumen. Feeds remain into the rumen for a longer time and rate of pass decrease. These factors caused to more utilization of nutrients. a+b or potential of degradability of dry matter in silage with 7.5% molasses was more than other treatments, these data showed that *in situ* ruminal DM digestibility of this treatments were higher than others that it due to more digestion nutrients (Nocek and Grant, 1987).

Aerobic Stability

Under aerobic situation some of species microorganisms analyses lactic acid to CO₂ and H₂O. This reaction caused to rise of pH silage. Then unfavorable microorganisms grow and degenerate silage (McDonald *et al.*, 1991). Among of experimental levels additive, additive 7.5% molasses was more positive effect on the inhibiting of raising temperature of silages, but generally all of the levels of additive had significant effect on increasing aerobic stability, which is similar to results of Umana *et al.* (1991) and Soderholm *et al.* (1998). Source energy silage additives decreased pH silage (McDonald *et al.*, 1991; Baytok *et al.*, 2005) and established unsuitable manner for unfavorable microorganisms therefore the degradation into silage was exhausted.

CONCLUSION

It was found that without additives, setaria can be turned into acceptable silage but the quality, nutrition value, DM digestibility and aerobic stability of the Foxtail millet silage was improved with the addition of 7.5% molasses. These results show that use of molasses as an additive can partially, but not totally, compensate for poor silo management practices. However, rapid filling of silos and achieving adequate packing densities to exclude excessive air should still be high priorities for making excellent quality silage.

ACKNOWLEDGMENT

The authors would like to thank the Gorgan University of Agricultural Science and Natural Resources.

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