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EVALUATION OF CHEMICAL TREATMENTS OF LOW
QUALITY ROUGHAGES FOR RUMINANTS.

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EVALUATION OF CHEMICAL TREATMENTS OF LOW QUALITY
ROUGHAGES FOR RUMINANTS

by

Sumer Hasimoglu

A THESIS

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Under the Supervision of Dr. T. J. Kopfenstein
and Dr. T. H. Doane

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Evaluation of Chemical Treatments of Low Quality Roughages

for Ruminants

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PREVIEW

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TABLE OF CONTENTS

	PAGE
INTRODUCTION.	1
LITERATURE REVIEW	4
Carbohydrate Constituents of Low Quality Forages.	4
Chemical Treatment of Low Quality Forages	9
Energy Requirements of the Ewes	15
EXPERIMENTAL PROCEDURES	19
Methods of Processing Ground Corn Cobs.	19
Ewe Feeding Trial I	21
Growth Trial I.	22
Growth Trial II	25
Rat Trial I	28
Rat Trial II.	31
Ewe Feeding Trial II.	31
Ewe Feeding Trial II, Milk Production and Milk Samples Analysis	36
Statistical Analysis.	38
RESULTS AND DISCUSSION.	39
Ewe Feeding Trial I	39
Growth Trial I.	65
Growth Trial II	73
Rat Trial	83
Ewe Feeding Trial II.	92
Second Ewe Trial, Milk Production and Milk Samples Analysis	106
SUMMARY	112
LITERATURE CITED.	117

INTRODUCTION

Corn cobs, which are a plant residue, represent a large reservoir of carbohydrate raw material. These carbohydrates, which are almost entirely of polysaccharide nature constitute the largest portion of the corn cob. The plant polysaccharides are found in the cell wall and are mainly cellulose, hemicellulose and nonpolysaccharide lignin. The non-cell wall constituents of the plant include sugars, starch, pectin, lipids and proteins.

Low quality forages, such as corn cobs and straw, contain less cell contents and more cell wall constituents as compared to higher quality forages. The cell wall portion of the plant contains lignin in small amounts as compared to cellulose and hemicellulose, but it binds cellulose and hemicellulose and makes indigestible complexes. The amounts of indigestible complexes increase as the forage plant matures. This low digestibility causes the plant residues to be considered as poor sources of energy.

Monogastric animals are incapable of utilizing the insoluble plant polysaccharides. The symbiotic cellulolytic rumen microorganisms allow the ruminant animals to utilize the high cellulolytic plant residues.

More economical rations for ruminant animals will depend largely on increasing forage carbohydrate utilization by the rumen microorganisms. The utilization of plant residues by ruminant animals would be beneficial to the grain producer. Increasing the digestibility of plant residues by chemical treatments will increase their economical value due

to their increased digestibility.

Previous studies at this institution have shown that the treatment of low quality forages with chemicals increased their digestibility and their utilization by the ruminants. Alkali (mostly sodium hydroxide) has been used to degrade the physical and chemical complex of lignin with cellulose and hemicellulose in low quality forage.

Sodium hydroxide has been used as a delignifying agent where the treatment involved soaking the roughage in large vats of sodium hydroxide solution and then washing it to remove residual alkali. The washing operation in this method causes heavy loss of nutrients. The neutralization of the excess alkali with hydrochloric or acetic acid is also one of the methods which has been used. This method is, however, expensive. A practical procedure is to ensile the sodium hydroxide treated straw and corn cobs which eliminates the majority of the spoilage. With this ensiling procedure, it is possible to produce and store treated low quality forages in large quantities.

This study was initiated to determine the practicality and economics of using sodium hydroxide treated, ensiled corn cobs for feeding a sheep flock during early gestation, late gestation and lactation periods for two years. Their performance was compared to sheep fed a medium quality chopped brome-alfalfa hay plus grain. In addition, two growth trials were conducted. The purpose of the first growth trial was to determine the effect of level and source of protein upon performance of lambs fed NaOH treated corn cob. The purpose of the second growth trial was to evaluate the complementary effects of certain natural

proteins with urea in NaOH treated cob rations. Two rat trials were also conducted to determine the effect of high sodium containing rations upon feed consumption, body weight performances and reproductive efficiency.

