RESEARCH ARTICLE

Effect of Silage Additives on Rumen Fermentation Pattern and Economics of Wheat Straw and Pasture Hay-based Green Maize Silage

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Abstract

The investigation was conducted to study the effect of *Lactobacillus* bacterial inoculants (*Lactobacillus fermentum*, *Lactobacillus plantarum)* and xylanase in maize silage along with different dry fodders like wheat straw and seasonal pasture hay and to evaluate rumen fermentation pattern, and the cost effectiveness of additives. Different silages were prepared using green maize fodder and wheat straw (WS) and pasture hay (PH) separately in the proportion of 10:0 & 7:3 ratio in plastic jar of 3 kg capacity by adding common salt @ 0.5%, urea @ 1% and molasses @ 1.5% in each silage with 9 different treatments, *viz.,* Control (green maize alone), WS, X, LF, LPLF, PH, XPH, LFPH and LPLFPH. Xylanase, LP and LF were used @ 1500 IU/g, 1 x 10^6 cfu/g and 2 x 10^6 cfu/g, respectively. All silages were evaluated in terms of their *in vitro* rumen fermentation parameters on 45 days of ensiling. Xylanase inoculated maize + wheat straw and maize + pasture hay silage significantly improved TVFA, total gas production, IVDMD, IVOMD and PF. LPLF added maize + WS silage significantly increased *in vitro* rumen ammonia nitrogen and total nitrogen content. Thus, it is concluded that Xylanase can be used as cost effective additive in silage production for maximum nutrient utilisation and overall economic benefits.

Key words: *In vitro* rumen fermentation, *Lactobacillus* inoculants, Maize silage, Pasture hay, Wheat straw, Xylanase.

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Introduction

The health and productivity of livestock are closely related to quantum of high satisfactory forage supplied to the animals. Previously, huge amount of green forage and dry forage were available in India. But, due to urbanization and increased animal population there is a gap between demand and supply of green forage and dry forage in India. As wheat straw and pasture hay are inexpensive and available locally, its use in silage along with additives may improve its quality and thereby its utilization in animal feeding. Silage additives have been used as a management tool to improve the nutritional value of silage. The main aim of using silage additives is to promote the growth of lactic acid producing bacteria during the fermentation cycle and improve the quality of the silage (Chauhan *et al.*, 2021).

The main function of the exogenous fibrolytic enzymes is to release maximum amount of nutrients from the digestible, potentially digestible and indigestible fractions of the plant cell wall (Suryanarayana and Kavitha, 2017; Patel *et al.,* 2025[ab]). Keeping the above facts in view, this experiment was planned to study the effect of *Lactobacillus* bacterial inoculants and xylanase on rumen fermentation pattern of wheat straw or pasture hay based maize silage and its cost effectiveness.

Materials and Methods

The present study was conducted at Department of Animal Nutrition, College of Veterinary Science and AH, Kamdhenu University, Junagadh, Gujarat (India) during the year 2022. All the experimental procedures were approved by Institutional Animal Ethics committee (IAEC) (Protocol no: KU-JVC-IAEC-LA-75-76/21) of the College.

Preparation of Silage

Different silages were prepared using green maize fodder (control) and wheat straw (WS) and pasture hay (PH) separately in the proportion of 10:0 & 7:3 ratio in plastic jar of 3 kg capacity (3 replication in each) by adding common salt @ 0.5%, urea @ 1% and molasses @ 1.5% in each silage with

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9 different treatments, *viz.,* Control (only green maize), WS (green maize and wheat straw in 7:3 ratio), X (WS added with Xylanase), LF (WS added with *Lactobacillus fermentum*), LPLF (WS added with both bacterial inoculants), PH (green maize and pasture hay in 7:3 ratio), XPH (PH added with Xylanase), LFPH (PH added with *Lactobacillus fermentum*), LPLFPH (PH added with both bacterial inoculants). Xylanase, *L. plantarum* and *L. fermentum* were used @ 1500 IU/g, 1 x 10^6 cfu/g and 2 x 10^6 cfu/g, respectively. Different additives were spread as per their application rate in different treatments and mixed thoroughly. Fodder mass along with different additives were packed in plastic jar having the capacity of 3 kg and designed with valve at the lid of the jar. Air from the jars was removed with the help of vacuum pump. Jars of different treatments were stored at room temperature for 45 days. The store house was disinfected and appropriate measures were taken to avoid the entry of rats, mice and birds

The samples of green maize fodder, wheat straw, pasture hay, mixture of green maize & wheat straw (7:3) and mixture of green maize & pasture hay (7:3) were analysed before and on 45 days of ensiling for proximate composition and cell wall constituents according to the methods of AOAC (2023) and Van Soest *et al*. (1991), respectively.

