The Effects of Different Moisture Content and Ensiling Time on Silo Degradation of Structural Carbohydrate of Orchardgrass

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ABSTRACT : This study determined the influence of moisture, ensiling time and their interactions on the losses of hemicellulose and cellulose during ensiling of orchardgrass. Orchardgrass containing 80 (HM), 70 (MM) and 55% (LM) moisture was ensiled in 3 laboratory silos of 500 ml capacity for 3, 7, 21 and 91 days. The dry matter (DM), water-soluble carbohydrates (WSC), hemicellulose and cellulose contents of the ensiled orchardgrass was lowered than that of the untreated grass regardless of moisture content. Ensiling orchardgrass for 91 days (d) decreased ($p<0.01$) hemicellulose contents from 19 to 15%, 20 to 15% and 18 to 12% and cellulose from 31 to 29%, 29 to 26% and 27 to 26% for LM, MM and HM silage, respectively. Results from fermentation of LM and MM silages were within acceptable guidelines except for butyric acid and ammonia after 3 weeks of ensiling of MM which appeared to be lower than ideal. The results of the fermentation of HM silages were poor showing higher concentration of acetic, propionic and butyric acids and traces of isovaleric, valeric and caproic acids with ammonia at all stage of time. While the DM losses from LM and MM silages over the ensiling period were acceptable, that for HM silage increased to 13% after 91 d ensiling, confirming a poor fermentation process occurred. The greatest WSC losses occurred within 7 d of ensiling and the lowest losses occurred after 3 weeks of ensiling. Except in HM silage, the hemicellulose and cellulose losses were highest ($p<0.01$) in the first 3 weeks of ensiling. Hemicellulose losses were between 19 and 22% and 4.2 and 5.9% up to 3 weeks and after 3 weeks of ensiling LM and MM silages, respectively. Cellulose losses were small. In contrast, hemicellulose losses after 3 weeks of ensiling of HM silage was about 50% higher than over the first 3 weeks possibly due to clostridial type fermentation. The results showed that increasing ensiling time of high moisture orchardgrass would result in the excessive losses of DM, WSC, hemicellulose and cellulose in the silage. ***(Asian-Aust. J. Anim. Sci. 2002. Vol 15, No. 2 : 213-217)***

Key Words : Ensiling, Moisture Content, Orchardgrass, Structural Carbohydrates, Losses

INTRODUCTION

Measurement of the breakdown of structural carbohydrate during ensiling is difficult and depends on many factors, which are not fully understood. Similarly, information available on the effect of moisture content on the breakdown during ensiling of the structural carbohydrates, hemicellulose and cellulose is scarce, relative to that for non-structural carbohydrates. Previous studies have reported that high moisture content lowers the critical pH value of silage providing an environment conducive to the growth of clostridial bacterial that are responsible for the undesirable breakdown of nutrients such as WSC (Perry et al., 1967; Sullivan, 1973 and Gary, 1992). However, little is known about the effect of moisture content and ensiling time on the degradation of cellulose and hemicellulose.

Most reports indicate that the acids produced during ensiling arise from the fermentation of WSC alone (McDonald and Whittenbury, 1977 and Butler et al., 1973). Recent studies revealed that other substances could also act as substrates (McDonald et al., 1991 and Yahaya et al., 2000). It is expected that cellulose and hemicellulose could be degraded by enzyme (cellulase and hemicellulase) bacterial action and organic acids produced during fermentation (Yahaya et al., 2001 and McDonald et al., 1991).

Generally, when forage is ensiled, plant cell respiration plus aerobic microbial fermentation and lactic acid bacterial activity take place before activity stabilises (Butler and Bailey, 1973; McDonald et al., 1991). Studies have shown that plant cell respiration plus aerobic microbial fermentation and the lactic acid bacterial activity stages are completed within 3 and 21 days of ensiling (Stephen and Micheal, 1960; Sullivan, 1973 and Gary, 1992). But little is known about the amounts of hemicellulose and cellulose lost during the 3 weeks during ensiling.

This study aimed to evaluate the extent of hemicellulose and cellulose losses in low (55%), medium (70%) and high (80%) moisture content orchardgrass forage after 3, 7, 21 and 91 days of ensiling. To examine whether any interactions between moisture content and ensiling time influenced the losses of hemicellulose and cellulose during ensiling.

