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Treatment of Low Quality Forages by Hydrogen Peroxide and(or) Anhydrous Ammonia and Their Utilization in Ruminant Nutrition

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TREATMENT OF LOW QUALITY FORAGES BY HYDROGEN PEROXIDE AND(OR) ANHYDROUS AMMONIA AND THEIR UTILIZATION IN RUMINANT NUTRITION

by

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A dissertation submitted in partial fulfillment
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DEDICATION

My mother Zhor,

You would not agree to be away from me for such a long time if it was not for a noble cause such as seeking knowledge.

My father Mohammed,

You have never refused any of my requests, especially when it concerned education.

To both of you,

May this work be a modest reward for your sacrifices.

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"If ye would count up the favors of God, never would ye be able to number them; for God is Oft-Forgiving, Most Merciful."

Qur'an 16:18
Translation of A. Yusuf Ali

"Say: 'Praise be to God, who begets no son, and has no partner in (His) dominion: nor (needs) He any to protect Him from humiliation: yea magnify Him for His greatness and glory.'"

Qur'an 17:111
Translation of A. Yusuf Ali

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Mohammed Diouri

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Treatment of Low Quality Forages by Hydrogen Peroxide and(or) Anhydrous Ammonia and Their Utilization in Ruminant Nutrition

by

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Three experiments were conducted to evaluate low quality forages treatment by anhydrous ammonia (NH_3) and(or) hydrogen peroxide (H_2O_2).

In experiment I, a control and three treatments of barley straw were compared: NH_3, NH_3 after rehydration of the straw with water (NH_3+H_2O), and NH_3 after rehydration with a H_2O_2 solution (NH_3+H_2O_2). Forages were fed, with a supplement, at two levels of intake (ad libitum and 75% of ad lib.). Treatments were administered in a split-plot in a Latin square design to 8 ewes. Total collections and rumen digesta measurements were made. Ammoniation, rehydration, and H_2O_2 increased primarily dry matter intake (DMI) but also digestibility of different nutrients. A similar trend was observed in rumen fermentation characteristics. Dry matter (DM) digestibility was slightly raised by limiting DMI.

Digestible DMI (DDMI) was correlated with both the increase of forage CP content and the decrease of forage neutral detergent fiber. Water intake and output were highly correlated with fiber intake and digestibility. Acid insoluble ash (AIA) estimated digestibility better than acid detergent lignin insoluble ash (ADLIA). Both markers were adequate in determining hemicellulose digestibility.

In experiment II, ten solutions were prepared to rehydrate wheat straw. Six solutions were adjusted (with NaOH) to a pH of 9, 11, or 13 and contained 2% H_2O_2. Four solutions had a pH of 7 or 11 and contained no H_2O_2. Half of the straws were treated with NH_3. In situ DM disappearance (DMD) of the different straws was measured at different times in 10 Holstein cows and three periods (incomplete block design). The evolution of DMD was slow and almost linear. The positive effect of ammoniation on DMD was consistently apparent at all pH levels though depressed at pH 11. The effect of H_2O_2 was minor, but was complementary with ammoniation.

In experiment III, mature baltic rush (*Juncus balticus Willd.*) was either treated or not with NH_3. Eight wethers had ad libitum access to the nonsupplemented forages in a cross-over design. Total collections and rumen digesta measurements were made. DMI and DDMI were not affected by treatment, but DM digestibility was decreased by ammoniation.

(84 pages)

INTRODUCTION

The demographic growth in the world, requiring an increase in food production, necessitates the expansion of the crop lands to satisfy the human food needs. The lands that used to be allocated primarily to grazing animals are therefore shrinking. Domestic animals will have to meet their nutritional requirements primarily in the feedstuffs that are not directly used by humans, namely range plants and crop residues and by-products.

Many of these plants and residues have a low nutritive value mainly because of the low content of crude protein and the low availability of the structural carbohydrates that constitute most of their dry matter. This low availability of structural carbohydrates is due primarily to the bonds linking lignin, which is virtually not digestible, with cellulose and hemicellulose.

Beside supplementation, different chemical and biological treatments have been studied to overcome the low nutrient availability of low quality forages. Anhydrous ammonia (NH_3) treatment has proven to be successful for several reasons. Its alkalinity makes cellulose and hemicellulose more available by partially dissolving them and by breaking the bonds between them and lignin. NH_3 also supplements the straw with nitrogen, and it is easy to use at the farm. Another chemical compound that has recently been used with successful results is hydrogen peroxide (H_2O_2). This oxidizing agent has greatly improved the utilization of different low quality forages both

in cattle and sheep. H_2O_2, however, acts optimally in alkaline milieus. Sodium hydroxide (NaOH) has been used to provide alkalinity for H_2O_2. This substance has the potential of sodium toxicity. Replacing it by NH_3 was expected not only to remedy this disadvantage but also to enhance the beneficial effects of NH_3 and H_2O_2.

This dissertation intends to contribute to the understanding and improvement of the treatment of different low quality forages with NH_3 alone or with other substances, as well as to the utilization of these forages in the ruminant nutrition.

CHAPTER I

NUTRITIONAL METABOLISM OF HYDROGEN PEROXIDE/ANHYDROUS AMMONIA-TREATED BARLEY STRAW IN EWE LAMBS

ABSTRACT

Combined treatments of low-quality forages with sodium hydroxide (NaOH) and hydrogen peroxide (H_2O_2) have been shown to increase energy availability. To keep the pH alkaline for a better action of H_2O_2, NaOH was replaced by anhydrous ammonia (NH_3). A control and three treatments of barley straw were compared: NH_3 (3% of DM), 3% NH_3 after rehydration of the straw to 15% moisture with water, and 3% NH_3 after rehydration to 15% moisture with a H_2O_2 solution (0.32% of DM). Forages were fed, with a faba bean supplement, at two levels of intake (ad libitum and 75% of ad lib.), to 8 ruminally cannulated ewes housed in elevated metabolism crates. Treatments were administered in a split-plot in a Latin square design. After a 14 d adaptation period, feeds, orts, and feces were measured and sampled during 5 d. On day 6, rumen fluid was sampled at 0, 2, 4, 6, 8, 12, and 16 h post-feeding to measure fermentation characteristics. At the ad libitum intake, digestible DM intake was 336, 455, 501, and 552 g/(h.d) for untreated, NH_3, NH_3+H_2O, and NH_3+H_2O_2-treated straws, respectively. The first, second, and fourth values were different from each other ($P < .01$), while the third one tended to be higher than the second ($P = .12$) and lower than

the fourth (P = .09). A similar trend was observed in digestibility and digestible intake of DM, OM, CP, cellulose, and hemicellulose. Total ruminal VFA and NH_3-N were increased by ammoniation and rehydration but not by H_2O_2. There was no difference among treatments in ruminal pH, proportion of propionate, or acetate/propionate ratio. The DM digestibility was raised from 56.7% at ad libitum intake to 58.2% at limited intake (P = .01). The combined NH_3-H_2O_2 treatment increases straw utilization by sheep compared with NH_3-H_2O.

INTRODUCTION

Besides supplementation (Wiedmeier et al., 1983), different chemical (Klopfenstein et al., 1979) and biological (Larwence and Abada, 1987; Wiedmeier et al., 1987) treatments have been studied to overcome the low nutrients availability of cereal straws. Anhydrous ammonia (NH_3) treatment has proved to be successful (Sundstol et al., 1978; Wiedmeier, 1988; Zorrilla-Rios et al., 1991) for several reasons. Its alkalinity makes cellulose and hemicellulose more available by partially dissolving them and by breaking the bonds between them and lignin. NH_3 also supplements the straw with nitrogen, although Males (1987) has estimated that only half of this added nitrogen is available to the animal as a protein source through rumen microbial synthesis. Ammoniation is also a cheap and easy treatment to use at the farm. Ammoniation has also some anti-toxin properties (Fremy et al., 1988; Frayssinet and Lafarge-Frayssinet, 1990; Kerr et al., 1990).

