EFFECT OF ALKALINE HYDROGEN PEROXIDE AND PERACETIC ACID ON IN SACCO RUMINAL DIGESTIBILITY OF ASPEN SAWDUST

K. H. Myung[1] and J. J. Kennelly[2]

Department of Dairy Science, Chonnam National University
Kwangju 500-757, Korea

Summary

The influence of alkaline hydrogen peroxide (AHP) and peracetic acid treatment on in sacco digestion of aspen was evaluated in three non-lactating ruminally cannulated Holstein cows fed a diet containing 90% forage and 10% concentrate on a DM basis. AHP treatment decreased lignin concentration by 40 to 60% resulting in increased concentrations of neutral detergent fiber (NDF), acid detergent fiber (ADF) and cellulose. Lignin concentrations in peracetic acid treated samples were less than 10% of values for control samples. In sacco disappearance rates of aspen DM, NDF, ADF and cellulose increased ($p < .05$) with AHP and peracetic acid treatment. Effective degradability of DM, NDF, ADF and cellulose were determined at a ruminal outflow rate of .05 h^{-1}. Effective degradabilities of AHP treated aspen were approximately three-fold greater and peracetic acid treated samples five-fold greater than untreated control samples. For all parameters measured, peracetic acid treatment resulted in higher ($p < .05$) digestion coefficients than AHP treated aspen. Results demonstrate that peracetic acid or AHP treatment can enhance the nutritive value of aspen sawdust for ruminants to a level comparable to that reported for many forages.

(Key Words: Alkaline Hydrogen Peroxide, Aspen, Rumen Digestibility)

Introduction

Wood has evoked little interest as a feedstuff for ruminants because of its low digestibility. The primary factors limiting digestion by ruminal cellulolytic microorganisms appear to be the close physical and chemical association between structural carbohydrates and lignin, and the high crystallinity of the cellulose in wood cell wall (Chambat et al., 1981; Jung and Fahey, 1983). Although relatively extensive research has been conducted on the effect of chemical and physical treatment on the feeding value of wood and bark (Butterbaugh and Johnson, 1974; Mellenberger et al., 1971; Millett et al., 1970 and Wilson and Pigden et al., 1964), treatment processes are generally either impractical on a large scale or do not increase *in vivo* digestibility of structural carbohydrates sufficiently to warrant commercial use.

Treatment of lignocellulosic materials, such as agricultural residues, with dilute alkaline hydrogen peroxide (AHP) solutions significantly increase the susceptibility of plant cell wall structural carbohydrates to ruminal digestion (Gould, 1984; Kerley et al., 1985). The mechanism by which AHP treatment enhances ruminal digestion appears to involve both a release of lignin from the lignocellulose matrix and an increase in the degree of hydration of the cellulose polymer (Kerley et al., 1985). Bacterial colonization of straw is limited, during the early stages of ruminal digestion, to those areas of the plant which have suffered physical damage during feed processing, chewing and rumination. In contrast, AHP treated straw is rapidly and extensively colonized by bacteria (Kerley et al., 1985). AHP treatment procedures which include washing of the substrate after treatment tend to result in substantial loss of cell wall hemicelluloses. pH-regulated solutions of AHP (Kerley et al., 1987) have been used to decrease the solubility and thus reduce the loss of hemicelluloses during the treatment process. Loss of soluble components can also be overcome by modifying the treatment procedure so that post-treatment washing of the sample is not necessary.

[1]Address reprint requests to Dr. K. H. Myung, Dairy Science Dept., Chonnam National University, Kwangju 500-757, Korea.

[2]Dept. of Animal Science, University of Alberta Edmonton, Alberta T6G 2P5, Canada.

Received October 25, 1991
Accepted June 1, 1992

Data on the efficacy of AHP or peracetic acid as a method of increasing digestibility of woody materials for ruminant animals is limited. Thus, the objectives of this experiment were to determine the influence of AHP or peracetic acid treatment on the chemical composition and in sacco digestion of aspen sawdust.

Materials and Methods

Three non-lactating ruminally cannulated Holstein cows were fed a diet with a forage to concentrate ratio (DM basis) of 90:10 (table 1).