MATERIALS AND METHODS

Silage preparation

Orchardgrass (*Dactylis glomerata L.*) was harvested during the early heading stage at the Obihiro University

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Farm, Japan. The grass was cut into 2 to 3 cm lengths using a mechanical forage cutter, and was thoroughly mixed before dividing into three equal parts. One part (327 g) fresh matter (80% moisture content) was ensiled in laboratory glass silo of 500 ml capacity. The second (285 g) and third (203 g) parts were wilted to moisture contents of 70% and 55%, respectively, and were ensiled in similar laboratory glass silos. Twele silos of each silage type were prepared. Three silos from the three groups representing 80%, 70% and 55% moisture content were opened after 3, 7, 21 and 91 days (d) of ensiling, and were weighed to determine the extent of losses during ensiling. A representative sample from each silo was mixed, and frozen at -15°C for chemical analysis.

Chemical analyses

The dry matter (DM) content of harvested material and silage from the three groups was determined by freeze drying for a minimum of 24 h. The chemical composition of fresh material orchardgrass is shown in table 1. The crude protein (CP) and ether extract (EE) were determined by standard procedures (AOAC, 1990). Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined as described by Goering and Van Soest (1970) (modified by Van Soest et al., 1991). Cellulose and hemicellulose contents were calculated by subtracting ADL from ADF and ADF from NDF respectively. Silage pH was immediately determined on a prepared extract from the silages, using a pH meter. Standard procedures were applied to estimate water-soluble carbohydrates (WSC; Deriaz, 1961), lactic acid (Baker and Summerson, 1961), volatile fatty acids (VFA; gas chromatography according to George and Melvin, 1979), and ammonia (Conway and O'Malley, 1942).

Table 1. Chemical composition (%) in fresh harvested orchardgrass

	LM	MM	HM
PH	6.35	6.23	6.28
Dry matter	44.29	32.88	20.35
	---------- % DM ----------		
Crude protein	13.42	12.85	11.3
Ether extract	2.99	3.22	3.34
Neutral detergent fiber	55.2	53.95	50.81
Acid detergent fiber	36.71	33.92	32.42
Acid detergent lignin	5.81	5.46	5.17
Water soluble carbohydrates	7.75	6.68	6.66
Hemicellulose	18.49	20.03	18.39
Cellulose	30.9	28.46	27.25

LM=Low moisture, MM=Medium moisture, HM=High moisture.

Statistical analysis

Data obtained from silage fermentation were treated by analysis of variance of two-way factorial randomized block design with moisture and ensiling time, respectively, at 3 and 4 levels. Means differences between ensiling time were determined using multiple range test procedures (Duncan, 1955 and Snedecor and Cochran, 1980).

RESULTS AND DISCUSSION

Silage chemical composition and fermentation quality

The chemical compositions of the silages are shown in table 2. The dry matter (DM), crude protein (CP), water-soluble carbohydrates (WSC), hemicellulose, and cellulose contents over the ensiling periods within the low moisture (LM), medium moisture (MM) and high moisture (HM) silages were lower than that of the fresh grass (table 1). Ensiling orchardgrass from 0 to 91 d resulted in decreased ($p<0.01$) hemicellulose content from 18 to 15%, 20 to 15% and 18 to 12% and cellulose from 31 to 29%, 28 to 26% and 27 to 26% respectively for LM, MM and HM silage. Similarly, comparing hemicellulose and cellulose contents at any ensiling time reveals a gradual decreased in hemicellolose and cellulose contents as the ensiling time and moisture content of the orchardgrass increased ($p<0.01$). In contrast to cellulose, the hemicellulose content fell from 18 to 12% after 91 d of ensiling in HM silage, compared with LM and MM silages (table 2) and the original herbage.

The fermentation qualities of the three silages are shown in table 3. Except in HM silage the pH values within LM and MM silages were progressively lowered ($p<0.01$), while lactic acid % increased as the ensiling process advanced. The results obtained from fermentation of LM and MM silages were within acceptable guidelines except for butyric acid and ammonia after 3 weeks of ensiling of MM which appeared to be lower than ideal (Butler and Bailey, 1973; McDonald et al., 1991 and Chamberlain and Wilkinson, 1996). Low moisture content probably influenced the relative growth of homofermentative and heterofermentative lactic acid bacteria during ensiling, which in turn lowered the pH of silage, minimizing deterioration (Wilkins et al., 1971; Morgan et al., 1980; McDonald et al., 1991 and Wayne et al., 1998). However, the result of fermentation of HM silage was poor due to clostridial type of fermentation indicated by the higher concentration of acetic, propionic and butyric acids compared to LM and MM silages (table 3). The concentration of ammonia, as % total nitrogen, in HM silage was always about 300% higher than that for LM and MM silages, indicating a defective type of preservation (Flynn, 1981). Similarly, no isovaleric, valeric and caproic acids were noticed in LM and MM silages, compared to