Another chemical compound that has recently been used with successful results is hydrogen peroxide (H_2O_2) (Gould, 1984). This oxidizing agent has greatly improved the utilization of different low-quality forages both in cattle and sheep (Atwell et al., 1991; Cameron et al., 1991a,b). H_2O_2 acts optimally in alkaline milieus (Gould, 1987).

Sodium hydroxide (NaOH) has been used to provide alkalinity for H_2O_2 (Kerley et al., 1986), but NaOH has the potential of sodium toxicity. Replacing it by NH_3 was expected

not only to remedy this disadvantage but also to enhance the beneficial effects of NH_3 and H_2O_2. The main objective of this study was to determine the impact, on straw nutritive value, of the combined NH_3/H_2O_2 treatment and the possible interaction or additivity of the two treatment components.

MATERIALS AND METHODS

Barley Straw Treatments

Four different treatments were compared:

1. Control
2. 3% NH_3
3. 3% NH_3 after rehydration of straw to 15% moisture with water
4. 3% NH_3 after rehydration of straw to 15% moisture with a H_2O_2 solution (to reach a level of .32% DM of H_2O_2).

Treatments 2 and 3 were used to isolate the effect of H_2O_2 and to confirm the results of former studies reporting the positive effect of moisture on straw ammoniation (Kernan et al., 1979).

Treatment Procedure

Ten 23 kg bales were randomly assigned to each treatment. Ten bales were set aside in a separate plastic bag for use as control. Ten bales were treated with enough water to bring the moisture level to 15%. This was accomplished by turning the cut edge of the bales up and pouring the water on slowly enough so there is no loss. The moisture content may have not

been uniform throughout the bale or among bales; but the ten bales were ground and mixed together after treatment. Ten bales were treated as above except a solution of H_2O_2 was used to rehydrate to 15% moisture instead of water. Three separate stacks (besides the control) were then made; one with straw bales that had not been rehydrated, one with straw bales rehydrated with water, and one with bales rehydrated with H_2O_2.

All four stacks were then placed in separately sealed 6 mm plastic bags. NH_3 was then introduced in the gaseous form into the appropriate bags through a perforated pipe at 3% of DM. After ammoniation, the bags were sealed for approximately 21 days and then opened to allow excess ammonia to escape. Straw was then ground, mixed in an auger-type grinder mixer, and stored in 30-gallon plastic containers until used.

Animal Model

Eight yearling ewe lambs (30 to 51 kg), fitted with ruminal fistulae, were used. The ewes were from three different breeds (4 Navajos, 2 Columbias, and 2 Black-faced) representing three major groups of sheep (carpet wool, dual purpose, and meat type).

The small number of sheep in each breed does not allow an accurate comparison of breed efficiencies (Schneider and Flatt, 1975; Pfister, 1983). Nevertheless, the diversified animal sample should allow a more confident generalization of the results.

Sample Collection and Analysis

After proper recovery from fistulation and adaptation to a straw-based diet, sheep were placed in elevated metabolism crates equipped with stainless steel feeders, water, and a feces-urine collection apparatus.

Treatments were administered in a split plot in a 4*4 Latin square design with repeated measures. Each of the 4 trial periods was composed of two subperiods. In the first subperiod, sheep had ad libitum access to straw (to determine the intake), allowing approximately 10% refusal. A supplement (whose nutrient composition is presented in Table 1) composed of ground faba bean (*Visia faba*) and fortified with vitamins and minerals as needed to meet nutrient requirements (NRC, 1985) was top dressed. The supplement intake was gradually adjusted to represent 25% of the ration in order to eliminate the possible negative associative effect between concentrate and roughage (Blaxter, 1969; Mould et al., 1983). Daily rations were given in 2 equal portions at 0800 and 1600. Diets were fed for a 14-d adaptation period followed by a 7-d collection period.

In the second subperiod, consisting of a 5-d adaptation and a 5-d collection, the sheep received the same diet but their ration was limited to 75% of each animal's ad libitum intake.

During d 1 to 5 of the collection periods, total fecal output was collected and weighed twice daily at the normal

feeding times. Fecal samples were dried at 60°C for 72 h and then ground to pass through a 2 mm screen using a Wiley mill (Thomas-Wiley Laboratories, Swedesboro, NJ). Dry matter content was calculated. Composites of fecal samples, within ewe and period, were then made. The amount of each sample that went into the composite was proportional to that sample's portion of the total output.

Feed (straw and supplement) was weighed at each feeding. Refusal was weighed back to determine intake. Starting 1 d before the beginning of the collection periods, samples of feed and refusals were taken at each feeding. These samples were ground to pass through a 1 mm screen and proportional composites were made as was described with feces samples.

All feed, ort, and fecal composite samples were analyzed (8 samples per treatment) for laboratory dry matter (DM), OM (AOAC, 1990), CP (Hach et al., 1985), ADF, NDF, and ADL (Van Soest, 1967).

Rumen measurements were made on days 6 and 7 of the ad libitum collection periods at 0, 2, 4, 6, 8, 12, and 16 h after the 1600 feeding of d 6. Volatile fatty acids (VFA), ammonia nitrogen (NH_3-N), and pH were measured. Approximately 50 ml of whole rumen liquor was taken from the ventral sac of the rumen using a soft polyethylene tube and a suction syringe. pH was taken immediately (using Fisher Accumet model, 425 digital pH/ion meter, Pittsburgh, PA). Approximately 25 ml of rumen fluid were strained through 2 layers of cheese cloth

and centrifuged for 20 min at 4°C and 20,000 g with a Sorvall RC-5B centrifuge (Dupont, Wilmington, DE). Eighteen ml of the supernatant were then added to 2 ml of 6N HCL and stored in a screw top vial at -20°C until later analysis for VFA and ammonia concentrations. Ammonia nitrogen was determined spectrophotometrically using Nessler's reagent (Hach et al., 1985). Volatile fatty acids were determined by gas chromatography (HP 5890, Hewlett-Packard Co., San Fernando, CA) using a 1.83 * 2 mm ID glass column packed with GP 10% SP-1200 / 1% H_3PO_4 on 80 / 100 120 C mesh Chromosorb W-AW (Supelco Inc., Bellefonte, PA).

Statistical Analysis

Data were analyzed by SAS (1988), using a general linear model including period, straw treatment, and sheep (appendix B). Subperiod was later included in the model to test the intake level effect. Sheep nested within breed was used as an error term to test the breed effect in a separate analysis. The least significant difference (LSD) multiple mean comparison was applied to the variables studied.

RESULTS AND DISCUSSION

The nutrient composition of the four straws and of the supplement is presented in Table 1. Crude protein was drastically increased by ammoniation, agreeing with Chestnut et al. (1987) and Brown (1988). The alkaline attack of ammonia on the ester bonds, linking lignin with cellulose and

hemicellulose (Chesson et al., 1983), supplied the straw with nitrogen. This alkaline attack also affected lignin and hemicellulose by partially dissolving them. This phenomenon was reported by Van Soest (1982). The positive effects of ammoniation were further increased by rehydration of the straw. This was probably due to the phenomenon of fiber swelling (Feist et al., 1970), making the fibers more available to NH_3. The presence of water increases the homogeneity of the reaction and provides an aqueous solution required by most chemical reactions. A similar positive effect was added by H_2O_2 treatment probably because of the oxidizing ability of this component.

Cellulose content was not affected by the treatments probably because of its location in an internal layer of the plant cell wall. This is consistent with data reported by Horton (1981). Nevertheless, digestible cellulose intake was increased by each additional treatment component (Table 2). This may mean that cellulose became more available to the rumen microorganisms.

Since many animals did not go through the second subperiod, only the ad libitum intake values will be used for treatment comparison.