TABLE 1. FORMULATION AND CHEMICAL COMPOSITION OF THE DIET FED TO COWS USED FOR THE IN SACCO STUDY

Items	DM basis (%)
Ingredients	
Whole crop oat silage	90.00
Barley grain, rolled	7.51
Canola meal	.57
Fish meal	.36
Wheat shorts	.55
Ground shelled corn	.90
Beet molasses	.55
Ammonium monophosphate	.01
Trace mineral premix[a]	.05
Chemical composition	
Crude protein	11.9
Neutral detergent fiber	50.3
Acid detergent fiber	34.5
Ash	7.5

[a] Contained 25 mg/kg Se, 40 mg/kg Co, .35% Mg, .25% Cu, .01% I and .75% Zn.

Animals were fed twice daily (07:30 and 17:00 h) in equal portions. Aspen sawdust was obtained from a sawmill that cuts bark-free logs of *Populus alba* × *Populus grandulosa*. After air drying for about 48 h, sawdust was ground through a 2 mm screen. Untreated aspen was used as a control. Treatment AHP-25 was prepared by suspending sawdust in a solution of H_2O_2 (1% w/v), pH adjusted to 11.5 with NaOH, for a period of 100 h at 25°C. Treatment AHP-120 was similar to AHP-25 except that reaction temperature was increased to 120°C for a period of 30 min. For the final treatment (PA) aspen was treated with a 1% NaOH solution at 120°C for 1 h followed by 20% peracetic acid at 120°C for 1 h (Toyama and Ogawa, 1975). Peracetic acid can be prepared using equal volumes of acetic anhydride and 35% hydrogen peroxide. After the specified reaction time, the insoluble residue was filtered, washed with distilled water until the pH of the filtrate was loss than 8, and dried at 60°C in a forced air oven for 48 h. Treated and untreated aspen sawdust were analyzed for proximate components (AOAC, 1975) and fiber fractions (Goering and Van Soest, 1970). All samples were ground through a 2 mm screen before being placed in nylon bags and incubated in the rumen.

Rumen bags were made of nylon cloth (Phentex, PE 3000 pore size 48 μm, B and SH Thomson Co. Ltd., Toronto, Ont.). Nylon cloth (14 × 22 cm) was folded in half and heat-sealed 3 mm from each of the two free edges. The final bag size exposed to ruminal fermentation was approximately 7 × 11 cm. Approximately 5 g (air dry) of test samples were placed in nylon bags. A total of 28 bags were placed into a polyester mesh bag (De Boer et al., 1987) and incubated in the rumen for 0, 6, 12, 24, 36, 48, 72 and 144 h. At the end of each incubation time, one bag per treatment per cow was removed from the rumen and mechanically washed (De Boer et al., 1987). After drying at 60°C in a forced-air oven for 72 h, the contents of each bag were subjected to DM (AOAC, 1975), NDF, ADF, and cellulose (Goering and Van Soest, 1970) analyses.

The percent disappearance of DM, NDF, ADF and cellulose at each incubation time was calculated from the proportion remaining after incubation in the rumen. The disappearance rate was fitted to the following equation (Robinson and Kennelly, 1988):

$$\text{Disappearance} = S + D\,(1 - e^{-k(t-\text{lag})})$$

Where S = soluble fraction (g kg^{-1}), D = potentially degradable fraction (g kg^{-1}), k = rate of degradation of D (h^{-1}), t = time of rumen incubation (h), lag = discrete time delay before degradation begins in the rumen (h). Nonlinear parameters S, D and k were estimated by an iterative least-square procedure. Effective degradability of DM (EDDM), NDF (EDNDF), ADF (EDADF) and cellulose (EDCE) were calculated

according to the equation (ørskov and McDonald, 1979):

EDDM, EDNDF, EDADF and EDCE = S + [(D × k)/(k + r)]

Where r = hypothetical rate of outflow from the rumen.

Analysis of variance were used to examine treatment differences (SAS, 1982). When F values were significant, treatment means were compared at probability of 0.05 using Student-Neuman-keuls' test (Steel and Torrie, 1960).

