SITE AND EXTENT OF NUTRIENT DIGESTION BY SHEEP FED ALKALINE HYDROGEN PEROXIDE-TREATED WHEAT STRAW-ALFALFA HAY COMBINATIONS AT RESTRICTED INTAKES[1]

D. G. Atwell[2], N. R. Merchen, E. H. Jaster, G. C. Fahey, Jr. and L. L. Berger

University of Illinois[3], Urbana 61801

ABSTRACT

Five crossbred wethers (58 kg) fitted with cannulas in the rumen, duodenum, and ileum were used in a 5 × 5 Latin square design to study effects of feeding combinations of alkaline hydrogen peroxide-treated wheat straw (AHP-WS) and alfalfa hay at restricted intakes on site and extent of nutrient digestion. Additionally, flows and disappearance of N and amino acids (AA) in the small intestine were regressed on alfalfa nitrogen intake (ANI) to estimate alfalfa's contribution to postruminal N and AA supplies. Diets consisted of 80:20 forage:concentrate mixtures; diet designations were 80:0, 80% AHP-WS and no alfalfa; 60:20, 60% AHP-WS and 20% alfalfa; 40:40, 40% AHP-WS and 40% alfalfa; 20:60, 20% AHP-WS and 60% alfalfa; and 0:80, no AHP-WS and 80% alfalfa. A modest positive quadratic ($P < .05$) response was noted for total tract digestibility (TTD) of OM; values were 76.5% for diets 80:0 and 0:80 vs 78% for diet 40:40. Ruminal digestibility (percentage of intake) of NDF and ADF increased in a quadratic manner from 43 and 30%, respectively, for diet 0:80 to 71 and 70%, respectively, for diet 80:0. Ruminal digestibility of fiber may have been enhanced due to linear ($P < .05$) decreases in liquid and particulate dilution rates, resulting in increased ruminal residence time of fiber as alfalfa hay replaced AHP-WS. Liquid and particulate dilution rates decreased linearly from 6.4 and 5.2%/h, respectively, for diet 80:0 to 5.4 and 3.4%/h, respectively, for diet 0:80. Regression analysis of N data indicated that alfalfa N had a ruminal escape value of 26%.

Key Words: Sheep, Digestibility, Alfalfa, Wheat Straw, Amino Acids

J. Anim. Sci. 1991. 69:1697–1706

Introduction

Treatment of wheat straw (WS) with alkaline solutions of hydrogen peroxide has dramatically improved both intake and digestibility of this crop residue (Kerley et al., 1986). However, due to the low concentration of N and several essential minerals in cereal straws (Preston and Leng, 1984), these nutrients need to be added to diets that contain large amounts of WS. Adequate supplementation is particularly important when feeding chemically treated WS in order to take advantage of the increased energy availability due to treatment (Ørskov and Grubb, 1978). One method of supplementing diets containing alkaline hydrogen peroxide-treated WS (AHP-WS) is through the incorporation of a forage, such as alfalfa. Alfalfa N is estimated to be 70% degraded in the rumen (NRC, 1985a); therefore, it would be an excellent source of ruminally degradable protein. Alfalfa also is a

[1]Research supported in part by a gift from a consortium of hydrogen peroxide manufacturers (Degussa Corp., Allendale, NJ; E. I. Dupont de Nemours and Co., Inc., Wilmington, DE; FMC Corp., Princeton, NJ). Appreciation also is extended to the Commodity Credit Corporation, USDA, for partial support of this project.

[2]Current address: Growmark, Inc., P. O. Box 2500, Bloomington, IL 61702.

[3]Dept. of Anim. Sci.

Received May 11, 1990.

Accepted October 24, 1990.

TABLE 1. INGREDIENT AND CHEMICAL COMPOSITION OF EXPERIMENTAL DIETS

Item[a]	AHP-WS[b]:alfalfa hay, % DM				
	80:0	60:20	40:40	20:60	0:80
Ingredient					
AHP-WS	80	60	40	20	—
Alfalfa	—	20	40	60	80
Ground corn	8.54	8.54	8.54	8.54	8.54
Soybean meal	8.13	8.13	8.13	8.13	8.13
Corn starch	—	1.98	2.71	2.98	2.98
Urea	1.5	.13	—	—	—
Dicalcium phosphate	.62	.36	.1	—	—
Calcium sulfate	.86	.51	.17	—	—
Trace mineral mix[c]	.25	.25	.25	.25	.25
Vitamin mix[d]	.1	.1	.1	.1	.1
Chemical composition					
DM	63.5	68.3	73.6	79.8	87
OM	84.8	86.2	87.4	88.4	89.1
N	1.8	1.8	2.5	3.1	3.7
NDF	53.9	50.2	46.8	43.2	39.8
ADF	40.5	36.3	32.4	28.3	24.4

[a]Ingredient and chemical composition are expressed as a percentage of diet DM.

[b]AHP-WS = alkaline hydrogen peroxide-treated wheat straw.