At the ad libitum intake, digestible DMI was increased by ammoniation ($P = .0006$), by rehydration ($P = .12$), and by H_2O_2 ($P = .09$). Comparing NH_3 with $NH_3+H_2O_2$ reveals a highly

Table 1. Nutrient Composition of Different Treated Straws and of the Faba Bean Supplement

Nutrients	Control	NH_3	NH_3+H_2O	$NH_3+H_2O_2$	Suppl.
DM	93.23	92.66	92.28	92.06	90.3
			% DM basis		
OM	92.37	92.03	92.49	92.34	96.03
CP	3.02	7.22	7.78	8.05	21.14
Cellulose	45.57	45.89	46.25	45.91	11.19
Hemicellulose	27.77	24.94	23.37	23.47	19.03
Lignin	8.37	7.72	7.58	7.16	2.24

significant difference (P = .003). These three treatment components had similar effects on the digestible intake of the other nutrients (Table 2). Although the DM digestibility was slightly increased, the improvement of digestible DMI was primarily obtained by an increase of the DMI. This increase in DMI was reported to be due to an increase in the rate and extent of fiber digestion (Chestnut et al., 1987). The fermentation characteristics (Table 3) were not improved enough (especially among the three treated straws) to allow a large increase in DM digestibility. The same pattern of

Table 2. Effect of Straw Treatments on Nutrient Utilization at the Ad Libitum Intake Level

Item	Control	NH_3	NH_3+H_2O	$NH_3+H_2O_2$	SE[a]
DMI, g/(h.d)	630.1[b]	811.1[c]	868.8[cd]	936.4[d]	32.2
DM digestibility, %	53.7[b]	56.3[c]	57.9[cd]	58.9[d]	.7
Digestible DMI, g/(h.d)	336.9[b]	454.7[c]	500.9[cd]	551.5[d]	21.3
Digestible OM intake, g/(h.d)	326.3[b]	437.1[c]	483.9[cd]	528.7[d]	20.1
Digestible CP intake, g/(h.d)	27.3[b]	51.8[c]	57.5[cd]	63.8[d]	2.4
Digestible cellulose intake, g/(h.d)	133.6[b]	183.4[c]	197.7[cd]	218.9[d]	9.1
Digestible hemicellulose intake, g/(h.d)	80.3[b]	121.5[c]	134.2[c]	151.6[d]	5.7

[a] Standard error of the LS means

[b,c,d] Means in a row lacking a common superscript differ ($P < .05$)

influence of ammoniation was reported by Llamas-Lamas and

Combs (1990). These authors obtained, however, a slightly higher improvement in DM digestibility than we did. This is probably because they used wheat straw, which is more responsive to ammoniation than barley straw (Horton, 1981).

Rumen ammonia nitrogen and total VFA (Table 3) confirmed the effect of ammoniation and that of rehydration. Propionate concentration was not affected by any of the treatment components. This disagrees with Morris and Mowat (1980) and Males and Gaskins (1982), who noted a higher propionate concentration when ammoniated straw or corn stover were fed.

The trend of the response of DM digestibility (Table 4) and of other digestion variables (data not shown) to the treatments was slightly different at limited intake. At this intake, some ewes did not go through all the treatments. This unbalanced design along with the high variability among ewes could have been the reason for this difference.

Overall, limiting the intake enhanced DM digestibility ($P = .005$) and other nutrients digestibility. This improvement was, however, minute. Similar findings were reported by Llamas-Lamas and Combs (1990).

The three breeds showed different DM intakes, which reflected their size differences. This difference disappeared when intake was divided by metabolic body weight. The only two variables that tended to be related to the breed are presented in Table 5.

Table 3. Effect of Straw Treatments on Fermentation Characteristics at the Ad Libitum Intake Level

Item[a]	Control	NH_3	NH_3+H_2O	$NH_3+H_2O_2$	SE[e]
pH	6.82	6.85	6.84	6.81	.04
NH_3-N, mg/dl	13.38[b]	19.77[c]	21.75[d]	19.85[c]	.64
Total VFA, mM	57.73[b]	65.03[c]	74.59[d]	71.46[cd]	2.47
BCVFA, mol/100 mol	1.95[b]	1.56[c]	1.79[bc]	1.88[b]	.1
Ac, mol/100 mol	71.13[b]	73.68[c]	72.92[c]	72.94[c]	.45
Pr, mol/100 mol	17.54	16.68	16.91	16.66	.44
Ac / Pr	4.08	4.43	4.34	4.39	.13

[a] VFA: volatile fatty acids, BCVFA: branch chained VFA, Ac: acetate, Pr: propionate

[b,c,d] Means in a row lacking a common superscript differ ($P < .05$)

[e] Standard error of the LS means

Table 4. Effect of 75% Limited Intake on DM Digestibility of Treated Straws

	Ad-libitum intake		75% limited intake		SE[c]	$P > F$
	n[a]	DMD[b], %	n[a]	DMD[b], %		
Control	8	53.66	6	53.71	.78	.75
NH_3	8	56.3	8	59.53	.76	.04
$NH_3 + H_2O$	8	57.88	5	61.27	.82	.02
$NH_3 + H_2O_2$	8	58.95	6	59.73	.79	.61
Average	32	56.7	25	58.21	.40	.01

[a] Number of observations

[b] Dry matter digestibility

[c] Standard error of the LS means

CONCLUSION

Although hydrogen peroxide tends to increase straw utilization, this increase does not seem large enough to be economically justifiable. Comparing our results with those using alkaline hydrogen peroxide, we can conclude that anhydrous ammonia cannot totally replace sodium hydroxide in providing alkalinity for the optimum action of hydrogen peroxide.

Table 5. Effect of the Breed on Total Water Consumed and Hemicellulose Digestibility at the Ad Libitum Intake (Average of All Treatments)

	Navajo	Columbia	Black Face	SE[a]
Water consumed, ml/kg MBW	122[b]	169[c]	129[bc]	12.55
Hemicellulose digestibility, %	63.8[de]	62.6[d]	68.4[e]	1.69

[a] Standard error of the LS means

[b,c] Means in a row lacking a common superscript differ ($P < .05$)

[d,e] Means in a row lacking a common superscript differ ($P < .1$)

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CHAPTER II

EFFECT OF TREATING WHEAT STRAW AT DIFFERENT PH LEVELS WITH HYDROGEN PEROXIDE AND(OR) ANHYDROUS AMMONIA ON IN SITU DRY MATTER DISAPPEARANCE IN HOLSTEIN COWS

ABSTRACT

Interactions between pH, anhydrous ammonia, and hydrogen peroxide treatments of wheat straw were investigated in this study. Ten solutions were prepared to bring the straw moisture content to 15%. Six solutions were adjusted (with NaOH) to a pH of 9, 11, or 13 and contained 2% hydrogen peroxide (H_2O_2). Four solutions had a pH of 7 or 11 and contained no H_2O_2. After rehydrating the straw with the appropriate solutions, half of the stacks were treated with 3.5% anhydrous ammonia. In situ DM disappearance (DMD) of the different treated straws was measured at 0, 6, 12, 24, and 48 h in nonlactating Holstein cows. Treatment were administered in an incomplete block design using ten cows in three different periods. The evolution of DMD was slow and almost linear. The effect of ammoniation on DMD was consistently apparent at all pH levels though it was depressed ($P < .1$) by raising the pH from 7 to 11. Hydrogen peroxide alone nonsignificantly increased DMD at pH 11. The effect of H_2O_2 on DMD did not change with the pH, but was improved by ammoniation. On the other hand, H_2O_2 improved ($P = .06$) the effect of ammoniation at pH 11. It is concluded that H_2O_2 and NH_3 have complementary mechanisms of

action on straw. Treatment with H_2O_2 and NH_3 at ph 13 had the highest DMD but did not significantly differ from straight ammoniation at pH 7. Compared with the treatments studied here, straight ammoniation remains a cheap and practical treatment.

INTRODUCTION

Diouri and Wiedmeier (1992) reported that the combined NH_3-H_2O_2 treatment tends to increase straw utilization by sheep. Gould (1987) and Kerley et al. (1987) showed that H_2O_2 requires an alkaline environment (pH around 11.5) to be optimally effective. NaOH has been used to provide this alkaline milieu. Because NH_3 is a weak base, it cannot raise the pH enough for H_2O_2 and therefore cannot totally replace NaOH. Measurements done in our laboratory showed that wheat straw pH was raised, by a 3% anhydrous ammonia treatment, from 5.97 (untreated) to 8.66 and 8.85 when the straw moisture content was 8% and 12%, respectively. Nevertheless, the use of ammonia treatment proved to be successful (Zorrilla-Rios et al., 1985; Brown et al., 1987; Wishmeyer, 1990). This beneficial effect of NH_3 is due not only to its ability to make energy more available, but also because it supplements the low quality forages with nitrogen, and because of its low cost and practicability on the farm.