Results

Chemical analyses of untreated and treated aspen are in table 2. Although CP and fat content of both untreated and treated sawdust were low, both were decreased ($p < .05$) by treatment. A similar effect was observed for nitrogen free extract. This suggests that treatments caused solubilization of non ND fiber organic matter. This is supported by increased ($p < .05$) concentrations of NDF, ADF, cellulose and crude fiber.

Disappearance of DM and NDF from nylon bags are a function of ruminal incubation time are summarized in table 3. Disappearance of treated aspen DM was greater ($p < .05$) than that of untreated aspen for all incubation times after 12 h. The PA sample had higher ($p < .05$) DM disappearance than the other treated samples at zero h incubation, reflecting the presence of more readily soluble components. There were no differences ($p < .05$) between PA and AHP-120 treatments after 36 h incubation. However, DM disappearance for AHP 25 was lower ($p < .05$) than PA at all incubation times.

Disappearance of NDF in untreated aspen was substantially less ($p < .05$) than observed for treated samples. Differences between treatments followed similar trends to that observed for DM with PA samples having the highest ($p < .05$) rate and extent of degradation. Disappearance of ADF and cellulose (table 4) were similar to that observed for DM and NDF. However, there were tendencies for greater treatment differences ($p < .05$) at earlier incubation times than observed for DM and NDF. In general, in sacco disappearance of DM and major structural carbohydrates in aspen increased more than threefold with AHP or PA treatment at almost all ruminal incubation times.

Nonlinear parameters for effective degradabilities of DM and NDF for aspen samples studied are summarized in table 5. The PA sample had a higher ($p < .05$) soluble DM fraction (parameter S) than all other aspen samples. The soluble DM

TABLE 2. CHEMICAL ANALYSES (%) OF TREATED AND UNTREATED ASPEN (*P. Alba* × *P. Grandulosa*)

Item	Untreated Control	Treated Aspen			SE[d]
		AHP-25[a]	AHP-120[b]	PA[c]	
Dry matter	93.00[e]	91.97[f]	91.67[f]	94.59[f]	0.15
Percent of dry matter					
Crude protein	.72[e]	.43[ef]	.26[f]	.25[f]	0.03
Crude fat	1.10[e]	.76[f]	.20[h]	.37[g]	0.01
Crude fiber	70.05[e]	83.68[f]	86.78[f]	88.74[f]	0.43
Nitrogen free extract	27.62[e]	14.04[fg]	11.14[gh]	10.23[h]	0.15
Ash	.51[e]	1.09[f]	1.62[g]	.41[e]	0.03
Neutral detergent fiber	79.42[e]	84.78[f]	86.35[fg]	89.47[g]	0.53
Acid detergent fiber	67.43[e]	74.29[f]	77.88[f]	81.43[g]	0.37
Hemicellulose	11.99[e]	9.86[f]	8.47[f]	8.04[f]	0.65
Cellulose	53.62[e]	65.13[f]	70.43[f]	87.12[g]	0.47
Acid detergent lignin	20.16[e]	12.46[f]	9.48[f]	1.75[g]	0.09

[a] Treated with alkaline hydrogen peroxide at 25°C for 100 h.

[b] Treated with alkaline hydrogen peroxide at 120°C for 30 min.

[c] Treated with 1% sodium hydroxide for 1 h at 120°C and 20% peracetic acid at 120°C for 1 h.

[d] Standard error of the mean.

[e,f,g,h] Means in the same row without common letter in the superscripts differ ($p < .05$).

