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Asian Journal of Animal and Veterinary Advances



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The Effects of Different Silage Additives on *in vitro* Gas Production, Digestibility and Energy Values of Sugar Beet Pulp Silage

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Abstract: The aim of this study was to evaluate the effects of additives on *in vitro* Gas Production (IVGP), IVGP kinetics, energy values, Organic Matter Digestibility (OMD) and Dry Matter Digestibility (DMD) of Sugar Beet Pulp Silage (SBPS). Eight different silages were prepared from the sugar beet pulp samples. Silage treatments included no additives (CONT), Artturi Imarin Virtanen solution (AIV), feed urea (UREA), Formic Acid (FAS), biological inoculants Maize All (MAL) and Sil All (SAL), F silofarm formiat dry (SFD) and F Silofarm Liquid (SLI). The effects of different silage additives were determined using IVGP technique and pepsin-cellulase method in this study. Three Sakiz x Karayaka rams aged 2 years with ruminal cannulas were used IVGP technique. *In vitro* dry matter digestibility (IVDMD), *in vitro* organic matter digestibility (IVOMD) and energy values were studied by using pepsin-cellulase method. The silage additives significantly affected the nutrient contents of SBPS ($p < 0.01$). The highest crude protein content was found in UREA treated silages. Highest values in terms of NFE were determined in FAS treated silages ($p < 0.01$). The IVGP of AIV treated silages were significantly lower than those of silages applied for other treatments at 24, 48, 72 and 96 h incubations ($p < 0.01$). Biological inoculants (MAL and SAL), SLI and SFD treatments resulted in the highest energy values and IVGP. In conclusion, the use of IVGP technique can be recommended in the estimation of ME and NE_L values of SBPS since this technique provides more reliable estimates as compared to pepsin-cellulase method.

Key words: Inoculant, formic acid, urea, aiv solution, formiat, enzyme method

INTRODUCTION

Sugar-beet pulp is a valuable by-product from the manufacture of beet sugar fresh or dried. It is a common ingredient in concentrates of cows. The fiber in sugar beet pulp is highly digestible and has an energy content close to that of starch. But it is a structural carbohydrate without the negative effect on rumen fermentation that is typical for non-structural carbohydrates like starch and sugars. For a longer storage it is recommended to preserve this feed in the form of silage. Provided that the technological conditions are maintained on an optimal level, the process of ensiling of pressed sugar-beet pulp usually runs without any marked problems. Consequently in most countries pulp is conserved by ensiling. Many experiments have been made to find the best ways to ensile beet pulp with

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the smallest possible losses. However, sugar beet pulp can easily be ensiled. After the sugar extracting factory process the pulp is quite wet with a Dry Matter (DM) content lower than 20%. By pressing the pulp and adding about 4% molasses to facilitate lactobacillus growth, the so called high pressure beet pulp of about 27% DM becomes very suitable for ensiling (Wyss, 2005).

Both fresh and ensiled sugar-beet pulp has a high feeding value and shows a positive dietetic effect on ruminal fermentation (Formigoni *et al.*, 1993). For a longer storage it is recommended to preserve this feed in the form of silage. Provided that the technological conditions are maintained on an optimal level, the process of ensiling of pressed sugar-beet pulp usually runs without any marked problems. Bulls fed sugar beet pulp silage gained daily 33.9% more than the bulls fed grass and maize silages. Beef was of high quality and the price of feed per t of weight gain was 35.2% lower when feeding bulls with sugar beet pulp silage (Bendikas *et al.*, 2007). Consequently in most countries pulp is conserved by ensiling. Many experiments have been made to find the best ways to ensile beet pulp with the smallest possible losses. Most of these experiments have been conducted on a practical scale. Studies showed that the addition of 2% (v/v) of AIV-acid resulted in smaller losses and was slightly better than 1% of molasses, which gave about the same result as 2% formic acid. Addition of 1, 2 or 3% molasses was also as good as, or even better than the use of AIV-acid (Olsen, 1951).

Silage additives can be divided into three general categories: (1) fermentation stimulants, such as bacterial inoculants and enzymes; (2) fermentation inhibitors, such as propionic, formic and sulfuric acids and (3) substrate or nutrient sources, such as molasses, urea and anhydrous ammonia (Bolsen *et al.*, 1996). Their main functions are to either increase nutritional value of silage or improve fermentation so that storage losses are reduced. Response to additives depend on what forage is being treated (Filya, 2001). Williams *et al.* (1997) and Schofield and Pell (1995) have investigated silage quality using gas production technique.

