# THE NUTRITIVE VALUE OF ACID HYDROLYZED WOOD RESIDUE IN RUMINANT RATIONS

Ву

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Bachelor of Science

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1970

Submitted to the Faculty of the Graduate College of the Oklahoma State University in partial fulfillment of the requirements for the Degree of MASTER OF SCIENCE July, 1972

Thesis 1972 B988n Cop. 2

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# THE NUTRITIVE VALUE OF ACID HYDROLYZED WOOD RESIDUE IN RUMINANT RATIONS

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#### ACKNOWLEDGMENTS

The author wishes to express his appreciation to Dr. R.R. Johnson, Professor of Animal Science and Biochemistry, for his guidance and assistance during the course of this study. Appreciation is also extended to Dr. J.E. McCroskey and Dr. L.J. Bush for their help in the preparation of this manuscript.

Grateful acknowledgment is also extended to Dr. R.D. Morrison, Professor of Mathematics and Statistics, for aid in conducting the statistical analysis of data.

Further acknowledgment is extended to fellow graduate students and Dr. H.E. Kiesling for their assistance and encouragement during the course of this study.

A very special thanks is given to the author's wife, Joy, for her most valuable help and understanding throughout this program of graduate study.

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#### CHAPTER I

#### INTRODUCTION

Through the years wood has evoked little interest in the diet of ruminants; only in times of national emergencies has any great quantity of wood or wood residue been fed to ruminants. The concept of feeding wood is very old, but until recently the practice has received little attention from researchers in developing the use of wood or wood by-products as an energy source for ruminants. Several factors have increased the interest in the utilization of wood. It is evident that the domestic animal will eventually compete with the human for cereal grain and other foodstuffs that are being produced for use by man; therefore, animals will be forced to utilize food supplies that cannot be utilized by humans. creased pressure to dispose of refuse from harvesting and milling practices in a manner that does not add increased pollution to our environment, the wood industry is in search of products that could utilize great quantities of low quality wood or wood residues.

Researchers have studied the incorporation of raw sawdust into ruminant rations but have been unable to incorporate more than minor quantities without depression

of intake and gain. Other workers have studied the use of chemical treatment of wood and poor quality forages by treating with alkaline reagents to release cellulose from the lignin-cellulose complex. Very little attention has been given to the use of acid hydrolyzed wood residue as an energy source for ruminants.

The purpose of this study was to compare the nutritive value of rations containing acid hydrolyzed wood residue to alfalfa meal diets. The methods used consisted of feeding trials, digestion trials and studies on in vivo volatile fatty acid concentrations. Data on digestion of cellulose in dacron bags suspended in the rumen were collected to determine if the level of hydrolyzed wood in the diet influenced the ability of rumen microorganisms to digest cellulose.

#### CHAPTER II

## REVIEW OF LITERATURE

## Introduction

Although wood contains a relatively high amount of carbohydrates, 70-80%, most of it is indigestible by the ruminant animal. Research workers have considered the problem from two viewpoints. Wood can be considered an inert ingredient that can be incorporated into ruminant rations in small amounts to serve as a roughage factor. The other view is that by chemically treating wood to improve its digestibility it may be considered a potential energy source for the ruminant animal. This review will cover the use of chemically treated and untreated wood in ruminant rations and the use of the dacron bag technique as a method for evaluating feedstuffs.

## Sawdust in Ruminant Rations

The utilization of untreated wood residue has been tried by several researchers. Most have found untreated wood to be nearly indigestible by the ruminant animal. The degree of digestibility of wood has been shown to be very species dependent. Millet et al. (1970) using in

<u>vitro</u> methods tested 24 species of wood and found that only three species were digested to any extent by rumen micro-organisms (aspen 32%, black ash 17%, soft maple 30%, and all other species 0-8%). Similar results have been obtained by Fiest, Baker and Tarkow (1970).

Satter, Baker and Millet (1970) have shown that aspen sawdust can replace 30% of a conventional ration in dairy cattle producing 20 kg of milk daily without reducing the intake of digestible dry matter or the production of milk. Cows consuming the aspen sawdust maintained a normal milk fat level. Mellenberger et al. (1970) obtained digestion coefficients of 41% when aspen was incorporated into a high-roughage ration and 28% when in a high-concentrate ration fed in the diets of goats.

In contrast to the data supporting the use of untreated aspen, Dinius et al. (1970) has confirmed that oak is essentially indigestible by sheep, thus making it a poor energy source. Researchers working with the utilization of indigestible woods have used them primarily as roughages or diluents in high concentrate rations with fair success.

Anthony and Cunningham (1969) reported that a ration containing 2.5% hardwood sawdust supported, a higher rate of gain than a basal ration or a basal ration plus 10% sawdust. In further work with oak sawdust it was found that sawdust included at 10% of the ration gave equal performance to rations containing an equal amount of coastal

bermudagrass; however, cattle fed 15% sawdust showed depressed gains and consumed slightly less feed than the coastal bermudagrass fed cattle. This led Anthony and Cunningham (1969) to conclude that sawdust could be added to steer rations up to a level of 10% with favorable results. Kitts et al. (1969) conducted several trials with alder sawdust and found similar results. Steers fed grass hay gained faster and had better feed efficiency than those fed untreated sawdust at 15 and 20% of the rations.

el-Sabban, Long and Baumgardt (1971) compared oak sawdust fed at 5% and 15% of the diet with oyster shells and ground timothy hay as roughages in cattle rations. Feedlot performance data indicated that sawdust could be used successfully at levels up to 15% of the ration without significantly effecting performance. Coarse ground sawdust gave better results than fine ground sawdust.

At stages in the life cycle of ruminants it may be advantageous to limit energy intake to prevent rapid growth or unwanted fattening. Dinius and Baumgardt (1970) used oak sawdust in the ration. Dilutions up to 35% allowed regulation of energy intake. Cody, Morill, and Hibbs (1968) reported that grain intake can be controlled by including 25-45% wood fiber with grain in rations of dairy animals.

Although it has been shown that limited quantities of wood or sawdust may be used as a roughage factor or a diluent in ruminant rations, it is evident that most

species of wood fed without chemical or physical treatment are unsatisfactory as major components in the ruminant diet. This has caused increased interest in the alteration of wood to make it more digestible by the ruminants.

## Chemical Treatment of Wood Residue

Before wood residue can receive any appreciable consideration as an energy source for ruminants some form of pretreatment is necessary to release wood carbohydrates. As stated by Guggolz, Kohler and Klopfenstein (1971), carbohydrate availability is related to one or more of three factors, (1) lignin acts as an inert barrier between the carbohydrate and the digesting enzyme, (2) the cellulose is too highly crystalline to be quickly available to enzyme action or (3) silica inhibits carbohydrate digestibility.

Research has concentrated on the removal of lignin and solubilization of some hemicellulose by treating with alkaline reagents, or on acid hydrolysis as a means of degrading wood carbohydrate to sugars without the removal of lignin. Because of the expense associated with conventional pulping methods they have not been extensively investigated although they ultimately remove lignin and yield cellulose in a usuable form.

## Alkaline Treatment of Wood Residue

The treatment of fibrous materials by dilute alkaline

reagents has been investigated by several researchers. The original process of NaOH treatment of roughages was patented by Beckman in 1918. The procedure was a modification of earlier practices which required heat in the digesting process. The Beckman process involved hydrolysis of forages with eight times its weight of 1.5 percent NaOH in open vats for approximately 3 hours. The liquor was then drained off and the remaining fiber was washed until neutral. Although this process increased the yield of digestible fibrous material, a considerable loss in soluble nutrients was apparent.

Much of the early work was conducted in Germany and was reviewed by Archibald (1924). An early explanation for the action of alkaline reagents is that given by Magnus in his "Theorie and Praxis der Strogayfshliessung" (30, p. 7-13) as cited by Archibald (1924).

Separation and solution of the silicic acid, which constitutes a portion of the incrusting substance of the straw and is present in most straws to the extent of 1 to 2 percent, while some straws contain as much as 5 percent.

Splitting off of the methoxy and acetyl groups from the lignin, of which they form a characteristic part. This results in the production of acetic acid with consequent neutralization of more or less of the alkali employed in the process. The lignin itself is also profoundly changed and is rendered more insoluble and inactive. It should be borne in mind, however, that complete, or nearly complete, solution and removal of the lignin can be brought about when desired by repeated treatment with alkali at higher temperatures than those successfully employed for straw hydrolysis. In paper manufacture this is what actually takes place.

Forcing or springing of the bonds which link the lignin and cellulose together. The theory of a linkage between these two substances in the fiber is advanced by Magnus, and he considers that the springing apart of these bonds is the most important and essential nature in the action of the alkali. As a result, the intestinal bacteria of animals are enabled to attack the cellulose and split it up into simpler substances, such as sugars and organic acids which can then be utilized by the animal organisms.

Holtzer (1952) reported that alkaline degradation appears to involve three separate stages. (1) The absorption of alkali at the lignin interface by acidic phenolic groups in the lignin complex. This process is rapid and does not influence the reaction rate. (2) Following absorption of alkali, a chemical combination takes place between lignin and the absorbed alkali. (3) In the final stage, chemical hydrolysis takes place and the alkali-lignin surface enters the liquid. The energy calculated to be necessary for the solution of one alkali-lignin complex is in the order of 5.3 x 10<sup>-23</sup> calories.

Tarkow and Fiest (1969) recently suggested that dilute alkali treatment of wood increased the digestibility
of wood by sponifying ester bonds on xylan chains acting
as cross links that restrain swelling. They also reported
the saponification of acetyl groups in wood. From this
they theorized that knowing the amount of esterified and
nonesterified uronic acid groups and acetyl groups in the
wood, it is possible to calculate the NaOH requirement for
saponification and therefore for maximum improvement in

digestibility. The increase in digestibility is due to the increased accessibility of the wood to hydrolytic attack by celluloytic organisms. Using their theoretical model and data on the chemical composition of oak and aspen, they calculated that it would require 5.1 grams of NaOH per 100 grams of wood for aspen and 4.5 grams of NaOH per 100 grams for oak. In vitro digestion trials indicated 5 to 6 grams of NaOH per 100 grams of wood were required to obtain maximum digestibility, and the minimum ratio for the maximum digestibility was independent of species. The maximum digestibility of aspen was about 50%, and the maximum with red oak was about 15%.

The problem of nutrient loss by solubilization may approach 25% by leaching during the washing operation as shown by Singh and Jackson (1971) and Saxena et al. (1971). Cost of processing and loss of nutrients are major reasons for only minor use of the technique. Recently Wilson and Pigden (1964) described a process for treatment of forages and wood that did not involve leaching after treatment with NaOH. The dry roughage was sprayed with 0 to 10% sodium hydroxide. It was then neutralized by exposing to atmospheric CO<sub>2</sub> or by mixing it with silages or acetic acid. Working with poplar wood they showed that the in vitro digestibility could be increased from 5 to approximately 40%. Other workers (Maeng, Mowat and Bilanski, 1971, Guggolz et al., 1971, Saxena et al., 1971, and Singh et al.,

quality forages.