PREVIEW

LITERATURE REVIEW

Carbohydrate Constituents of Low Quality Forages

The ruminant animal has a unique ability to utilize cellulose material as a major source of energy. In order to take advantage of this unique ability of the ruminant animals, it is necessary to understand the chemical composition of hay, straw, corn cobs or other fibrous by-products which consist of complex carbohydrates.

The forage carbohydrates are the most important energy sources in rations of ruminant animals. The principal products of photosynthesis in plants are stored as such or may be converted to other substances. Through the metabolic activities of the plant, the primary sugars are transformed 1) to the polysaccharides such as starch and fructans, 2) to the substances which make up the structure of the cell wall such as cellulose, hemicellulose, lignin and pectins, 3) to plant proteins, 4) to the fats and waxes (Hansen *et al.*, 1958). Quantitatively, polysaccharides are the most important nutrients in feeds of plant origin according to Maynard and Loosli (1962).

Plant constituents are classified by Van Soest (1963) and principally divided into two groups according to their solubility in neutral detergent. First is the cell wall constituents which include: a) hemicellulose, b) cellulose, c) heat damaged protein, d) lignin; and second is cell contents which include: a) sugars, soluble carbohydrates, b) starch, c) pectin, d) protein, e) lipids, f) other solubles. Whistler and Smart (1957) indicated that the majority of plant polysaccharides

are components of the cell wall.

The cell contents are the soluble portion of the plant and are readily digestible, but the cell wall carbohydrate constituents are not available to the higher animals except ruminants.

The cell wall is composed of varying amounts of cellulose, hemicellulose and lignin. In all cell walls the chain (of glucose units) runs in the plane of the wall. Mature cell walls consist of numerous layers, which comprise primary and secondary walls. Between the primary walls of adjoining cells is the middle lamella, which consists primarily of pectins. The primary and secondary walls are mainly cellulose but may also contain pectins, lignins, polyuronides and silica. Cellulose once laid down in the cell wall is not considered available as an energy source for the plant. Cellulases of higher plant origin have been reported only in germinating seeds (Whistler and Smart, 1957).

The term holocellulose was devised to include cellulose and hemicellulose of plant cell polysaccharides (Hansen et al., 1958). Flanders (1952a) analyzed low quality forages for their holocellulose contents as follows: barley straw 78.4%, oat and wheat straw 78.0%. Corn cobs were found to contain 81.5% holocellulose by Whistler et al. (1948).

The qualitative relationship between cellulose and glucose was first discovered by Braconnot in 1819 (Wise and John, 1952) who found that the complete hydrolysis of cellulose by acid gave only glucose.

The hemicellulose and lignin may be in some form of combination, and the digestion of the hemicelluloses certainly will depend on the breakage of the linkage with lignin (Norman and Shrikhande, 1935).

Flanders (1952b) gave the holocellulose and hemicellulose percentages of some hay and straw: alfalfa hay 85%, 34.1%; red clover hay 77.5%, 42.5%; barley straw 78.7%, 50.7%; bucket wheat straw 79.4%, 47.0%. Van Soest (1967) found that amounts of hemicellulose found in grasses may be four times the quantity found in legumes.

Another group of cell wall materials is that of the pectic substances, derivatives of pectic acid, which in turn is made up of long chains of galacturonic acid residues. The galacturonic acid residues all possess the 6-membered pyranose ring structure. The chain structure of pectin and protopectin is similar to that of pectic acid, except that the carboxyl groups are methylated (Hansen et al., 1958). They also indicate that pectic acid in the form of its calcium and magnesium salts appears to make up much of the middle lamella of the cell wall and promotes the adhesion of adjacent cells.

The experimental evidences indicate that hemicellulose, cellulose and pectin digestion are influenced by the maturity of the forage and suggest that this effect is the result of lignin forming a physical barrier between the plant hemicellulose, cellulose or pectin and the rumen bacteria.