In this study, the effects of different silage additive added ensiling period was investigated on the silages quality, *in vitro* gas production, gas production kinetics, energy values, organic matter digestibility and dry matter digestibility of sugar beet pulp silage.

MATERIALS AND METHODS

This study was conducted over the period from November 2004 to March 2007 at University of Ondokuz Mayıs in Samsun-Turkey. In this study sugar beet pulp samples (SBPS) were obtained from Samsun sugar factory (dry matter contents 13.98-14.12%). Fresh pulp samples were wilted for 24 h in the laboratory then the samples were ensiled in experimental jar (1.5 L glass jar) silos in quadruplicate. Eight different silages were prepared from the sugar beet pulp samples. Silage treatments included control (no additives) (CONT). The AIV (Artturi Imartti Virtanen) concludes 1 part H₂SO₄, 1 part HCl and 6 part water which it was applied at a 80 g kg⁻¹ of the fresh weight of the sugar beet pulp (AIV) and urea was applied 1% of the fresh weight of the sugar beet pulp (UREA). Formic acid was applied at 2.2-2.5 kg ton⁻¹ of the fresh weight of the sugar beet pulp (FAS), 10 g t⁻¹ microbial inoculant (Maize All and Sil All obtained from Alltech-Pioneer). As recommend by the manufacturer, inoculant was added at 1.0×10¹¹ cfu g⁻¹ of fresh forage (MAL and SAL). Maize All from Alltech is formulated with three lactic acid-producing bacteria (*Lactobacillus plantarum*, *Pediococcus acidilactici* and *Lactobacillus salivarius*) and high level amylase. Sil All from Alltech is formulated with four lactic acid-producing bacteria (*Lactobacillus plantarum*,

Enterococcus faecium, *Pediococcus acidilactici* and *Lactobacillus salivarius*) and four enzyme (Cellulase, Hemicellulase, Pentosanase and Amylase). 0.5 kg ton⁻¹ F silofarm formiat dry (sodium formiat) (SFD) and 5-7 kg ton⁻¹ F silofarm liquid (formic acid, sodium format and water) (SLI) as recommend by the manufacturer (from Farmavet), was added at 1.0×10¹¹ cfu g⁻¹ of fresh forage. These silos were covered to measure entering oxygen with nylon.

In the cellulase method, enzyme cellulase obtained from the microorganism *Trichoderma viridae*, hemicellulase from the microorganism *Aspergillus niger*, α -amylase from the porcine pancrease and the enzyme pepsin were used.

After two months (60 days) storage, the silos were opened. Representative samples were dried at 48°C in a forced-air oven for 72 h. After drying, silage samples were ground through a 1 mm screen for chemical analysis. The DM was determined by drying the samples at 105°C overnight. Organic matter content was determined by ashing in a muffle furnace at 550°C for 8 h. Nitrogen (N) content was determined using Kjeldahl method (AOAC, 1998). Crude protein was calculated as N×6.25. Crude Fibre (CF) and Ether Extract (EE) were determined by the methods described by AOAC (1998). Volatile fatty acid and NH₃-N contents in rumen fluid were determined using Markham (1942). Furthermore, quality analyses (Kilic, 1986), were made in silages and NFE was determined by calculation. All cemical analysis was carried out in triplicate. Total points (DLG) were determined and Fleig points were calculated in silages according to Kilic (1986) (Fleig point = 220+(2×% dry matter-15)-40×pH).

The pH of each sample was determined in triplicate using approximately 25 g wet ensilage added to 100 mL of distilled water. After hydration for 10 min using blender, pH was determined using digital pH meter (HANNA INSTRUMENTS 1332 model).