Mellenberger et al. (1971) used alkali concentration only slightly greater than the amount needed for saponification of acetyl and carboxyl groups. The fiber was than washed once with a resulting weight loss of 5%. The in vitro dry matter digestibility was increased from 41% to 52% using aspen wood as the test material.

Many modications are used in the treatment of forages and wood fibers with alkaline reagents. The effectiveness of treating with alkali to improve digestibility is closely related to the species of wood or forage being used as suggested by Tarkow to the species of wood. (1969).

## Acid Treatment of Wood Residue

The acid hydrolysis of wood is used in the production of wood alcohol and wood molasses. Limited research has been conducted on the adaptation of the process to the production of animal feeds. Acid hydrolysis of wood involves the interaction of water with the various components of wood. At ordinary temperature and pressure, the reaction with water is so slow that it is not detected by usual analytical procedures. The action may be catalyzed by heat, acids or pressure so that the reactions occur more rapidly. (Harris, 1952)

Hydrolysis of the components of wood require varying conditions. Conditions which hydrolyzed hemicellulose and the lignin to soluble products leave the cellulose un-

changed. The resistance of cellulose to hydrolysis is assumed to be due to the crystalline nature of cellulose rather than the type of linkage between the glucose molecules. Cellulose requires temperature corresponding to 150 psi steam pressure with 0.8 percent sulfuric acid for 20 to 30 minutes to convert half of the cellulose into simple sugars. (Harris, 1952)

Early work in the development of dilute acid hydrolysis of wood into an animal feed was conducted by Sherrard and Blanco (1921). Their procedure treated sawdust in the same manner as for the production of ethyl alcohol. Sawdust was digested with 0.8 percent sulfuric acid for 15 or 20 minutes under 120 pounds of steam pressure. acid and soluble sugars were extracted and neutralized with The sugars were then evaporated and combined with the residue remaining after hydrolysis. Analysis revealed that the finished product yield was 90 to 94% of the original wood. Loss in weight was largely accounted for by the conversion of the pentosans of the hemicellulose fraction to furfural and volatile acids. The original wood was found to be 56% cellulose. The cellulose remaining after hydrolysis calculated upon the weight of the original wood was 34 percent. This indicated that approximately 40 percent of the cellulose was hydrolyzed. From this a yield of 15% total reducing sugar was produced (based on weight of original wood). It is of interest to note that the total quantity of lignin contained in the wood (31%) was

not altered by this method of acid hydrolysis.

Feeding trials were conducted by several investigators using the hydrolyzed wood produced by Sherrard (1921). Archibald (1926) conducted digestibility trials to evaluate the feeding value of hydrolyzed Douglas fir sawdust, Eastern white pine and the residue of Eastern white pine minus the liquor produced. The residue was fed to evaluate the assumption that it was probably without food value. Results of the digestion trials revealed that hydrolyzed Douglas fir sawdust was about 33% digestible. Dry matter of hydrolyzed Eastern white pine (residue and liquor) was found to be 46% digestible. Archibald noted that the residue from the Eastern white pine when fed to lambs caused digestive disturbances and yielded dry matter digestion coefficient of 18%. (Average of four trials varying from 1.69 to 32.41%.)

The most promising approach to the utilization of acid hydrolyzed wood residue is the hydrolysis of the polysaccharides to soluble sugars and the preparation for use in the form of molasses. Improvements in hydrolysis and evaporating equipment have made it possible to evaporate low yield sugar solutions to molasses. Several workers have found wood molasses to be comparable to cane molasses in feeding value as stated by Jones (1949). Harris (1952) reported the hydrolysis of hardwood, such as aspen, would yield about 1 ton of 50% sugar solution per ton of wood. These processes are limited by economics and the

use of the remaining residue after hydrolysis.

The problem of using the residue for an animal feed is compounded by methods of hydrolysis. The formation of carbohydrate decomposition products during the hydrolysis of wood may increase the indigestible residue which may be detected as a resinous humic material with lignin-like properties. Furfural, the decomposition product of xylose, may be produced at the extent of 2 to 3 percent of the weight of the wood. In the presence of dilute mineral acids at elevated temperature, hexose sugars undergo decomposition, producing among other substances hydroxymethyl-furfural and eventually levulinic acid. Many of these products are not metabolizable or are toxic to microorganisms.

## Nylon Bag Technique

Research workers have investigated methods for obtaining an estimate of forage nutritive value by the use of small sample methods. The majority of these procedures are based on the use of in vitro techniques; however, work has been conducted on the use of small sample in vivo techniques by the use of cotton threads, dialysis membrane bags, and nylon or dacron bags suspended in the rumen of the test animal.

Hoflund, Quin and Clark (1948) used cotton threads to study the rate of cellulose digestion when placed in the rumen of fistulated sheep. Balch and Johnson (1950) used cotton threads to determine that cellulose is broken

down faster in the ventral sac of the rumen than in the dorsal sac.

Pettyjohn, Leatherwood and Mochrie (1964) used a dialysis membrane bag inside a plastic cylinder to simulate rumen conditions. The nature of the bag prevents entrance of nondialyzable molecules or particles, and allows diffusion of end products such as volatile fatty acids without escape of small feed particles. This procedure has several disadvantages in that the membrane bag will deteriorate after extended periods of time in the rumen. The dialysis membrane bag method requires an in vitro type preparation as the digesting media, including inoculum since the microorganisms of the rumen from the test animal cannot penetrate the membrane.

The most widely used methods for small sample <u>in vivo</u> evaluation of forages are based on the use of the weighted nylon or dacron bag suspended in the rumen of a fistulated sheep. The pores in the nylon allow movement of rumen liquor in and out of the bag without allowing escape of undigested particles of the feed or forage being evaluated. Several workers have investigated the use of the nylon bag technique as a means of relating estimates of digestion coefficients to coefficients obtained by conventional digestion trials. Quin, Van der Wath and Mybrugh (1938) observed disintegration of feed samples by suspending small hylon bags in the rumen of fistulated sheep. Van Keuren and Heineman (1962) evaluated the nylon bag technique and were able

to obtain good repeatability between samples taken over a series of days. Anthony et al. (1969) used the nylon bag technique to determine dry matter digestibility of rations containing 10 and 15% sawdust. His data were corrected by using forages of known digestibility placed in the rumen with the test ration.

One major problem encountered by researchers using dry matter digestibility in the nylon bag was the effect of washing time on the dry matter disappearance. Van Dyne (1962) reported that rinsing of nylon bags upon removal from the animal as compared to thorough repeated washing and soaking resulted in large (highly significant) differences in estimates of dry matter digestion. Other workers have also found washing time to be a factor effecting dry matter digestibility coefficients obtained by the nylon bag technique (Manta, 1969).

Cellulose digestion in small nylon bags as an estimate of actual cellulose digestion has been used successfully by Lusk, Browning and Miles (1962). They found that a correlation coefficient of +0.83 (P <.05) was obtained when the 48 hour legume and 72 hour grass hay nylon bag digestion coefficients were compared with results from conventional digestion trials. Significant positive correlations between cellulose digestion measured by two methods indicated that the nylon bag technique used with a regression equation might provide a valid estimate of cellulose digestion in nylon bags were small although significant.

Van Dyne (1962), evaluating factors influencing digestion coefficients obtained by the nylon bag technique, found that sample size was inversely related to in vivo cellulose digestion in the range of sample sizes from 2 to 10 grams of ground forage. Length of fermentation period appeared to have a linear relationship to cellulose digestion between 24 and 72 hours when annual range forage was evaluated. Van Keuren et al. (1962), Neathery (1969), and Erwin and Ellistan (1959) have reported dry matter digestion coefficients obtained by the nylon bag technique were decreased by increasing sample size (from 2 to 24 grams) and directly influenced by length of the time in the rumen.

The dietary regime of the test animal has been shown to be a major source of variation in estimating digestion coefficients with the nylon bag as demonstrated by Neathery (1969), Van Keuren et al. (1962), and Van Dyne (1962). Hopson, Johnson and Dehority (1963) reported the dietary regime of the test animal had a significant effect on the digestion coefficients obtained from the nylon bag technique. It was also noted that the nylon bag technique is not well adapted to the estimation of in vivo digestion coefficients when using incubation times less than 42 hours.

Due to the sources of variation associated with the nylon bag technique, Johnson (1966) suggested the nylon bag technique would find better application in determining

the effects of various ration treatments on the rate of digestion within the rumen than in determining estimated digestion coefficients.

The use of the nylon bag to demonstrate inhibition of cellulose digestion was used by el-Shazly, Dehority and Johnson (1961) by placing shredded filter paper in nylon bags and determining cellulose disappearance when the corn level in hay and corn diets were changed. Lambert and Jacobson (1958) used nylon bags containing cellulose to measure the inhibition of cellulose digestion caused by chlorotetracycline in rations fed to cattle.

## CHAPTER III

## MATERIALS AND METHODS

#### Introduction

Five trials were conducted to evaluate the nutritive value of acid hydrolyzed wood residue. Two degrees of dilute sulfuric acid hydrolysis were evaluated. Low acid hydrolyzed wood residue (LA-HWR) was produced by treating the wood material with 0.8%  $\mathrm{H}_2\mathrm{SO}_A$  (dry wood basis) for 60 seconds at 4.2185 x 10<sup>4</sup> g/cm<sup>2</sup> pressure. High acid hydrolyzed wood residue (HA-HWR) was produced by treating the wood material with 2.3%  ${\rm H_2SO}_{\Lambda}$  (dry wood basis) for 40 seconds at  $4.2185 \times 10^4$  g/cm<sup>2</sup> pressure. No products of the acid hydrolysis were removed during the processing of the wood residue. After hydrolysis both treatments were neutralized with anhydrous ammonia to a pH of approximately 6.5. The addition of ammonia gave the resulting product a crude protein value of 15%. The dry matter contents of the LA-HWR and HA-HWR were 60% and 50%, respectively. The wood residue used in both acid treatments consisted approximately of 80% hardwood and 20% pine material.

Trial l was a growth, feed efficiency, and digestibility trial in which give rations containing LA-HWR were fed to evaluate the performance of growing lambs fed high levels of hydrolyzed wood residue. These rations were compared to a basal control ration composed of alfalfa meal. Rations containing 25, 50, and 75% LA-HWR were balanced with soybean meal to be isonitrogenous with the alfalfa meal. Two rations containing 50 and 75% LA-HWR were formulated with no additional nitrogen supplement.

Trial 2 was used to determine if the level of LA-HWR used in the diet influenced the ability of rumen microorganisms to digest cellulose. The six rations used in
trial 1 were used in this study. Dacron bags (suspended in the rumen) containing soybean hulls were used to detect differences in cellulose digestion caused by the treatments applied. Differences in rumen volatile fatty acid
(VFA) concentrations were also measured.

Trial 3 was a growth, feed efficiency and digestibility trial, similar to trial 1, using HA-HWR. Due to severe depression in palability caused by the high acid hydrolyzed wood residue, HA-HWR was incorporated into the basal diet at 20 and 35% with added soybean meal and at 20 and 35% with no additional nitrogen supplement. A basal control ration of alfalfa meal was also fed in this trial.