The woody parts of plants, such as cobs, hulls and the fibrous portion of roots, stems and leaves, contain a complex indigestible substance called lignin. Lignin is not carbohydrate in nature. Hansen et al. (1958) indicated that it is a high molecular weight condensation product of one or more types of aromatic compounds intermixed in the cell wall with cellulose and other constituents. Woody plants may

contain up to 50% lignin, but roughages will contain 2% and up to 20%, depending on the stage of maturity. Pigden (1953) showed that the amount of the lignin and the distribution of the lignin throughout the grasses may vary.

The new methods of forage evaluation introduced by Van Soest have reduced some of the limitations of forage analysis (Krause, 1969). The neutral detergent analysis introduced by Van Soest (1963) eliminates the problems of the Weende system by dividing the plant cell into cell soluble and cell wall constituents.

The lignin in relatively poor forages is practically indigestible. Kamstra (1958) indicated that the indigestibility and variations in the digestibility of lignins can be explained by its lack of homogeneity. Hale et al. (1940) showed that lignin is primarily digested after leaving the rumen.

It has been reported by Sullivan (1959) that lignin might influence digestibility by acting as an "encrusting substance" and preventing the digestive enzymes of the rumen microorganisms and of the intestinal juices from attacking the plant nutrients. Tomlin et al. (1965) suggested that lignification linearly relates to cellulose digestibility as the grasses matured. Lancaster (1944) and Sullivan (1959) showed that lignin in the forages decreased the dry matter digestibility. Van Soest (1964) considered that lignin exerts its effect by either encrustation or chemical combination with holocellulose in the plant. Ball milling can split c-o and c-c bonds and breaks the linkage between "lignin" and "cellulose" exposing the latter to enzymatic action (Dehority, 1961).

It is evident that lignin does not go through the digestive tract as an inert substance, and if it does not give evidence of quantitative digestion, it does at least undergo qualitative changes.

During the growth of the plant tissues, the carbohydrate constituents are formed first followed by the formation of lignin and the spaces previously existing between the polysaccharide fibers are gradually filled in with lignin. Lignin serves as a cement for holding the cellulose fibers together and it protects the plant from physical and chemical damages (Shubert et al., 1957). Work by Armstrong et al. (1950) indicated that many species of grasses had the same lignin content in the dry matter at the same stages of growth. In most species a marked increase in lignin content was found to occur at the flowering stage.

The most important factors in the dry matter digestibility of feeds are the proportion of the available soluble cell contents and the concentration of lignin which controls the digestibility of cell wall constituents (Van Soest, 1965). A significant negative correlation was found by Sullivan (1959) between the percentage of lignin in a forage and the digestibility of the dry matter and crude fiber.

Lignin is normally high in low quality forages, and it exerts a controlling influence on the digestibility of holocellulose which is also characteristically high (Koers, 1970). Chemical treatments to increase the soluble fraction of forages or to reduce the depressing effect upon digestion by lignin have, therefore, been investigated.

Chemical Treatment of Low Quality Forages

Polysaccharides are hydrolyzed by adding strong mineral acids to water soluble simple compounds (Brauns, 1952). Lignin is readily attacked by oxidizing agents and is decomposed to water soluble products.

Kelner and Kohler (1900) showed that pulp prepared for making the product of sulphide process was highly digestible by ruminants (88.3%). Lehman (1902) treated straw with a caustic soda solution, and by this treatment the digestibility was increased by about one-half.

Godden (1920) treated chopped oat straw with a 1.5% sodium hydroxide solution, steamed for an hour and fed. The dry matter of the treated straw had 1.5 times the value of the original dry matter. Archibald et al. (1924) treated grain hulls with 1, 1.5 and 3% dilute sodium hydroxide solution and found that with a treatment of 1.5% sodium hydroxide, the digestibility of the oat hulls and barley hulls was markedly increased, the feeding value of oat hulls being doubled. The dissolution of the straw lignin seemed to be dependent upon the presence of hydroxyl ions (Brauns, 1952).