The main objective of this study was to optimize the use, with a simple procedure, of NH_3-H_2O_2 after adjusting the pH with NaOH and to detect the possible interaction or additivity between H_2O_2, NH_3, and NaOH.

MATERIALS AND METHODS

Wheat straw bales were randomly assigned to ten stacks of ten bales each. After determination of the straw moisture

content, ten solutions were prepared to treat the straw and raise its moisture content to 15% (Kernan et al., 1979). These solutions contained either 0 (NP) or 2% (P) of H_2O_2 and were adjusted, using sodium hydroxide, to pH 7, 9, 11, or 13 (Table 6). Each solution was poured evenly on the cut edge of the bales of the corresponding stack. The stacks were then placed in separately sealed 6 mm plastic bags. Anhydrous ammonia (A) was introduced into half of the stacks, through a perforated pipe, at 3.5% of the straw DM. The perforation was immediately sealed. After 3 wk, all the plastic bags were opened for one wk. A representative core sample was taken from each stack. All the samples were ground twice to pass through a 1 cm screen using a Wiley mill (Thomas-Wiley Laboratories, Swedesboro, NJ).

White polyester monofilament bags (Bar Diamond Inc. Parma, ID) with an average pore size of 50 μm, and dimensions of 12 x 5 cm were used. Approximately 2 g of treated straw were placed in each bag before heat sealing. This sample size to bag surface area was calculated to represent the recommendation of Nocek (1988). Bags were then contained in perforated rigid plastic tubes.

Ten Holstein nonlactating cows, equipped with rumen fistulae, were all adapted to the same diet for 3 wk. This diet consisted of 2.3 kg/(h.d) of alfalfa hay pellets and an ad libitum access to an ammoniated wheat straw and a mineral-vitamin block (NRC, 1989).

Table 6. Nature of the Ten Straw Treatments

Treatment[a]	Treatment component		
	pH[b]	H_2O_2[c]	NH_3[d]
9PNA	9	2	0
11PNA	11	2	0
13PNA	13	2	0
9PA	9	2	3.5
11PA	11	2	3.5
13PA	13	2	3.5
7NPNA	7	0	0
11NPNA	11	0	0
7NPA	7	0	3.5
11NPA	11	0	3.5

[a] Treatments: PA: hydrogen peroxide and ammonia, PNA: hydrogen peroxide and no ammonia, NPA: no hydrogen peroxide and ammonia, NPNA: no hydrogen peroxide and no ammonia

[b] pH of the treatment solution

[c] H_2O_2, in % of the treatment solution

[d] NH_3, in % of the straw DM, applied to the straw

Each cow was randomly assigned one tube containing 12 bags and representing three different treatments. One bag from each treatment was collected at 6, 12, 24, and 48 h after placement of the tubes in the rumen. This procedure was repeated three times with the same cows. Once removed, the bags were frozen at -20°C. Three bags were prepared from each treatment, to represent time 0, and frozen without entering the rumen. After the bags from the three periods were all collected, they were rinsed twice for 2 min in a conventional washing machine as suggested by Cherney et al. (1990). The bags were then dried at 60°C for 72 h.

Data were analyzed using the general linear model of SAS (1988). Treatment, with ten different levels, and cow (considered as block), with 30 levels (10 cows in three periods) were the main effects. The contrast option of this procedure was used to compare the different treatment means. The incomplete block design was used as described by Cochran and Cox (1957).

RESULTS AND DISCUSSION

Figure 1 shows a slow degradability rate in all the treatments, caused by the high fiber content of the straw. Marinucci et al. (1992) also showed that when bags are incubated inside a rigid plastic container, DM disappearance was reduced compared with bags suspended freely in the rumen. The present results should be used only to compare the

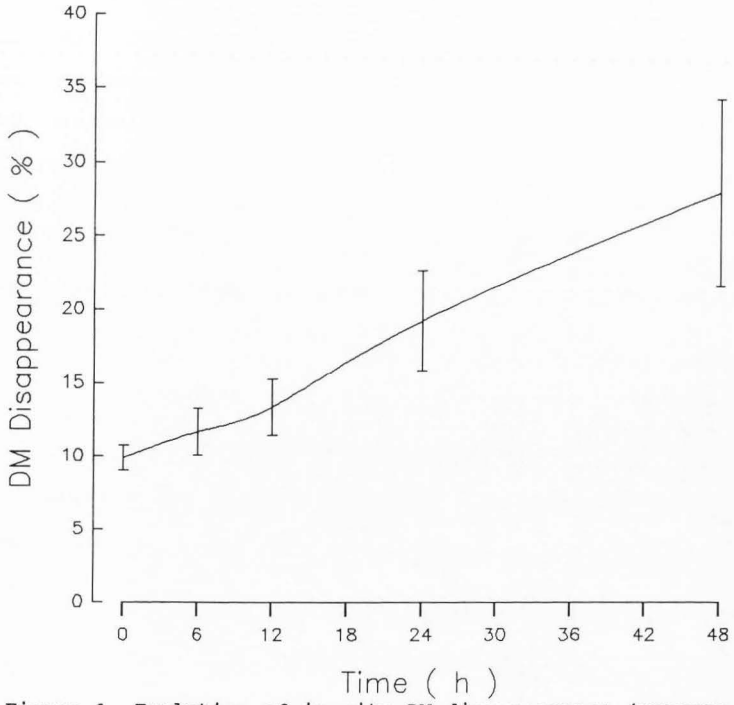

Figure 1. Evolution of in situ DM disappearance (average of all treatments). The error bars represent the standard deviations.

different treatments. The evolution of DM disappearance (Figure 1) was almost linear. Logarithmic transformation of the data improved the correlation coefficient by 1 percentage unit only. The degradability rate was then expected to follow the same pattern as the DM disappearance at 48 h (DMD). For this reason, only the latter variable was used as a criterion of treatment comparison. The variability in DMD (represented in Figure 1 by the standard deviation) was the highest at 48 h, reflecting the segregation of the different treatments. This rate of degradability suggests that it would have been better to measure the DM disappearance at a later time (e.g., 72 h).

The effect of treatments on DMD is summarized in Figure 2. The effect of ammoniation is consistently apparent at every pH whether used alone or in association with H_2O_2, though it was influenced by the other treatment components. Comparing the five ammoniated with the five nonammoniated straws shows a big improvement (P = .005) due to ammoniation. This agrees with the findings of several authors (Herrera-Saldana et al., 1982, 1983; Grings and Males, 1987) who demonstrated the positive effect of anhydrous ammonia treatment.

It is interesting to note the depression (P = .01) in DMD when elevating the ammonia treatment pH from 7 to 11. This cannot be totally explained by a change in the ammonia form--because of the pka--or by a competition between ammonia and

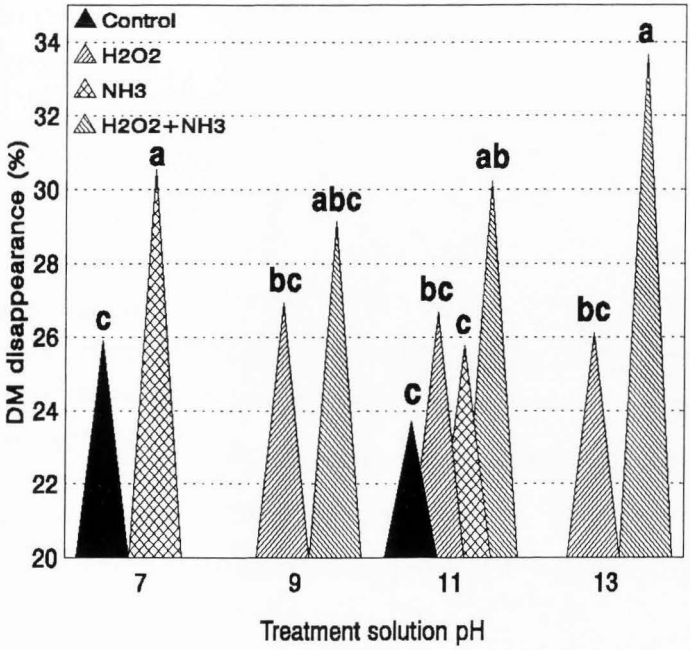

Figure 2. Effect of treatment on in situ DM disappearance at 48 h. Means lacking a common letter differ ($P < .1$)

NaOH, since a similar (but nonsignificant) pattern was observed in the control. Sodium hydroxide was used in several studies as an efficient treatment of low quality forages (Levy et al., 1977; Lesoing et al., 1980; El-Yassin et al., 1991). However, all of these studies used high concentrations of this agent (around 4% of forage DM). Thus it appears that the effect of NaOH is not linearly proportional to its concentration.