Trial 4 was designed to study differences in the cellulose digestion using the dacron bag technique when HA-HWR was incorporated into the diet at 0, 25, and 35%. Ruminal ammonia-nitrogen and volatile fatty acid concentrations

#### were also measured.

Trial 5 was used to evaluate the total concentration of volatile fatty acids and ammonia-nitrogen present in the rumen of steers consuming rations containing 25 and 50% HA-HWR as compared to an alfalfa meal diet.

## Experimental Procedure

## Trial 1

Thirty-four wether lambs (Hampshire x Western and Dorset) weighing approximately 20 kg were randomly placed in individual pens (140 cms x 84 cms) with slotted floors. The lambs were maintained on a high roughage ration until allotment to treatment.

Feed and water were withheld for 16 hours prior to taking the initial weight of the lambs. On the basis of the initial weight, the lambs were divided into three weight groups (heavy, medium and light). The lambs were then randomly allotted to 6 ration treatments (Table I). Two lambs from each weight group were allotted to each ration treatment (6 lambs per treatment) with the exception of rations 1 and 5 which were allotted 5 lambs per treatment.

The lambs were allowed to eat <u>ad libitum</u>. Refused feed was weighed and recorded to determine daily feed consumption. Free access to water was allowed at all times. After 89 days on their assigned treatments the lambs were

TABLE I

COMPOSITION OF EXPERIMENTAL RATIONS USED IN TRIALS 1 AND 2

DRY MATTER BASIS

**************************************	Ration							
	1	2	3	4	5	<u>6</u> %		
Alfalfa meal, 17%	% 99 <b>.</b> 5	% 70 <b>.</b> 60	% 40.5	% 11.1	% 48.7	% 23.0		
LA-HWR	-	25.0	50.0	75.0	50.0	75.0		
Soybean meal, 44%	-	3.9	8.2	12.4	-	-		
T.M. salt	0.5	0.5	0.5	0.5	0.5	0.5		
Dicalcium phosphate	-	-	0.8	1.0	0.8	1.5		
Vit. D supp.	+	+	+	+	+	+		

withheld from feed and water for 16 hours and weighed.

The effects of the treatment imposed were measured by weight gain and feed efficiency data. In addition, the amount of non-wood dry matter per kg of gain was calculated to determine if LA-HWR was replacing any of the basal feed required to support gain.

All lambs were used concurrently in a digestion study. The thirty-four lambs were fitted with canvas collection bags and total collections of feces were made over seven day periods starting on days 14 and 49 of the feeding period.

All feed used in trials 1 and 2, and all subsequent trials were mixed in 100 kg batches. Feed and feces samples were chemically analyzed for dry matter, organic matter, cellulose, and nitrogen. Digestion coefficients of the rations were calculated for the respective fractions. Digestion coefficients of the LA-HWR were calculated by regression of the ration digestion coefficients.

## Trial 2

Twelve lambs fitted with permanent rumen cannulas were randomly allotted to the 6 LA-HWR treatments used in trial 1 (Table I) using two lambs per treatment. All lambs were kept in individual pens and allowed to eat ad libitum with free access to water.

Dacron bags, similar to those used by Hopson (1963), measuring 5 cms  $\times$  10 cms were constructed from lining

material with a thread count of  $40 \times 50$  threads per cm<sup>2</sup>. The bags were then tied securely with 38 cm lengths of nylon cord, which were used to attach them to the cannula.

On days 14, 35, and 56, six dacron bags, each containing 1 gram of soybean hulls, were placed in the rumen of each lamb via the rumen cannula, 4 hours post feeding. Two bags were removed from each lamb at 15, 30 and 48 hours after being placed in the rumen.

After the nylon bags were removed from the rumen, they were washed of adhering rumen contents and frozen for later analysis for cellulose.

Samples of rumen contents were taken before feeding and at 1, 2 and 4 hours post feeding on days 39 and 60. Rumen ingesta was obtained by opening the rumen cannula and allowing approximately 300 ml to flow freely into a flask.

## Trial 3

Thirty wether lambs (Dorset x Rambouillet) weighing approximately 24 kg were allotted to pens similar to those used in trial 1. After the initial weighing and allotment to three weight groups (heavy, medium and light), two lambs from each group were randomly allotted to 5 treatments (Table II) with rations containing high acid hydrolyzed wood residue (HA-HWR).

The lambs were maintained on their assigned treatment for 90 days and were then withheld from feed and water for

TABLE II

COMPOSITION OF EXPERIMENTAL RATIONS USED IN TRIALS 3 AND 4

DRY MATTER BASIS

		Ration		
1	2	3	4	5
% 99 <b>.</b> 5	% 72 <b>.</b> 85	% 52 <b>.</b> 79	% 79 <b>.</b> 50	% 64.00
-	20.00	35.00	20.00	35.00
-	6.24	11.51	-	-
0.5	0.5	0.5	0.5	0.5
-	c	0.2	-	0.5
0.5	0.5	0.5	0.5	0.5
	99.5	7 7 7 7 7 7 99.5 72.85 - 20.00 - 6.24 0.5 0.5	1     2     3       %     %     %       99.5     72.85     52.79       -     20.00     35.00       -     6.24     11.51       0.5     0.5     0.5       -     0.2	%       %       %       %       %       79.50         -       20.00       35.00       20.00         -       6.24       11.51       -         0.5       0.5       0.5       0.5         -       0.2       -

16 hours and weighed.

The parameters used to evaluate growth and efficiency of feed utilization were total weight gain, average daily gain and dry matter per kg of gain. Non-wood dry matter per kg of gain was calculated to determine the amount of basal feed replaced by feeding different levels of HA-HWR.

A digestion trial similar to that conducted in trial 1 was conducted concurrently with the thirty lambs in trial 3, starting on day 14 and day 49 of the growth trial.

## Trial 4

Twelve lambs (of similar weight and breeding as those used in trial 3) fitted with permanent rumen cannulae were randomly allotted to treatments 1, 2 and 3 (Table II) of the treatments used in trial 3 with HA-HWR. Four lambs were allotted per treatment and randomly allotted to individual pens.

Dacron bags similar to those used in trial 2 with LA-HWR were filled with 1 gram of soybean hulls. Six bags were tied to a weighted and sealed 12 cm length of tygon tubing. A 38 cm length of nylon cord was attached to one end of the tubing and connected to the cannula.

On days 14, 35 and 56 the weighted bars and six attached bags were placed in the rumen of each lamb via the rumen cannula at 4 hours post feeding. As in trial 2, two bags each were removed at 15, 30 and 48 hours after being placed in the rumen and were washed and frozen as described

## for trial 2.

Rumen content samples were taken before feeding and at 1, 2, 4 and 8 hours post-feeding on days 18, 39 and 60 for VFA and rumen ammonia analysis.

## Trial 5

Three mature hereford steers fitted with rumen cannulas were used in a 3 x 3 latin square design using rations containing 0, 25 and 50% HA-HWR (Table III). A 21 day adaptation period was allowed between each ration change. On days 21 and 25 of each period, rumen liquor samples were taken before feeding and at 1, 2, 4 and 8 hours post-feeding. Fifty grams of polyethylene glycol (PEG), M.W. 4,000, dissolved in 100 ml of water was administered via the rumen cannula after the first rumen fluid sample was taken each day. Rumen fluid samples were analyzed for VFA concentration, rumen ammonia-nitrogen and PEG. PEG was used to measure rumen volume by regressing the values obtained to zero time. By calculating the rumen volume the total concentrations of VFA and ammonia were obtained for each steer on each ration.

## Preparation of Samples and Chemical Analysis

Feces collected during the digestion trials were weighed daily and 10% aliquots were composited and stored at  $-4^{\circ}$ C. until analyzed. Wet fecal samples were used for nitrogen analysis. Eighty grams of wet feces were

TABLE III

COMPOSITION OF EXPERIMENTAL
RATIONS USED IN TRIAL 5
DRY MATTER BASIS

	Ration					
	1	2	3			
	%	%	%			
Alfalfa meal	99.5	74.5	49.0			
HA-HWR	0.0	25.0	50.0			
T.M. salt	0.5	0.5	0.5			
Dicalcium phosphate	0.0	0.0	0.0			

mixed with 400 ml of distilled water in a Waring blender. An aliquot of this was analyzed for nitrogen by the Kjeldahl procedure. The remaining fecal sample was than dried at 60°C. in a forced air oven for 24 hours, allowed to equilbrate to atmospheric conditions for 24 hours, then ground through a 1 mm screen in a Wiley mill. The air dried samples were stored in sealed containers. Final dry matter and ash were determined by standard A.O.A.C. (1960) methods. Cellulose was determined by procedures described by Crampton and Maynard (1938).

Soybean hulls (49.10% cellulose) were used as a source of cellulose in the dacron bags because they contain a highly digestible form of cellulose. The particle size of the soybean hulls prevents undigested material from escaping through the pores of the bag. The soybean hulls remaining in the dacron bag after digestion in the rumen were removed by washing into a centrifuge tube. The residue remaining after removal of excess water was subjected to cellulose analysis.

Samples of rumen fluid obtained for ammonia-nitrogen, VFA and PEG analysis were strained through four layers of cheesecloth and mixed with mercuric chloride (HgCl<sub>2</sub>). Volatile fatty acid analysis was completed by the procedure of Erwin et al. (1961) with a Bendix Series 2,500 Gas Chromatograph. Column length was 183.0 cm with an

<sup>&</sup>lt;sup>1</sup>The Bendix Corporation, Ronceverte, W. Va.

inside diameter of 2 mm. The column packing material used was 10% SP 1,200 on Chromsorb W, acid washed, 80/100 mesh.<sup>2</sup> Nitrogen, carrier gas, flow was maintained at 60 cc/min. and hydrogen flow at 40 cc/min. Air flow was regulated to a flow rate of 1.6 cc/min. Column temperature was maintained at 120°C. Calculation of VFA data was by the rectangular method suggested by Carroll (1961). Rumen ammonian nitrogen was measured by procedures recommended for the Oxford<sup>3</sup> ammonianitrogen analysis. Polyethylene glycol (PEG) analysis was conducted according to the Hyden procedures as modified by Davis<sup>4</sup> for polyethylene glycol determination in rumen liquor samples.

# Statistical Analysis

All statistical analyses were made by a computer system entitled "Statistical Analysis System" developed by Anthony J. Barr and James Howard Goodnight of the Department of Statistics, North Carolina State University, Raleigh, North Carolina.

The analysis of variance suboption was used to test for differences in response due to ration treatments.

Differences between individual rations were tested by the

<sup>&</sup>lt;sup>2</sup>Supelco, Inc., Bellefonte, Pa.

<sup>&</sup>lt;sup>3</sup>Oxford Laboratories, San Mateo, Calif.

 $<sup>^{4}\</sup>text{C.L.}$  Davis, University of Illinois, personal communication.

use of the least significant difference test. (Steel and Torrie, 1960)

Growth and feed efficiency parameters used in trials 1 and 3 were analyzed as a randomized block design. The digestion studies in trials 1 and 3 were analyzed as a split split plot with rations as main plots and weight groups and periods within the trial as subplots. Trials 2 and 4 were analyzed as a split plot with rations as main plots and periods as subplots. The fifth trial was a split plot with the main plot a latin square and days within each cell of the latin square the subplot.