[c]Trace mineral mix composition (g/100 g); NaCl, 95 to 99; Mn, > .2; Fe, > .3; Cu, >.033; Zn, >.01; I, >.007; Co, >.003.

[d]Provided (International Units/kilogram of diet DM) A, 3,300; D, 330; E, 44.

good source of Ca and P and generally is highly acceptable by ruminants. Additionally, several studies have indicated that feeding chemically treated crop residues in combination with alfalfa resulted in positive associative effects on both nutrient intake and digestibility (Maeng et al., 1971; Paterson et al., 1982; Brandt and Klopfenstein, 1986).

The objectives of this experiment were to evaluate the effects of substituting increasing levels of alfalfa hay for AHP-WS in high-forage diets fed to sheep on site and extent of nutrient digestion, ruminal characteristics, and postruminal supply and disappearance of N and amino acids (AA).

Materials and Methods

Animals and Diets. Five mature crossbred wethers (average weight 58 kg) fitted with permanent ruminal and t-type duodenal and ileal cannulas were used as experimental animals in a 5 × 5 Latin square design. Surgery was performed in a sterile environment under general anesthesia as described by Hsu et al. (1991) and followed a protocol approved by the University of Illinois Laboratory Animal Care Advisory Committee. Diets (Table 1) consisted of an 80:20 forage to concentrate ratio (DM basis). Dietary designations, based on DM ratios of AHP-WS:alfalfa hay in the diet, were the following: **80:0**, 80% AHP-WS and no alfalfa hay; **60:20**, 60% AHP-WS and 20% alfalfa hay; **40:40**, 40% AHP-WS and 40% alfalfa hay; **20:60**, 20% AHP-WS and 60% alfalfa hay; and **0:80**, no AHP-WS and 80% alfalfa hay. Treated WS was prepared as described by Cecava et al. (1990). The final product contained 59% DM; OM, CP, NDF, and ADF content averaged 84, 2, 66, and 50% of DM, respectively. Alfalfa hay was cut in the prebloom stage, sun-cured, harvested as bales and ground through a 1.9-cm screen. It contained 87% DM; OM, CP, NDF, and ADF content of the alfalfa hay averaged 88, 23, 48, and 30% of DM, respectively. Constituents of concentrate portions were formulated such that source and percentage of true protein in all concentrates were constant for all diets (Table 1). Differences in protein content among the diets predominantly reflect different levels of alfalfa hay. Diets were formulated to meet or exceed NRC (1985b) requirements for CP, Ca, P, S, K, trace minerals, and vitamins A, D, and E for replacement ram lambs (average weight 60 kg). Ad libitum intake for diet 80:0 was determined during a 10-d prefeeding period. Throughout the experiment, DMI for each animal was held at 90% of its ad libitum intake of diet 80:0 determined during the prefeeding

period. Concentrate and forage portions of the diet were weighed and hand-mixed daily for each animal and fed in equal portions at 0800 and 2000. At each feeding, wethers were given a bolus containing 1.5 g of chromic oxide (Cr_2O_3) via the ruminal cannula to provide a nonabsorbable marker for measurement of digesta flow. Water was available continuously and wethers were tethered in elevated mesh-bottom pens (2.6 m × 2.6 m) in a temperature-controlled room (21°C) under continuous lighting.

Sampling Procedures. Experimental periods were 16 d in length with a 10-d adaptation phase followed by a 6-d collection phase. Samples of the forages and concentrates were taken daily on d 9 to 16 of each period and analyzed to determine nutrient intake and digestibility. Animals were fitted with canvas collection bags to allow total collection of feces. A 10% aliquot of daily fecal excretion was taken on d 11 through 16 and composited for each animal in each period. Feces were dried at 55°C and ground (2-mm screen) before analyses.

Approximately 75 ml of duodenal and ileal digesta were collected at 1, 3, and 5 h after the morning feeding on d 11, 13, and 15 and 7, 9, and 11 h after the morning feeding on d 12, 14, and 16 of each period. Duodenal and ileal samples were composited by animal and period, lyophilized, and ground (2-mm screen) before analyses.

On d 15 of each period, the morning feeding was sprayed with an aqueous solution containing 200 mg of ytterbium (Yb as $YbCl_3 \cdot 6H_2O$) and wethers were given an intraruminal dose of 100 mg of cobalt as CoEDTA (Uden et al., 1980). Sampling scheme and analyses for determination of particulate and fluid dilution rates are identical to those used by Merchen et al. (1986), with the exception that a 20-h sample was not taken and Yb was extracted from ruminal particulate samples as described by Hart and Polan (1984). Whole ruminal contents (150 to 200 g) were collected once daily on d 11 through 16 of each period such that a sample was taken at each 2-h interval between the morning and evening feeding. Whole ruminal contents were squeezed through six layers of cheesecloth to obtain 50-ml samples of ruminal fluid for measurement of pH with a Beckman[4] Model Φ31 pH meter. Subsequent to pH measurement, samples were acidified with 2 ml of 6 *N* HCl and frozen for analysis of VFA and ammonia N (NH_3 N). The remaining sample was diluted with approximately 50 to 100 ml of .9% saline and homogenized for 1.5 min in a Waring blender to detach particle-associated bacteria. Homogenized samples were squeezed through six layers of cheesecloth; the filtrate, composited by wether and period, was frozen. Composited filtrates were thawed and a bacteria-rich fraction was isolated according to the method of Merchen and Satter (1983). This fraction was lyophilized before analyses.