Estimation of *In Vitro* Rumen Fermentation Pattern

The dried samples were used as substrate for determining the *in vitro* dry matter degradability (IVDMD), *in vitro* organic matter degradability (IVOMD) and *in vitro* total gas production. IVDMD and IVOMD were analyzed as per the method described by Tilley and Terry (1963).

Total gas production was determined by method of Menke and Steingass (1988). After 24-h, total gas production was measured and suitable aliquot was taken from glass syringe for determination of pH, total volatile fatty acids (TVFA), ammonia nitrogen (NH_3-N) and total nitrogen. The rumen pH was measured by pen-type pH meter. While, TVFA and NH_3-N were analyzed using standard protocols, and the total nitrogen content was measured as per the Kjeldahl method (AOAC, 2023).

Statistical Analysis

The data was analysed for descriptive statistics (mean and standard error), and treatment effects on different parameters were analyzed by one way ANOVA according to Snedecor and Cochran (1994). Pair wise mean differences between groups were compared by Duncan's new multiple range test for the significance at $p<0.05$.

Results and Discussion

Different experimental silages were analysed for proximate composition and cell wall constituents before and after ensiling and the results are presented in Table 1. Data of Table 1 indicate that by incorporating either wheat straw or pasture hay in silage making at 7:3 ratio does not affect nutritional value of silage when compared with maize silage alone. The results on *in vitro* rumen fermentation pattern of different experimental silages are presented in Table 2 and 3.

The IVDMD was not affected significantly however, numerically higher values were observed in Xylanase added Wheat straw and Pasture hay Silage. IVOMD ($p<0.01$) was found to be significantly higher in Xylanase treated wheat straw silage as compared to all other silages. Similar results on IVDMD were reported by Chen *et al.* (2019), Gang *et al.* (2020) and Huo *et al*. (2021) but, in present study IVDMD was numerically improved. In disparity, Lee *et al.* (2020) noticed non-significantly lower IVDMD. Regarding IVOMD, similar results were noted by Dakore (2018) and Yadav (2018). However, Filya (2003) and Khota *et al*. (2017) observed numerically increased IVOMD.

In vitro total gas production was significantly ($p<0.01$) higher in xylanase inoculated wheat straw and pasture hay silage. Similar findings were observed by Dakore (2018),

Table 1: Proximate composition and cell wall constituents of experimental fodders before and after ensiling (% DM basis)

Parameters	Before ensiling			After ensiling		
	Green maize	GM:WS-7:3	GM:PH-7:3	Green maize	GM:WS-7:3	GM:PH-7:3
DM	33.29±0.38	42.01±1.45	40.95 ± 0.12	31.39±0.32	41.02±0.22	40.52±0.37
OM	90.90±0.20	88.70±0.19	90.90 ± 0.25	84.59±0.23	87.57±0.64	84.76±0.32
CP	9.10±0.55	6.33±0.83	7.23 ± 0.69	8.06±0.17	7.15±0.40	8.25±0.54
EE	1.64±0.05	1.35±0.07	1.44 ± 0.14	1.08±0.05	0.90±0.03	0.94±0.08
CF	32.90±1.02	38.72±0.42	39.52 ± 1.36	36.06±2.39	45.11±3.69	39.08±0.60
TA	9.10±0.20	11.30±0.19	9.10 ± 0.25	15.40±0.23	12.42±0.64	15.23±0.32
NFE	47.26±1.42	42.30±0.67	42.71 ± 2.44	39.40±2.61	33.37±4.65	36.47±0.84
NDF	68.08±0.27	71.79±0.59	71.08 ± 0.56	67.41±1.74	69.89±0.84	70.37±0.38
ADF	41.94±1.05	48.11±0.09	50.9 ± 0.08	47.41±1.70	49.95±0.47	50.49±0.50
Cellulose	33.95±1.54	36.64±0.35	38.57 ± 0.07	33.49±0.63	33.58±0.42	33.99±0.94
Hemicellulose	26.14±1.32	23.68±0.50	20.18 ±0.48	20.00±0.70	19.94±0.90	19.87±0.71