#### CHAPTER IV

## RESULTS AND DISCUSSION

## Trial 1

The performance of the lambs in the growth study with low acid hydrolyzed wood residue (LA-HWR) is shown in Table IV and Figures 1, 2 and 3. Sources of variation and degrees of freedom associated with the analysis of variance are given in Appendix Table XV. The lambs consuming 25% LA-HWR with supplemental soybean meal (SBM) gained significantly faster (P < .05) than lambs consuming all other rations except the basal alfalfa ration. Weight gain was decreased, in relation to the basal ration, when LA-HWR comprised greater than 25% of the ration fed. This decrease in weight gain was significant (P < .05) when LA-HWR was incorporated into the ration in amounts greater than 50%, with or without SBM supplementation.

Lambs fed the rations containing LA-HWR consumed more dry matter daily than those fed the basal ration, but these differences were not significantly different (P < .05). The highest average daily dry matter intake was 1.81 kg for lambs consuming 25% LA-HWR with SBM, and the lowest was 1.42 kg for the basal ration.

TABLE IV

THE EFFECT ON GROWTH AND FEED EFFICIENCY OF ADDING LOW ACID HYDROLYZED WOOD RESIDUE TO LAMB RATIONS, WITH AND WITHOUT SOYBEAN MEAL SUPPLEMENTATION

Item			Rati	on <sup>2</sup>			s.E. <sup>1</sup>
	1	2	3	4	5	6	
Weight gain, kg	18.8 <sup>ab</sup>	20.3ª	16.5 <sup>bc</sup>	15.1°	15.7 <sup>bc</sup>	10.3 <sup>d</sup>	0.99
Average daily gain, kg/day	0.20 <sup>ab</sup>	0.23 <sup>a</sup>	0.19 <sup>bc</sup>	0.17 <sup>c</sup>	0.18 <sup>bc</sup>	0.12 <sup>d</sup>	0.01
Dry matter intake kg/kg gain	6.99 <sup>a</sup>	7.90 <sup>a</sup>	9.31 <sup>b</sup>	10.37 <sup>b</sup>	9.43 <sup>b</sup>	12.60 <sup>c</sup>	0.16
Average daily dry matter consumption, kg	1.42 <sup>a</sup>	1.81 <sup>a</sup>	1.72 <sup>a</sup>	1.76 <sup>a</sup>	1.64 <sup>a</sup>	1.45 <sup>a</sup>	0.11
Non-wood dry matter/kg gain, kg	6.99 <sup>a</sup>	5.93 <sup>a</sup>	4.66 <sup>b</sup>	2.59 <sup>c</sup>	4.72 <sup>b</sup>	3.15 <sup>c</sup>	0.08

<sup>&</sup>lt;sup>1</sup>Standard error

<sup>&</sup>lt;sup>2</sup>Ration: 1 = basal ration; 2 = 25% LA-HWR + SBM; 3 = 50% LA-HWR + SBM; 4 = 75% LA-HWR + SBM; 5 = 50% LA-HWR; 6 = 75% LA-HWR

 $<sup>^{</sup>m abcd}$  means on the same line with differing superscripts differ significantly (P .05)

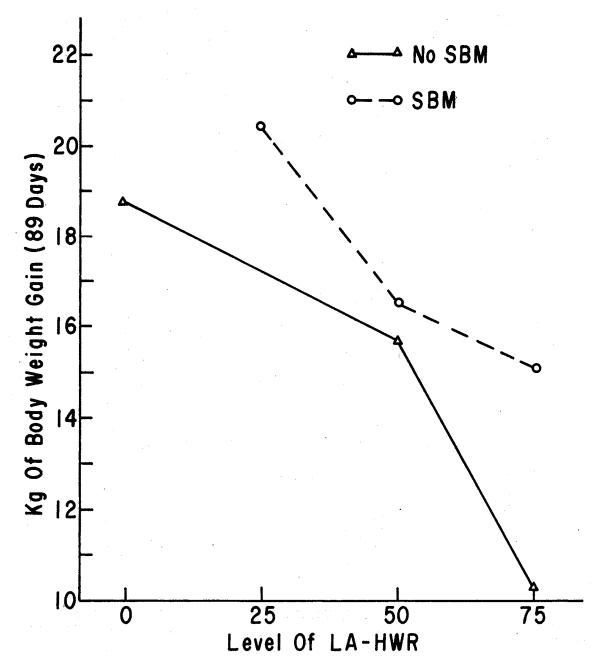


Figure 1. Body Weight Gain of Lambs Consuming LA-HWR With and Without SBM Supplement

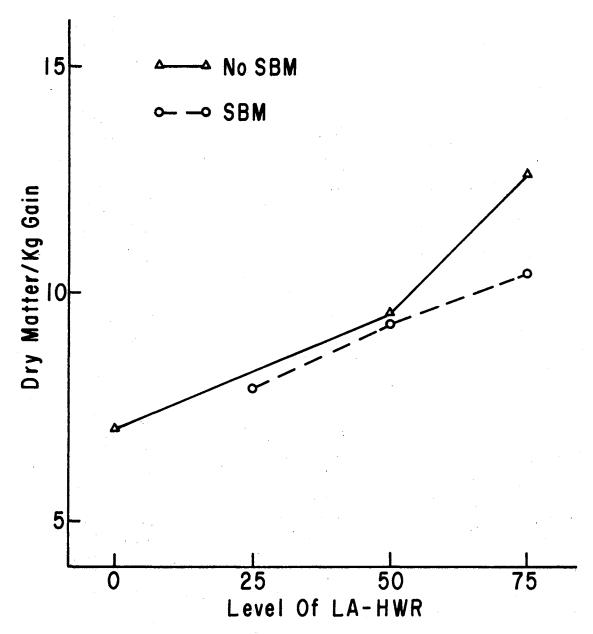


Figure 2. Dry Matter/kg of Gain Requirements of Lambs Consuming LA-HWR With and Without SBM Supplement

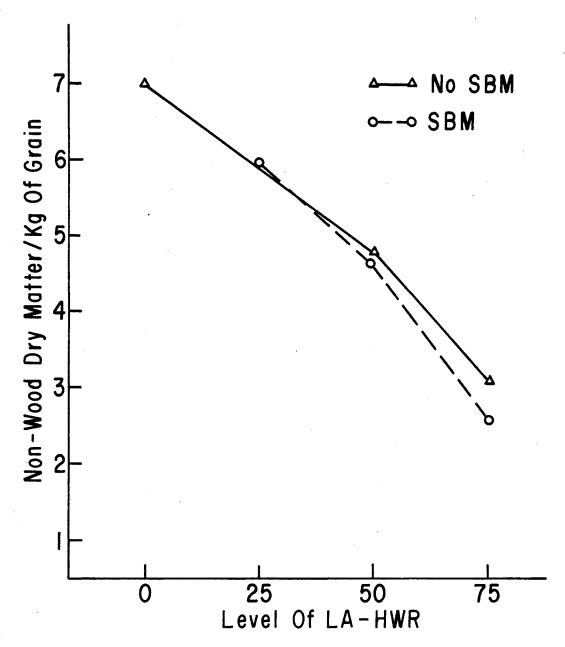


Figure 3. Non-wood Dry Matter/kg of Body Weight
Gain of Lambs Consuming LA-HWR With
and Without SBM Supplement

Less feed per kg of gain was required by lambs consuming the basal rations than by lambs consuming rations containing LA-HWR. The differences between lambs fed the basal ration and the lambs fed rations with 50 or 75% LA-HWR were significant (P < .05). Less non-wood dry matter per kg of gain was needed with increasing amounts of LA-HWR, (Figure 1). These data suggest; therefore, that LA-HWR was supporting maintenance and gain but with less efficiency than the ration containing only alfalfa meal.

The data from the lambs consuming rations containing 50 and 75% LA-HWR, with and without SBM, were analyzed using a 2 x 2 factorial to determine the effects of SBM supplementation to rations containing LA-HWR. An interaction approaching significance at the P<.05 level, showed that as the level of LA-HWR increased in the ration the addition of SBM increased body weight gain (Figure 2) and lowered dry matter per kg of gain requirements, when compared to rations containing the same levels of LA-HWR and no SBM.

Digestion coefficients determined using the rations and lambs in the growth study with LA-HWR are presented in Table V. Sources of variation and degrees of freedom associated with the analysis of variance are given in Appendix Table XVI. Periods 1 and 2 of this study refer to the two seven-day fecal collection trials starting on days 14 and 49, respectively. There was a significant period effect in the digestion of dry matter and organic matter.

TABLE V

DIGESTION COEFFICIENTS OF RATIONS CONTAINING
LOW ACID HYDROLYZED WOOD RESIDUE

Item	$Period^3$			Rat:	ion <sup>2</sup>			$s.e.^1$
		1	2	3	4	5	6	
Dry matter digestibility	1 2 Mean	55.8 59.4 57.6	48.7 53.7 51.2	44.6 45.6 45.1	36.9 40.7 38.8	37.6 40.0 38.8	36.4 37.2 36.8	1.6
Organic matter digestibility	1 2 Mean	55.0 60.4 57.8	47.4 54.2 50.8	44.1 46.0 45.1	36.7 40.2 38.4	37.2 39.5 38.3	35.9 37.0 36.4	1.6
Cellulose digestibility	1 2 Mean	38.3 36.9 37.6	30.1 47.2 38.6	30.4 40.7 35.6	31.0 48.6 39.8	32.4 35.6 34.0	36.3 43.4 39.9	2.7
Nitrogen digestibility	1 2 Mean	57.1 65.4 61.2	59.1 59.4 59.3	62.9 55.7 59.3	62.9 53.8 58.4	52.4 50.1 51.3	51.5 39.2 45.4	1.7

<sup>&</sup>lt;sup>1</sup>Standard error

<sup>&</sup>lt;sup>2</sup>Ration: 1 = basal ration; 2 = 25% LA-HWR + SBM; 3 = 50% LA-HWR + SBM; 4 = 75% LA-HWR + SBM; 5 = 50% LA-HWR; 6 = 75% LA-HWR

<sup>&</sup>lt;sup>3</sup>Period: Period 1 = days 14-21; Period 2 = days 49-56 of the growth trial abcd means on the same line with differing superscripts differ significantly (P<.05)

However, since there was no significant period x ration interaction, ration effects will be discussed across periods. The dry matter and organic matter digestibilities of the basal ration were significantly higher (P < .05) than the rations containing LA-HWR. The addition of increasing amounts of LA-HWR significantly lowered the dry matter digestibility of the rations with each 25% increase in LA-HWR (Figure 4). Extrapolation of the regression line calculated from the regression of the dry matter digestibilities of rations containing 25, 50 and 75% LA-HWR was used to estimate the dry matter digestibility at 100% LA-HWR in a (The dry matter digestibility of LA-HWR obtained ration. by this procedure was approximately 32%). This value is in agreement with the work of Archibald (1926) using Douglas fir with a similar acid hydrolysis. It is important in making this comparison to point out the improvement of wood digestibility by chemical treatment is very species dependent.