***In vitro* Gas Production**

Three Sakiz x Karayaka rams aged 2 years with ruminal cannulas were used in gas production technique. Rumen fluid was obtained from three fistulated sheep fed twice daily (08.30-16.30) with a diet containing grass hay (60%) and concentrate (40%). The samples (milled through a 1 mm sieve) were incubated *in vitro* rumen fluid in calibrated glass syringes following the procedures of Menke and Steingass (1988). Approximately 200 mg dry weight of sample was weighed in triplicate into calibrated glass syringes of 100 mL. Rumen fluid was collected before the morning feeding. The syringes were prewarmed at 39°C before the injection of 30 mL rumen fluid-buffer mixture consisting of 10 mL rumen fluid and 20 mL digestion media into each syringe followed by incubation in a water bath at 39°C. Triplicates of each sample were used in two separate runs. Readings of gas production recorded before incubation and 3, 6, 9, 12, 24, 48, 72 and 96 h after incubation. Total gas production values corrected for blank incubation. Cumulative gas production data were fitted to the model of Orskov (1979) by NEWAY computer package programme.

$$y = a + b(1 - e^{-ct})$$

where, a is the gas production from the immediately soluble fraction (mL), b is the gas production from the insoluble fraction (mL), c is the gas production rate constant for the insoluble fraction (mL h⁻¹), a+b is the potential gas production (mL), t is incubation time (h), y is gas produced at time t.

Organic matter digestibility (Menke *et al.*, 1979), ME (Menke *et al.*, 1979) and NE_L (Menke and Steingass, 1988) contents of forages were estimated using equations given as:

$$\begin{aligned}\text{OMD (\%)} &= 14.88 + 0.8893 \text{ GP} + 0.448 \text{ CP} + 0.651 \text{ A} \\ \text{ME (MJ kg}^{-1} \text{ DM)} &= 2.20 + 0.136 \text{ GP} + 0.057 \text{ CP} + 0.002859 \text{ EE}^2 \\ \text{NEL (MJ kg}^{-1} \text{ DM)} &= 0.101 \text{ GP} + 0.051 \text{ CP} + 0.11 \text{ EE}\end{aligned}$$

Where:

GP = 24 h net gas production (mL/200 mg DM)

CP = Crude protein (%)

EE = Ether extract (%)

A = Ash content (%)

Pepsin-Cellulase Method

In vitro digestibility of DM and OM were determining according to Alcicek and Wagener (1995) as follow; ONUZUKA cellulase enzyme was used in this study.

$$\begin{aligned}\text{DMD (\%)} &= (S1 (T1-T0)/S1) \times 100 \\ \text{OMD (\%)} &= 1 - ((T1-T2)/(S1-A1)) \times 100\end{aligned}$$

Where:

S1 = Sample amount (as DM)

T0 = Weight of crucible (105°C, 48 h)

T1 = Dry sample (105°C, 24 h) + T0

T2 = Ashed sample (550°C, 4 h) + T0

A1 = Crude ash amount of sample (g)

Total Energy (TE), Digestible Energy (DE), Metabolisable Energy (ME), net energy lactation (NE_L), net energy fat (NE_F) and net energy maintenance (NE_M) value were calculated according to the enzyme method (Jarrige, 1989; Malossini *et al.*, 1993). Calculated values were converted to MJ kg⁻¹ DM.

$$\begin{aligned}\text{TE (kcal kg}^{-1} \text{ DM)} &= 5.99\text{CP} + 6.71\text{EE} + 4.28\text{CF} + 4.73\text{NFE} \\ \text{DE (kcal kg}^{-1} \text{ DM)} &= (\text{TEXOMD})/100 \\ \text{ME (kcal kg}^{-1} \text{ DM)} &= [(86.82 - 0.0099\text{CF} - 0.0196\text{CP})\text{DE}]/100 \\ q &= \text{ME/TE} \\ \text{NE}_L \text{ (kcal kg}^{-1} \text{ DM)} &= kx\text{ME}, (k = 0.60 + 0.24 (q - 0.57)) \\ \text{NE}_F \text{ (kcal kg}^{-1} \text{ DM)} &= kx\text{ME}, (k = 0.78q + 0.006) \\ \text{NE}_M \text{ (kcal kg}^{-1} \text{ DM)} &= kx\text{ME}, (k = 0.287q + 0.554)\end{aligned}$$

Statistical Analysis

One-way analysis of variance was carried out to compare gas production, gas production parameters, Energy values, DMD and OMD values using General Linear Model (GLM) of SPSS 10.0 package programs. Significance between individual means were identified using the Duncan's multiple range test (Duncan, 1955).

