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Impact of Biological and Chemical Additives on Fermentation Characteristics of Seasonal Pasture Hay and Green Maize Based Silage

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ABSTRACT

The present study was envisaged to study the impact of biological and chemical additives on fermentation characteristics of seasonal pasture hay (PH) and green maize based silage. Different silages were prepared by using green maize fodder and seasonal pasture hay in the proportion of 10:0 & 7:3 ratioin plastic jars of 3 kg capacity by adding common salt @ 0.5%, urea @1% and molasses @ 1.5% in each silages with seven different treatments *viz.* Control (only green maize), PH (green maize and seasonal pasture hay in 7:3 ratio), X (PH added with xylanase), LP (PH added with *L. plantarum*), LF (PH added with *L. fermentum*), LPLF (PH added with both bacterial inoculants) and XLPLF (PH added with xylanase and both bacterial inoculants). Xylanase, *L. plantarum* and *L. fermentum* were used @ 1500 IU/g, 1 x 10^6cfu/g and 2 x 10^6cfu/g, respectively. All silages were evaluated for their fermentation characteristics after 45 days of ensiling. Silage pH was reduced significantly ($p<0.001$) in all inoculated silages. Levels of lactic acid and TVFA were significantly ($p<0.001$) higher in all inoculated silages as compared to PH silage. While, WSC content was significantly ($P<0.01$) decreased in all inoculated silages. Additives had no significant effect on NH_3-N content of silages. Total N content was found significantly ($p<0.01$) higher in LP silage as compared to PH silage. It was concluded that xylanase and LAB inoculants improve fermentation quality of silage.

KEYWORDS: Bacterial inoculants, Green maize, Seasonal pasture hay, Silage, Xylanase

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INTRODUCTION

India faces deficiency of green and dry fodder due to urbanization and reduction in cultivated land area. There is a gap between demand and supply of green forage, dry forage and concentrate. An overall deficit of green fodder, dry fodder and concentrate was 11.24, 23.4 and 28.9%, respectively (Roy et al., 2019). Alternatives to overcome the shortage in fodder requirement are to increase the area under fodder cultivation or the judicious use of existing fodder resources or preservation of forage when in surplus. In India, surplus green fodders are available in rainy season, which can be preserved in the form of silage for their utilization as green fodder in later date. In India, low quality feedstuff like seasonal pasture hay is available in huge amount andits use in silage may improve its quality and there by its utilization in animal feeding. Maize (*Zea mays*) is the most suitable crop for silage preparation because of its relatively constant nutritive value, high yield and having high concentration of soluble carbohydrates for fermentation to lactic acid (Hundal et al., 2019).

The primary goal of making silage is to maximize the preservation of original nutrients in the forage crop for the feeding at later date. However, considerable amount of nutrient losses occur during ensiling process. Silage additives can be used to improve the ensiling process and reduce the nutrient losses with subsequent improvements in animal performance. Silage additives arenatural or industrial products added in smaller quantities to the forage or grain mass that will promote rapid fermentation process, reduce fermentation losses by limiting the extent of fermentation and improve nutrient composition of silage (Yitbarek and Tamir, 2014). The most commonly used silage additives are molasses, which is a source of fermentable carbohydrate and urea that provides fermentable nitrogen for microorganisms in the silage and rumen of the animal. Urea also has buffering capacity by raising the pH of the silage at early stage of fermentation that might inhibit yeast growth (Kebede et al, 2018).

Biological additives like bacterial inoculants are used to increase the rate of acidification of ensiled forages (Weinberg and Muck, 1996)by accelerating the initial phase of the ensiling process via the rapid fermentation of water soluble carbohydrate (WSC) into lactic acid, along with a subsequent rapid decrease in pH (Hashemzadeh et al., 2014). Chemical additives like enzymes may improve digestibility of nutrients via numbers of mechanism that include direct hydrolysis of sugar, change in gut viscosity and change in the site of digestion (Kung Jr, 2010). The main function of the exogenous fibrolytic enzymes is to supply maximum amount of nutrients from the digestible, potentially digestible and indigestible fractions of the cell wall (Mocherla et al., 2017). Considering huge availability of seasonal pasture hay at cheaper price in India, and role of biological and chemical additives in silage production, the present study was envisaged to study the effects of biological and chemical additives on quality and *in vitro* rumen fermentation pattern of seasonal pasture hay and green maize based silage.

