Kafkas Üniv Vet Fak Derg
15 (1): 19-24, 2009
DOI:10.9775/kvfd.2008.44-A

Effects of Different Additives on the Quality of Grass Silage and Rumen Degradability and Rumen Parameters of the Grass Silage in Rams [1]

İsmail KAYA * ✎ Yücel ÜNAL* Dilek AKSU ELMALI*

[1] *This study was supported by DPT (96 K-121250-10)*

* Department of Animal Nutrition and Nutritional Diseases, Faculty of Veterinary Medicine, University of Kafkas, Kars - TURKEY

Yayın Kodu (Article Code): 2008/44-A

Summary

This study was carried out to determine the effects of ensiling grass with different silages additives on dry matter (DM), nutrient content, pH and rumen degradability and the effects of grass silage on the digestibility and rumen parameters in the rams. Fifteen silages were ensiled as grass (with no additives), grass + 2.5% barley mixture, grass + 5% barley mixture, grass + 2.5% barley + 1% molasses + 0.5% salt mixture and grass + 5% barley + 2% molasses + 1% salt mixture with three replicates. Additionally grass with no additives was ensiled in the plastic barrels to determine the effects of this silage on the digestibility, rumen pH, ammonia nitrogen and volatile fatty acids by using three rams. The highest crude protein (CP) contents were determined in the barley, molasses and salt mixture of grass silages (10.41, 10.86%) and the highest nitrogen free extract (NFE) contents were determined in the barley mixture grass silages (44.41, 47.17%). The pH value of silages was decreased according to the amount and type of silage additives. DM and nutrient degradabilities of grass silages with or without additives left rumen incubation at different hours were found statistically different with aspect to hours and types of silage. ($P<0.05$). CP and DM digestibilities of grass silage in the rams were determined as 64.49 and 53.21%. Total volatile fatty acids were determined as 69.73 mmol/L. Grass silage, mixtured with barley, molasses and salt were determined the best silage with respect to the nutrient content and pH.

Keywords: *Grass Silage, Nutrient Contents, Degradability, Rumen Parameters*

Farklı Katkıların Çayır Otu Silajının Kalitesi ve Rumen Yıkılabilirliği ile Ot Silajının Koçlarda Rumen Parametrelerine Etkileri

Özet

Bu çalışma, çayır otunun farklı katkı maddeleriyle silolanmasının kuru madde (KM), besin madde içerikleri, pH ve rumende yıkılabilirlik ile yalnız ot silajının koçlarda sindirilebilirlik ve rumen parametrelerine etkisini incelemek amacıyla yapılmıştır. Araştırmada katkısız çayır otu, çayır otu + %2.5 arpa karışımı, çayır otu + %5 arpa karışımı, çayır otu + %2.5 arpa + %1 melas + %0.5 tuz karışımı ve çayır otu + %5 arpa + %2 melas + %1 tuz karışımından oluşan üç tekerrürlü 15 adet silaj yapıldı. Ayrıca plastik varillere yalnız çayır otu silajı yapılarak, besin madde sindirilebilirliği ile rumen pH, amonyak azotu ve uçucu yağ asitlerine olan etkileri üç baş koç üzerinde belirlenmiştir. Ham protein (HP) oranı arpa, melas ve tuz karışımlı silajlarda en yüksek değerde (%10.41, 10.86) bulunurken, en yüksek azotsuz öz madde (NÖM) içeriği ise arpa karışımlı silajlarda (%44.41, 47.17) tespit edilmiştir. Silajların pH değeri, katkı maddelerinin çeşit ve miktarına göre azalmıştır. Rumende değişik saatlerde inkubasyona bırakılan çayır otu ve katkılarla hazırlanan ot silajlarının KM ve besin madde yıkılabilirlikleri, silaj türü ve inkübasyon süresine göre önemli farklılık göstermiştir ($P<0.05$). Ot silajının koçlardaki KM ve HP sindirilebilirliği ise %64.49 ve 53.21 olarak saptanmıştır. Rumen sıvısı uçucu yağ asitlerinin toplam miktarı ise 69.73 mmol/L olarak bulunmuştur. Araştırmada, çayır otu silajlarına katkı maddesi olarak arpa, melas ve tuz katılmasının besin maddeleri ve pH üzerine olumlu etkileri tespit edilmiştir.