RESULTS

Chemical composition contents of SBPS were given in Table 1, the silage additives significantly affected the nutrient contents ($p < 0.01$). The addition of AIV resulted in the highest EE and ash values in silages ($p < 0.01$). The highest content of CP was found with

UREA, whereas that of NFE was determined with FAS ($p < 0.01$). Rumen pH, ammonia N ($\text{NH}_3\text{-N}$) and total volatile fatty acid (VFA) contents determined for rumen liquid using *in vitro* gas production technique were; 6.20 (5.92-6.29), 310 mg L^{-1} (280-427 mg L^{-1}) and 119 mmol L^{-1} (90-136 mmol L^{-1}), respectively.

Silage qualities of SBPS were given in Table 2, based on Flieg point evaluation, all the silages were classified as excellent quality silages. The quality of MAL, SAL and SLI silages was better than the others when total point (DLG) was taken into account. The lowest silage pH was determined with AIV.

The highest Total Energy (TE) occurred with SAL addition (Table 3), whereas the lowest TE values were found with AIV ($p < 0.01$). Total energy of all the silages differed significantly ($p < 0.01$). The highest DE, ME and NE_L values were determined with SAL, MAL and SLI ($p < 0.01$).

Table 1: The effects of different silage additives on chemical composition of SBPS (g kg^{-1} DM)

Treatments	DM	EE	Ash	CP	CF	NFE
CONT	129.1	10.2 ^b	67.1 ^c	115.8 ^{bcd}	253.4 ^{bc}	553.5 ^b
AIV	125.4	30.6 ^a	167.8 ^a	99.4 ^{de}	259.8 ^{bc}	442.5 ^d
FAS	140.1	12.5 ^b	62.1 ^c	88.8 ^e	223.9 ^d	612.7 ^a
MAL	115.8	10.3 ^b	57.8 ^e	121.7 ^{bc}	263.9 ^b	546.3 ^b
SAL	119.2	12.7 ^b	54.8 ^e	118.5 ^{bc}	249.9 ^c	564.0 ^b
SFD	109.5	11.2 ^b	71.6 ^c	127.4 ^b	279.6 ^a	510.2 ^c
SLI	139.0	12.7 ^b	77.5 ^c	105.5 ^{de}	229.4 ^d	574.7 ^b
UREA	129.2	11.7 ^b	107.0 ^b	212.3 ^a	249.4 ^c	419.7 ^d
SEM		0.116	0.565	0.578	0.283	0.969
Significant		**	**	**	**	**

DM: Dry matter, EE: Ether extracts, CP: Crude protein, CF: Crude fibre, NFE: Nitrogen free extracts. SEM: Standard error of means. Means within the same column with differing superscript letters are different * $p < 0.05$; ** $p < 0.01$

Table 2: The effects of different silage additives on silage pH and silage qualities

Treatments	pH	Smell	Structure	Colour	DLG point	Quality	Flieg point	Quality
CONT	3.44	9.00	3.00	1.50	13.50	Moderate	93.36	Excellent
AIV	1.45	8.00	3.00	1.50	12.50	Moderate	172.09	Excellent
FAS	3.08	9.00	3.00	1.50	13.50	Moderate	109.66	Excellent
MAL	3.35	10.00	3.00	1.50	14.50	Good	94.29	Excellent
SAL	3.38	11.00	3.00	1.50	15.50	Good	93.51	Excellent
SFD	3.55	9.00	3.00	1.50	13.50	Moderate	84.90	Excellent
SLI	3.32	10.00	3.00	1.50	14.50	Good	100.15	Excellent
UREA	3.17	9.00	3.00	1.50	13.50	Moderate	103.84	Excellent

Table 3: The effects of different silage additives on energy values (kcal kg^{-1} DM and MJ kg^{-1} DM), DMD (%) and OMD (%) of sugar beet pulp silage by using cellulase method