Table 2: *In vitro* rumen fermentation parameters of different experimental silages

Treatments	Parameters			
	IVDMD (%)	IVOMD** (%)	Total gas production** (mL/200mg)	PF**(mg/mL)
Control	52.81 ±3.17	53.43^{ab} ±1.31	20.41^{b} ±0.95	4.33^{c} ±0.06
WS	50.89 ±0.50	51.00^{a} ±1.86	17.00^{a} ±0.51	4.42^{c} ±0.02
X	57.67 ±1.64	59.52^{d} ±0.95	25.21^{d} ±0.51	3.81^{a} ±0.07
LF	53.49 ±2.32	54.22^{ab} ±0.23	21.31^{bc} ±0.46	4.23^{c} ±0.04
LPLF	54.70 ±2.46	55.86^{bcd} ±0.78	23.16^{cd} ±0.39	4.02^{b} ±0.05
PH	49.25 ±0.64	51.27^{a} ±0.95	16.37^{a} ±0.84	4.33^{c} ±0.08
XPH	57.32 ±1.93	58.28^{cd} ±1.56	24.90^{d} ±0.92	3.66^{a} ±0.06
LFPH	54.37 ±1.87	54.62^{abc} ±1.48	21.08^{bc} ±1.43	4.02^{b} ±0.08
LPLFPH	55.46 ±1.83	56.34^{bcd} ±1.36	22.29^{bc} ±0.67	3.82^{a} ±0.05
p value	0.077	<0.001	<0.001	<0.001

^{abcd}Means with different superscript in a column differ significantly from each other (**p<0.01).

Table 3: *In vitro* rumen fermentation parameters of different experimental silages

Treatments	Parameters			
	pH	TVFA** (mMol/dL)	NH_3-N** (mg/dL)	Total N** (mg/dL)
C	6.70 ±0.02	6.95^{abc} ±0.15	41.66^{a} ±2.10	83.16^{bc} ±0.90
WS	6.68 ±0.03	5.88^{ab} ±0.68	39.16^{a} ±1.53	74.66^{a} ±1.60
X	6.71 ±0.03	9.54^{d} ±0.34	42.50^{a} ±1.11	81.00^{b} ±1.26
LF	6.76 ±0.03	7.79^{cd} ±0.66	43.33^{a} ±1.05	85.83^{cd} ±1.40
LPLF	6.80 ±0.04	8.69^{cd} ±1.16	51.66^{b} ±1.05	90.66^{e} ±2.04
PH	6.76 ±0.03	5.36^{a} ±0.21	41.16^{a} ±0.94	82.68^{bc} ±1.00
XPH	6.70 ±0.02	9.02^{d} ±0.39	43.16^{a} ±1.49	89.12^{de} ±1.25
LFPH	6.76 ±0.03	7.10^{bc} ±0.25	43.00^{a} ±0.96	85.45^{cd} ±1.29
LPLFPH	6.73 ±0.03	8.25^{cd} ±0.41	41.00^{a} ±0.96	80.21^{b} ±1.19
p value	0.194	<0.001	<0.001	<0.001

^{abcd}Means with different superscript in a column differ significantly from each other (**p<0.01).

Yadav (2018) and Huo *et al.* (2021). In distinction, Oskoueian *et al.* (2021) noticed significantly lower total gas production. Partitioning factor was found significantly lower in xylanase treated wheat straw silage (X) followed by xylanase treated pasture hay silage (XPH) and *Lactobacillus fermentum* and *Lactobacillus plantarum* treated pasture hay silage (LPLFPH). Similar findings were recorded by Dakore (2018) and Yadav (2018).

In vitro rumen pH was found to be statistical similar with each other (Table 3). In agreement to present study, Chen *et al.* (2019) and Oskoueian *et al.* (2021), also noticed non-significant difference in pH. In dissimilarity to present findings, Ce *et al.* (2016), Gang *et al.* (2020) and Huo *et al.* (2021) recorded non-significantly lower pH.

The TVFA content was found significantly (p<0.01) higher in xylanase added wheat straw and pasture hay silage. The results were supported by findings of Gang *et al.* (2020), Huo *et al.* (2021) and Oskoueian *et al.* (2021), while in contrast, Ce *et al.* (2016) observed non-significant reduction in TVFA.

