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# Fox,

Susan M.

1989

# A COMPARISON OF CHEMICAL COMPOSITION AND FERMENTATION PATTERNS OF ALTERNATIVE SILAGES TO WHOLE PLANT CORN SILAGE

A Thesis

Presented to the Faculty of the Department of Agriculture Western Kentucky University Bowling Green, Kentucky

In Partial Fullfillment of the Requirements for the Degree Master of Science

by

Susan M. Fox July 1989

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# A COMPARISON OF CHEMICAL COMPOSITION AND FERMENTATION PATTERNS OF ALTERNATIVE SILAGES TO WHOLE PLANT CORN SILAGE

Recommended June 23, 1555 David a. Stile Luther B. Hughes Jr. Director of Thesis

Approved July 17, 1989 (date) Elmer Bray Dean of the Graduate School

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# A COMPARISON OF CHEMICAL COMPOSITION AND FERMENTATION PATTERNS OF ALTERNATIVE SILAGES TO WHOLE PLANT CORN SILAGE

Susan M. FoxJuly 198947 pagesDirected by:David A. Stiles, L. B. Hughes, Jr., and J. P. WorthingtonDepartment of AgricultureWestern Kentucky University

A comparison of the relationship of fermentation to chemical composition was made for forages which were wilted and ensiled at 35 to 45 percent dry matter. Trial I consisted of three forages ensiled in October, 1981: interseeded soybeans and grain sorghum, whole plant corn with added anhydrous ammonia, and whole plant corn with shelled corn added at a rate of 150 kg/t of fresh forage. Trial II consisted of two forages ensiled in 1982: interseeded soybeans and pearlmillet, and wheat. Temperatures of fermentation were collected, and chemical composition during the first 25 days of fermentation analyzed.

Production data were also collected in Trial I. The cash expenses and yields do not indicate a significant advantage to either crop in this study in terms of yields and return over variables. In terms of plant nutrient content there was an advantage for soybean/grain sorgum silage in protein yield of 854 kilograms per hectare as compared to whole plant corn silage.

Temperature data collected on the forages in Trial I covered 57 days from October 8 through December 4. The maximum recorded temperatures for soybean/grain sorghum, corn silage with added anhydrous ammonia and the corn silage control were 37, 37, and 24 degrees Celcius, achieved by day 11, 4, and 11 of ensilement respectively. The rapid temperature increase for material treated with anhydrous ammonia confirmed previous reports. Small fluctuations occurred in silage temperatures but these were not correlated to ambient temperature. Temperatures declined very slowly in all forages, with the lowest reading for silages by day 57 recorded at 19 C for soybean/grain sorghum silage. Ambient temperature was not reached in the ensiled mass during the 57 day period in which data were recorded.

The soybean/pearlmillet ensiled in Trial II quickly reached a high peak temperature of 44 C at 4 days of ensilement, gradually decreasing over a 57 day period to 35 degrees on October 19. The silage did not reach a desireable pH. Initial forage pH was high, 7.4, dropped rapidly to 5.1 on day 5, but did not decline further in succeeding samples. Samples were low in lactic acid and high in acetic acid content. Initial buffering capacities for both of the soybean mixture forages were considerably higher than the other forage materials. Buffering capacities at day 0 for whole plant corn, whole plant corn treated with anhydrous ammonia, soybean/grain sorghum, soybean/pearlmillet and wheat forages were 19.4, 20.0, 35.3, 35.3. 38.6 and 22.2 milliequivalents/100 g of dry matter respectively. Increases in buffering capacities during fermentation were smaller for soybean/grain sorgum and soybean/pearlmillet . Buffering capacity increases for the forages were 133.5, 290.5, 69.1, 64.2 and 87.4%.