	TE	DE	ME	NE_L	NE_F	NE_M	DMD	OMD
CONT	4464.98 ^d (18.69)	1670.96 ^{bc} (7.00)	1370.88 ^{bc} (5.74)	736.34 ^{bc} (3.08)	337.61 ^c (1.41)	880.66 ^{bc} (3.69)	44.02 ^a	37.42 ^b
AIV	3996.34 ^b (16.73)	1123.32 ^d (4.70)	924.55 ^d (3.87)	481.52 ^d (2.02)	178.68 ^d (0.75)	575.90 ^d (2.41)	36.63 ^{bc}	28.11 ^c
FAS	4471.90 ^c (18.72)	1696.11 ^{bc} (7.10)	1405.46 ^{bc} (5.88)	757.14 ^{bc} (3.17)	353.35 ^{bc} (1.48)	905.54 ^{bc} (3.79)	36.61 ^{bc}	37.93 ^b
MAL	4511.77 ^b (18.89)	1964.45 ^a (8.22)	1607.37 ^a (6.73)	882.05 ^a (3.69)	456.56 ^a (1.91)	1054.93 ^a (4.42)	32.14 ^b	43.54 ^a
SAL	4532.70 ^a (18.98)	1910.59 ^b (8.00)	1567.11 ^b (6.56)	855.96 ^b (3.58)	432.16 ^b (1.81)	1023.73 ^b (4.29)	38.62 ^b	42.15 ^b
SFD	4447.93 ^c (18.62)	1699.48 ^{bc} (7.12)	1386.01 ^{bc} (5.80)	746.38 ^{bc} (3.12)	347.56 ^{bc} (1.46)	892.67 ^{bc} (3.74)	33.58 ^{bc}	38.21 ^{bc}
SLI	4418.55 ^c (18.50)	1736.69 ^{bc} (7.27)	1432.44 ^{bc} (6.00)	775.33 ^{bc} (3.25)	372.02 ^{bc} (1.56)	927.29 ^{bc} (3.88)	41.71 ^b	39.30 ^{bc}
UREA	4402.64 ^c (18.43)	1600.86 ^c (6.70)	1283.73 ^c (5.37)	684.56 ^c (2.87)	300.01 ^c (1.26)	818.74 ^c (3.43)	40.42 ^{bc}	36.36 ^b
SEM	33.36	54.72	44.95	26.39	18.48	31.56	1.21	1.06
Sig.	**	**	**	**	**	**	*	**

TE: Total energy, DE: Digestible energy, ME: Metabolisable energy, NE_L : Net energy lactation, NE_F : Net energy fat, NE_M : Net energy maintenance, DMD: Dry matter digestibility, OMD: Organic matter digestibility. SEM: Standard error of means. Energy values were given in the parenthesis as MJ kg^{-1} DM. Means within the same column with differing superscript letters are different * $p < 0.05$; ** $p < 0.01$

Table 4: The effects of different silage additives on *in vitro* gas production and estimated parameters of sugar beet pulp silage

Incubation time (h)	CONT	AIV	FAS	MAL	SAL	SFD	SLI	UREA	SEM	Sig.
Gas production, mL/200 mg DM										
3	8.91 ^b	12.50 ^{ab}	11.27 ^{ab}	11.79 ^{ab}	10.63 ^{ab}	9.13 ^b	15.34 ^a	8.71 ^b	0.62	*
6	16.82 ^{ab}	14.86 ^b	15.58 ^b	19.68 ^{ab}	18.73 ^{ab}	16.20 ^{ab}	21.88 ^a	15.68 ^b	0.72	*
9	23.71 ^{abc}	16.89 ^c	19.54 ^{bc}	26.46 ^{ab}	25.72 ^{ab}	22.43 ^{abc}	27.63 ^a	21.86 ^{abc}	0.92	*
12	29.73 ^{ab}	18.65 ^c	23.18 ^{bc}	32.27 ^a	31.76 ^a	27.90 ^{ab}	32.70 ^a	27.35 ^{ab}	1.13	**
24	47.05 ^a	23.62 ^c	35.04 ^b	48.54 ^a	48.80 ^a	44.00 ^{ab}	47.57 ^a	43.77 ^{ab}	1.76	**
48	63.15 ^a	28.06 ^c	49.62 ^b	62.80 ^a	63.89 ^a	59.50 ^a	61.98 ^a	60.33 ^a	2.30	**
72	68.88 ^a	29.54 ^c	57.11 ^b	67.54 ^{ab}	68.91 ^a	65.12 ^{ab}	67.33 ^{ab}	66.77 ^{ab}	2.48	**
96	70.99 ^a	30.06 ^b	60.99 ^a	69.19 ^a	70.67 ^a	67.16 ^a	69.35 ^a	69.28 ^a	2.55	**
96 h pH	6.61 ^a	6.36 ^b	6.57 ^a	6.60 ^a	6.54 ^{ab}	6.59 ^a	6.54 ^{ab}	6.57 ^a	0.02	*
Gas production parameters										
a (mL)	-0.18 ^c	9.78 ^a	6.58 ^{ab}	2.58 ^{bc}	1.24 ^c	1.10 ^c	7.90 ^a	0.85 ^c	0.80	**
b (mL)	72.55 ^a	20.57 ^d	58.62 ^c	67.54 ^{abc}	70.50 ^{ab}	67.24 ^{abc}	62.75 ^{bc}	70.08 ^{ab}	3.01	**
c (mL h ⁻¹)	0.04 ^{ab}	0.05 ^a	0.03 ^b	0.05 ^a	0.05 ^a	0.04 ^{ab}	0.04 ^{ab}	0.04 ^{ab}	0.001	*
Estimated parameters										
ME (MJ kg ⁻¹ DM)	9.26 ^a	5.98 ^c	7.48 ^b	9.50 ^a	9.52 ^a	8.92 ^a	9.28 ^a	9.37 ^a	0.25	**
NE _L (MJ kg ⁻¹ DM)	5.46 ^c	3.23 ^b	4.13 ^b	5.64 ^a	5.67 ^a	5.22 ^a	5.49 ^a	5.63 ^a	0.18	**
OMD (%)	62.35 ^a	41.44 ^c	50.43 ^b	63.88 ^a	63.95 ^a	60.19 ^a	62.42 ^a	64.04 ^a	1.63	**