MATERIALS AND METHODS

The present study was conducted atDepartment of Animal Nutrition, College of Veterinary Science and A. H., Kamdhenu University, Junagadh, Gujarat.)

Preparation of silage

Different silages were prepared by using green maize fodder and seasonal pasture hay in the proportion of 10:0 & 7:3 ratio in plastic jars of 3 kg capacity (3 replication in each) by adding common salt @ 0.5%, urea @ 1% and molasses @ 1.5% in each silages with seven different treatments *viz.* Control (only green maize), PH (green maize and seasonal pasture hay in 7:3 ratio), X (PH added with xylanase), LP (PH added with *L. plantarum*), LF (PH added with *L. fermentum*), LPLF (PH added with both bacterial inoculants) and XLPLF (PH added with xylanase and both bacterial inoculants). Xylanase, *L. plantarum* and *L. fermentum*were used @ 1500 IU/g, 1 x 10^6cfu/g and 2 x 10^6cfu/g, respectively. All silages were evaluated for their fermentation characteristics and *in vitro* rumen fermentation pattern after 45 days of ensiling.

Estimation of silage characteristics

Organoleptic criteria are the most important way to judging the silage quality. For this colours of silageswere observed visually. For estimation of silage pH, total volatile fatty acid (TVFA) and ammonia nitrogen(NH_3-N), water extracts of silageswere prepared by adding 90 ml of distilled water to 10 g fresh silage sample in a beaker and homogenize by mechanical homogenizer. A few drops of 0.1% mercuric chloride were added into the extract and kept in refrigerator at 4!. After 48 hrs, material was filtered through four layer muslin cloth and stored at 4!. The pH of silage was measured by pentype pH meter. TVFA and NH_3-N were analyzed as per the methods given by Barnett and Reid (1957),and Conway(1957), respectively. Lactic acid was analyzed as per the method of Barker and Summerson (1941). Silage samples were oven dried at 60±5 deg c. The dried samples were ground to pass through a 1 mm screen for analysis of WSC as per the method of Yemm and Willis (1954).

Statistical analysis

The data were analyzed for descriptive statistics (mean and standard error). Treatment effects on different parameters were analyzed by one way analysis of variance according to Snedecor and Cochran (1994). Pair wise mean difference between groups were compared by Duncan's New Multiple Range Test as modified by Kramer (1957) for the significance at $p<0.05$.

RESULTS AND DISCUSSION

The colour of all silages was golden yellow. Data pertaining to pH, lactic acid, WSC, TVFA, NH_3-N and total nitrogen (N) content of silages after 45 days of ensiling are presented in Table 1.

Table 1. Effects of bacterial inoculants and xylanase on silage characteristics of different experimental silages

Treatments	Parameters					
	pH	LA (g/100g silage)	WSC (g/100g DM)	TVFA (mmol/100g DM)	NH_3-N (g/kg silage)	Total N (g/100g DM)
Control	4.41^{d} ± 0.13	4.73^{b} ± 0.06	1.46^{a} ± 0.22	19.76^{a} ± 1.48	3.30^{b} ± 0.03	1.38^{a} ± 0.11
PH	4.31^{cd} ± 0.04	4.33^{a} ± 0.07	2.14^{b} ± 0.05	21.61^{a} ± 0.85	3.13^{ab} ± 0.04	1.32^{a} ± 0.08
X	4.08^{bc} ± 0.07	5.42^{c} ± 0.15	1.59^{a} ± 0.07	43.24^{b} ± 1.79	3.08^{a} ± 0.03	1.44^{a} ± 0.13
LP	3.75^{a} ± 0.16	6.37^{e} ± 0.13	1.40^{a} ± 0.12	48.60^{c} ± 1.91	3.06^{a} ± 0.09	1.84^{b} ± 0.11
LF	3.95^{ab} ± 0.04	5.71^{d} ± 0.10	1.49^{a} ± 0.10	49.43^{c} ± 2.11	3.16^{ab} ± 0.08	1.43^{a} ± 0.11
LPLF	3.98^{ab} ± 0.03	5.79^{d} ± 0.10	1.46^{a} ± 0.19	41.56^{b} ± 1.95	3.21^{ab} ± 0.10	1.21^{a} ± 0.04
XLPLF	3.87^{ab} ± 0.07	5.88^{d} ± 0.07	1.43^{a} ± 0.03	53.60^{c} ± 1.84	3.08^{a} ± 0.03	1.44^{a} ± 0.09
P value	<0.001	<0.001	0.008	<0.001	0.166	0.009