TABLE 3. DRY MATTER AND NDF DISAPPEARANCE (%) FROM TREATED AND UNTREATED ASPEN (*P. Alba* × *P. Grandulosa*) FROM NYLON BAG, AS A FUNCTION OF INCUBATION TIME

Incubation time (h)	Untreated Control	Treated Aspen[a]			
		AHP-25	AHP-120	PA	SE[b]
Dry matter					
0	8.8[c]	8.6[c]	5.8[c]	27.8[d]	.8
6	9.5[c]	9.3[c]	8.4[c]	32.2[d]	1.8
12	10.8[c]	18.6[d]	23.9[d]	48.5[e]	2.7
24	14.7[c]	42.8[d]	42.4[d]	81.9[e]	1.7
36	17.0[c]	61.5[d]	66.5[d]	94.9[e]	2.4
48	19.4[c]	75.3[d]	81.1[d]	99.0[e]	2.1
72	24.2[c]	82.4[d]	85.2[d]	99.7[e]	1.8
144	29.9[c]	85.5[d]	87.2[d]	99.9[e]	1.2
Neutral detergent fiber					
0	8.7[d]	9.0[d]	6.3[c]	28.0[e]	1.5
6	9.1[c]	13.2[d]	12.5[d]	34.2[e]	1.7
12	10.4[c]	22.8[d]	29.1[d]	53.4[e]	.7
24	13.6[c]	46.5[d]	47.6[d]	84.7[e]	.7
36	15.4[c]	67.5[d]	74.6[e]	97.6[f]	1.9
48	18.3[c]	79.3[d]	88.2[e]	99.1[f]	1.7
72	22.7[c]	86.7[d]	88.3[d]	99 4[e]	3.4
144	25.3[c]	88.4[d]	90.3[d]	99.9[e]	1.3

[a] See table 2 for details of treatments.

[b] Standard error of the mean.

[c,d,e,f] Means in the same row without a common letter in their superscripts differ ($p < .05$).

fraction for AHP-25 was similar to that observed for AHP-120. The soluble NDF fraction for PA was higher ($p < .05$) than observed for AHP-25 and AHP-120. The potentially degradable DM and NDF fractions (sum of parameters S + D) was least ($p < .05$) for the untreated sample and greatest ($p < .05$) for PA. The fractional rate constant (k) of both DM and NDF was largest for PA, intermediate for AHP-25 and AHP-120 and least ($p < .05$) for the untreated sample. The EDDM and EDNDF values were three to fivefold higher for treated samples reflecting the dramatic effect of treatment on in sacco disappearance of aspen. The nonlinear parameters for EDADF and EDCE (table 6) followed similar trends to that observed for DM and NDF. Although lag values obtained in this experiment were not always significant ($p < .05$), there was a trend for decreased lag values for treated samples.

Discussion

Untreated aspen sawdust has limited potential as a feedstuff for ruminants due to its low ruminal digestibility (Butterbaugh and Johnson, 1974; Millett et al., 1970; Scott et al., 1969). Millett et al. (1970) reported a 60% improvement in DM digestibility when aspen was treated with 20% (W/W) NaOH. Although NaOH treatment results in partial delignification and increases the susceptibility of aspen cellulose to bacterial digestion, a substantial proportion of the cellulose resists ruminal digestion. Lignin has been implicated as the primary compound limiting digestion of low quality roughages (Jung and Fahey, 1983; Kerley et al., 1985; Lewis et al., 1987b). The use of oxidative agents such as hydrogen peroxide in combination with alkaline hydrolysis result in solubilization of up to 60% of lignin. This appears to be sufficient to allow almost complete digestion of cellulose. In this study, AHP treatment resulted in a loss of 40 to 50% of lignin which is comparable to that reported for wheat straw (Lewis et al., 1987b). The extent and rate of ruminal digestion was markedly increased by AHP treatment. The mechanism of action of AHP

TABLE 4. ACID DETERGENT FIBER AND CELLLLOSE DISAPPEARANCE (%) FROM TREATED AND UNTREATED ASPEN (*P. Alba* × *P. Grandulosa*) FROM NYLON BAG, AS A FUNCTION OF INCUBATION TIME