Hydrogen peroxide alone had a little (nonsignificant) improvement of the degradability (e.g., 11PNA vs 11NPNA). With H_2O_2 alone, DMD was not improved by the pH. This disagrees with Kerley et al. (1985, 1986, 1987) who demonstrated a large improvement of straw nutrient availability by H_2O_2 treatment at a pH around 11.5. This difference can be explained by two factors. First, in these studies, straw was totally soaked in a large volume of pH-regulated H_2O_2 solution. In our case, the small amount of solution may have been unable to raise the pH of straw (which was not measured) to the optimum level. Second, to raise the pH of this large solution to 11.5, a big amount of NaOH is needed, especially with the presence of H_2O_2. The ratio of NaOH to the straw DM may have been high enough to contribute to that result.

Ammoniation improved (P = .02) the effect of H_2O_2 (comparison of the three PNA vs the three PA). The additivity of ammonia alkalinity does not seem to be an explanation for that, when we compare for example 9PA and 11PA with 11PNA and

13PNA. This effect cannot either be explained by the nitrogen supply of ammonia because nitrogen--in a better form--was provided by the alfalfa hay pellets supplement. It appears, therefore, that the mechanisms of action of NH_3 and that of H_2O_2 are complementary. This conclusion is confirmed by comparing 11PA with 11NPA ($P = .06$).

Compared with straight ammoniation at pH 7 (7NPA), the only treatment which gave a better DMD is 13PA. This difference, beside its nonsignificance, was very costly (cost of NaOH and H_2O_2).

IMPLICATION

Unless an economical analysis justifies the cost of alkaline hydrogen peroxide treatment, straight ammoniation remains an adequate and practical treatment of straw compared to the other treatments studied here. This is especially true when the moisture content of the straw is high enough (15 to 20%).

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CHAPTER III

EFFECT OF ANHYDROUS AMMONIA TREATMENT OF BALTIC RUSH (*JUNCUS BALTICUS WILLD.*) ON INTAKE, DIGESTIBILITY, AND RUMINAL FERMENTATION CHARACTERISTICS IN SHEEP

ABSTRACT

Rushes (*Juncus Sp.*) produce large amounts of biomass in wetlands where other forage plants have difficulty competing; however, nutrient availability is relatively low for ruminants. Ammoniation of this biomass to improve nutrient availability was investigated. Mature rush biomass was harvested and baled. Half of the bales were treated (T) with anhydrous ammonia, the other half served as a control (UT). Forages were then ground through a 5 cm screen and fed to 8 ruminally cannulated wethers housed in elevated metabolism crates in a crossover design. Animals had an ad libitum access to the nonsupplemented forages for a 14-d adaptation period followed by a 5-d collection period. Total collections were made on days 1-5. On day 6 of the second collection period, ruminal digesta were sampled at 0, 2, 4, 6, and 8 h post-feeding to measure fermentation characteristics. Dry matter (DM) and digestible DM intakes were similar at 845.9, 880.7 and 497.2, 469.7 g/(h.d) for UT and T biomass, respectively. DM digestibility decreased (P = .05) from 58.4 to 54.7% by ammoniation. Ruminal pH, volatile fatty acid (VFA) and

ammonia-nitrogen (NH_3-N) concentrations were 6.67, 6.36 (P = .03); 87.2, 94.6 mM (P = .05); 4.5, 8.3 mg/dl (P = .004) for UT vs T rush. Molar proportions of acetate, propionate, and butyrate were 73.8, 74.1; 17.3, 17.4; 7.3, 7.6% for UT vs T rush, respectively, and were not different (P > .05). Ammoniation of rush did not improve nutrient availability as compared to Poaceae species possibly due to differences in lignin-carbohydrate bonding.

INTRODUCTION

Rushes (*Juncus Sp.*) produce large amounts of biomass in flood plane areas where other forage plants have difficulty competing (Smith, 1984; Catling and Spicer, 1987; Manning et al., 1989). The utilization of this biomass in livestock nutrition has been studied by others (Hartley, 1983; Buchsbaum and Valiela, 1987; Kirby et al., 1989). Since this biomass is sometimes baled and fed to ruminants, chemical treatment was investigated to improve its relatively low nutrient availability.

Anhydrous ammonia treatment has been reported to increase low-quality forage utilization by ruminants (Sundstol et al., 1978; Wiedmeier, 1988; Zorrilla-Rios et al., 1991). This positive effect of ammoniation is especially apparent in Poaceae (grasses). Fabaceae (legumes) species have also been ammoniated (Fremy et al., 1988; Frayssinet and Lafarge-Frayssinet, 1990). However, this treatment was used for purposes other than improving the nutrients availability (e.g., against molds and their toxins). The main cause of the unsuccessful ammoniation results in Fabaceae is the different lignin-carbohydrate bonding type. Baltic rush belongs to a family (Juncaceae) that is taxonomically close to Poaceae and was expected to show a similar response to anhydrous ammonia treatment. The main objective of this study was to determine the effect of anhydrous ammonia treatment on rush nutrient utilization by sheep.

MATERIALS AND METHODS

Rush bales were randomly assigned to one of two stacks and covered by a 6 mm plastic. One stack was not treated to serve as a control. NH_3 was then introduced into the second stack through a perforated pipe at 3% of DM. After ammoniation, the perforation was sealed for approximately 21 d, and then the stacks were uncovered. Rush was then ground, mixed in an auger-type grinder mixer, and stored in 30-gallon plastic containers until used.

Four Navajo and four Saint Croix yearling wethers, fitted with ruminal fistulae, were used. After proper recovery from fistulation, sheep were placed in elevated metabolism crates equipped with stainless steel feeders, water, and a feces-urine collection apparatus.

Treatments were administered in a cross-over design, with two periods. Daily rations were given in 2 equal portions at 0800 and 1600. The wethers had ad libitum access to one of the two forages constituting their whole diets. These diets were fed for a 14-d adaptation period followed by a 5-d collection period.

During d 1 to 5 of the collection periods, total fecal output was collected and weighed twice daily at the normal feeding times. Fecal samples were dried at 60°C for 72 h and then ground to pass through a 2 mm screen using a Wiley mill (Thomas-Wiley Laboratories, Swedesboro, NJ). Dry matter content was calculated. Composites of fecal samples, within

animal and period, were then made. The amount of each sample that went into the composite was proportional to that sample's portion of the total output.

Feed was weighed at each feeding. Refusal was weighed back to determine intake. Starting 1 d before the beginning of the collection periods, samples of feed and refusals were taken at each feeding. These samples were ground to pass through a 1 mm screen and proportional composites were made as was described with feces samples.

All feed, ort, and fecal composite samples were analyzed for laboratory dry matter (DM), OM (AOAC, 1990), CP (Hach et al., 1985), ADF, and NDF (Van Soest, 1967).