Anahtar sözcükler: *Çayır otu silajı, Besin maddeleri, Yıkılabilirlik, Rumen parametreleri*

✎ İletişim (Correspondence)
☎ +90 474 242 68 07/1133
✉ ismayilkaya@hotmail.com

INTRODUCTION

Grasses are among the forages which are ensilageable at medium quality as far as their gramineae and leguminosea contents are concerned [1]. Different forages which go under fermentation easily including ground grains and molasses are used mostly to enhance the silage quality of green forages and to ensile them more easily [1,2]. Moreover, organic and inorganic acids are added to silage to lower pH, and inorganic salts are used to destroy bacterial toxins which may potentially occur during the silage process [1-4].

Dry matter (DM) and nutrient contents of the grass silage and its pH level vary depending on the kind, vegetation period and silage additives [3-7]. Thus, DM and nutrients degradabilitiy rates of the grass silage increase in rumen as incubation period lasts longer [3,5,8]. Increasing consumption of the grass silage by rams has had no essential difference among the acetic, propionic and butyric asit amounts in the rumen fluid [9].

Grasland comprise 34.7% of the Kars district [10], which is used as grazing and hay production areas. Usage of the grass silage as forage in this district particularly in winter season is extremely important to increase animal production.

In regard to that, this study was conducted to determine the dry matter, nutrient contents, pH and rumen degradability ratios of the ensiled grass with different silage additives, and to reveal the effects of feeding with grass silage on digestibilty and rumen parameters of rams.

MATERIAL and METHODS

Grass was harvested at the end of the July of 2002 and cropped into pieces roughly 3 cm in length. For the study, grass (silage 1=S1), grass + 2.5% barley mixture (silage 2=S2), grass + 5% barley mixture (silage 3=S3), grass + 2.5% barley + 1% molasses + 0.5% salt mixture (silage 4=S4) and grass + 5% barley + 2% molasses + 1% salt mixture (silage 5=S5) were prepared in glass jars with the volume of 1 kg. Barley was ground roughly and molasses was added to the mixture samples after diluted by three fold water of the molasses weight. The addivites prepared on the basis of percentage to wet weights of grass. Glass jars were closed with proper caps and wrapped up by the parafilm to avoid air entrance. Three samples were prepared for each silage type. Grass silage was also prepared in the plastic barrels with 50 kg capacity to use in the digestibility trials.

The silage samples were opened in the April of 2003, and analyses and trials planned were performed.

Determination of the Nutrient Contents and pH

Silage samples were dried in air forced oven at 60°C for 48 h. Then, contents of the dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), crude fiber (CF), ash and nitrogen free extract (NFE) were determined in accordance with the AOAC, [11]. Moreover, values of the DM, OM, CP and CF of the incubated rumen and fecal samples by the same method.

To determine the pH, 25 g silage sample and 100 ml distile water were put in a beaker and after stirring up the content for 10 min the pH was measured by the indicated method Polan et al.[12].

Degradability of the Silage Samples in the Rumen

To use on rumen degradability trials, 3 rams with average 50 kg body weights have been had rumen fistula opened and cannula placed. The animals were fed 900 g gras hay and 200 g concentrate daily maintenance requirements. Grass hay was medium quality and concentrate composed of 50% barley, 25% cotten seed meal, 22% wheat bran, 1% salt, 1% limestone, and 1% vitamin-mineral premix (Mn 10.000 mg, Zn 10.000 mg, Fe 10.000 mg, Cu 5.000, Co 100 mg, I 100 mg, Se 100 mg, Vit A 10.000.000 IU, Vit D_3 2.000.000, Vit E 15 mg). The animals were fed in individual cages.

Nylon bags with pore diameter of 45 µm and size of 9x14 cm were used in the study, in accordance with the suggestions of Bhargava and Ørskov [13]. Dried silage samples weighing 2.5-3 g were incubated in the rumen for 8, 16, 24 and 48 hours of periods. Each sample and incubation period was tripled. After the incubation period, the nylon bags were taken out of the rumen and, to cease the microbial activity, embedded immediately in the water. The bags then were rinsed in a pail

until the water became clear. Later, they were dried in oven at 60°C for 48 h to reach their constant weight, were put in dessicator to cool, and were weighed. The samples were analyzed for DM, OM, CP and CF according to AOAC [11]. Finally, the analyses and calculations were performed on the samples.