Table 2. Chemical composition (%) of orchardgrass silages

	LM				MM				HM				SEM	Contrast		
	3 d	7 d	21 d	91 d	3 d	7 d	21 d	91 d	3 d	7 d	21 d	91 d		L	M	L×M
														P		
Dry matter	44.1	43.5	43.1	42.9	32.6	32.3	32.3	31.6	20.4	19.8	19.2	18.3	0.18	++	++	++
Crude protein[1]	12.7	12.1	11.6	11.2	12.1	11.6	11.0	10.5	10.9	10.5	10.4	10.3	0.06	NS	++	++
Ether extract	2.0	2.0	2.5	2.6	1.9	2.5	2.7	3.0	2.0	2.6	2.8	3.3	0.14	++	++	+
Neutral detergent fiber	54.9	52.4	50.6	49.4	53.4	50.5	48.3	46.5	50.3	48.3	46.7	43.9	0.23	++	++	++
Acid detergent fiber	36.7	35.2	35.0	34.5	33.8	32.6	32.1	32.3	32.3	31.6	31.4	32.1	0.23	++	++	++
Acid detergent lignin	6.1	5.8	5.7	5.4	5.9	5.7	5.5	5.3	5.3	5.4	5.6	5.8	0.15	+	+	+
Water soluble carbohydrate	7.1	5.9	4.6	4.7	5.0	3.1	2.9	1.4	1.2	1.0	1.0	1.1	0.15	++	++	++
Hemicellulose	18.1	17.1	15.6	15.0	19.6	17.9	16.1	15.2	17.9	16.7	15.3	11.9	0.27	++	++	++
Cellulose	30.7	29.5	29.4	29.1	27.9	26.9	26.7	25.9	27.0	26.2	25.8	26.3	0.23	++	++	++

L=Ensiling time, LM=Low moisture, MM=Medium moisture, HM=High moisture, M=Moisture content, SEM=Standard error of means. L×M=Interaction of length of ensiling and moisture content, d=day of ensiling, Each value in the table represent a mean over three silo. NS=Not significant. $^{*}p<0.05$, $^{**}p<0.01$.

Table 3. Fermentation quality of orchardgrass silages (%)

	LM				MM				HM				SEM	Contrast		
	3 d	7 d	21 d	91 d	3 d	7 d	21 d	91 d	3 d	7 d	21 d	91 d		L	M	L×M
														P		
pH	6.30	6.22	5.71	5.10	6.15	5.46	4.97	4.73	5.48	5.52	5.80	5.86	0.04	++	++	++
Lactic acid	1.10	1.29	4.08	4.34	5.17	5.69	6.74	7.16	2.58	1.77	1.84	1.84	0.34	++	++	++
Acetic acid	0.02	0.09	0.17	0.20	0.09	0.16	0.15	0.30	0.30	0.53	1.35	1.91	0.06	++	++	++
Propionic acid	0.03	0.01	0.04	0.06	0.06	0.05	0.24	0.19	0.09	0.30	0.67	0.93	0.03	++	++	++
Butyric acid	0.00	0.00	0.00	0.00	0.00	0.06	1.63	1.12	0.33	2.38	3.67	4.47	0.14	++	++	++
Isovaleric acid	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.04	0.37	0.49	0.03	++	++	++
Valeric acid	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.12	0.23	<0.01	++	++	++
Caproic acid	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.07	0.69	0.93	0.01	++	++	++
Ammonia (% Total N)	3.46	3.82	5.76	9.54	6.75	8.96	10.97	21.67	10.91	21.13	51.51	64.03	0.69	++	++	++

L=Ensiling time, LM=Low moisture, MM=Medium moisture, HM=High moisture, M=Moisture content, SEM=Standard error of means. L×M=Interaction of length of ensiling and moisture content, d=day of ensiling, Each value in the table represent a mean over three silo. NS=Not significant. $^{*}p<0.05$, $^{**}p<0.01$.

HM sialge. This was probably because the pH of HM silage was not sufficient to prevent clostridial growths, as they are more sensitive to low pH than are lactic acid bacteria and yeast (Wieringa. 1958) (table 3). High moisture content favored clostidia and other microorganisms resulting in excessive utilization of plant WSC. hemicellulose. cellulose, as energy sources to degrade lactic acid to acetic, butyric, propionic acids and other VFAs (Stephen and Micheal. 1960; Perry et al., 1967; Sullivan. 1973; Morgan et al., 1980 and Gary. 1992).