Rumen measurements were made on d 6 of the second collection period at 0, 2, 4, 6, and 8 h after the 0800 feeding. Volatile fatty acids (VFA), ammonia nitrogen (NH_3-N), and pH were measured. Approximately 50 ml of whole rumen liquor was taken from the ventral sac using a soft polyethylene tube and a suction syringe. pH was taken immediately (using Fisher Accumet model, 425 digital pH/ion meter, Pittsburgh, PA). Approximately 25 ml of rumen fluid were then be strained through 2 layers of cheese cloth and centrifuged for 20 min at 4°C and 20,000 g with a Sorvall RC-5B centrifuge (Dupont, Wilmington, DE). Eighteen ml of the supernatant were then added to 2 ml of 6N HCL and stored in a screw top vial at -20°C until later analysis for VFA and ammonia concentrations. Ammonia nitrogen was determined

spectrophotometrically using Nessler's reagent (Hach et al., 1985). Volatile fatty acids were determined by gas chromatography (HP 5890, Hewlett-Packard Co., San Fernando, CA) using a 1.83 * 2 mm ID glass column packed with GP 10% SP-1200 / 1% H_3PO_4 on 80 / 100 120 C mesh Chromosorb W-AW (Supelco Inc., Bellefonte, PA).

Data were analyzed by SAS (1988) using a general linear model, including period, rush treatment, and sheep as main effects. The breed was not included in the model because it did not show any significant effect. Period was not in the model for the rumen fermentation characteristics analysis, since these parameters were measured only in the second period. Although the cross-over is not designed to test the treatment*period interaction, this effect was separately introduced in the model and was found highly significant ($P < .01$).

RESULTS AND DISCUSSION

Unlike cereal straws (Horton, 1981), ammoniation did not dissolve rush fiber (Table 7). This solubilization is important in making structural carbohydrates more available to rumen microorganisms.

The increase in CP content may have been caused by mere absorption of ammonia, or by chemical reaction between NH_3 and some cell wall components. The first hypothesis is favored by the unchanged fiber content.

Table 7. Effect of Treatment on Rush Nutrient Composition

	Untreated	Treated
DM, %	92.18	91.72
	------- DM basis -------	
OM, %	92.75	92.99
CP, %	10.66	12.89
NDF, %	74.91	74.87
ADF, %	48.74	48.29

Table 8 shows that, except for CP, the digestibility of all the nutrients was depressed ($P < .1$) by ammoniation. Ammoniation of cereal straws has always increased nutrient digestibility (Klopfenstein et al., 1979; Horton, 1981), though this increase is sometimes minor. This decrease in digestibility was compensated by a nonsignificant enhancement of intake leading to a similar digestible intake of the different nutrients. Digestible CP intake was the only exception to this rule, and was increased by ammoniation. This was probably because of the critical level of the control rumen NH_3-N (Table 9). Ruminal pH ($P = .033$) decreased with ammoniation; total VFA and NH_3-N increased ($P = .052$ and $P = .004$, respectively). These changes generally indicate an improvement in nutrient utilization. The rumen measurements

Table 8. Effect of Treatment on Nutrient Utilization

Item	Untreated	Treated	SE[a]	*P* value
DMI, g/(h.d)	845.9	880.7	34.9	.507
DM Dig[b], %	58.4	54.7	1.1	.05
DM DI[c], g/(h.d)	497.2	469.7	17.8	.316
OM dig[b], %	59.7	56.9	.98	.094
OM DI[c], g/(h.d)	472.9	460.2	17.6	.629
NDF dig[b], %	55.49	51.48	1.2	.051
NDF DI[c], g/(h.d)	346.2	322.3	18.1	.385
ADF dig[b], %	60.5	56.9	1.2	.086
ADF DI[c], g/(h.d)	251.7	240.8	10.3	.486
CP dig[b], %	63.3	68.1	1.6	.074
CP DI[c], g/(h.d)	64.4	88.3	1.4	.0001

[a] Standard error of the Ls mean

[b] Dig = digestibility

[c] DI = digestible intake

Table 9. Effect of Treatment on Rumen Fermentation Characteristics[a]

Item	UT	T	SE[b]	*P* value
pH	6.67	6.36	.07	.033
NH_3-N, mg/dl	4.47	8.26	0.44	.004
Total VFA, mM	87.2	94.57	1.90	.052
Acetate[c]	73.79	74.08	1.14	.868
Propionate[c]	17.26	17.38	1.06	.939
Butyrate[c]	7.33	7.56	.59	.594
Isobutyrate[c]	.51	.22	.05	.019
Valerate[c]	.59	.58	.08	.913
Isovalerate[c]	.53	.18	.03	.001

[a] Data from period 2 only.

[b] Standard error of the LS mean.

[c] In mol / 100 mol

were made only in the second collection period, where nutrient utilization followed a similar pattern as the rumen fermentation characteristics (appendix E).

CONCLUSION

Although there was a highly significant treatment * period interaction, the overall present results show that anhydrous ammonia treatment of rush does not improve nutrient utilization. Juncaceae species may have a different lignin-carbohydrate bonding type than Poaceae. Some of the rush components (especially silica) may also have interfered with the treatment.

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CHAPTER IV

WATER BALANCE, USE OF INTERNAL MARKERS, AND PREDICTION OF NUTRIENT UTILIZATION IN SHEEP FED STRAW-BASED DIETS

ABSTRACT

The efficiency of acid insoluble ash (AIA) and ADL insoluble ash (ADLIA) to predict digestibility was investigated, as well as the correlation of nutrient utilization with water balance and with treated straw composition. A control and three treatments of barley straw were used: NH_3 (3% of DM), 3% NH_3 after rehydration of the straw to 15% moisture with water, and 3% NH_3 after rehydration to 15% moisture with a H_2O_2 solution (.32% of DM). Forages were fed, with a faba bean supplement, at two levels of intake (ad libitum and 75% of ad lib.) to 8 ewes housed in elevated metabolism crates. Treatments were administered in a split-plot in a Latin square design. After an adaptation period, feeds, orts, feces, and urine were measured and sampled during 5 d. Water intake and evaporation were also monitored. AIA estimated digestibility better than ADLIA. Both markers were adequate in determining hemicellulose digestibility (R = .96 and .87 for AIA and ADLIA, respectively). Water intake and insensible water loss (in ml/kg MBW) were both positively correlated with digestible cellulose intake (R = .57 and .69). The fraction of water intake lost through feces was negatively correlated (R = -.70) with NDF digestibility, and negatively

correlated with the fraction of water intake lost through urine (R = -.70). Digestible DMI (DDMI) was correlated with both the increase of forage CP content (R = .74) and the decrease of forage NDF (R = .75). Only the latter was used to establish an equation predicting DDMI and, in turn, estimating straw treatment efficiency.

INTRODUCTION

Treated straws are commonly used in animal nutrition (Zorilla-Rios et al., 1985; Kerley et al., 1986; El-Yassin et al., 1991). In a previous study (chapter I), intake and in vivo digestibility of treated barley straw-based diets were measured as well as rumen fermentation characteristics. In such diets, we were interested in three other measurements which make the main objectives of the present study:

1. to assess the efficiency of acid insoluble ash (AIA) and ADL insoluble ash (ADLIA) as internal markers for digestibility determination.

2. to correlate water intake and use with different nutrients utilization.

3. to predict nutrient utilization from the change of forage composition caused by chemical treatment.

MATERIAL AND METHODS

Four different barley straw treatments were used:

1. Control (untreated)

2. 3% NH_3 (anhydrous ammonia at 3% of straw DM)

3. 3% NH_3 after rehydration of straw to 15% moisture with water

4. 3% NH_3 after rehydration of straw to 15% moisture with a H_2O_2 solution (to reach a level of .32% DM of H_2O_2).

After treatment, straw was chopped, mixed in an auger-type grinder mixer, and stored until used.

Eight (4 Navajos, 2 Columbias, and 2 Black-faced) yearling ewes (30 to 51 kg) were used. After adaptation to a straw-based diet, sheep were placed in elevated metabolism crates equipped with stainless steel feeders, water, and a feces-urine collection apparatus.

Treatments were administered in a split plot in a 4*4 Latin square design with repeated measures. Each of the 4 trial periods was composed of two subperiods. In the first subperiod, sheep had ad libitum access to straw (to determine the intake) allowing approximately 10% refusal. A supplement (whose nutrient composition is presented in Table 1) composed of ground faba bean (*Visia faba*) and fortified with vitamins and minerals as needed to meet nutrient requirements (NRC, 1985) was top dressed. The supplement intake was gradually adjusted to represent 25% of the ration in order to eliminate the possible negative associative effect between concentrate and roughage (Blaxter, 1969; Mould et al., 1983). Daily rations were given in 2 equal portions at 0800 and 1600. Diets were fed for a 14-d adaptation period followed by a 5-d collection period.