Determination of Digestibility and Rumen Parameters

To determine the digestibility and rumen parameters of the grass silage, 3 rams with average 70 kg body weight were used in the study. The animals housed in individual cages were fed 3 kg grass silage on a daily base at 08:30 in the morning and at 17:00 in the evening as two meals with water ad libitum. Trial lasted for 20 days; 15 days for adaptation, 5 days for sample collection. For digestibility trial, each animal's feces was weighed daily and a 10% aliquot retained, composited and frozen. Composited samples were subsequently dried in a forced air oven at 60°C at 48 h. Apparent dry matter, crude protein, crude fiber and nitrogen free extract digestibility were determined.

On the final day of the experiment, an amount of 40 ml ruminal fluid samples was obtained from each animal through the rumen tube 2 h after the morning feeding. The pH was determined with pH meter (Accumet, Fischer Scientific, USA) immediately. Then, the samples were divided into two different 20 ml bottles.

Rumen ammonia N was determined by description of Markham [14] from the 20 ml rumen fluid samples. The remaining of the 20 ml of ruminal fluid samples was treated with 1 ml of a 25% (w/v) dilution of metaphosphoric acid per 4 ml of ruminal fluid, and was stored at -20°C for volatile fatty acids analysis, as indicated by Horney et al.[15]. Volatile fatty acid concentrations were also analyzed in gas chromatography (Agilent 6980N, USA) with using 30 m x 0.53 mm (i.d.) capillary colon (Restek Corp. Canada).

ANOVA was used to determine the differences between the nutrinet contents of the grass silage prepared by different additives and degraded nutrient amounts in the rumen on an hourly base. Differences between the groups were defined by the Duncan test. Data were represented as mean ± S.E.M (Standard error of mean). Descriptive Statistics analysis was done on digestibility and rumen parameters. Minitab [16] was used all the calculations.

RESULTS

Nutrients contents and pH values of the grass silages prepared were displayed in the *Table 1*. Rumen degradability ratios of the DM, OM, CP, and CF of the incubated and dried silage samples were depicted in the *Table 2*. The data obtained from the grass silage sample were also shown in the *Table 3*.

Table 1. *Nutrient contents and pH values of the grass silages without - or with different additives*
Tablo 1. *Farklı katkı maddeleri ilave edilen veya edilmeyen ot silajlarının pH ve besin madde içerikleri*

Nutrient contents and pH	Grass silage (S1)	Grass + 2.5% barley silage (S2)	Grass + 5% barley silage (S3)	Grass + 2.5% barley + 1% molasses + 0.5% salt silage (S4)	Grass + 5% barley + 2% molasses +1% salt silage (S5)
Dry matter	29.51±2.13 ^{b}	34.20±1.05 ^{a}	29.43±0.76 ^{b}	31.45±0.61 ^{ab}	29.16±1.70 ^{b}
Ash	9.40±0.09 ^{c}	9.53±0.22 ^{c}	11.25±0.20 ^{b}	10.89±0.33 ^{b}	12.93±0.45 ^{a}
Organic matter	90.60±0.09 ^{a}	90.48±0.21 ^{a}	88.75±0.48 ^{b}	89.11±0.78 ^{ab}	87.07±0.45 ^{c}
Crude protein	9.77±0.49 ^{a}	7.75±0.65 ^{b}	8.14±0.06 ^{b}	10.41±0.52 ^{a}	10.86±0.46 ^{a}
Crude fiber	37.69±1.18 ^{a}	35.10±1.53 ^{ab}	30.42±0.41 ^{c}	36.43±0.89 ^{a}	33.12±0.39 ^{bc}
Ether extract	4.39±0.63 ^{ab}	3.22±0.20 ^{bc}	3.02±0.14 ^{c}	4.14±0.41 ^{abc}	4.49±0.20 ^{a}
Nitrogen free extarct	38.75±2.13 ^{b}	44.41±2.17 ^{a}	47.17±1.48 ^{a}	38.13±0.58 ^{b}	38.60±0.24 ^{b}
pH	4.48±0.03 ^{a}	4.10±0.09 ^{b}	4.02±0.02 ^{b}	4.07±0.04 ^{b}	3.97±0.03 ^{b}

a, b, c: *Differences between values having different letters in the same line are statistically significant (P<0.05)*