Beckman (1922) converted chopped straw into a feedstuff by treatment for at least 24 hr with 1.5% sodium hydroxide at the temperature and pressure of the atmosphere; 100 kg of straw yielded from 75 to 80 kg of feedstuff with a nutrient value equal to 56 kg of starch. An experiment with animals showed that 66.86% of the untreated straw and 86.24% of the treated straw were found to be utilized. It was also indicated that sodium carbonate or milk lime may be used instead of sodium hydroxide, but this process was less satisfactory.

Stone et al. (1965) treated alfalfa, rice straw and bagasse with 1.5% alkali for 24 hr at room temperature, then either washed the alkali free or neutralized the system with a mixture of volatile fatty acids. No benefits were obtained from any treatment of alfalfa. The digestibility of rice straw increased from 20.0 to 49.8% and digestibility of bagasse was increased from 16.7 to 69.3% with the 2.0% alkali-wash treatments.

Wheat straw was boiled by Woodman and Evans (1947) with 5.9% caustic soda solution for 7 hr under a pressure of 70 lb/sq in. The alkali was then removed and the residual fodder cellulose washed with water until free from alkali, pressed, dried and finally obtained in the form of a coarse meal. Fodder cellulose formed 38% of the total daily food for digestion trials with sheep. The digestion coefficients of the constituents of the fodder cellulose obtained were: crude fiber, 90.6%; NFE, 38.6%; ether extract, 68.5; total organic matter, 76.5% and total cellulose, 81.8%.

McAnally (1942) found that the rate of digestion of cellulose in straw declines considerably and 30-40% of cellulose remains undigested even after a week's digestion in vitro. Soaking the straw in 1.5% NaOH for 24 hr then washing with cold water until free from alkali increased digestibility in vitro.

Wilson and Pigden (1964) objected to the method described by Beckman (1922) in that it required large volumes of water to wash out unreacted alkali and this washing process removed soluble nutrients. Instead of washing out the unreacted alkali sodium hydroxide, treated

straw was mixed with corn silage and ground alfalfa and neutralized with acetic acid. The dry matter digestibility with alkali treatment up to about 9% caused marked increases in in vitro digestibility, but above this level no further increases were obtained.

A sodium hydroxide spray treatment of wheat straw was evaluated in experiments on male calves by Singh and Jackson (1971). Sodium hydroxide solutions of 0, 3.3, 6.7 and 10.0% concentration were sprayed on ground wheat straw at the rate of 1000 liters per ton of straw. The treated material was fed either dry or wet by fortifying with ground nut cake, molasses and a mineral supplement. The effectiveness of this spray treatment in terms of increased nutritive value was comparable with that of the older Beckman method of alkali treatment.

Donefer (1968) ground oat straw and mixed it with a 13.3% NaOH solution at a ratio of 60 liters of solution per 100 kg straw. After 24 hr, 16.7 liters of 50% acetic acid were added to neutralize excess alkali and the mixture air dried. Alkali treatment increased dry matter digestibility from 45.1 to 61.0%.

The treatment of forages to remove lignin should lead to an improved utilization of the fibrous constituents of forages. Sullivan and Hersberger (1959) used chlorine dioxide in order to treat three forages and measured the digestibilities of orchard grass, reed canary grass and wheat straw which were increased from 36 to 46%; 30 to 46% and 22 to 52.2%, respectively. Chlorine dioxide treatment resulted in a marked decrease in the acid-insoluble lignin content of the forages. Brauns (1952) indicated that ozone and hydrogen peroxide have been used

in neutral solution for the oxidation of lignin. Wise *et al.* (1959) used anhydrous ethanolamine in order to delignify wood lignin. Chandra and Jackson (1971) used the spraying method with NaOH as a delignifying agent on corn cobs. The lignin content of ground corn cobs was reduced by 26% and rumen DMD increased by more than 100%. Bleaching powder was as effective as NaOH in removing lignin, but residual chloride inhibited rumen microbial digestion.