Sample Analyses. Feed, feces, duodenal and ileal digesta samples, and ruminal bacteria were analyzed for DM, OM and Kjeldahl N (AOAC, 1984), NDF (Robertson and Van Soest, 1977), and ADF (Goering and Van Soest, 1970). Samples of feeds, duodenal and ileal digesta, and bacteria were analyzed for AA composition as described by Merchen et al. (1986). Duodenal digesta and lyophilized ruminal bacteria were analyzed for total purines by the method of Zinn and Owens (1986). Ruminal fluid was analyzed for NH_3 N (Chaney and Marbach, 1962) and VFA (Merchen et al., 1986).

Calculations. The proportion of total duodenal N provided by bacterial N was calculated by dividing the N:purine ratio of ruminal bacteria by the N:purine ratio of duodenal digesta. Because differences due to treatment were not detected for bacterial N:purine ratio and AA composition, values were averaged across all observations and used in all calculations. Bacterial N and AA flow to the ileum were calculated based on the indigestibilities of bacterial constituents reported by Storm et al. (1983). Chromium content of duodenal and ileal digesta was determined by the method of Williams et al. (1962). Dry matter flow (grams/day) at the duodenum and ileum was calculated by dividing Cr intake (milligrams) by the Cr concentration (milligrams/gram) in duodenal and ileal digesta. Nutrient flows were calculated by multiplying DM flow by the concentration of the nutrient in duodenal, ileal, or fecal DM.

Statistical Analyses. Data were analyzed using the GLM procedure of SAS (1985). The

[4]Beckman Instruments, Palo Alto, CA.

model included animal, period, and diet effects. Sums of squares for diet were further partitioned into orthogonal contrasts for linear, quadratic, and cubic effects of dietary forage source. Plots of data collected at different times postfeeding (ruminal pH, VFA, and NH_3 N) indicated that differences among diets were relatively constant at each sampling time; therefore, means across times were calculated and analyzed as above. Data presented on intestinal supply and disappearance of N and individual AA were analyzed using a model that included animal and period effects and alfalfa N intake (ANI) as a regression variable. The intercept of the regression equation was calculated by assuming that animal and period effects summed to zero. Linearity of response to ANI is an inherent assumption of this model. The intercept is assumed to be the expected value when sheep were fed diet 80:0 and the slope represents the change in the dependent variable for each gram of ANI.

Results and Discussion

A linear ($P < .05$) increase in OM intake was observed as level of alfalfa increased (Table 2). This was due to a modest linear increase in DM intake with increasing proportion of alfalfa and the higher OM content of alfalfa relative to AHP-WS. Organic matter digestion (OMD) in the stomach (prior to the duodenum) (grams/day), whether apparent or true, was similar among diets, averaging 532 and 730 g/d, respectively. However, when OMD was expressed as a percentage of OMI, both apparent and true digestibility increased linearly ($P < .05$) with increasing level of AHP-WS in the diet. Higher extent of OMD in the stomach of wethers fed diets high in AHP-WS can be explained by the fermentable nature of the fiber fraction of the treated WS. This conclusion is supported by the high-fiber digestibility data in Table 2, as well as the work of Gould (1984) and Kerley et al. (1986).

TABLE 2. DRY MATTER (DM), ORGANIC MATTER (OM), NEUTRAL DETERGENT FIBER, ACID DETERGENT FIBER, AND CELL CONTENT (CC)[a] DIGESTION BY SHEEP FED COMBINATIONS OF AHP-WS[b] AND ALFALFA HAY

	AHP-WS:alfalfa hay, % DM					
Item	80:0	60:20	40:40	20:60	0:80	SE
Intake						
DM, g/d[c]	1,177	1,189	1,194	1,204	1,212	1.8
OM, g/d[c]	998	1,025	1,044	1,064	1,079	3.7
NDF, g/d[c]	635	597	559	520	482	6.7
ADF, g/d[c]	477	431	387	341	293	8.0
CC, g/d[c]	364	428	485	544	597	10.3
OM digestibility in stomach, g/d						
Apparent	528	531	536	552	514	14.8
True	736	746	738	727	706	14.2
OM digestibility in stomach, % of OM intake						
Apparent[c]	53.1	52.0	51.4	52.2	48.1	1.37
True[c]	74.1	72.9	70.9	68.6	65.8	1.32
NDF apparently digested in stomach						
% of NDF intake[d]	71.3	67.4	60.4	56.0	42.6	1.82
% of Total tract digestibility[c]	87.9	85.8	80.1	79.5	67.7	2.24
ADF apparently digested in stomach						
% of ADF intake[d]	70.5	67.2	57.9	51.4	29.7	2.32
% of Total tract digestibility[d]	87.7	87.4	78.9	77.9	55.1	3.39
Apparent total tract digestibility						
DM, %[d]	74.2	75.6	76.2	75.8	75.1	.52
OM, %[d]	76.7	77.8	77.9	77.3	76.4	.48
NDF, %[d]	81.1	78.6	75.5	70.5	62.7	.80
ADF, %[d]	80.3	77.0	73.6	66.1	53.6	1.19
CC, %[d]	69.0	76.6	80.7	83.8	87.4	.42

[a]Cell content = OM − NDF.