In the second subperiod, consisting of a 5-d adaptation and a 5-d collection, the sheep received the same diet but their ration was limited to 75% of each animal's ad libitum intake.

During the collection periods, total urine and fecal output was separated and collected. Urine volumes and fecal

weights were recorded twice daily at the normal feeding times. Urine was stabilized in the collection container with mercuric chloride ($HgCl_2$). Fecal samples were dried at 60°C for 72 h and then ground to pass through a 2 mm screen using a Wiley mill (Thomas-Wiley Laboratories, Swedesboro, NJ). Dry matter content was calculated. Composites of fecal samples, within ewe and period, were then made. The amount of each sample that went into the composite was proportional to that sample's portion of the total output. Other fecal composites were made to simulate experiments where internal markers are measured. These composites were not proportional (NP) to the fecal outputs, and contained equal amounts of each fecal sample.

Urine samples were placed in whirl-pak bags, stored at -20°C, composited, then freeze dried (Labconco Lyph-Lock 12, Kansas City, MO).

Feed (straw and supplement) was weighed at each feeding. Refusal was weighed back to determine intake. Starting 1 d before the beginning of the collection periods, samples of feed and refusals were taken at each feeding. These samples were ground to pass through a 1 mm screen and proportional composites were made as was described with feces samples.

All feed, ort, and fecal composite samples were analyzed for laboratory dry matter (DM), OM (AOAC, 1990), CP (Hach et al., 1985), ADF, NDF, ADL (Van Soest, 1967), and AIA (Van Keulen and Young, 1977). ADLIA was the ash remaining after ADL analysis.

Water intake was determined by measuring water given and water remaining at each feeding. To account for evaporation, a container, exactly like those used by the animals, was charged with 2 L of water and left in the open. This water was measured again after a fixed period of time. Representative fresh fecal and urine samples were also left in the open and remeasured to determine water evaporation from feces and urine per unit of time.

Data were analyzed by SAS (1988) using regression analysis to determine the relationship between the different variables. When multiple regression was used, the correlation between the independent variables was tested with the collinearity option.

RESULTS AND DISCUSSION

Internal Markers

Only results of the limited intake are used for marker comparisons. Total-collection digestibility was better estimated by AIA than ADLIA (Table 10). ADLIA is determined from a smaller sample size (1 g vs. 5 g) and requires more analytical steps than AIA. This could lead to more analysis errors.

Both markers gave better estimation of the digestibility of cellulose and hemicellulose than that of DM or organic matter. The same trend was reported by Undersander et al. (1987). In hemicellulose digestibility, not only the

correlation was high, but also the intercept was close to zero and the slope was close to one (AIA estimation). The hemicellulose digestibilities determined by AIA can not be used only for comparison purposes, but also to accurately estimate total collection digestibilities.

Van Keulen and Young (1977) did not detect any diurnal AIA excretion pattern. Comparison of AIAp and AIAnp suggests that AIA excretion is not different from one day to another after an adequate adaptation period. Therefore, as long as the diet has a high marker content (Sherrod et al., 1978; Sunvold and Cochran, 1991) (which was our case) and the grab samples are representative (Thonney et al., 1985), AIA can serve as a satisfactory internal marker. However, care must be taken when using this marker in grazing animals mainly because of the ingestion of soil containing large amounts of insoluble ash (Van Keulen and Young, 1977).

Water Balance

Water utilization parameters were correlated with different nutrient utilization parameters. The pattern of correlation was similar in the two levels of intake. Table 11 presents the highest correlations averaged over the two intake levels.

Water intake (from feed and drinking water) was best correlated with digestible cellulose intake. The correlation coefficient is not very high, probably because of the variability due to animals breed and size. Nevertheless, this

Table 10. Regression Parameters of In Vivo Digestibility vs. Digestibility Determined by Internal Markers[a]

		R	Intercept	Pi[b]	Slope	Ps[b]
DMD[c]	ADLIAp[d]	.68	48.19	.0001	.20	.0002
	AIAp[d]	.79	21.97	.001	.56	.0001
	AIAnp[d]	.79	19.31	.005	.59	.0001
OMD[c]	ADLIAp	.71	48.82	.0001	.21	.0001
	AIAp	.82	21.39	.0009	.59	.0001
	AIAnp	.82	18.64	.005	.62	.0001
CPD[c]	ADLIAp	.48	49.16	.0001	.22	.02
	AIAp	.67	4.81	.713	.84	.0002
CLD[c]	ADLIAp	.84	42.64	.0001	.37	.0001
	AIAp	.93	8.52	.075	.80	.0001
HCD[c]	ADLIAp	.87	34.80	.0001	.50	.0001
	AIAp	.96	-.28	.94	.92	.0001

[a] Data from the limited intake periods

[b] Probability that intercept (Pi) or slope (Ps) equal zero

[c] Digestibility of: dry matter (DMD), organic matter (OMD), crude protein (CPD), cellulose (CLD), and hemicellulose (HCD)

[d] internal marker: ADLIA = ADL insoluble ash, AIA = acid insoluble ash; fecal samples composited proportionally (AIAp) or nonproportionally (AIAnp) to the fecal outputs

Table 11. Water Utilization Parameters and Their Best Correlations with Nutrients Utilization

Dep. var.[a]	Mean1 ± SE[b]	Mean2 ± SE[b]	Best ind[c]	R
WI	135.53 ± 6.01	111.75 ± 7.43	DCI	.57
%WF	38.86 ± 1.48	33.27 ± 2.05	NDFD	-.70
%WU	24.69 ± 1.53	31.87 ± 2.09	%WF	-.70
IWL	49.56 ± 3.12	38.01 ± 2.18	DCI	.69

[a] Dep. var = dependent variable: WI = Water intake (ml/(d.kg mbw)); %WF = percentage of water intake lost in feces; %WU = percentage of water intake lost in urine; IWL = insensible water loss (ml/(d.kg mbw))

[b] Mean ± Standard error of the dep. var. at voluntary (Mean1) or limited intake (Mean2)

[c] Best ind. = the independent variable that has the best correlation with the dependent variable: DCI = digestible cellulose intake; NDFD = NDF digestibility

correlation shows that cellulose metabolism requires relatively high amounts of water. Jacques et al. (1989) reported also that water intake is increased by an increased DM intake.

Water intake goes out of the body through feces, urine, or insensible loss (calculated here by difference). The portion of water lost through feces is negatively correlated

with NDF digestibility and with water lost through urine. The more NDF is digestible, the more water is needed to transport its products to the circulatory system instead of being lost in the feces. This water will, in turn be excreted in the urine. The portion of water intake in insensible loss depends primarily on environmental conditions, and was not highly correlated with nutrients digestibility. The absolute amount of insensible water loss, however, was positively correlated with digestible cellulose intake, probably because of the difference in metabolism level with different intake levels (Silanikove, 1989; Shalit et al., 1991). It is important to note that the different water utilization parameters are not independent, probably because of the interference of water secretion at different parts of the GI tract (Sklan and Hurwitz, 1985; Jacques et al., 1989).

Efficiency of Straw Treatment

The treatment components used in this study increase nutrient utilization by dissolving forage NDF and(or) increasing forage CP. The increase in straw CP ($\hat{\Delta}$CP) and the decrease in straw NDF ($\hat{\Delta}$NDF) should, therefore, approximately indicate straw nutrient availability.

Digestible DMI (DDMI), at ad libitum intake level, was correlated (R = .75) with $\hat{\Delta}$NDF through the following equation:

$$DDMI/MBW = 22.96 + 1.52 * \hat{\Delta}NDF$$

where: DDMI = digestible DM intake (g/(h.d))

MBW = metabolic body weight ($Kg^{.75}$)

ΔNDF = decrease in forage NDF content (%)

ΔCP was also correlated with DDMI (R = .74). The introduction of this parameter in the model increased R^2 from .56 to .62. This improvement was minor mainly because of the high correlation (R = .79) between ΔCP and ΔNDF.