Table 2. *Rumen degradabilities of the grass silages without - or with different additives with hours, %*
Tablo 2. *Farklı katkı maddeleri ilave edilen veya edilmeyen ot silajlarının rumende saatlere göre yıkılabilirlikleri %*

Hour	Nutrient contents	Grass silage (S1)	Grass + 2.5% barley silage (S2)	Grass + 5% barley silage (S3)	Grass + 2.5% barley + 1% molasses + 0.5% salt silage (S4)	Grass + 5% barley + 2% molasses +1% salt silage (S5)
8	Dry matter	34.91±1.72 ^{b}	40.13±0.42 ^{a}	41.38±0.57 ^{a}	41.78±1.50 ^{a}	42.62±1.95 ^{a}
	Organic matter	32.11±1.56 ^{b}	37.54±0.68 ^{a}	38.17±0.44 ^{a}	38.56±1.27 ^{a}	39.25±1.84 ^{a}
	Crude protein	59.57±1.09 ^{b}	59.08±0.80 ^{b}	58.67±0.73 ^{b}	63.74±1.98 ^{a}	65.93±0.66 ^{a}
	Crude fiber	17.58±1.30	20.98±1.52	19.13±2.27	21.65±1.09	22.70±1.96
16	Dry matter	44.60±0.75 ^{c}	47.47±0.40 ^{ab}	48.64±0.48 ^{ab}	47.02±1.24 ^{b}	49.80±0.32 ^{a}
	Organic matter	41.55±0.65 ^{b}	44.69±0.39 ^{a}	45.00±0.76 ^{a}	44.32±1.48 ^{a}	46.52±0.36 ^{a}
	Crude protein	63.46±0.98 ^{c}	63.30±0.46 ^{c}	67.07±0.86 ^{b}	68.42±1.16 ^{b}	71.54±0.31 ^{a}
	Crude fiber	29.22±1.54 ^{b}	30.18±1.78 ^{ab}	30.83±0.92 ^{ab}	31.46±0.99 ^{ab}	33.44±0.43 ^{a}
24	Dry matter	55.28 ±0.97 ^{b}	56.88±1.02 ^{ab}	56.65±0.81 ^{ab}	57.14±0.73 ^{ab}	58.98±0.13 ^{a}
	Organic matter	52.52±2.06	53.14±1.77	53.96±0.85	54.59±1.03	56.45±0.34
	Crude protein	67.20±0.98 ^{c}	67.91±1.23 ^{c}	69.96±1.08 bc	73.33±1.35 ^{ab}	75.81±0.85 ^{a}
	Crude fiber	47.39±0.88 ^{ab}	47.86±0.40 ^{ab}	45.88±1.09 ^{b}	50.31±1.24 ^{a}	49.72±1.02 ^{a}
48	Dry matter	67.78±2.18 ^{c}	69.67±0.02 ^{bc}	74.74±0.20 ^{a}	72.54±1.22 ^{ab}	73.64±1.73 ^{ab}
	Organic matter	66.29±2.93 ^{b}	68.80±0.09 ^{a}	72.86±0.31 ^{a}	71.33±1.22 ^{a}	71.76±1.83 ^{a}
	Crude protein	71.63 ±1.29 ^{c}	74.29±0.43 ^{bc}	79.97±0.81 ^{a}	77.12±1.08 ^{ab}	80.78±1.93 ^{a}
	Crude fiber	69.87±1.73 ^{b}	71.41±0.38 ^{ab}	72.04±0.23 ^{ab}	71.73±1.19 ^{ab}	74.98±1.39 ^{a}

a, b, c: Differences between values having different letters in the same line are statistically significant (P<0.05)

Table 3. *Dry matter and nutrient digestibilities of the grass silage in rams, %*
Tablo 3. *Ot silajının koçlarda kuru madde ve besin madde sindirilebilirlikleri %*

Dry matter	Organic matter	Crude protein	Crude fiber	Ether extract	Nitrogen free extract
64.49±1.94	65.74±1.78	53.21±6.10	63.95±0.51	66.38±2.93	64.68±2.53