[b]AHP-WS = alkaline hydrogen peroxide-treated wheat straw.

[c]Linear effect of diet ($P < .05$).

[d]Quadratic effect of diet ($P < .05$).

Apparent total tract digestibility (TTD) of OM exhibited a quadratic ($P < .05$) effect due to diet, with diets containing combinations of the two forages (60:20, 40:40, and 20:60) having slightly higher OMD. Total tract digestibility of OM ranged from approximately 76.5% for diets 80:0 and 0:80 to 78% for diet 40:40. Although these results may indicate some positive associative action between the two forage sources, the increase of only 1 percentage unit is quite modest.

Intakes of NDF and ADF (Table 2) increased linearly with increasing AHP-WS in the diet. This is not surprising given the higher concentration of these fractions in AHP-WS (Table 1). Apparent digestibility of NDF in the stomach, expressed as a percentage of NDF intake, increased quadratically from 42.6 to 71.3% with an increasing level of AHP-WS. When expressed as a percentage of TTD, a linear ($P < .05$) increase from 67.7 to 87.9% with an increasing level of AHP-WS was observed. Apparent digestibility of ADF in the stomach, whether expressed as a percentage of ADF intake or TTD, increased in a quadratic ($P < .05$) fashion with increasing levels of AHP-WS. Apparent digestibility of ADF in the stomach increased by 137% from diet 0:80 to 80:0 when expressed as a percentage of ADF intake. When expressed as a percentage of TTD of ADF, there was a 59% increase between these two diets. Ruminal digestibilities of NDF and ADF in diets containing combinations of the two forages were slightly higher than would be expected given the digestibilities of NDF and ADF observed for diets 80:0 and 0:80. Based on NDF digestibilities for AHP-WS (diet 80:0) and alfalfa hay (diet 0:80) and amounts from each source in each diet, digestibilities (percentage of NDF intake) for diets 60:20, 40:40, and 20:60 should have been 65.8, 59.4, and 52%, respectively. Based on ADF digestibilities observed for diets 80:0 and 0:80, digestibilities (percentage of ADF intake) for diets 60:20, 40:40, and 20:60 should have been 63.7, 55, and 44%, respectively. Even though differences between observed and predicted values were small, averaging 2.2% for NDF and 4.6% for ADF, this may indicate a positive associative effect on ruminal fiber digestibility for combinations of the two forages. This slight improvement may explain the higher TTD of OM observed for diets containing combinations of the two forages. Total tract digestibility of NDF and ADF increased in a quadratic ($P < .05$) manner from 62.7 and 53.6%, respectively, for diet 0:80 to 81.1 and 80.3%, respectively, for diet 80:0.

When WS is treated with AHP, approximately 20 percentage units of the material analyzed as NDF is solubilized. Generally, WS has a NDF value of approximately 85%, compared with 65% for AHP-WS. Therefore, it is important to determine the digestibility of this material when evaluating the effects of the AHP treatment process. In the present study, TTD of OM-NDF or cell content (CC) decreased linearly ($P < .05$) with increasing substitution of AHP-WS for alfalfa, from 87.4 to 69%, respectively, for diets 0:80 and 80:0. This reduced digestibility of a fraction normally highly digestible tempers the improvements in fiber digestion brought about by the treatment process. Reduction in CC digestion in part explains the similar TTD of OM among diets having drastically different fiber digestibilities.

Nitrogen intake (Table 3) increased quadratically ($P < .05$) with increasing levels of alfalfa. This result is due to a greater N content in the diets with more alfalfa (Table 1). Total and nonbacterial (NBT) N flows (grams/day) at the duodenum increased with increasing levels of alfalfa due to undegraded alfalfa N reaching the duodenum. Bacterial N flow was not affected by diet, averaging 14.7 g/d. Similarly, efficiency of bacterial N synthesis was unaffected by diet, averaging 2.93 and 2.02 g of N per 100 g of OM apparently and truly digested in the stomach, respectively. Availability of energy and N for bacterial growth was similar among diets. Organic matter digested in the rumen was similar among diets (Table 2), and ruminal fluid NH_3 N levels were above the 5 mg/dl level reported by Satter and Slyter (1974) as necessary to maximize bacterial flows from a chemostat. Apparent N digestibility in the small intestine was similar across diets. However, a greater ($P < .05$) proportion of total tract N digestibility occurred in the small intestine of animals fed diets high in AHP-WS. This is indicative of the extensive ruminal degradation of alfalfa CP and lower total tract digestion of diets high in AHP-WS. Currently, NRC (1985a) estimates that approximately 72% of alfalfa CP is degraded in the rumen. Diets containing higher amounts of alfalfa (20:60 and 0:80) had apparent N disappearances in the stomach of