There were no statistically significant differences between rations in the digestibility of cellulose. A general trend for all rations containing LA-HWR to improve in cellulose digestibility during the second period suggested that some adaptation of cellulolytic microorganisms have occurred within the rumen. This improvement was not consistant between rations and resulted in significant period and ration x period effect. The rations showing the most improvement in cellulose digestibility from period

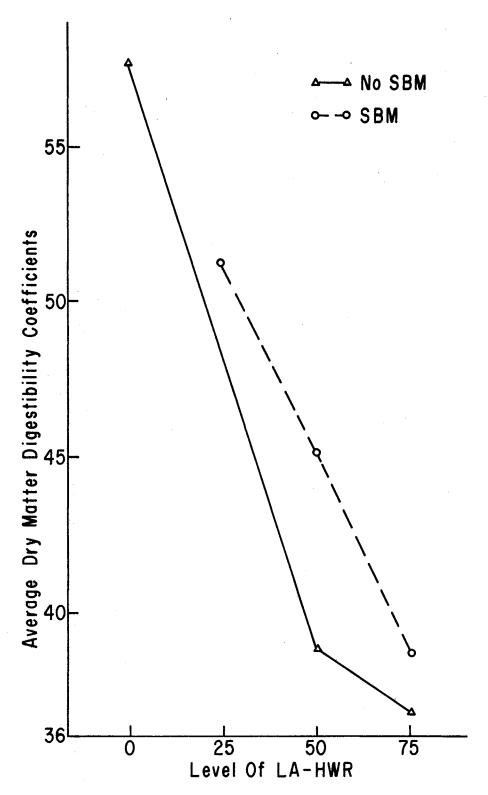


Figure 4. Dry Matter Digestion Coefficients of Rations Containing LA-HWR

1 to period 2 as a percent of period 1 (Figure 5) were those containing SBM and LA-HWR. Improvements of 57%, 34% and 57% in cellulose digestion were observed for 25, 50 and 75% LA-HWR, respectively.

Nitrogen digestion coefficients are presented in Table V. Due to significant (P < .01) interactions for weight group x ration and ration x period (A.O.V. in Appendix Table XVI), main effects of rations on nitrogen digestibility are difficult to interpret. Percent changes in nitrogen digestibility are presented in Figure 6. The basal ration showed an increased nitrogen digestibility from period 1 to period 2 (14.46% as a percent of period 1). Rations containing LA-HWR with and without SBM showed marked declines in nitrogen digestibility from period 1 to period 2. Hudson (1971) reported that hydrolyzed pine residue substituted for 15% of an alfalfa meal diet decreased the crude protein digestibility 5.8%. Supplementation with protein and alkaline materials tended to alleviate the depression in crude protein digestibility. The results of period l indicate that addition of SBM was effective in preventing depression of crude protein digestibility in rations containing LA-HWR. The reason for the decline in nitrogen digestibility in the second period with rations containing 50 and 75% LA-HWR even with SBM is not apparent.

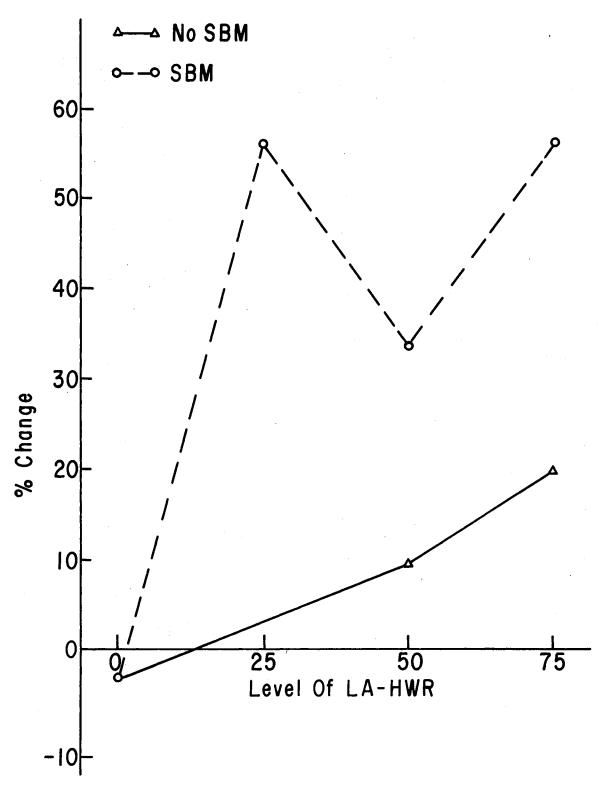


Figure 5. Percent Change in Cellulose Digestibility from Period 1 to Period 2 of Rations Containing LA-HWR

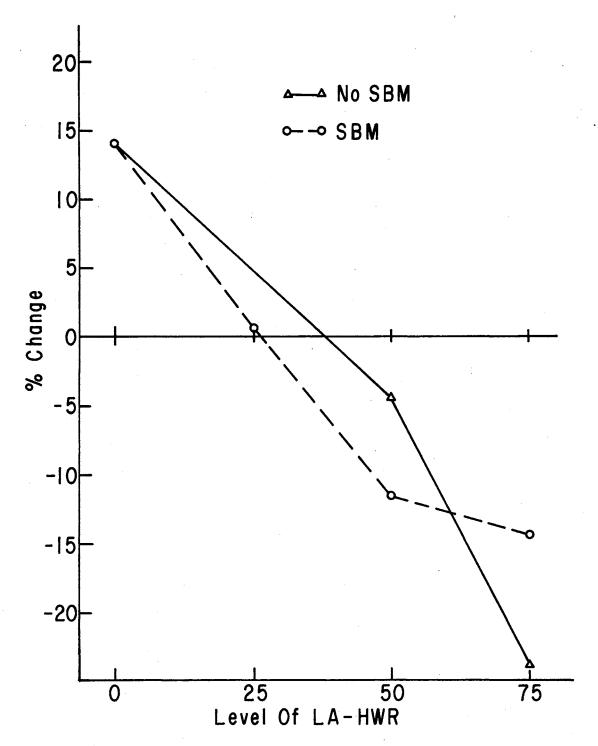


Figure 6. Percent Change in Nitrogen Digestibility from Period 1 to Period 2 of Rations Containing LA-HWR

## Trial 2

The results of the dacron bag study with the rations containing LA-HWR are presented in Table VI. Periods 1, 2 and 3 correspond to the 4 days following the placement of the dacron bags in the rumen of the lambs on days 14, 35 and 56, respectively. Sources of variation and degrees of freedom for trial 2 are presented in the Appendix Table XVII. The mean digestibilities appeared to be lower for rations containing LA-HWR and SBM when compared with the basal ration. However, there were no statistically significant ration effects on cellulose digestion at the three time intervals the bags were removed from the rumen (15, 30 and 48 hours after being placed in the rumen). Period effects were significant (P < .05) across all times and ration x period effects were significant (P<.05) at 15 hours. There was a large amount of variation in the data as shown by the standard errors.

Attempts at making inferences about ration effects on cellulose digestion at 15 hours were futile because of the large animal variation within rations and the unexplainable differences between periods.

Cellulose digestion at 30 and 48 hours, although not significant, was generally the highest with lambs consuming the basal ration (73.5 and 92.2% at 30 and 48 hours, respectively.

Rations containing 50 and 75% LA-HWR with SBM

TABLE VI

DIGESTION OF CELLULOSE IN DACRON BAGS SUSPENDED IN THE RUMEN OF LAMBS CONSUMING RATIONS CONTAINING LOW ACID HYDROLYZED WOOD RESIDUE

Time <sup>4</sup>	$\mathtt{Period}^3$			Ra	tion <sup>2</sup>			Period	s.E. <sup>1</sup>
		1	2	3	4	5	6	Mean	·
15 hours	1	31.9	17.9	33.9	17.1	46.3	22.0	28.2	
•	2	30.2	9.0	12.4	16.6	35.5	17.4	20.2	
	3	42.2	43.5	44.2	18.6	29.5	39.2	36.2	
	Mean	34.8	23.5	30.2	17.4	37.1	38.6		13.4
30 hours	1	85.4	64.3	64.4	45.0	71.4	48.3	63.2	
	2	60.3	34.7	29.0	32.2	62.7	28.8	48.2	
	3	74.8	78.6	73.0	26.5	52.8	69.9	62.6	
	Mean	73.5	59.2	55.5	34.6	62.3	49.0		19.5
48 hours	1	97.0	92.1	87.1	70.6	86.3	66.3	83.2	
	2	86.1	81.2	55.2	60.8	93.2	52.2	71.5	
	3	93.5	90.2	90.2	49.9	79.6	88.9	82.1	
	Mean	92.2	87.8	77.5	60.4	86.4	69.1		14.8

<sup>&</sup>lt;sup>1</sup>Standard error

<sup>&</sup>lt;sup>2</sup>Rations: 1 = basal ration; 2 = 25% LA-HWR + SBM; 3 = 50% LA-HWR + SBM; 4 = 75% LA-HWR + SBM; 5 = 50% LA-HWR; 6 = 75% LA-HWR

 $<sup>^3</sup>$ Period: Period 1 - days 14-18; Period 2 - days 35-39; Period 3 - days 56-60 of trial 2

<sup>4</sup>Length of time after being placed in the rumen that dacron bags were removed

supplement gave consistantly lower cellulose digestion values than similar rations without SBM. A decrease in in vivo cellulose digestion has usually resulted when a source of readily available carbohydrates is added to high fiber rations (Chappel and Fontennot, 1965) and this may partly explain this observation.

Rumen fluid volatile fatty acid concentrations and molar percent data are presented in Table VII. Significant period effects similar to those encountered with the dacron bag study makes the interpretation of these data difficult.

Acetic, butyric and total volatile fatty acid concentrations were significantly higher prior to feeding (0 hour sample) in rumen contents from lambs fed rations 1 and 2 than in contents from lambs fed rations containing 50 and 75% LA-HWR. There were no significant differences in volatile fatty acid concentration due to rations at sample times of 1, 2 and 4 hours post-feeding.

Rumen contents of the lambs fed the basal ration were significantly higher (P<.05) in molar percent acetic and less in molar percent propionic at the zero hour sampling than the rumen contents of lambs fed rations containing 50 and 75% LA-HWR without SBM.