Table 4. *Rumen fluid parameters in rams feed grass silage*
Tablo 4. *Ot silajı verilen koçlarda rumen parametreleri*

pH	NH3-N (mg/l)	Volatile fatty acids (mmol/l)				
		Acetic acid	Propionic acid	Butyric acid	Isovaleric acid	Valeric acid
6.81±0.09	165±20.0	51.17±10.23	10.51±0.85	5.22±2.03	1.56±1.10	1.27±0.28

DISCUSSION

Nutrients and dry matter contents of the silage samples prepared in this study varied significantly in regard with the botanical variation, additives put in the silage samples, and distrubition ratios of the leaf, trunk, and stem of the grass. The lowest dry matter content was in the S5 (grass+5% barley +2% molasses+1% salt silage) with a percentage of 29.16% while the highest level was in the S2 added 2.5% barley. Like wise, the crude protein levels were low in the S2 and S3, both made of grass+barley mixture (7.75 and 8.14%, respectively while those in the S4 and S5, both made of grass +barley+molasses+salt were high (10.41 and 10.86%, respectively). Thus, nitrogen free extract was found to be high in the S2 and S3 (44.42 and 47.17% respectively).

In our study, DM and CP values determined in the grass silages prepared with different mixtures are in parallel with those of the literatures [8,17,18]. Thus, the DM results are similar to those reported by [3,5,7] while the CP results found in our study are either similar to or lower than the values indicated in these research reports. Another study [6] supporting

our results has documented the DM and CP values in three different grass silage samples to be 34.3, 29.9, 38.8% and 8.12, 9.37, 11.87%, respectively. The values of the DM, CP, ash, CF, EE in our silage samples have been foud to be close to the results of Gurdogan et al.[18] who have prepared silages with whole crop barley. The NFE values of the S2 and S4 silage samples are also similar to the reports of those studies. Differences in the nutrient contents of the grass silage samples may be explained by the fact that the grass used in our study comprises plants belonging to gramineae family at very high levels, just as indicated by the literature Kaya et al.[19].

The pH values of the silage samples have decreased regarding to the amount and kind of the additives used. The highest level has been measured in the S1 (4.48) while it is the lowest in the S5 (3.97). The pH value of Rinne et al.[7] in the grass silage (4.10) is in parallel with our results, just as the results of More et al.[6] and Cone et al.[5]. Like wise, another study [6] has found the pH values in three grass silages to be 4.6, 4.6, and 4.4.

Rumen degradabilities of the DM and nutrients of the silage samples made of grass and mixtures, and incubated for different periods, have shown significant variations with regard to the silage type and incubation period ($P<0.05$). CP degradability ratios of the silage samples containing grass and 5% barley and grass, 5% barley, molasses and salt, incubated particularly for 24 and 48 h have been determined to be relatively higher *(Table 2)*. These results are similar to the findings of Baytok and Muruz [3] who have measured timely CP degradability ratios of the grass silages incubated in the rumen for 4-48 h. Besides, timely OM and CP degradability ratios are in-between the results of the Cone et al.[5] and Keyselingk et al.[8]. Finally, these ratios are also similar to the results of Kaya et al.[20].

Digestibility ratios of the DM, CP, CF and NFE in the our grass silage samples in rams have been found to be 64.49, 53.21, 63.95 and 64.68%, respectively. These results are in the range of the findings on the whole crop barley preserved with different techniques [18]. However, DM and CF digestibility ratios (52.3%, 58%) of the grass silages in lambs have been reported [6] to be lower than our results. Moreover, CP and OM digestibility ratios (66.2 and 72.3%) of the silage samples made of grass harvested during the late vegatation period have been reported [7] to be higher than our results. Furthermore, rumen pH value of the cattle fed by these samples has been measured as 6.29, and the values of the acetic, propionic, butyric, valeric and isovaleric acids have been determined as 659, 161, 135, 15.8, 12.9 mmol/mol, respectively [7]. These results have showed relative similarities to our results. Friggens et al.[9] has also found the levels of the acetic, propionic and butyric acid to be 68.04, 20.54 and 7.21 mol/100 mol, respectively, in the rumen fluids of the rams fed by 800 g grass silage, weighed on the basis of DM. The results are higher than the findings of our study.

Consequently, different silage types, either alone or by adding barley, molasses, or salt, can be made through using the grass produced in Kars district. However, it has been determined that, feedstuff rich in carbohydrate such as barley and molasses have increased the quality of grass silage.