TABLE 3. NITROGEN DIGESTION BY SHEEP FED COMBINATIONS OF AHP-WS[a] AND ALFALFA HAY

Item	AHP-WS:alfalfa hay, % DM					SE
	80:0	60:20	40:40	20:60	0:80	
N intake, g/d[b]	21.3	21.9	29.4	37.2	45.2	1.16
Alfalfa N intake, g/d[c]	0	8.8	17.6	26.5	35.5	1.65
N at duodenum						
Total flow, g/d[c]	25.5	27.3	27.8	29.8	32.2	1.12
Bacterial, g/d	15.5	16	15	13	14.2	.9
Nonbacterial, g/d[c]	10	11.3	12.8	16.8	18	1.07
% of N intake[c]	46.2	52.4	43.7	43.9	39.5	2.71
Ileal N flow, g/d[d]	9.7	10.9	11.6	10.9	13.8	.41
Fecal N excretion, g/d	6.9	6.5	6.4	6.7	6.6	.18
Apparent N digestibility						
Small intestine						
% of duodenal N	62	60	58.1	62.7	56.6	2.07
% of total tract digestibility[c]	110.8	107.3	70.8	60.7	47.8	5.91
Total tract, %[d]	67.9	70.3	78.5	81.9	85.5	.58
Bacterial N synthesis						
g N/100 g OMD_A[e]	3.11	3.15	2.93	2.47	2.97	.23
g N/100 g OMD_T[f]	2.11	2.14	2.04	1.78	2.02	.12

[a]AHP-WS = alkaline hydrogen peroxide-treated wheat straw.

[b]Quadratic effect of diet ($P < .05$).

[c]Linear effect of diet ($P < .05$).

[d]Cubic effect of diet ($P < .05$).

[e]OM apparently digested in the stomach.

[f]OM truly digested in the stomach.

20 and 29%, respectively, presumably due to losses of NH_3 N from the rumen. The cubic increase ($P < .05$) in apparent TTD of N with increasing levels of alfalfa is largely an artifact of increased N intake diluting the effect of endogenous N on digestibility calculations.

Intercepts generated from the regression of small intestinal supply and disappearance of N on ANI (Table 4) are estimates of values for flow and disappearance of N from the small intestine when animals received no N from alfalfa (diet 80:0). Total, bacterial, and NBT N flows at the duodenum for animals fed diet 80:0 were estimated to be 25.1, 15.8, and 9.2 g/d, respectively. Slopes from Table 4 indicate that for every gram of ANI, there is a corresponding change of .19 ± .031, –.06 ± .031, and .26 ± .027 g of total, bacterial, and NBT-N, respectively, reaching the duodenum. The slightly negative slope for bacterial N flow indicates little response in bacterial N synthesis to additional N supplied by alfalfa. Additionally, a slope of .26 for NBT N flow to the duodenum indicates that N from alfalfa had a ruminal escape value of 26%, compared with the estimate of 28% given by NRC (1985a). Total ruminal output of N was equal to 19% of ANI due to the negative effect of bacterial N. Slopes for small intestinal disappearance of total and NBT N (.1 ± .034; .15 ± .032) indicate the quantity of those forms of N provided per gram of ANI.

Intercepts in Tables 5 and 6 provide estimates of daily flows of total and NBT AA, respectively, supplied to and disappearing from the small intestine of sheep fed diet 80:0. Slopes provide an estimate of additional flows

TABLE 4. SMALL INTESTINAL SUPPLY AND DISAPPEARANCE OF NITROGEN IN SHEEP FED INCREASING LEVELS OF ALFALFA HAY

Nitrogen	Intercept, g/d	Slope, g/g ANI[b]	SE[a]
Flow to duodenum			
Total	25.1	.19	.031
Bacterial	15.8	–.06	.031
NBT[c]	9.2	.26	.027
SI[d] disappearance			
Total	15.4	.1	.034
NBT	2.4	.15	.032

[a]Standard error of estimate.

[b]Alfalfa N intake.

[c]Nonbacterial.

[d]Small intestine.

TABLE 5. AMINO ACIDS (AA) SUPPLIED TO AND DISAPPEARING FROM THE SMALL INTESTINE OF SHEEP FED INCREASING LEVELS OF ALFALFA HAY

Amino acid	AA supplied			AA disappearing		
	Intercept, g/d	Slope, g/g ANI[b]	SE[a]	Intercept, g/d	Slope, g/g ANI	SE
Thr	6.4	.038	.009	4.0	.024	.008
Val	6.8	.060	.009	4.4	.046	.009
Met	1.3	.016	.004	.6	.014	.004
Ile	5.9	.052	.007	4.2	.039	.008
Leu	10.1	.096	.015	6.1	.090	.019
Phe	5.5	.073	.008	4.1	.058	.008
His	2.3	.015	.004	1.7	.006	.004
Lys	9.6	.029	.011	7.3	.014	.012
Arg	4.8	.054	.007	3.8	.041	.007
EAA[c]	52.7	.431	.068	36.2	.350	.073
NEAA[d]	56.7	.360	.072	36.4	.254	.074
Total AA	109.4	.791	.139	72.4	.634	.145

[a]Standard error of estimate.