The equation given above is very limited. It depends, among other things, on the type and amount of supplement, animal species and breed, and the type of straw. Moreover, it cannot detect toxicities or deficiencies. Therefore, it does not determine DDMI per se, but rather compares the efficiency of different treatments (mainly ammonia treatments).

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CONCLUSION

Ammoniation of low-quality forages, especially cereal straws, makes energy more available. It improves intake primarily, and also digestibility of different nutrients.

Rehydration of straw, as well as the addition of hydrogen peroxide (H_2O_2), increases the efficiency of ammoniation. These improvements are, however, small. Another small improvement is added by raising the treatment solution pH.

To take advantage of the big potentialities of straw, large amounts of H_2O_2 alkaline solution are needed in which to soak the forage. Beside the risk of toxicity, such treatment should be compared with ammoniation on the basis of practicability on the farm. Although centralization may solve this problem in some regions, the cost of treatment remains the major criterion of comparison.

More research is needed to evaluate the different chemical treatments in different plant species and to improve their efficiency.

It is my opinion that, unless the economical situation dictates otherwise, the chemical treatment should be as simple as possible. The appropriate chemical treatment should, however, be complemented by microbiological treatments, selection of the forage and the animal, and physiological control of digestion and metabolism.

APPENDICES

Appendix A.

Appendices Abbreviations:

DF ..... degrees of freedom

MS ..... mean square

* ..... P<.1

** ..... P<.05

*** ..... P<.01

ADF ..... acid detergent fiber

NDF ..... neutral detergent fiber

Appendix B.

Analysis of Variance Table for Chapter I.

Dry matter intake, g/(h.d)

Source	DF	MS	
Treatment	3	138141.69	***
Period	3	55844.67	***
Breed	2	195950.72	***
Sheep(breed)	5	47418.68	***
Error	18	8301.02	

Dry matter digestibility, %

Source	DF	MS	
Treatment	3	42.26	***
Period	3	20.33	***
Breed	2	.76	
Sheep(breed)	5	8.32	*
Error	18	3.30	

Digestible dry matter intake, g/(h.d)

Source	DF	MS	
Treatment	3	67804.71	***
Period	3	10470.44	**
Breed	2	65439.20	***
Sheep(breed)	5	12527.01	**
Error	18	3278.78	

Digestible organic matter intake, g/(h.d)

Source	DF	MS	
Treatment	3	61019.03	***
Period	3	9092.22	*
Breed	2	61069.89	***
Sheep(breed)	5	11209.04	**
Error	18	2912.79	

Digestible crude protein intake, g/(h.d)

Source	DF	MS	
Treatment	3	2040.84	***
Period	3	136.78	**
Breed	2	956.31	***
Sheep(breed)	5	179.73	***
Error	18	42.08	

Digestible cellulose intake, g/(h.d)

Source	DF	MS	
Treatment	3	10536.25	***
Period	3	1303.03	
Breed	2	12521.73	***
Sheep(breed)	5	1876.58	**
Error	18	665.67	

Digestible hemicellulose intake, g/(h.d)

Source	DF	MS	
Treatment	3	7372.26	***
Period	3	2600.15	***
Breed	2	5208.76	***
Sheep(breed)	5	1747.09	***
Error	18	263.60	

Hemicellulose digestibility, %

Source	DF	MS	
Treatment	3	684.37	***
Period	3	329.36	***
Breed	2	78.75	**
Sheep(breed)	5	31.21	
Error	18	18.81	

Water intake per metabolic body weight, ml/Kg$^{.75}$

Source	DF	MS
Treatment	3	820.39
Period	3	1738.58 **
Breed	2	5993.99 ***
Sheep(breed)	5	1728.79 **
Error	18	416.52

Rumen fluid pH

Source	DF	MS
Treatment	3	.003
Period	3	.185 ***
Sheep	7	.032 **
Error	18	.012

Rumen fluid ammonia nitrogen, mg/dl

Source	DF	MS
Treatment	3	106.70 ***
Period	3	10.56 **
Sheep	7	12.25 **
Error	18	3.30

Total volatile fatty acids in rumen fluid, mM

Source	DF	MS
Treatment	3	445.44 ***
Period	3	413.98 ***
Sheep	7	350.53 ***
Error	18	48.72

Branch chained volatile fatty acids in rumen fluid, mol/100 mol

Source	DF	MS
Treatment	3	.230 *
Period	3	.476 ***
Sheep	7	.279 **
Error	18	.075

Rumen acetate, mol/100 mol

Source	DF	MS
Treatment	3	9.37 ***
Period	3	2.59
Sheep	7	1.22
Error	18	1.66

Rumen propionate, mol/100 mol

Source	DF	MS
Treatment	3	1.34
Period	3	1.15
Sheep	7	1.31
Error	18	1.57

Acetate/propionate ratio in rumen fluid

Source	DF	MS
Treatment	3	.201
Period	3	.080
Sheep	7	.082
Error	18	.135

Appendix C.

Analysis of Variance Table for Chapter II.

In situ dry matter disappearance at 48 h, %

Source	DF	MS
Cow	29	74.18 ***
Treatment	9	43.13 **
Error	51	19.75

Appendix D.

Analysis of Variance Table for Chapter III.

Dry matter intake, g/(h.d)

Source	DF	MS	
Period	1	135111.38	***
Treatment	1	4836.51	
Sheep	7	97503.10	***
Error	6	9753.13	

Dry matter digestibility, %

Source	DF	MS	
Period	1	202.35	***
Treatment	1	53.88	**
Sheep	7	8.88	
Error	6	8.94	

Digestible dry matter intake, g/(h.d)

Source	DF	MS	
Period	1	5639.26	
Treatment	1	3022.80	
Sheep	7	30357.39	***
Error	6	2531.28	

Organic matter digestibility, %

Source	DF	MS	
Period	1	186.32	***
Treatment	1	30.09	*
Sheep	7	8.53	
Error	6	7.61	

Digestible organic matter intake

Source	DF	MS	
Period	1	6174.42	
Treatment	1	645.29	
Sheep	7	28158.22	***
Error	6	2486.37	

Crude protein digestibility, %

Source	DF	MS	
Period	1	34.43	
Treatment	1	95.40	*
Sheep	7	23.32	
Error	6	20.43	

Digestible crude protein intake, g/(h.d)

Source	DF	MS
Period	1	1071.91 ***
Treatment	1	2296.33 ***
Sheep	7	591.70 ***
Error	6	15.91

NDF digestibility, %

Source	DF	MS
Period	1	245.47 ***
Treatment	1	64.36 *
Sheep	7	29.34
Error	6	10.91

Digestible NDF intake, g/(h.d)

Source	DF	MS
Period	1	390.56
Treatment	1	2283.65
Sheep	7	20915.95 **
Error	6	2605.42

ADF digestibility, %

Source	DF	MS	
Period	1	117.61	**
Treatment	1	50.27	*
Sheep	7	45.41	*
Error	6	11.89	

Digestible ADF intake, g/(h.d)

Source	DF	MS	
Period	1	6958.48	**
Treatment	1	470.35	
Sheep	7	3851.90	**
Error	6	852.45	

Appendix E.

Effect of Ammoniation on Rush Nutrient Utilization (Period 2)

Item	Untreated	treated	SE[a]	P value
DMI, g/(h.d)	776.2	1134.2	72.6	.013
DM dig[b]	55.6	50.5	1.2	.025
DM DI[c]	431.4	573	40	.046
OM dig[b]	56.9	52.9	1.2	.056
OM DI[c]	411.2	561.2	39.5	.036
NDF dig[b]	51	48.1	2.3	.404
NDF DI[c]	285.4	393	37.5	.089
ADF dig[b]	61.5	50.5	1.4	.002
ADF DI[c]	256.9	277.3	17.6	.446
CP dig[b]	62.3	66.2	1.9	.181
CP DI[c]	57.8	111.3	4.5	.0001

[a] Standard error of the LS means

[b] Dig = digestibility (%)

[c] DI = digestible intake (g/(h.d))

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AWARDS

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