ME: Metabolisable energy, NE_L: Net energy lactation, OMD: Organic matter digestibility, SEM: Standard error of means. Means within the same rows with differing superscript letters are different *p<0.05; **p<0.01

As seen Table 3, only DMD of MAL was different from that of CONT (p<0.05). The OMD of AIV was found the lowest (p<0.01) and all the treatments significantly differed. In addition, a significant difference was found between the OMD of UREA and MAL (p<0.01).

AIV resulted in the lowest *in vitro* gas production values at 24, 48, 72 and 96 h and differed significantly from other treatments (p<0.01). However, AIV addition, which had high a-value, had the lowest b-value (p<0.01). The rates of gas production (c value) of MAL, SAL and AIV were significantly higher than that of FAS (p<0.05) and no significant differences were found between other treatments (Table 4).

AIV resulted in the lowest values of ME and OMD estimated from *in vitro* gas production values, which was followed by FAS (p<0.01). No differences were observed between other feedstuffs (p>0.05). NE_L of AIV did not differ from that of FAS, however these treatments caused lower values than did the other additives (p<0.01).

DISCUSSION

The quality (total point and Flieg point) of silage did not decrease in UREA treated silage with the highest CP content. However, NFE content decreased due to the increase in CP content. In spite of the reduction in NFE content, UREA treated SBPS as roughage source would like to be preferred in ruminant nutrition as compared to other additive treated silages. The CP content of FAS was similar to the values reported for FAS by Sahin *et al.* (1999). However, the CP contents of other silages were higher. EE, ash, CF, NFE and DM contents of SBPS were also similar to those reported by Sahin *et al.* (1999). The silage pH of AIV was found higher than the pH value (3.66-4.10) of the silage reported by Dolezal *et al.* (2005). However the pH of silages with other additives were found similar to the reported values. The DM contents of silages determined (17.81-20.78) by Dolezal *et al.* (2005) were higher than the DM content values in the present study. The pH, CP, EE, CF, ash and NFE values of the silages ensiled with different additives are in accordance with pH (3,72-4,31), CP (2,30-9,20), EE (0,30-2,80), CF (24,70-32,40), ash (5,70-11,00) and NFE (45,40-59,80) values of SBPS made by Deniz *et al.* (2001). The differences in nutrient composition of SBPS between the results of the present study and other studies can be attributed to the nature of the additives used.

Kamarloiy and Yansari (2008) reported that digestibilities of DM and nutrients of silages were significantly increased and pH values of silages significantly reduced by microbial inoculation. Kung *et al.* (1993) reported that digestibility may be greater in some silages treated with microbial inoculants as silage additives. Likewise, pH values of silages which treated with microbial inoculants lower than control group in this study. Beside, it showed that DMD varied compared to control group by different microbial inoculants.

It is known that the quality of silage can change depending upon the type of additive (Kilic, 1986; Filya, 2001). In the present study, all the SBPS were classified as excellent silages based on Flieg point evaluation. When the total point is considered, MAL, SAL and SLI were better quality. Silage fermentation is thought to be benefited from biological inoculants containing lactic acid bacteria and SLI.