[b]Alfalfa nitrogen intake.

[c]Essential AA = threonine, valine, methionine, isoleucine, leucine, phenylalanine, histidine, lysine, and arginine.

[d]Nonessential AA = alanine, aspartic acid, glutamic acid, glycine, proline, serine, and tyrosine.

of AA supplied to and disappearing from the small intestine with each additional gram of ANI. Slopes presented in Table 6 can be used to calculate amounts of each AA entering and disappearing from the small intestine from alfalfa. Slopes are presented as grams of AA/grams of ANI, so the amount of any AA entering and disappearing from the small intestine for each kilogram of alfalfa CP is equal to the slope multiplied by 1,000 and divided by 6.25. For example, alfalfa would supply (.062 g threonine/g ANI (Table 6) × 1,000 g/kg × 1 g N/6.25 g CP) or 9.9 g threonine/kg alfalfa CP to the proximal duodenum. Similarly, the amount of threonine disappearing in the small intestine would be 7.0 g/kg alfalfa CP. Slopes for total (.038 ± .009) and NBT threonine (.062 ± .008) supplied to the small intestine differ due to the decreasing supply of bacterial threonine. This

TABLE 6. NONBACTERIAL AMINO ACIDS (NBT AA) SUPPLIED TO AND DISAPPEARING FROM THE SMALL INTESTINE OF SHEEP FED INCREASING LEVELS OF ALFALFA HAY

Amino acid	NBT-AA supplied			NBT-AA disappearing		
	Intercept, g/d	Slope, g/g ANI[b]	SE[a]	Intercept, g/d	Slope, g/g ANI	SE
Thr	.54	.062	.008	−.97	.044	.008
Val	−.21	.098	.01	−1.38	.069	.009
Met	−1.14	.026	.006	−1.57	.023	.006
Ile	−.46	.078	.01	−1.25	.059	.01
Leu	.33	.136	.018	−2.35	.124	.02
Phe	.08	.096	.009	−1.16	.079	.009
His	.12	.023	.003	.25	.012	.004
Lys	.07	.067	.015	−.46	.047	.014
Arg	−.5	.075	.009	−.88	.06	.009
EAA[c]	−1.66	.65	.085	−10.98	.52	.086
NEAA[d]	2.26	.58	.08	−10	.44	.079
Total AA	.6	1.23	.165	−20.59	.98	.169

[a]Standard error of estimate.

[b]Alfalfa N intake.

[c]Essential AA = threonine, valine, methionine, isoleucine, leucine, phenylalanine, histidine, lysine, and arginine.

[d]Nonessential AA = alanine, aspartic acid, glutamic acid, glycine, proline, serine, and tyrosine.

TABLE 7. RUMINAL CHARACTERISTICS OF SHEEP FED COMBINATIONS OF AHP-WS[a] AND ALFALFA HAY

Item	AHP-WS:alfalfa hay, % DM					SE
	80:0[b]	60:20	40:40	20:60	0:80	
VFA						
Total, m*M*	84.7	95.2	89.3	91.5	88.6	3.16
Mol/100 mol						
Acetate[c]	71.7	72.1	71.1	70.1	68.1	.43
Propionate[c]	17.4	15.4	14.6	14.4	15.6	.19
Isobutyrate[d]	.76	1.02	1.26	1.47	1.91	.102
Butyrate[c]	8.8	9.9	11.1	11.2	10.8	.39
Isovalerate[d]	.81	.91	1.18	1.72	2.18	.13
Valerate[c]	.56	.65	.78	1.01	1.39	.069
Ruminal pH[d]	6.67	6.56	6.50	6.46	6.45	.032
Ammonia N, mg/dl[e]	13.0	11.3	17.5	25.2	32.8	.73
Liquid dilution rate (Co), %/h[d]	6.4	6.0	5.7	5.4	5.4	.31
Particulate dilution rate (Yb), %/h[d]	5.2	4.9	4.3	4.0	3.4	.17
Ruminal volume, liters[d]	10.8	10.3	9.6	9.5	9.4	.34

[a]AHP-WS = alkaline hydrogen peroxide-treated wheat straw.

[b]Diet 80:0 had one missing cell for liquid and particulate dilution rates and ruminal volume; consequently, SEM for that diet were as follows: liquid dilution rate (percentage/hour), .37; particulate dilution rate (percentage/hour), .20; ruminal volume (liters), .41.

[c]Quadratic effect of diet ($P < .05$).

[d]Linear effect of diet ($P < .05$).