## Trial 3

The results of the growth trial using high acid hydrolyzed wood residue (HA-HWR) are presented in Table VIII

TABLE VII

RUMEN FLUID VOLATILE FATTY ACIDS FOR TRIAL 2

1	2	Acet	ic	Propi	onic	Butyr	ic	<del></del> <del></del>
Ration	Time	umoles/	mole %	umoles/ ml	mole %	umoles/ ml		Total VFA umoles/ml
1	0	34.9	68.7	9.2	15.47	5.0	10.7	51.4
	1	49.9	62.6	15.4	18.7	8.9	12.9	77.8
	2	67.3	70.2	20.8	21.2	7.0	7.4	96.3
	4	40.9	61.1	12.7	21.3	8.0	13.8	63.3
2	0	39.2	66.5	12.0	20.2	6.0	10.5	58.7
	1	60.9	71.2	17.8	20.8	6.3	7.0	85.9
	2	52.7	69.5	15.7	21.2	6.6	8.5	75.7
	4	35.4	69.1	10.7	20.3	4.9	9.4	51.6
3	0	23.2	64.5	9.3	25.5	2.4	6.1	36.4
	1	53.7	72.0	16.4	21.8	3.9	4.8	75.1
	2	51.6	71.1	15.9	21.9	4.6	6.0	72.9
	4	42.5	69.3	13.5	23.1	4.3	6.8	60.8
4	0	12.0	59.1	5.9	30.7	1.3	6.1	20.0
	1	53.4	75.2	12.2	17.2	4.0	6.0	70.8
	2	38.5	70.8	11.9	20.6	3.7	6.5	55.0
	4	35.0	69.1	10.3	21.8	3.8	7.5	49.8

TABLE VII (Continued)

1	2	Acet	ic	Pro	pionic	Butyr	ic	
Ration	Time	umoles/		umole	-	umoles/		Total VFA
		ml_	%	m1	%	m1	%	umoles/ml
5	0	16.4	57.5	8.7	31.5	1.6	6.2	28.1
	1	51.0	67.6	17.4	23.2	4.8	6.5	75.0
	2 .	39.7	64.3	14.8	23.6	5.7	9.4	61.9
	4	36.1	68.6	11.9	22.7	4.0	7.5	52.7
6	0	12.3	54.1	8.7	36.7	1.3	5.7	23.1
	1	57.6	75.5	15.8	19.8	2.8	3.5	77.07
	2	49.4	70.8	15.2	22.6	3.7	5.3	69.2
	. 4	31.2	67.7	12.5	24.1	3.3	6.7	47.8

 $<sup>^{1}\</sup>text{Ration: } 1 = \text{basal ration; } 2 = 25\% \text{ LA-HWR} + \text{SBM; } 3 = 50\% \text{ LA-HWR} + \text{SBM; } 4 = 75\% \text{ LA-HWR} + \text{SBM; } 5 = 50\% \text{ LA-HWR; } 6 = 75\% \text{ LA-HWR}$ 

 $<sup>^{2}\</sup>mathrm{Hours}$  post feeding that rumen contents samples were taken

and Figures 7, 8 and 9. Sources of variation and degrees of freedom are presented in Appendix Table XVIII. Lambs consuming the basal ration gained significantly more weight (P < .05) and consumed less dry matter per kg of gain than those lambs fed rations containing HA-HWR at 20 and 35% of the ration either with or without SBM supplement. There were no significant differences in average daily dry matter comsumption at the levels fed in this trial but palatability problems prevented the incorporation of HA-HWR at higher levels than 35% of the ration.

There were no significant differences in non-wood dry matter per kg of gain between lambs fed the basal ration and those fed rations containing HA-HWR. The basal ration required 8.17 and 7.50 kg at 20 and 35% HA-HWR with SBM, respectively. Lambs consuming rations of 20 and 35% HA-HWR without SBM required 8.82 and 7.99 kg of non-wood dry matter per kg of gain, respectively. These data suggest that HA-HWR was very poorly utilized as an energy source in the diets of growing lambs. The hydrolysis with 2.3%  ${
m H}_2{
m SO}_4$ (dry weight of the wood basis) may have resulted in the formation of non-digestible carbohydrate degradation pro-Examination of the HA-HWR revealed that the apparent ducts. lignin content was increased by hydrolysis from 21% in the raw wood to 60% in the HA-HWR. This observation indicates that a large fraction of the HA-HWR was probably indigestible.

TABLE VIII

THE EFFECT ON GROWTH AND FEED EFFICIENCY OF ADDING HIGH ACID HYDROLYZED WOOD RESIDUE TO LAMB RATIONS, WITH AND WITHOUT SOYBEAN MEAL SUPPLEMENTATION

Item			Ration <sup>2</sup>			S.E. <sup>1</sup>
	1	_2_	3	4	5 .	
Weight gain, kg	15.2 <sup>a</sup>	12.2 <sup>b</sup>	11.3 <sup>b</sup>	11.4 <sup>b</sup>	10.1 <sup>b</sup>	.81
Average daily gain, kg/day	.17 <sup>a</sup>	.14 <sup>b</sup>	.13 <sup>b</sup>	.13 <sup>b</sup>	.11 <sup>b</sup>	.01
Dry matter intake kg/kg gain	7.97 <sup>a</sup>	10.21 <sup>b</sup>	11.50 <sup>bc</sup>	11.04 <sup>bc</sup>	12.16 <sup>c</sup>	.26
Average daily dry matter consumption, kg	1.34 <sup>a</sup>	1.34 <sup>a</sup>	1.40 <sup>a</sup>	1.36 <sup>a</sup>	1.38 <sup>a</sup>	.06
Non-wood dry matter/kg gain, kg	7.97 <sup>a</sup>	8.17 <sup>a</sup>	7.50 <sup>a</sup>	8.82 <sup>a</sup>	7.99 <sup>a</sup>	.20

Standard error

<sup>&</sup>lt;sup>2</sup>Ration: 1 = basal alfalfa meal; 2 = 20% HA-HWR + SBM; 3 = 35% HA-HWR + SBM; 4 = 20% HA-HWR; 5 = 35% HA-HWR

 $<sup>^{\</sup>rm abcd}_{\rm means}$  on the same line with differing superscripts differ significantly (P < .05)

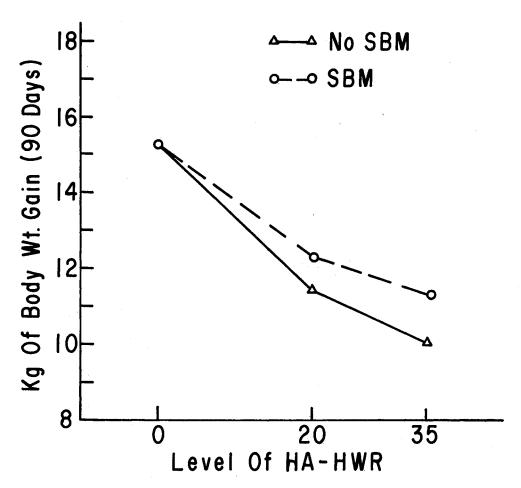


Figure 7. Body Weight Gain of Lambs Consuming HA-HWR With and Without SBM Supplement

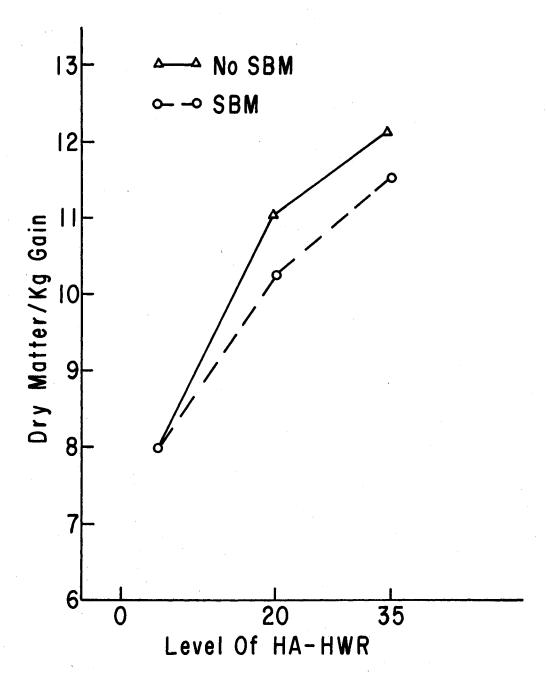


Figure 8. Dry Matter/kg of Gain Requirements of Lambs Consuming HA-HWR With and Without SBM Supplement

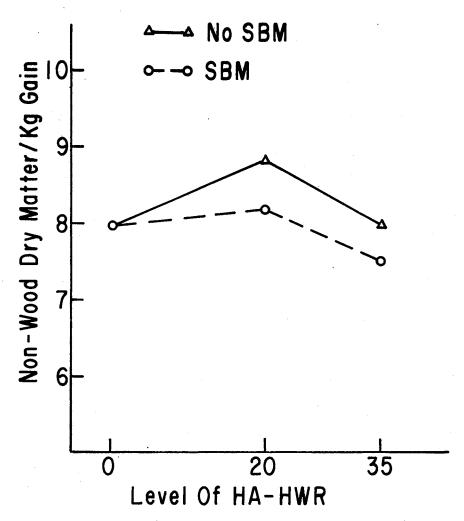


Figure 9. Non-wood Dry Matter/kg of Body
Weight Gain of Lambs Consuming
HA-HWR With and Without SBM
Supplement

Digestion coefficients for dry matter, organic matter, cellulose and nitrogen for the rations and lambs used in the growth study with HA-HWR are presented in Table IX.

Dry matter and organic digestibilities of rations were decreased when HA-HWR was incorporated into the rations, in comparison to the basal ration. These decreases in digestibility were significant for all rations containing HA-HWR, except when HA-HWR was incorporated into the ration at 20% with SBM supplement.

Figure 11 presents the change in cellulose digestibilities from period 1 to period 2 as a percent of period 1. Rations containing HA-HWR and SBM showed larger percent increases in the digestibility of cellulose than the basal ration or rations containing HA-HWR and no SBM.

Cellulose digestibility was significantly lower for the ration containing 35% HA-HWR and SBM (ration 3) when compared to all other rations. With the exception of ration 3, no significant difference existed between the other rations, although cellulose digestibility of the rations tended to be decreased by the addition of HA-HWR.

Nitrogen digestibility was significantly depressed by the addition of 20 and 35% HA-HWR without SBM. Although the actual nitrogen digestion coefficients for the 20% and 35% HA-HWR rations with SBM tended to be lower than that of the basal ration, these differences were not statistically significant.

TABLE IX

DIGESTION COEFFICIENTS OF RATIONS CONTAINING
HIGH ACID HYDROLYZED WOOD RESIDUE

Item	$Period^3$		Ra	tion <sup>2</sup>			Period Mean	$s.e.^1$
·	· · · · · · · · · · · · · · · · · · ·	1	2	3	4	5		*
Dry matter	. 1	55.2	50.3	45.6	49.8	46.2	49.4	
digestibility	2	54.4	50.7	45.7	47.7	40.2.	47.7	
	Mean	54.8ª	50.7 50.5 <sup>ab</sup>	45.7 <sup>cd</sup>	47.7 48.8 <sup>bc</sup>	43.2 <sup>d</sup>	<i>,</i>	1.69
Organic matter	1	54.3	50.0	45.5	49.9	46.9	49.3	
digestibility	2	54.0	50.2.	44.7 cd	46.8 <sub>bc</sub>	39.1.	47.0	
	Mean	54.0 54.2	50.2 50.1	45.1 <sup>cd</sup>	48.4 <sup>DC</sup>	43.0 <sup>d</sup>		1.73
Cellulose	1	41.9	35.7	23.5	32.5	43.7	35.5	
digestibility	2	42.9	43.6	35.7.	45.8	41.1	41.8	
	Mean	42.9 42.4 <sup>a</sup>	39.7 <sup>a</sup>	35.7 <sub>b</sub>	39.2ª	42.2 <sup>a</sup>		1.76
Nitrogen	1	59.6	59.4	53.5	52.1	52.1	55.3	
digestibility	2	53.5	50.9	50.3.	46.8	39.8	48.3	
	Mean	56.6 <sup>a</sup>	55.2ª	50.3 51.9 <sup>ab</sup>	46.8 49.5 <sup>bc</sup>	46.0 <sup>c</sup>	51.8	1.73