High ammonia nitrogen levels were found in samples of whole plant corn silage with added anhydrous ammonia obtained on days 10 and 20 of ensilement. These contained .282 and .351% ammonia-nitrogen and tested 18.0 and 20.1% crude protein. The increase in buffering capacity which occurs with addition of anhydrous ammonia was confirmed by the higher acetic acid and pH levels, with corresponding decreases in lactic acid from samples taken days 10 and 25. Crude protein levels were 17.9 and 17.4 percent while the other samples contained only 10-12% crude protein. It appeared that application of anhydrous ammonia was not uniform throughout the silage. Where high concentrations of ammonia occurred fermentation was prolonged as indicated by excessive amounts of lactic and acetic acids and a high pH in these samples. Values for L(+) lactic acid ranged from .2 to .523 in fresh forage to 5.66% of dry matter on day 25. Concentrations of ammonia-nitrogen in the other silages ranged from .061 to .131%.

The low buffering capacity of whole plant corn silage was reflected by normal concentrations of lactate and acetate but a more acidic pH when compared to the other forages. Lactic and acetic acid production for soybean/grain sorghum silage was similar to that of the corn silage control. Corn silage pH was lower, however, throughout fermentation and reached a stable pH by day 5 of fermentation. The wheat silage went through a gradual fermentation with low lactic acid production, and an intermediary ending pH of 4.0.

Fermentation was essentially complete by day 10 in all silages as indicated by pH, buffering capacity, and lactic acid production; however, there was a tendency for buffering capacity and acetic acid content to increase in all of the ensiled materials throughout the 25 day collection period.

#### INTRODUCTION

Whole plant corn has become the standard high energy, high yield crop harvested for use as silage in the United States. The use of other cereal and forage crops for silage is of interest for several reasons. The differing seasonal distributions of production among species allows for evaluation of a total systems approach in a farming operation (27). Crop rotations aid in breaking plant disease and insect pest cycles (27) and play an important part in conservation plans. The ability to be harvested as silage allows for an alternative to harvesting as a grain crop, extends the harvest period, and maximizes yield of nutrients and cropping flexibility, including the ability to salvage drought or frost damaged crops (20).

To maximize production, double-cropping systems are used which allow more than one harvest per unit of land over a growing season. Frequently in the upper southern states a drought period will occur (10). Crops which can be harvested earlier in the growing season by cutting for silage allow earlier planting of a second crop. This generally provides more optimum soil moisture conditions for germination and growth, and lengthens the period in which the crop can grow and reach maturity before a frost. Corn is often planted as a second crop following harvest of a small grain crop. However, corn is lacking in drought tolerance (17). Summer annuals such as grain sorghum and pearlmillet offer greater resistance to drought stress (16).

Whole plant corn silage is also low in protein, calcium and phosphorus (19). Since protein supplements very often represent a large portion of the cost of purchased feeds, the addition of non-protein nitrogen to whole plant corn at ensiling can be an economical means of increasing the nitrogen content of the silage (19) while allowing for a reduction in protein feed supplements.

Our purpose was to obtain data on the fermentation and chemical composition of various alternative crops as compared to whole plant corn when ensiled. The crops investigated were whole plant corn as the control, whole plant corn silage with added anhydrous ammonia, soybeans interseeded with grain sorghum, soybeans interseeded with pearlmillet, and winter wheat. Chemical data presented covers a fermentation period of twenty-five days; data collected on the temperature of fermentation encompassed a sixty day period.

#### **REVIEW OF LITERATURE**

#### Factors Affecting Fermentation and Nutritive Value of Silage

#### **Overview of Factors**

Ensilement continues to increase in use as a major means of storing forages for dairy animals because it offers several advantages over other means of storage: a higher level of total nutrients preserved; less hindrance from unfavorable weather conditions; increased mechanization of harvesting, storing, and feeding; reduced field losses; and preservation for extended periods of time (43). These factors continue to gain in importance as dairy herd size and milk production levels continue to increase.

A complex interrelationship of factors exists between fermentation of forages in the silo and dry matter intake (DMI) and utilization by ruminants. Silage quality, forage intake and dry matter digestibility (DMD) for dairy cattle are important in maximizing production for the least cost.

Preservation of forages as silage is a result of an anaerobic environment and a lowering of pH through bacterial fermentation of sugars to lactic and acetic acids (42). The extent and type of ensilement is determined principally by the interactions of three major factors.