The evaluation of the effect of oxidizing agents on alfalfa stems and corn cobs *in vitro* was reported by Jones and Klopfenstein (1967). NaOH, Na_2O_2 and $\text{Na}_2\text{O}_2\text{-H}_2\text{O}_2$ treatments significantly increased apparent *in vivo* dry matter disappearance of both alfalfa stems and corn cobs. Treatments of Na_2O_2 were superior to NaOH treatment of corn cobs and corn cobs were more susceptible to oxidation than were alfalfa stems. Concentrations of 4% NaOH, 4% Na_2O_2 and 4% $\text{Na}_2\text{O}_2\text{-3% H}_2\text{O}_2$ mix resulted in maximum increases of 7.8, 8.7 and 6.4 percentage units, respectively, over the control for alfalfa stems and 19.5, 20.1 and 16.4 percentage units for corn cob treatments. Chemical treatments increased the amount of nitrogen which was retained in the digestion trials with lambs.

The effect of 4% NaOH treatment on the digestibility and nutritive efficiency of wheat straw fed with ground wheat and urea or soybean meal was studied by Hasimoglu, Klopfenstein and Doane (1969). The organic matter digestibility of the wheat straw increased from 49% to 60% by 4% NaOH treatment in SBM supplemented rations and was increased from 51 to 59% in NaOH treated urea supplemented wheat straw rations. *In vitro* dry matter digestion of straw increased from 46 to 61% by NaOH

treatment. The NaOH treatment significantly increased the nitrogen retention in both urea or SBM supplemented rations compared to untreated urea or SBM supplemented straw rations.

A similar experiment was conducted by Saxena et al. (1971) who soaked ground oat straw for 22 hr in a 1.5% s/v solution of NaOH. It was then washed with water. Oat straw and treated straw were then supplemented with SBM, urea or diammonium phosphate and fed to the lambs. Acetic acid was added at a rate of 33 ml/kg DM to the treated oat straw to neutralize residual alkali. Approximately 25% of the straw dry matter was lost in the treatment and the washing. Lambs fed different sources of nitrogen gained at significantly different rates when treated straw rations were used. They found that animals fed treated straw had lower levels of rumen ammonia and blood urea indicating that straw treatment was effective in bringing about the release of energy to stimulate bacteria growth in accord with the NPN present.

Krause (1969) ensiled whole corn plant, corn stover and corn cobs which were treated with 0, 3, 4 or 5% NaOH of the dry weight of the plant. In vivo energy digestibility of the 0 and 4% NaOH treated corn silage fed to lambs was 64.2 and 68.6%, respectively. The corresponding in vitro digestibilities were 66.0 and 75.7%. The rations containing 0 to 5% NaOH treated stover silage were fed to growing lambs. In vivo organic matter digestion was increased from 44.6% to 55.1%. In vitro dry matter disappearance of the alfalfa plus 0% NaOH treated stover silage and the alfalfa plus 5% NaOH treated stover silage was 56.0 and 67.8%, respectively. In vivo organic matter digestibilities increase

due to NaOH treatment nearly equaled in vitro dry matter disappearance.

Potassium chloride supplementation of NaOH treated rations was investigated by Koers, Woods and Klopfenstein (1970). The treatments were different supplemental levels of potassium which had no influence upon the performance of the cattle. Four percent treated corn cobs made up 80% of the ration increase and average daily gain from 0.66 to 1.61 lb. Efficiency of conversion was increased considerably.

Another procedure is the high pressure and temperature treatment of plant residues with and without alkali. Klopfenstein and Bolsen (1971) found that in vitro dry matter disappearance of crop residues was increased by steam treatment at 28 kg/cm^2 . Addition of 0.1% HCl or 3% NaOH (% of dry matter) lowered the optimum pressure for treatment of all: corn cobs, wheat straw and milo stubble from 21 kg/cm^2 to 14 to 17.5 kg/cm^2 .

Guggolz et al. (1971) analyzed samples of grass straws in detail. They were given laboratory high pressure steam treatments with and without alkali. Steam treatment for 3 minutes at 28 kg/cm^2 increased the digestibility of grass straws by an average of more than 50%. Digestibility of most of the straws was more than doubled when 3% NaOH was added with the steam treatment.

An important conclusion could be drawn from the above review. Although chemical treatments increased forage digestibility, its utilization still may not be economical. There seems to be some practical application in the addition of sodium hydroxide to plant residues then

ensiling the mixture.

Energy Requirements of the Ewes

Maintenance of the ewe is a continuous cost in sheep production. Her maintenance nutrient requirements must be met throughout the year, and the largest portion of this continuous cost is protein and energy.