Incubation time (h)	Untreated Control	Treated Aspen[a]			
		AHP-25	AHP-120	PA	SE[b]
Acid Detergent Fiber					
0	8.5[c]	9.2[c]	7.8[c]	25.9[d]	.8
6	9.6[c]	13.3[d]	8.8[c]	31.2[e]	.3
12	11.5[c]	23.6[d]	23.7[d]	42.2[e]	1.5
24	15.2[c]	43.0[d]	39.6[d]	80.3[e]	2.2
36	16.9[c]	59.3[d]	64.3[e]	94.0[f]	2.5
48	19.2[c]	75.2[d]	79.1[e]	98.7[f]	1.8
72	24.3[c]	85.6[d]	82.6[e]	99.9[f]	1.8
144	29.3[c]	88.4[d]	84.7[d]	99.9[e]	1.4
Cellulose					
0	9.0[c]	8.6[c]	7.9[c]	31.4[d]	1.4
6	13.1[c]	13.0[c]	14.0[c]	44.4[d]	1.9
12	14.1[c]	20.7[d]	30.2[e]	57.5[f]	1.5
24	16.7[c]	48.7[d]	48.0[d]	83.1[e]	2.4
36	18.1[c]	65.1[d]	75.2[e]	98.3[f]	2.9
48	22.7[c]	79.0[d]	84.3[e]	99.9[f]	2.5
72	26.9[c]	86.4[d]	88.1[d]	99.9[e]	1.7
144	33.1[c]	88.7[d]	89.7[d]	99.9[e]	2.2

[a] See table 2 for details of treatments.

[b] Standard error of the mean.

[c,d,e,f] Means in the same row without common letter in their superscripts differ ($p < .05$).

has not been fully elucidated; however, there is evidence that partial solubilization of lignin, and physical disruption of cell walls facilitates rapid attachment of bacteria and subsequent digestion of cellulose by bacterial enzymes (Kerley et al., 1985; Kerley et al., 1988; Lewis et al., 1988). Although AHP treatment is likely to partially disrupt the crystalline structure of the fiber source, this disruption does not appear to be a primary factor in enhancing digestibility. Fiber sources which are extremely crystalline, but contain low concentrations of lignin and hemicellulose are not markedly influenced by AHP treatment (Lewis et al., 1988). Substrates which are highly lignified; such as aspen sawdust, are good candidates for AHP treatment. Gould (1985) reported that AHP is more effective in promoting digestion of monocotyledonous plants than digestion of dicotyledonous plants. The higher frequency of P-hydroxyphenylpropane units in the lignin of monocotyledonous plants has been implicated as a causative factor. AHP treatment markedly increased the degradable fraction and rate of degradation for NDF, ADF and cellulose (tables 5 and 6). In addition, the lag time required prior to digestion was significantly less for ADP treated than control samples. A decreased lag time could be due to more rapid bacterial colonization of treated substrates. Once colonized, digestion would proceed at a more rapid rate due to the partial removal of lignin and physical disruptions of the cell wall.

In this study, peracetic acid was the most efficacious treatment in maximizing in sacco digestion of aspen sawdust. Peracetic acid appears to exert its effect by degrading the aromatic nuclei of lignin to water soluble muconic acid structures (Sarkanen and Suzuki, 1965; Browning, 1967). Delignification with AHP can proceed at a relatively rapid rate at room temperature (Gould, 1984). In contrast, effective peracetic acid treatment appears to require higher temperatures or extended reaction times (Toyama and Ogawa, 1975). As a result, the use of peracetic acid is

TABLE 5. NONLINEAR PARAMETERS[a] AND EFFECTIVE DEGRADABILTY OF DRY MATTER (EDDM) AND NEUTRAL DETERGENT FIBER (EDNDF) FOR TREATED AND UNTREATED ASPEN (*P. Alba* × *P. Grandulosa*)

Parameter	Untreated Control	Treated Aspen[b]			
		AHP-25	AHP 120	PA	SE[c]
Fraction of dry matter					
soluble (S) (% of total)	8.9[d]	9.0[d]	6.7[d]	28.7[e]	1.15
degradable (D) (% of total)	25.0[d]	78.2[f]	83.4[g]	72.2[e]	1.41
degradation rate of the degradable fraction (k), (h^{-1})	.01[d]	.04[e]	.04[e]	.08[e]	.001
lag (l), (h)	5.4	9.3	7.1	7.2	1.1
EDDM	14.2[d]	45.1[e]	44.9[e]	72.5[f]	1.17
Fraction of NDF					
soluble (S) (% of total)	8.9[e]	10.5[e]	6.3[d]	28.0[f]	0.72
degradable (D) (% of total)	18.5[d]	80.4[f]	87.4[g]	73.4[e]	1.11
degradation rate of the degradable fraction (*k*), (h^{-1})	.02[d]	.04[e]	.04[e]	.08[f]	.002
lag (l), (h)	8.5	8.0	4.9	5.2	.7
EDNDF	13.9[d]	47.2[e]	47.3[e]	72.2[f]	.81