<sup>&</sup>lt;sup>1</sup>Standard error

<sup>&</sup>lt;sup>2</sup>Ration: 1 = basal alfalfa meal; 2 = 20% HA-HWR + SBM; 3 = 35% HA-HWR + SBM: 4 = 20% HA-HWR; 5 = 35% HA-HWR

 $<sup>^{3}</sup>$ Period: Period 1 = days 14-21; Period 2 = days 49-56 of the growth trial with HA-HWR

means on the same line with differing superscripts differ significantly (P<.05)

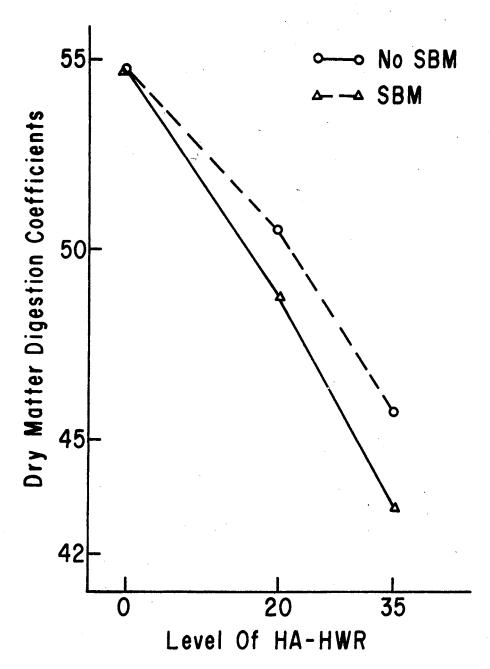


Figure 10. Dry Matter Digestion Coefficients of Rations Containing HA-HWR

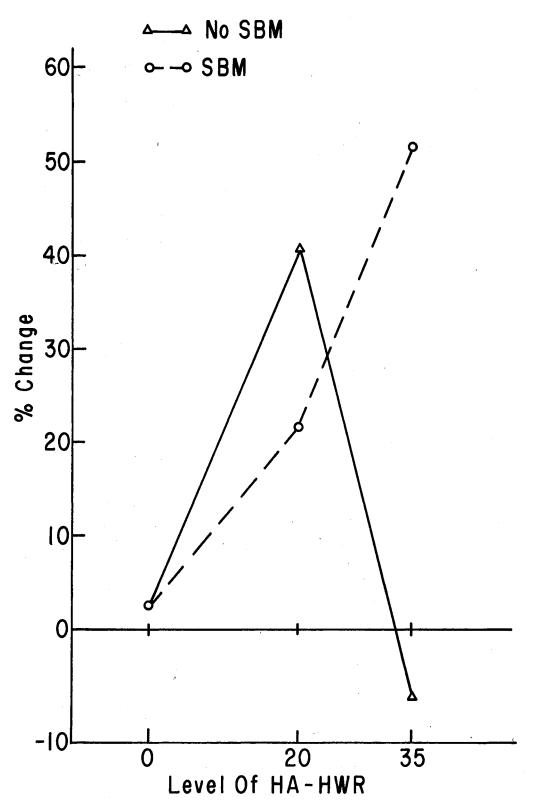


Figure 11. Percent Change in Cellulose Digestibility From Period 1 to Period 2 of Rations Containing HA-HWR

#### Trial 4

Cellulose digested in dacron bags suspended in the rumen of lambs consuming rations 1, 2 and 3 of trial 3 with HA-HWR are presented in Table X. Sources of variation and degrees of freedom for the analysis of variance of the data are presented in Appendix Table XX. There were no significant differences in the rate of cellulose digestion as influenced by the dietary regime of the lambs at 15 and 48 hours. Cellulose digested at 30 hours was significantly higher for lambs consuming the basal ration and the ration containing 20% HA-HWR compared to lambs fed the ration containing 35% HA-HWR and SBM. These data are in agreement with the results of trial 3 with respect to the depression of cellulose digestibility when 25% HA-HWR with SBM is fed to lambs. These data suggest that the combination of SBM and HA-HWR have an adverse effect on cellulose digesting microorganisms when they are fed in combination at the level used in this trial. Decreased cellulose utilization may be the result of the utilization of readily available carbohydrates contained in the HA-HWR and the SBM.

Rumen ammonia nitrogen expressed as ug/ml of rumen fluid at various times after feeding are presented in Table XI. Ammonia nitrogen was significantly higher in rumen contents of lambs consuming rations containing HA-HWR than in lambs consuming the basal ration at all sampling times except the 8 hour post-feeding sample. This indicates that

TABLE X

DIGESTION OF CELLULOSE IN DACRON BAGS SUSPENDED IN THE RUMEN OF LAMBS CONSUMING RATIONS CONTAINING HIGH ACID HYDROLYZED WOOD RESIDUE

Time <sup>4</sup>	Period <sup>3</sup>		Ration <sup>2</sup>		Period	-
		1	2	3	Mean	S.E.
		% Ce	llulose Dige	sted		
15 hours	1	19.7	18.6	11.8	16.7	
	2 3	27.6	24.8	17.5	23.3	
	3	24.1	40.4	27.9	30.8	
	Mean	23.8	27.9	19.0		7.0
30 hours	1	81.2	65.4	49.1	65.2	
		78.7	79.5	54.2	70.8	
	2 3	71.1	97.9	69.1	79.4	
	Mean	77.0 <sup>a</sup>	80.9 <sup>a</sup>	57.5 <sup>b</sup>		9.7
48 hours	1	95.4	90.5	78.6	88.2	
	2	94.8	97.4	81.9	91.4	
	2 3	93.4	97.9	92.9	94.8	
	Mean	94.5	95.3	84.5		9.0

<sup>&</sup>lt;sup>1</sup>Standard error

<sup>&</sup>lt;sup>2</sup>Ration: 1 = basal alfalfa meal; 2 = 20% HA-HWR + SBM; 3 = 35% HA-HWR + SBM

 $<sup>^3</sup>$ Period: 1 = Period 1 - days 14-18; Period 2 = days 35-39; Period 3 - days 56-60 of trial 4

<sup>&</sup>lt;sup>4</sup>Length of time after being placed in the rumen that bags were removed

TABLE XI RUMEN AMMONIA NITROGEN CONCENTRATION OF LAMBS CONSUMING HA-HWR IN TRIAL 4

Time	l ug/ml	Ration <sup>2</sup> 2 ug/ml	3 ug/ml
0	90.5	146.5	155.1
1	109.7	234.4	341.7
2	66.6	137.5	182.3
4	43.0	96.7	135.7
8	62.7	92.2	121.3

Ration: 1 = basal alfalfa meal 2 = 20% HA-HWR + SBM 3 = 35% HA-HWR + SBM

at least part of the ammonia used in the neutralization of the HA-HWR during production is rapidly liberated in the rumen shortly after feeding. Results of research with non-protein nitrogen sources indicate that when ammonia concentrations in the rumen fluid exceed a level of 50 mg/100 ml, the rate of absorption is so great that the liver cannot completely remove ammonia, the excess of which passes to the general circulation. The toxicity of ammonia involves, as a direct effect of the ammonium ion, a disturbance of the acid base status and changes in the electrolyte balance (Lewis, 1961). Rumen pH samples were not obtained in this trial. Levels of ammonia nitrogen reached 341.6 µg/ml (34 mg/100 ml) at 1 hour post-feeding when HA-HWR was incorporated into the diet at 35% of the ration. This level of rumen ammonia although not at the toxic level may have been influential in the alteration of microbial action in the rumen, resulting in lower cellulose digestion and decreased nitrogen 'utilization in rations containing HA-HWR.

Rumen volatile fatty acid concentrations for trial 4 are given in Table XII. There were no significant differences in total VFA concentrations; however, the rumen contents of lambs consuming the basal ration were consistently higher in volatile fatty acids across all sampling times.

TABLE XII

RUMEN VOLATILE FATTY ACID CONCENTRATION OF LAMBS CONSUMING HA-HWR

1	2	Acet	ic	Propi	onic	Butyr	ic	
Ration	Time <sup>2</sup>	umoļes/	mole	umoles/		umoles/	mole	Total VFA
<del> </del>		m1	%	ml_	%	<u>m1</u>	%	umoles/ml
1	0	21.8	74.4	4.8	15.8	2.1	6.7	29.6
<u>.</u>	1	63.6	76.5	13.5	16.5	4.9	5.9	82.8
•								
	2	56.7	74.3	13.4	18.3	4.7	6.4	75.4
	4	45.0	74.4	10.1	17.0	4.6	7.7	60.3
	8	30.5	75.9	6.4	15.0	2.8	6.8	40.5
2	0	17.9	72.7	12.7	12.7	1.8	7.1	25.0
	1	54.4	75.5	11.2	15.4	4.9	6.6	72.4
	2	45.7	75.2	9.0	15.4	4.4	7.0	60.6
	4	40.1	74.5	7.9	15.1	4.1	7.5	53.6
	8	29.9	75.9	5.5	13.4	3.2	7.1	39.8
3	0	20.8	70.6	4.8	15.3	2.3	7.7	29.8
•	1.	57 <b>.</b> 7	76.0	10.5	14.0	5.6	7.5	75.7
	2	46.2	76.4	7.9	14.0	4.4	7.4	59.6
	4	39.5	74.3	7.3	14.1	4.3	8.3	51.9
	8	27.5	73.3	5.4	15.2	2.8	7.5	36.5

 $<sup>^{1}</sup>$ Ration: 1 = basal alfalfa meal; 2 = 20% HA-HWR + SBM; 3 = 35% HA-HWR + SBM

<sup>&</sup>lt;sup>2</sup>Hours post feeding that rumen contents samples were taken

## Trial 5

The results of this trial are presented in Tables XIII and XIV. Sources of variation and degrees of freedom for the analysis of variance are presented in Appendix Table Three steers fed the basal diet and HA-HWR with no XXI. added SBM showed irregular response in rumen volume between sampling days within each period of the latin square. Consequently, there were no significant differences in volatile fatty acid concentrations and rumen ammonia data when the parameters were evaluated on total rumen concentrations. The rations were fed on constant dry matter intake basis. Rations containing HA-HWR were poorly consumed and therefore were administered to the steers via the rumen cannula to maintain the desired intake. Total VFA concentrations and rumen ammonia concentrations were higher in the rumen contents of steers consuming 50% HA-HWR than in the rumen contents of steers consuming the basal ration or the 25% HA-HWR ration.