Lack of energy is probably the most common manifestation of nutritional deficiency of sheep. This may result from lack of sufficient feed or from too little net energy available to the animal from the feed consumed. Poor-quality roughages are not only poorly digested but are also consumed in small quantities.

The maintenance energy requirements of pen-fed sheep were determined by Coop (1962) with four trials. Nutrient requirements for wool growth were allowed. Live weight change was the measure of energy gain or loss. Energy maintenance requirements were found to be .8 to 1.0 lb digestible organic matter (DOM) for a 100 lb pen-fed sheep.

The maintenance energy requirements of sheep (Phillipson, 1958) were found to be 0.9 to 1.0 lb DOM/day for a 100 lb sheep which is 60 to 70% of the USDA standards. The figure .92 lb DOM/day found for stall-fed sheep is in agreement with the above view.

The study of Coop and Hill (1962) determined the maintenance energy requirement of grazing sheep by using the chromium oxide-nitrogen method in three trials. The energy maintenance requirements were found to be 1.48 lb; 1.63 lb and 1.36 lb DOM/day for a 100 lb grazing sheep.

The figures for the maintenance requirement of grazing sheep are 50 to 80% above the pen feeding figures. Lambourne (1955, 1961) indicated that maintenance requirements of the grazing sheep were in all cases higher than those of pen-fed sheep of the same weight. The increase was only 10 to 30% for heavy sheep grazing in good pasture, but this amount increased to 50 to 100% for the medium and low weight sheep maintaining weight in progressively poorer pastures. It is obvious that there will be energy cost of grazing.

Langlands et al. (1963) measured the maintenance requirement with direct calorimetric method of the mature 45.4 kg (100 lb) housed sheep and found it to be 973 k cal/24 hr and this value is estimated to be 0.79 lb DOM which is lower than Coop's 1962 value. In the second indoor experiment, it was found to be 0.82 lb DOM sheep/day.

In another study, Langlands (1963) with grazing sheep found the energy requirement to be 1.02 lb DOM daily which is 24% higher than the corresponding value for housed sheep. The NRC recommendation is 1.15 lb DOM/day for 100 lb sheep which is higher than the values found by the researchers indicated above.

In practice most pregnant ewes must be given a sufficient energy allowance to enable them to gain some weight during the pregnancy period as a whole, with special attention to the last quarter when the specific needs are substantial.

In a two year experiment, Wright et al. (1962) used 87 white faced western ewes to evaluate the adequacy of varying levels of energy and protein in the ration of gestating and lactating ewes which were fed

Timothy hay, oat straw, shelled corn, oats and soybean meal. It was shown that .18 lb; .22 lb; 0.29 lb of protein levels were adequate during early gestation, the last six weeks of gestation and the first eight weeks of lactation, respectively. It also appeared that 1.7 lb, 2.4 lb and 2.8 lb TDN fed during early and late gestation and the first eight weeks of lactation, respectively, were adequate. An increase in the energy intake above this requirement did not result in greater lamb production. They pointed out that .14 lb/day should be the minimum weight gain during late gestation in ewes of 130-140 lb.

Two experiments were conducted by Gardner and Hogue (1963, 1964). The levels of TDN used were those recommended by the NRC for 130 lb ewes during the last six weeks of gestation (1.77 lb TDN/100 lb body weight) and the first 8-10 weeks of lactation (2.31 lb TDN/100 lb body weight) and 125% of these values. Varying TDN levels for ewes during the last six weeks of gestation did not affect single lamb birth weight, but feeding higher levels significantly increased the average 90 day weight of twin lambs in both experiments. They interpreted the data to indicate that the present NRC-TDN level for late gestation is high enough for ewes pregnant with single lambs but not for ewes with twin lambs.

The energy requirement for the lactating ewe during the first 8 to 10 weeks of lactation is based by NRC (1957) on a 45 kg ewe that produces 1.36 kg of milk per day is 1.22 kg TDN. Hogue (1968) indicated that the NRC requirement is not adequate to meet the nutritional needs of the lactating ewe.