[e]Cubic effect of diet ($P < .05$).

mirrors what was seen with the nitrogenous fractions (Table 4). Negative intercepts found in Table 6 are indicative of the relatively minor contribution of NBT AA to duodenal flows for diet 80:0. This is not surprising because the two primary sources of N in this diet were urea and soybean meal (Table 1), both of which are extensively degraded in the rumen (NRC, 1985a).

Total concentrations of ruminal VFA (Table 7) were not affected by diet, averaging 90 m*M*. Molar proportions of acetate and propionate increased from 68.1 and 15.6, respectively, for diet 0:80 to 71.7 and 17.4, respectively, for diet 80:0 (quadratic effect, $P < .05$). Molar proportions of the branched-chain VFA (isobutyrate and isovalerate) increased linearly ($P < .05$) with increasing levels of alfalfa. Isobutyrate and isovalerate molar proportions increased from .76 and .81, respectively, for diet 80:0 to 1.91 and 2.18, respectively, for diet 0:80. Molar proportions of butyrate and valerate increased in a quadratic ($P < .05$) manner with increasing alfalfa in the diet. Butyrate and valerate molar proportions increased from 8.8 and .56, respectively, for diet 80:0 to 10.8 and 1.39, respectively, for diet 0:80. The quadratic increase ($P < .05$) in molar proportion of propionate observed in this study with increasing levels of AHP-WS, and in other studies when straws have been chemically treated (Ololade and Mowat, 1975), may be due to the more fermentable nature of the treated material (Table 2). Additionally, because the WS is not washed subsequent to treatment, hemicellulosic sugars solubilized by the AHP process presumably are readily available for ruminal fermentation. The fermentation of AHP WS in the rumen probably results in the generation of an excess of reducing equivalents. Under these conditions pyruvate would be converted to propionate, as observed for high-concentrate diets (Owens and Goetsch, 1988), as a rapid means of disposing of reducing equivalents. Linear ($P < .05$) increases in molar proportions of isobutyrate and isovalerate with increasing levels of alfalfa in the diet is indicative of the extensive ruminal fermentation of alfalfa protein (Owens and Goetsch, 1988). The linear ($P < .05$) increase in pH with increasing level of AHP WS from 6.45 on diet 0:80 to 6.67 on diet 80:0 was expected, given the alkaline nature of the treated WS. Ruminal NH_3 N increased ($P < .05$) from 13 mg/dl in diet 80:0 to 32.8 mg/dl in diet 0:80 in response to increasing levels of ruminally degraded protein from alfalfa. Ruminal liquid dilution rate

(LDR), particulate dilution rate (PDR), and ruminal volume all decreased linearly ($P < .05$) with increasing levels of alfalfa hay in the diet. Liquid and PDR decreased from 6.4 and 5.2%/h, respectively, for diet 80:0 to 5.4 and 3.4%/h, respectively, for diet 0:80. Ruminal volume decreased linearly ($P < .05$) from 10.8 liters in diet 80:0 to 9.4 liters in diet 0:80. Decreases in ruminal DR due to alfalfa addition may have been responsible for the modest but nonsignificant positive associative effects observed for ruminal fiber digestibility. Increased ruminal residence time would allow longer exposure of AHP WS to fibrolytic microorganisms. Work done by Berger et al. (1980) indicated that higher NaOH treatment levels of crop residues increased rates of passage and decreased fiber digestibilities. In the present work, alfalfa may have decreased both LDR and PDR and reduced ruminal volume by diluting the Na content of the experimental diets. Water intake is often positively correlated to ruminal passage rates (Merchen, 1988) and has been shown to increase when animals are fed NaOH-treated forages (Maeng et al., 1971). Alfalfa addition would reduce Na intake of sheep fed diets containing combinations of the two forages. This would result in decreased water intake and, therefore, LDR, PDR, and ruminal volume would decrease. Similar suggestions have been made by Paterson et al. (1982).

Implications

The alkaline hydrogen peroxide treatment process increased the availability of the fiber fraction of crop residues. However, total tract digestibilities of cell contents decreased in response to the treatment process. Total tract organic matter digestibility of a diet containing 80% treated wheat straw was identical to that observed when sheep were fed 80% high-quality alfalfa. Modest positive associative effects on digestibilities were noted when combinations of the forages were fed. Ruminal escape of alfalfa protein was approximately 25%. The supply of absorbable amino acids from alfalfa hay to the small intestine of sheep was quantified.

Literature Cited

AOAC. 1984. Official Methods of Analysis (14th Ed.). Association of Official Analytical Chemists, Washington, DC.

Berger, L. L., T. J. Klopfenstein and R. A. Britton. 1980. Effect of sodium hydroxide treatment on rate of passage and rate of ruminal fiber digestion. J. Anim. Sci. 50:745.

Brandt, R. T. and T. J. Klopfenstein. 1986. Evaluation of alfalfa-corn cob associative action. I. Interactions between alfalfa hay and ruminal escape protein on growth of lambs and steers. J. Anim. Sci. 63:894.

Cecava, M. J., N. R. Merchen, L. L. Berger and G. C. Fahey, Jr. 1990. Intestinal supply of amino acids in sheep fed alkaline hydrogen peroxide-treated wheat straw-based diets supplemented with soybean meal or combinations of corn gluten meal and blood meal. J. Anim. Sci. 68:467.