[a] S, D and k are non-linear parameters. l is discreet lag before degradation began. EDDM and EDNDF are calculated on the basis of .05 h^{-1} solid outflow rate.

[b] See table 2 for details of treatments.

[c] Standard error of the mean.

[d,e,f,g] Means in the same row without a common letter in their superscripts differ ($p < .05$).

TABLE 6. NONLINEAR PARAMETERS[a] AND EFFECTIVE DEGRADABILITY OF ACID DETERGENT FIBER (EDADF) AND CELLULOSE (EDCE) FOR TREATED AND UNTREATED ASPEN (*P. Alba* × *P. Grandulosa*)

Parameter	Untreated Control	Treated Aspen[b]			
		AHP-25	AHP-120	PA	SE[c]
Fraction of ADF					
soluble (S) (% of total)	8.5[d]	9.2[d]	8.3[d]	27.2[e]	1.12
degradable (D) (% of total)	24.9[d]	83.0[f]	79.1[f]	73.8[e]	1.73
degradation rate of the degradable fraction (k), (h^{-1})	.01[d]	.03[e]	.04[e]	.08[f]	.001
lag (l), (h)	8.3	8.1	5.4	3.9	1.0
EDADF	13.9[d]	41.7[e]	44.7[e]	72.5[f]	1.52
Fraction of cellulose					
soluble (S) (% of total)	8.6[d]	10.0[d]	7.9[d]	31.4[e]	.71
degradable (D) (% of total)	33.8[d]	80.9[e]	84.9[e]	70.6[e]	3.32
degradation rate of the degradable fraction (k), (h^{-1})	.01[d]	.04[e]	.04[e]	.07[f]	.001
lag (l), (h)	8.2[d]	4.8[f]	3.5[e]	3.4[e]	1.22
EDCE	14.3[d]	47.5[e]	47.3[e]	71.7[f]	1.22

[a] See table 5 for details of parameters.

[b] See table 2 for details of treatments.

[c] Standard error of the mean.

[d,e,f] Means in the same row without common letter in their superscripts differ ($p < .05$).

more likely to be limited by treatment cost than AHP (Kamstra et al., 1980) even though it appears to be one of the most potent delignifying agents available (Toyama and Ogawa, 1975).

On the basis of data presented here, it is concluded that peracetic acid or AHP treatment permits extensive digestion of aspen sawdust by bacterial enzymes possibly by removing the physical and chemical barriers to digestion attributed to the close association between lignin, cellulose, and hemicellulose.

Literature Cited

AOAC. 1975. Official Methods of Analysis (12th Ed.). Association of Official Analytical Chemists. Washington, D.C.

Butterbaugh, J. W. and R. R. Johnson. 1974. Nutritive value of acid hydrolyzed wood residue in ruminant rations. J. Anim. Sci. 38:394.

Chambat, G., J. P. Joseleau and F. Barnoud. 1981. The carbohydrate constituents of the cell wall of suspension cultures of *Rosa glauca*. Phytochemistry 10:241.

De Boer, G., J. J. Murphy and J. J. Kennelly. 1987. A modified method for determination of in situ rumen degradation of feedstuffs. Can. J. Anim. Sci. 67:93.

Gellerstedt, G. and R. Agmemo. 1980. The reaction of lignin with alkaline hydrogen peroxide. Part 4. The oxidation of conjugated carbonyl structures. Acta. Chem. Scan. B34:275.

Goering, H. K. and P. J. Van Soest. 1975. Forage fiber analyses (apparatus, reagents, procedures and some applications). Agric. Handbook 379. ARS, USDA, Washington, D. C.