The NH_3-N was found significantly (p<0.01) higher in LPLF added wheat straw silage as compared to all other experimental silage. Present findings collaborated with the observations of Marbun *et al.* (2020), Lee *et al.* (2020) and Oskoueian *et al.* (2021).

The total nitrogen content was significantly (p<0.01) higher in LPLF added wheat straw silage. It is not possible to compare the present findings due to the unavailability of literature on this parameter.

Patel *et al.* (2025[a]) evaluated the effect of xylanase on *in vitro* rumen fermentation on total mixed ration and they found significant effect of xylanase on rumen fermentation pattern. In their other study (Patel *et al.*, 2025[b]) also they observed significant effect of xylanase on *in vitro* degradability of TMR. Though, these findings of both the studies for effect of xylanase are on rumen fermentation in TMR, but concurred with the present findings on silage, except IVDMD, which was only numerically improved in the current study.

Table 4: Comparative economics of silage production

Items	Price (₹/kg or ₹/dose/ kg)	Treatments								
		C (10:0)	WS (7:3)	X (7:3)	LF (7:3)	LPLF (7:3)	PH (7:3)	XPH (7:3)	LFPH (7:3)	LPLFPH (7:3)
Green maize fodder	5	50	35	35	35	35	35	35	35	35
Wheat straw	1.25	0	3.75	3.75	3.75	3.75	-	-	-	-
Pasture hay	1.5	0	-	-	-	-	4.5	4.5	4.5	4.5
Salt	3	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Urea	5.35	0.53	0.53	0.53	0.53	0.53	0.53	0.53	0.53	0.53
Molasses	9	1.35	1.35	1.35	1.35	1.35	1.35	1.35	1.35	1.35
LP	0.80	-	-	-	-	8	-		-	8
LF	0.80	-	-	-	8	8	-		8	8
Xylanase	100	-	-	7.5	-	-	-	7.5	-	-
Total price (₹ /10kg)	-	52.03	40.78	48.28	48.78	56.78	41.53	49.03	49.53	57.53
Total price (₹ / kg)	-	5.20	4.07	4.82	4.87	5.67	4.15	4.90	4.95	5.75
Cost saving (₹/kg)	-	-	+1.13	+0.38	+0.33	-0.47	+1.05	+0.30	+0.25	-0.55
% Cost saving	-	-	+21.73	+7.30	+6.34	-9.03	+20.19	+5.76	+4.80	-10.57

Economics of Silage Production

The most crucial factor in dairy business is fodder cost. Hence, in the present experiment, prevailing market price of green maize fodder, wheat straw, pasture hay, salt, molasses, urea, xylanase and bacterial inoculants were taken into consideration to calculate cost of silage production. Green maize fodder was purchased from local market of Junagadh city @ 5 ₹/ kg. However, wheat straw is generally not utilised as a feed by the farmer and thrown as a waste material. But, for calculation of economics, farm prevailing price of wheat straw and pasture hay was considered @ 1.25 ₹/kg and 1.50 ₹/kg, respectively. For salt, urea and molasses supplements purchasing price of cattle breeding farm was taken into consideration @ 3 ₹/kg, 5.35 ₹/kg and 9 ₹/kg, respectively.

Enzyme xylanase was procured from the standard manufacturer company @ 100 ₹/kg. LP and LF were procured from National Collection of Dairy Cultures (NCDC), National Dairy Research Institute (NDRI), Karnal. As, LP and LF are culture media, it can be grown in laboratory for subsequent uses, but for calculating production cost of LP and LF was decided @ 0.80 ₹/kg in the consultation with Department of Veterinary Microbiology of the College.

Statistical data revealed that cost of silage production did not differ significantly among treatment groups. However, percent cost saving was found to be higher in xylanase added wheat straw silage as compared to all other additive inoculated and pasture hay silage. Xylanase showed effective utilisation of nutrients during *in vitro* rumen fermentation and was cost effective when used in green maize and wheat straw silage in the ration of 7:3.

Conclusion

From the study, it is concluded that xylanase can be used as cost effective additive when green maize fodder and wheat straw are principal fodder in the ratio of 7:3 for economical silage production and to ensure maximum nutrient utilisation and overall economic benefits.

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