First, the composition of plant material when ensiled. Plant composition is related to plant species, maturity, field fertility and harvest conditions. Chemical constituents of greatest concern are carbohydrate levels, buffering capacity, and dry matter (DM). Carbohydrates are utilized in formation of acids for silage preservation. Buffering capacity effects the level of acids, and thus carbohydrates, required to adequately lower forage pH. Dry matter determines the type and course of fermentation and final parameters required for quality silage. Secondly, the amount of air entrapped or allowed to enter the silage mass is very important. The third factor is the species and levels of native bacteria on the plant material (42).

Van Soest (62) states that "the best forages are those in which the original forage is least altered". High quality silage will be forage harvested at the proper stage of growth, with a pH of 4.2 or less for direct cut and 4.5 or less for wilted silages, between 5-9% lactic acid content in high-moisture silages, free from molds and objectionable odors such

as ammonia, butyric acid and mustiness, show no carmelization or tobacco odor, be of a green color and firm in texture without sliminess. Length of chop suggested is .6 to 2.5 cm for unwilted material and .6 to 1.2 cm for wilted material (43).

#### Pre-seal phase

The stages of ensilement can be regarded as four time periods: pre-seal, fermentation, infiltration, and feed-out (9). During the pre-seal phase aerobic respiration is influenced by the plant composition, DM, length of chop, and the rate of silo filling. Slower rates of filling allow longer exposure to oxygen and extended aerobic activity. In aerobic respiration water soluble carbohydrates are oxidated to produce water, carbon dioxide and heat (62).

In a study by Fenner et al. (13) delayed filling of a silo with whole plant corn forage for 18 hours resulted in lower dry matter (DM), non-protein nitrogen (NPN), ether extract (EE), and nitrogen-free extract (NFE) of 6.1, 25.1, 27.2, and 12.7%, and increased crude protein (CP), true protein, crude fiber (CF), and ash at 10.7, 2.1, 6.8, and 5.2%. The pH and volatile fatty acids (VFA) were increased while lactic acid decreased 51.7%. In three silages the lactic acid to VFA ratio averaged 5.71 while for the delayed-fill silo the ratio was 1.50. A good ratio for quality silage has been suggested to be 4 or higher (42).

Oxygen is entrapped in the forage as the silo is filled, with the level of inclusion affected by forage porosity and rate of filling. Increasing density of the silage mass decreases porosity and slows air movement into the silage (9, 46). Density of packed forage is determined by the DM content of the forage, length of chop and height of forage in the silo.

The DM content of forage material depends upon maturity of the plant material and extent of wilting before chopping. During this phase oxygen infiltration occurs into forage exposed to air, plant and aerobic organisms continue to respire resulting in forage DM loss, temperature increases, and a delay in pH decline (9). Dry matter losses are the result of use of rapidly fermentable carbohydrate in respiration, which decreases substrate available for production of lactic acid by anaerobic organisms and allows an increase in detrimental plant and microbial activity (41, 50).

#### Fermentation Phase

The second phase begins with the sealing of the silo. Negative processes affecting crop ensilement include aerobic microbial activity, plant respiration, proteolytic enzyme activity, and anaerobic clostridial activity. Aerobic microorganisms, cheifly yeasts with some activity by bacteria, use a wide range of substrates, most importantly the rapidly available carbohydrates, organic acids, and proteins (41). Yeast produce ethanol which does not contribute to a lower pH and silage preservation. This is a disadvantage as they compete with the pH reducing, lactic acid bacteria for water soluble carbohydrates (50).

During the initial aerobic phase of fermentation plant enzymes use the readily available carbohydrates and oxygen to produce heat, water, and carbon dioxide. This early depletion of sugars can significantly reduce substrate available for fermentation in lower energy crops, preventing adequate presevation (41). High oxygen levels result in lower lactate, higher acetate, pH, volatile nitrogen, and possibly increased butyrate levels (50).

Plant enzyme activity is affected by pH, time, DM content and temperature of forage material (36, 41). The extent of respiration is dependent on the oxygen supply and available carbohydrates but the rate is affected by temperature (36, 38). Temperature increases respiration exponentially from 0-30 C; above 30 C the rate of increase decreases slowly (38). Reducing moisture content to reduce the level of plant respiration has produced variable results although in general respiration decreases as DM levels increase (40). Plant and microbial respiration cease when the silo oxygen is depleted, approximately 48 hours after ensilement (46).