Chaney, A. L. and E. P. Marbach. 1962. Modified reagents for determination of urea and ammonia. Clin. Chem. 8: 130.

Goering, H. K. and P. J. Van Soest. 1970. Forage fiber analyses (apparatus, reagents, procedures, and some applications). Agric. Handbook 379. ARS USDA, Washington, DC.

Gould, J. M. 1984. Alkaline peroxide delignification of agricultural residues to enhance enzymatic saccharification. Biotechnol. Bioeng. 26:46.

Hart, S. P. and C. E. Polan. 1984. Simultaneous extraction and determination of ytterbium and cobalt ethylenediaminetetraacetate complex in feces. J. Dairy Sci. 67: 888.

Hsu, J. T., G. C. Fahey, Jr., L. L. Berger, R. I. Mackie and N. R. Merchen. 1991. Manipulation of nitrogen digestion by sheep using defaunation and various nitrogen supplementation regimens. J. Anim. Sci. (In press).

Kerley, M. S., G. C. Fahey, Jr., L. L. Berger, N. R. Merchen and J. M. Gould. 1986. Effects of alkaline hydrogen peroxide treatment of wheat straw on site and extent of digestion in sheep. J. Anim. Sci. 63:868.

Maeng, W. J., D. N. Mowat and W. K. Bilanski. 1971. Digestibility of sodium hydroxide-treated straw fed alone or in combination with alfalfa silage. Can. J. Anim. Sci. 51:743.

Merchen, N. R. 1988. Digestion, absorption and excretion in ruminants. In: D. C. Church (ed.) The Ruminant Animal: Digestive Physiology and Nutrition. pp 172–201. Prentice-Hall, Englewood Cliffs, NJ.

Merchen, N. R., J. L. Firkins and L. L. Berger. 1986. Effect of intake and forage level on ruminal turnover rates, bacterial protein synthesis and duodenal amino acid flows in sheep. J. Anim. Sci. 62:216.

Merchen, N. R. and L. D. Satter. 1983. Digestion of nitrogen by lambs fed alfalfa conserved as baled hay or as low-moisture silage. J. Anim. Sci. 56:943.

NRC. 1985a. Ruminant Nitrogen Usage. National Academy Press, Washington, DC.

NRC. 1985b. Nutrient Requirements of Sheep (6th Ed.). National Academy Press, Washington, DC.

Ololade, B. G. and D. N. Mowat. 1975. Influence of whole-plant barley reconstituted with sodium hydroxide on digestibility, rumen fluid and plasma metabolism of sheep. J. Anim. Sci. 40:351.

Ørskov, E. R. and D. A. Grubb. 1978. Validation of new systems for protein evaluation in ruminants by testing the effects of urea supplementation on intake and digestibility of straw with or without sodium hydroxide treatment. J. Agric. Sci. (Camb.) 91:483.

Owens, F. N. and A. L. Goetsch. 1988. Ruminal fermentation. In: D. C. Church (Ed.) The Ruminant Animal: Digestive Physiology and Nutrition. pp 145–171.

Prentice-Hall, Englewood Cliffs, NJ.

Paterson, J. A., T. J. Klopfenstein and R. A. Britton. 1982. Digestibility of sodium hydroxide-treated crop residues when fed with alfalfa hay. J. Anim. Sci. 54: 1056.

Preston, T. R. and R. A. Leng. 1984. Supplementation of diets based on fibrous residues and byproducts. In: F. Sundstøl and E. Owen (Ed.) Straw and Other Fibrous Byproducts as Feed. pp 373–413. Elsevier, Amsterdam, The Netherlands.

Robertson, J. B. and P. J. Van Soest. 1977. Dietary fiber estimation in concentrate feedstuffs. J. Anim. Sci. 45(Suppl. 1):439 (Abstr.).

SAS. 1985. SAS User's Guide: Statistics. SAS Inst., Inc., Cary, NC.

Satter, L. D. and L. L. Slyter. 1974. Effect of ammonia concentration on rumen microbial production in vitro. Br. J. Nutr. 32:199.

Storm, E., D. S. Brown and E. R. Ørskov. 1983. The nutritive value of rumen microorganisms in ruminants. 3. The digestion of microbial amino and nucleic acid in, and losses of endogenous nitrogen from, the small intestine of sheep. Br. J. Nutr. 50:479.

Uden, P., P. E. Colucci and P. J. Van Soest. 1980. Investigation of chromium, cerium and cobalt as markers in digesta rate of passage studies. J. Sci. Food Agric. 31:625.

Williams, C. J., D. J. David and O. Iismaa. 1962. The determination of chromic oxide in faeces samples by atomic absorption spectrophotometry. J. Agric. Sci. (Camb.) 59:381.

Zinn, R. A. and F. N. Owens. 1986. A rapid procedure for purine measurement and its use for estimating net ruminal protein synthesis. Can. J. Anim. Sci. 66:157.