REFERENCES

1. **Akyıldız R:** Yemler Bilgisi ve Teknolojisi. Ankara Üniv Zir Fak Yay, No: 974. Ankara, 1986.
2. **Jacobs JL, Morris RJ, Zorrilla-Rios J:** Effect of ensiling whole barley grain with pasture on silage quality and effluent production and the performance of growing cattle. *Aust J Exp Agric,* 35, 731-738, 1995.
3. **Baytok E, Muruz M:** The effects of formic acid or formic acid plus molasses additives on the fermentation quality and DM and ADF degradabilities of grass silage. *Turk J Vet Anim Sci*, 27, 425-431, 2003.
4. **Haigh PM, Parker JWG:** Effect of silage additives and wilting on silage fermentation, digestibility and intake, and on liveweight change of young cattle. *Grass and Forage Sci,* 40, 429-436, 1985.
5. **Cone JW, Van Gelder AH, Soliman IA, De Visser H, Van Vuuren AM:** Different technigues to study rumen fermentation characteristics of maturing grass and grass silage. *J Dairy Sci,* 82, 957-966, 1999.
6. **Moore KJ, Lemenager RP, Lechtenberg VL, Hendrix KS, Risk JE:** Digestion and utilization of ammoniated grass-legume silage. *J Anim Sci,* 62, 235-243,1986.
7. **Rinne M, Huhtanen P, Jaakkola S:** Digestive processes of dairy cows fed silages harvested at four stages of grass maturity. *J Anim Sci,* 80, 1986–1998, 2002.
8. **Keyserlingk von MAG, Swift M L, Puchala R, Shelford JA:** Degradability characteristics of dry matter and crude protein of forages in ruminants. *Anim Feed Sci Technol*, 57, 291-311, 1996.
9. **Friggens NCJ, Oldham D, Dewhurst RJ, Horgan G:** Proportions of volatile fatty acids in relation to the chemical composition of feeds based on grass silage. *J Dairy Sci,* 81, 1331–1344, 1998.
10. **TIM:** Tarım İl Müdürlüğü Kayıtları. İstatistik Şubesi, Hayvancılık İstatistikleri, Kars, 1997.

11. **AOAC:** Official Methods of Analysis 14th. ed, Arlington, Virginia, U.S.A., 1990.
12. **Polan CE, Stive DE, Garrett JL:** Protein preservation and ruminal degradation of ensiled forage treated with heat, formic acid, ammonia, or microbial inoculant. *J Dairy Sci,* 81, 765-776, 1998.
13. **Bhargava PK, Ørskov ER:** Manual for the use of nylon bag technique in the evaluation of feedstuff. The Rowett Research Institute, 1-20, Bucksburn, 1987.
14. **Markham RA:** Steam distilation apparatus suitable for micro-kjeldahl analysis. *Biochem J,* 36, 790-796, 1942.
15. **Horney MR, DelCurto T, Stamm MM, Bailey RK, Brandyberry SD:** Early-vegetative tall fescue hay vs alfalfa hay as a supplement for cattle consuming low-quality roughages. *J Anim Sci,* 74, 1959–1966, 1996.
16. **Minitab:** Minitab Reference Manual Release 10. for Windows. Minitab Inc, 1994.
17. **Keyserlingk Von MAG, Shelford JA, Puchala R, Swift ML, Fisher LJ:** In situ disapperance of amino acids from grass silages in the rumen and intestine of cattle. *J Dairy Sci,* 81, 140-149, 1998.
18. **Gurdogan F, Tatlı P, Cerci İH, Azman MA:** The effects on food intake and nutrient digestibility of different conservation methods of whole crop barley harvested at boot stage in lambs. *Turk J Vet Anim Sci,* 26, 1237-1242, 2002.
19. **Kaya I, Oncuer A, Unal Y, Yıldız S:** Nutritive value of pastures in kars district I.botanical and nutrient composition at different stages of maturity. *Turk J Vet Anim Sci,* 28, 275-280, 2004.
20. **Kaya I,Yıldız S, Kaya S, Oncuer A:** Nutritive value of pastures in Kars district II. Degradation kinetics in the rumen with respect to stages of maturity. *Turk J Vet Anim Sci,* 28, 280-287, 2004.