Similarly. extensive degradation of protein occurred in HM silage was reflected by high contents of ammonia and low CP (McDonald and Whittenbury. 1977; Wayne et al., 1998 and Micheal. 1984).

The results of the silage fermentation at any stage of ensiling period. showed that increasing ensiling time of high moisture orchardgrass would result in the excessive losses of DM. WSC. hemicellulose and cellulose in the silage.

Losses during ensiling

The DM. WSC. hemicellulose and cellulose losses are shown in table 4. The DM losses increased ($p<0.01$) across the length of ensiling and appeared highest after 91 d of

Table 4. Losses of dry matter (DM), water soluble carbohydrates (WSC) and structural carbohydrates during ensiling (%)

	LM				MM				HM					Contrast		
	3d	7d	21d	91d	3d	7d	21d	91d	3d	7d	21d	91d	SEM	L	M	L×M
															P	
DM	0.7	2.1	3.4	4.5	1.2	2.7	3.5	5.7	0.7	4.6	8.2	12.7	0.6	++	++	++
WSC	8.6	25.4	42.2	42.4	47.5	56.2	60.2	81.0	82.3	85.7	86.3	86.2	1.7	++	++	++
Hemicellulose	2.7	9.3	18.5	22.7	3.3	12.9	22.3	28.2	3.2	13	24	44	1.6	++	++	++
Cellulose	1.4	6.7	8.2	10.2	3.1	8.1	9.6	14.1	1.6	8.2	13	16	0.9	++	++	++

L=Ensiling time, LM=Low moisture, MM=Medium moisture, HM=High moisture, M=Moisture content, SEM=Standard error of means. L×M=Interaction of length of ensiling and moisutre content, Each value in the table represent a mean over three silo. NS=Not significant. ¯p<0.05, ¯¯p<0.01

ensiling regardless of moisture content. Except in HM silage, the DM losses in LM and MM silages over the ensiling period were small and within the acceptable range (McDonald et al., 1991). At all ensiling times DM losses for the HM silage increased by about 4% reaching 12% by 91 d, confirming a poor fermentation process (Xiccoto et al., 1998). Higher DM losses (18.6%) was obtained in unwilted silages (Watson and Nash, 1960). This indicated that higher moisture contents and increasing ensiling times were associated with higher DM losses (Wayne et al., 1998).

The WSC losses for all silages evaluated increased with increasing ensiling time. The greater losses of WSC occurred within 0 to 7 d of ensiling and least after 21 to 91 d of ensiling. Increasing the ensiling time of a high moisture crop resulted in the loss of almost all the WSC confirming the finding of Matsuoka et al. (1993). Higher moisture provided an environment conducive to clostridial type of fermentation likely resulting in excessive utilization of WSC as the main substrate for energy for microbial growth (Gary, 1992). However, the WSC loss recorded in HM silage in this study was similar to that obtained (86%) for lucerne after 22 d ensiling (Ayako et al., 2000). But lower than the 83 and 92.4% in orchardgrass ensiled for 35 d respectively, by Yahaya et al. (2001) and Matsuoka et al. (1997).

Hemicellulose and cellulose losses increased ($p<0.05$) with ensiling time as a result of microbial activity during fermentation (Dewar et al., 1963). Losses of hemicellulose and cellulose in LM and MM silages were higher during first 3 weeks of ensiling compared with losses from d 21 to 91 after ensiling. This could have been due to the cumulative activity of plant cell respiration, enzymes (hemicellulase and cellulase) and aerobic and anaerobic facultative bacteria in the fresh ensiled forage (McDonald et al., 1962). The cellulose losses were small compared to hemicellulose and the amount lost was 8 to 10% during the first 21 d and 2.0 to 4.5% during 21 to 91 d of ensiling LM and MM silages, respectively. The hemicellulose and cellulose losses after 21 to 91 d of ensiling in LM and MM silages were small, probably due to organic acid hydrolysis during fermentation (McDonald et al., 1960). However, comparing the hemicellulose and cellulose in HM silage, showed the value obtained over 21 to 91 d was about 50% lower than during 0 to 21 d of ensiling. This indicates a clostridial type fermentation (Dewar et al., 1963, table 3).

CONCLUSION

The DM, CP and WSC contents were lowered during ensiling regardless of silage moisture content. However, the losses of DM, WSC, hemicellulose and cellulose were increased as ensiling time and silage moisture content increased. Similarly, at all ensiling time losses from HM silages were highest causing a significant interaction between moisture content and ensiling time. The results revealed that increasing the ensiling time of high moisture orchardgrass would result in the excessive losses of DM, WSC, hemicellulose and cellulose and higher levels of ammonia (% total nitrogen) in the silage.

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