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

Gould, J. M. and S. N. Freer. 1984. High-efficiency ethanol production from lignocellulosic residues pretreated with alkaline H_2O_2. Biotechnol. Bioeng. 26:628.

Gould, J. M. 1985. Studies on the mechanism of alkaline peroxide delignification of agricultural residues. Biotechnol. Bioeng. 27:225.

Jung, H. G. and G. C. Fahey, Jr. 1983. Nutritional implications of phenolic monomers and lignin: A review. J. Anim. Sci. 57:206.

Kerley, M. S., G. C. Fahey, Jr., L. L. Berger, J. M. Gould and F. L. Baker. 1985. Alkaline hydrogen peroxide treatment unlocks energy in agricultural by-products. Science 230:820.

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.

Kerley, M. S., G. C. Fahey, Jr., L. L. Berger Jr., N. R. Merchen and J. M. Gould. 1987. Effects of treating wheat straw with pH-regulated solutions of alkaline hydrogen peroxide on nutrient digestion by sheep. J. Dairy Sci. 70:2078.

Kerley, M. S., G. C. Fahey, Jr., J. M. Gould and E. L. Innotti. 1988. Effects of lignification, cellulose crystallinity and enzyme accessible space on the digestibility of plant cell wall carbohydrates by the ruminant. Food Microstructure 7:59.

Lewis, S. M., D. P. Holzgraefe, L. L. Berger, G. C. Fahey, Jr., J. M. Gould and G. F. Fanta. 1987a. Alkaline hydrogen peroxide treatments of crop residues to increase ruminal dry matter disappearance in sacco. Anim. Feed Sci. Technol. 17:179.

Lewis, S. M., M. S. Kerley, G. C. Fahey, Jr., L. L. Berger and J. M. Gould. 1987b. Use of alkaline hydrogen peroxide-treated wheat straw as an energy source for the growing ruminant. Nutr. Rep. Int. 35:1093.

Lewis, S. M., L. Montgomery, K. A. Garleb, L. L. Berger and G. C. Fahey, Jr. 1988. Effects of alkaline hydrogen peroxide treatment on *in vitro* degradation of cellulosic substrates by mixed ruminal microorganisms and bacteroides succinogenes S85. Appl. Environ. Microbiol. 54:1163.

Mellenberger, R. W., I. D. Satter, M. A. Millett and A. J. Baker. 1971. Digestion of aspen, alkali-treated aspen, and aspen bark by goats. J. Anim. Sci. 32:756.

Millett, M. A., A. J. Baker, W. C. Feist, R. W. Mellenberger and I. D. Satter. 1970. Modifing wood to increase its *in vitro* digestibility. J. Anim. Sci. 31:781.

Ørskov, E. R. and I. McDonald. 1979. The estimation of protein digestibility in the rumen from incubation measurments weighted according to rate of passage. J. Agri. Sci. (Camb.) 92:499.

Robinson, P. H. and J. J. Kennelly. 1988. Ammonia or sulfur dioxide treatment of high-moisture barley on in situ rumen degradability and in situ whole-tract digestibility. Can. J. Anim. Sci. 68:779.

SAS. 1982. SAS User's guide: Statistics. SAS Inst., Inc., Cary, NC.

Scott, R. W., M. A. Millett and G. J. Hajny. 1969. Wood wastes for animal feeding. Forest. Prod. J. 19(4):14.

Steel, R. G. D. and J. H. Torrie. 1960. Principles and Procedures of Statistics. McGraw-Hill Book Co., New York.

Toyama, N. and K. Ogawa. 1975. Sugar production from agricultural woody waste by saccharification with *Trichoderma viride* celluase. In: C. R. Wilke (Ed.). Symposium #5: Cellulose as a Chemical and Energy Resource. John Wiley and Sons Inc., New York. pp. 225-244.

Wilson, R. K. and W. J. Pigden. 1964. Effect of sodium hydroxide treatment on the utilization of wheat straw and poplar wood by rumen microorganisms. Can. J. Anim. Sci. 44:122.