Values with different superscripts[a-e] within a column significantly varied ($p<0.05$).
LA- lactic acid; WSC- water soluble carbohydrate; TVFA- total volatile fatty acid; NH_3-N- ammonia nitrogen

Results from the present investigation denoted thatafter 45 days of ensiling, silage pH was significantly ($P<0.001$) reduced in all inoculated silages as compared to control (4.41± 0.13) being lowest in *L. plantarum* (3.75 ± 0.16) added silage.

All silage additives significantly ($P<0.01$) increased lactic acid content in silage as compared to PH (4.33 ± 0.07) and control (4.73± 0.06), being highest in *L. plantarum* (6.37 ± 0.13) added silage while, contents of WSC in all inoculated silageswere significantly ($p<0.01$) lower as compared to PH silage and werecomparable with control. Similar findings were reported in previous studies (Xing et al., 2009; Nkosi et al., 2012; Yadav, 2018; Su et al., 2019) wherethepH of silage and content of WSC were decreased, and content of lactic acid was increased significantly in the enzyme and bacterial inoculants added silages. The reduction in pH value of inoculants treated silages may be due to production of more lactic acid by bacterial inoculants (McDonald et al., 1991) or may be due to the fact that addition of fibrolytic enzymes in forage at the time of ensiling degrade the cell wall content of silage leading to increased availability of WSC that serve as substrate for LAB and more lactic acid production is expected (Sheperd and Kung, 1996).

All bacterial inoculants and xylanase had significant effect on TVFA content of silages. The content of TVFA (m mol/100 gm) in all additives added silages was significantly ($P<0.001$) higher as compared to PH (21.6 ± 0.85) and control (19.7 ± 1.48), being highest in XLPLF (53.60 ± 1.84) followed by LF (49.4 ± 2.11) and LP (48.6 ± 1.91) silages. Similar results were reported by Dakore (2018) and Yadav (2018) in their studies where TVFA content was significantly increased in bacterial inoculants and enzyme added silages at 30 days of ensiling. Bacterial inoculants and xylanase had no significanteffect on NH_3-N content of silages, however it was numerically lower in all inoculated silages as compared to PH silage except LPLF silage.Dakore (2018) and Yadav (2018) reported a significant decreased in value of NH_3-N content in enzyme and bacterial inoculated silages. According to McDonald et al. (2002), well-preserved silages should contain less than 100 g NH_3-N/kg TN. In present study, lower concentration of NH_3-N in additives treated silages is indicative of well-preserved silage. It has been reported that inoculation of bacteria reduced proteolysis during ensiling and resulted in improved efficiency of silage protein utilization and reduced nitrogenlosses (McDonald et al., 2002) possibly sequent of reduction in pH. Data on total N content in silage shows that *L. plantarum*(1.84 ± 0.11) significantly ($p<0.01$) increased total N in silage as compared to others. Similar increase in total N content in silage inoculated with *L. plantarum* had been reported by Dakore (2018).

CONCLUSIONS

It is concluded that all additives improve fermentation quality of silage. Among all additives, *L. plantarum*is the best silage additive which improved silage fermentation quality when seasonal pasture hay is used with green maize in silage making at 3:7 ratio.

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