TABLE XIII

TOTAL MOLES OF VOLATILE FATTY ACIDS PRESENT AT 0, 1, 2, 4 AND 8 HOURS POST FEEDING IN THE RUMEN OF STEERS CONSUMING HA-HWR

1	<b></b> 2	Acet	ic	Propi	onic	Buty	ric	Total	
Ration	Time <sup>2</sup>	Total	Mole	Total	Mole	Total	Mole	Moles	
		moles	%	moles	%	moles	%	VFA	
1	0	1.179	68.9	.332	19.4	.113	6.6	1.712	
	1	1.980	70.5	.588	20.9	.165	5.9	2.810	
	2	2.247	69.5	.704	21.8	.197	6.1	3.232	
	4	2.287	70.4	.681	21.0	.212	6.5	3.249	
	8	1.875	70.4	.534	20.0	.194	7.3	2.665	
2	0	1.086	70.8	.279	18.2	.103	6.7	1.534	
	1	1.667	69.5	.479	20.0	.179	7.5	2.399	
	2	1.857	67.2	.589	21.3	.231	8.4	2.764	
	4	1.981	68.5	.588	20.3	.243	8.4	2.892	
	8	1.482	69.5	.403	18.9	.171	8.0	2.135	
3	0	1.592	71.6	.369	16.6	.169	7.6	2.225	
	1	2.952	76.5	。532	13.8	. 265	6.9	3.857	
	2	3.204	73.0	.677	15.4	.371	8.4	4.392	
	4	3.251	72.8	.587	13.1	،465	10.4	4.468	
	8	2.894	67.6	.698	16.3	。507	11.8	4.284	

 $<sup>^{1}</sup>$ Ration: 1 = basal alfalfa meal; 2 = 25% LA-HWR; 3 = 50% HA-HWR

<sup>&</sup>lt;sup>2</sup>Hours post feeding that rumen samples were taken

Time		Ration <sup>2</sup>	
Time	1	2	3
. 0	4.529	3.189	5,258
1,	4.388	6.067	31.926
2	4.700	6.752	30.735
4	2.187	4.563	27.694
8	2.160	2.510	17.688

lation: l = basal alfalfa meal; 2 = 25% HA-HWR; 3 = 50% HA-HWR

# General Discussion

The results of trials 1 and 2 with LA-HWR indicate that it would be nutritionally feasible to use large quantities of wood residue in maintenance diets of ruminants. Results of the growth and feed efficiency study with LA-HWR indicate that it may be incorporated into the diet as high as 75% without palatability problems or toxic effects and still produce body weight gain in lambs. Digestion studies revealed that LA-HWR has a dry matter digestion coefficient of approximately 32%. This is in agreement with the dry matter digestion coefficient obtained by Archibald (1926) for Douglas fir (33% dry matter digestibility) hydrolyzed with 0.8  $\mathrm{H}_2\mathrm{SO}_4$  with 120 pounds of pressure for 20 minutes. Roughage materials with this degree of dry matter digestibility are comparable to the dry matter digestibility coefficient of many dormant range grasses, which serve as major energy sources in beef cow diets under range con-Therefore, it is a matter of economics on the production of LA-HWR as to whether this product can compete as an energy source in ruminant rations.

Examination of the LA-HWR indicated that the cellulose content was decreased from 57% in the raw wood to 48% in the LA-HWR, while the lignin content was raised from 21% in the raw wood to 29% in the LA-HWR. The increases in lignin content may be attributed to the formation of carbohydrate degradation products. Hardwoods may contain as

much as 20% pentosans, which are easily converted to furfural by dilute mineral acids. Hydroxymethlyfurfural may
be formed from the degradation of hexoses formed in the
hydrolysis of cellulose (Harris, 1952). Other wood constituents such as tannins and resins may be a part of this
indigestible artifact lignin.

Hudson (1971) stated that crude protein digestibility was decreased by the incorporation of 15% pine residue hydrolyzed under similar conditions used in this study with LA-HWR. It was thought that hydroxymethylfurfural (HMF) was responsible for the inhibition of nitrogen utilization. His results indicated that when (HMF) was added to the diet it had no significant effect on crude protein digestibility or animal performance. In further work with hydrolyzed pine residue he found that increasing the crude protein levels and adding alkaline material reduced the detrimental effect on crude protein digestibility. Hudson (1971) hypothesized that the high levels of tannins in wood may be responsible for the depression in crude protein utilization. Hatfield et al. (1969) and Peters et al. (1970) reported using approximately one percent tannins to coat protein to make it partially indigestible in the rumen.

The results of the first period of the digestion trials with LA-HWR substantiate the findings about the benefit from adding protein to the hydrolyzed wood residue rations. When SBM was added to LA-HWR rations, nitrogen digestibility was equal to or slightly better than the nitrogen

digestibility of the basal ration. Reduction of nitrogen digestibility during the second period (after 49 days on feed) may have been due to a build up in the level of tannins in the rumen or in the ration forming complexes with SBM protein and rendering it indigestible.

The results of trials 3, 4 and 5 indicate that the utilization of HA-HWR for an energy source in ruminant diets is not feasible. Palatability problems limit the incorporation of high percentages in the diet of ruminants. Trial 3 results showed that there was no significant difference in the amount of non-wood dry matter/kg of gain, indicating that the HA-HWR is probably non-digestible material.

Hydrolysis with 2.3% H<sub>2</sub>SO<sub>4</sub> reduced the cellulose content from 57% in the raw wood to 11% in the HA-HWR. However, the lignin content was raised from 21% in the raw wood to 60% in the HA-HWR. Hydrolysis under the conditions used to produce HA-HWR were optimum conditions for the production of furfural. In addition, it appears that large amounts of hydroxymethylfurfural and other artifact lignin may have been produced, reducing the digestibility and palatibility of high acid hydrolyzed wood residue.

<sup>&</sup>lt;sup>1</sup>H. Dale Turner, Dierks Division, Weyerhaeuser Co., 810 Whittington Ave., Hot Springs, Arkansas. Personal communication.

It is the author's opinion that varying degrees of acid hydrolysis, between the two methods described in this study, need to be tested and evaluated to determine the optimum point at which digestibility and growth responses are maximum.

#### CHAPTER V

### SUMMARY AND CONCLUSIONS

This study was conducted to evaluate the nutritive value of acid hydrolyzed wood residue (composed of 80% hardwood and 20% pine material) in ruminant rations. Two degrees of dilute sulfuric acid hydrolysis were evaluated. Low acid hydrolyzed wood residue (LA-HWR) was produced by treating the wood material with 0.8% H<sub>2</sub>SO<sub>4</sub> (dry wood basis) under pressure. High acid hydrolyzed wood residue (HA-HWR) was produced by treatment with 2.3% H<sub>2</sub>SO<sub>4</sub> and pressure. After hydrolysis the products were neutralized to a pH of 6.5 with anhydrous ammonia. The products were mixed in varying proportions with a basal ration of alfalfa meal and fed to lambs in growth and digestion trials. Rations with hydrolyzed wood were tested with and without addition of soybean meal to make them isonitrogenous with the basal alfalfa ration.

No palatability problems were experienced with LA-HWR when incorporated as high as 75% of an alfalfa meal ration. There were no significant differences in weight gain when lambs fed 25 or 50% LA-HWR rations were compared to lambs fed the basal ration. Dry matter/kg gain was significantly increased as levels of LA-HWR increased in the diet over

25%. Supplementation of 75% LA-HWR with soybean meal significantly decreased dry matter/kg of gain requirements and increased weight gain over rations containing 75% LA-HWR and no SBM. Digestion studies with LA-HWR indicate it had a dry matter digestibility of 32%. The rate of cellulose digestion in small dacron bags, suspended in the rumen of the test lambs, was not significantly different for any of the ration treatments with LA-HWR. Rumen volatile fatty acid analyses failed to show any significant effect due to the LA-HWR diets. These data suggest that LA-HWR is comparable to a low quality roughage such as weathered range grass and could possibly be used at high levels in maintenance diets for ruminants.

Palatability factors limited the utilization of HA-HWR at more than 35% of the ration. Growth and digestion studies indicated that HA-HWR was poorly utilized as an energy source in ruminant rations. The incorporation of HA-HWR at the 20 and 35% levels significantly reduced weight gain compared to the basal ration. When compared to the basal ration, dry matter digestibility was significantly decreased in all HA-HWR rations except the 20% HA-HWR with SBM. Cellulose digestion was significantly lower in the ration containing 35% HA-HWR and SBM. Dacron bag studies revealed that rate of cellulose digestion was significantly depressed at the 30 hour sampling period by the addition of 35% HA-HWR and SBM to a basal ration. No significant differences were noted at 15 or 48 hours.

Attempts to evaluate the total concentration of volatile fatty acids and ammonia nitrogen in the rumen of steers consuming HA-HWR failed to show any significant differences although ammonia production was markedly higher in the 50% HA-HWR ration.

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APPENDIX

TABLE XV

SOURCES OF VARIATION AND DEGREES OF FREEDOM ASSOCIATED WITH THE ANALYSIS OF VARIANCE OF GROWTH AND FEED EFFICIENCY PARAMETERS EVALUATED IN TRIAL 1

Source	Degrees of Freedom
Ration	5
Weight group	2
Ration x weight group	10
Error A	16
Corrected total	33

TABLE XVI

SOURCES OF VARIATION AND DEGREES OF FREEDOM ASSOCIATED WITH THE ANALYSIS OF VARIANCE OF DIGESTIBILITY PARAMETERS EVALUATED IN TRIAL 1

	Damasa af
Source	Degrees of Freedom
Ration	5
Weight group	2
Ration x weight group	10
Error A	16
Period	1
Ration x period	5
Weight group x period	2
Ration x weight group x period	10
Error B	16
Corrected trial	67

TABLE XVII

SOURCES OF VARIATION AND DEGREES OF FREEDOM ASSOCIATED WITH THE ANALYSIS OF VARIANCE OF DACRON BAG AND VOLATILE FATTY ACID DATA EVALUATED IN TRIAL 2

Source	Degrees of Freedom
Ration	5
Error A	6
Period	2
Ration x period	10
Error B	12
Corrected trial	35

TABLE XVIII

SOURCES OF VARIATION AND DEGREES OF FREEDOM ASSOCIATED
WITH THE ANALYSIS OF VARIANCE OF GROWTH AND FEED
EFFICIENCY PARAMETERS EVALUATED IN TRIAL 3

Source	Degrees of Freedom
Ration	4
Weight group	2
Ration x weight group	8
Error A	15
Corrected total	29

TABLE XIX

SOURCES OF VARIATION AND DEGREES OF FREEDOM ASSOCIATED
WITH THE ANALYSIS OF DIGESTIBILITY
PARAMETERS EVALUATED IN TRIAL 3

	D
Course	Degrees of
Source	Freedom
Ration	4
Weight group	2
Weight group x ration	8
Error A	15
Period	1
Ration x period	4
Weight group x period	2
Ration x weight group x period	8
Error B	15
Corrected total	59

TABLE XX

SOURCES OF VARIATION AND DEGREES OF FREEDOM ASSOCIATED WITH THE ANALYSIS OF VARIANCE OF DACRON BAG, RUMEN AMMONIA, AND VOLATILE FATTY ACID DATA EVALUATED IN TRIAL 4

Source	Degrees of Freedom
Ration	2
Error	9
Period	2
Ration x period	4
Error B	18
Corrected total	35

TABLE XXI

SOURCES OF VARIATION AND DEGREES OF FREEDOM ASSOCIATED WITH THE ANALYSIS OF VARIANCE OF RUMEN AMMONIA AND VOLATILE FATTY ACID DATA EVALUATED IN TRIAL 5

Source	Degrees of Freedom
Animal	2
Period	2
Ration	2
Error A	2
Day	1
Animal x day	2
Period x day	2
Ration x day	2
Error B	2
Corrected total	17

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