The lowest DLG value (total point) was observed in AIV treated silage with its smell worse than other silages. It is probable that this silage would be less preferred by the animals due to its smell, low NFE and high ash contents. The increase in ash content of AIV treated silages is thought to due to HCl and H₂SO₄. In addition, the intake of AIV treated silages an cause physiological problems due to their very low pH (1.45). Therefore these silages should be offered together with other roughages, which would increase the pH.

The lowest energy value estimated with cellulase method and OMD value were found in AIV treated silages (p<0.01). This can be attributed to the highest ash and the lowest NFE contents. The differences in total energy of all silage treatments are thought to depend on the nutrient contents of silages.

Dry matter digestibility (Table 3) of MAL added silages was lower than that of the CONT silages (p<0.05), whereas no difference was found between the other silages (p>0.05). Of the silages with the highest DLG points, MAL, SAL and SLI groups had generally higher energy values. The lowest values were determined with AIV silages, which also had the lowest DLG point. This suggests that energy values determined in this way can be predicted from DLG.

Gas production values at 24, 48, 72 and 96 h incubations were lowest in AIV treated silages and differed significantly from other silages (p<0.01). The decrease in the pH of incubations after 96 h could have caused such a difference. Low silage pH probably prevented acetic acid formation and reduced *in vitro* gas production (Wolin, 1960). However, the variations in the nutrient compositions of feedstuffs are also known to influence *in vitro* gas production and related parameters (Kilic and Saricicek, 2006). The increase in ash and NFE content reduces gas production (Menke and Steingass, 1988). Lower gas production in AIV and FAS groups can be associated with high ash content of AIV silage and high NFE content of FAS.

The gas production parameters, a-value (the gas production from the immediately soluble fraction) of CONT group was negative. Like in *in vitro* gas production, the lowest b-value (the gas production from the insoluble fraction) was determined with AIV group. On the other hand, the c-value (the gas production rate constant for the insoluble fraction) of FAS treated silage was lower than the other silages. This indicates that the addition of FAS would slow down the rate of gas production, thus provide environmental benefits such as a reduction in methane production when given together with feeds with a high rate of gas production.

The lowest ME and OMD values determined using *in vitro* gas production technique of AIV treatment and NE_L of AIV and FAS additions can be associated with high ash content. The comparison of the ME and NE_L values estimated with cellulase method and *in vitro* gas production technique indicates that the values estimated using *in vitro* gas production technique were higher and more reliable than the values estimated with cellulase method. The energy values estimated using *in vitro* gas production technique are similar to those calculated by using nutrient contents (Karabulut and Canbolat, 2005).

However, Arbabi *et al.* (2008) reported that dried sugar beet pulp (DSP), which is the appropriate water absorbent material and have extra sugar, utilize as the additives to silage high moisture forage. The researchers demonstrated that amount of CP in the treated silage with DSP was more than others. According to above findings sugar beet pulp can be use as a silage additives.

CONCLUSIONS

Addition of AIV increased ash content of the SBP silage but decreased sensorial observation based quality, energy, OMD and *in vitro* gas production values. In spite of high Flieg point, addition of AIV to SBPS is not recommended. However, addition of UREA increased the CP content of silages but did not affect silage quality. The *in vitro* gas production and energy values and OMD contents of the silages were high. UREA can be used as an additive for SBPS due to the increment in the CP content in spite of a reduction in NFE content.

The addition of FAS decreased CP content but increased NFE content of the silage. In addition, FAS added silages had high quality points with a low rate of gas production. Therefore this silage would not cause a rapid gas production and could be fed with feedstuffs with high gas production. MAL and SAL biological inoculants showed the highest energy values and gas productions. SLI and SFD had high gas production, energy and OMD values. The use of *in vitro* gas production technique can be recommended in the estimation of ME and NE_L values of SBPS since this technique provides more reliable estimates as compared to cellulase method.

In conclusion, suitable additive should be selected following the consideration of the rate and amount of roughage and concentrate feeds offered to the animals. In this respect, *in vivo* trials are also required in the future.

ACKNOWLEDGMENT

We are grateful to Ondokuz Mayıs University Research Fund of Turkey for their financial support (Project No. Z-415).

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