Proteolytic enzymes convert protein N to peptides and free amino acids, lowering the feeding value (21, 62). Plant proteolytic activity decreased with lowered pH, increased DM, and over time but increases with increasing temperature (62). An air by temperature interaction increases NPN levels although temperature is a greater factor than oxygen levels (25). The greatest activity occurs on day one of ensilement and then declines over a course of approximately 5 days (45, 67). For silages greater than 50% DM decreasing silage pH rapidly and maintaining low silage temperatures reduces proteolysis (18).

Carbon dioxide forms rapidly and, after reaching a maximum level in a few days, gradually decreased to approximately 20% of the gasses present (42). As anaerobic conditions prevail lactic acid bacteria, found initially in small numbers on the plant, increase in three or four days to several hundred million colony forming units per gram of forage. A level of 10<sup>8</sup> cfu/g of lactic acid bacteria in forage is required to begin a pH decline (41). The bacteria may be heterolytic or homolytic and act on readily available carbohydrates to produce lactic acid and some acetic, propionic, butyric, formic and succinic acids. Homolactic fermentation dominates the first few days of ensilement and heterolactic fermentation dominates the to two moles of lactic acid is efficient, with virtually no energy loss and 0-33% losses in silage DM (36, 39). Heterofermentative fermentation of glucose produces one mole each of lactic acid, acetic acid and carbon dioxide with a 24.0

and 1.7% loss of DM and energy. Energy recovery in low DM silages may be 99% while DM recovery is 51-100%, thus heterolactic fermentation can having a concentrating effect on gross energy with increases of 6-10% (39). Normally heterolactic fermentation accounts for less than half of the end products in a natural silage fermentation (41).

Certain forages such as grasses and legumes may be deficient in readily available carbohydrates which are necessary for production of lactic acid at levels sufficient for adequate preservation. High carbohydrate feedstuffs may be added to the forage at the time of ensiling to ensure adequate substrate (36).

#### Temperature Effects

Temperature increases during ensilement of forages are dependent upon the extent of respiration, which is controlled by oxygen entrapped in the silo and available carbohydrates, the rate of respiration, insulation of the silage and its specific heat (38). Temperature rise is mainly due to aerobic respiration (38, 65). Temperature increases cease when oxygen within the system is depleted followed by a gradual decline over an extended period of time. Reports in one review indicated a period of 103 days occurred before ambient temperature was reached with wilted ryegrass (46). Total increase in temperature has been reported at 3-5 C for 43% DM silage (46)

For homofermentative bacteria 30-35 C is an optimum growth temperature while heterofermentative bacteria prefer temperatures between 15-45 C (18). Good quality silage can be made at temperatures of less than 26 degrees Celcius (37, 38). In a study by McDonald and Henderson (38) grass silage was ensiled at 16-17% DM. In silage maintained at a temperature of 42 C a clostridial fermentation resulted whereas in similar silage fermented at 20 C a lactic acid fermentation occurred. The high temperature silage reached a final pH of 4.57, and the low temperature silage a pH of 3.72. Values for percent lactic, acetic, propionic, and butyric acids in silage DM were .45, 4.53, .3, and .81 for the 42 C silage, and 7.93, 2.67, .09, with no measureable butyric acid for the latter silage. High-moisture direct-cut silages heated more rapidly than did wilted silages as enzyme activity was greater. Plant enzyme activity produced the most heat; production from microbial activity would be small in the early stages of ensilement. Overheating of wilted material was attributed to improper consolidation and re-entry of air.

Oxygen limitation becomes more important for proper preservation as silage moisture decreases. At temperatures above 40 C, a selective effect prevents growth of many lactic acid forming bacteria. Researchers (35, 65) have suggested that the optimum temperature for lactic acid producing bacteria is 27-38 C but that this is also optimum for other bacteria which produce volatile fatty acids and breakdown protein. Maximum temperature recommended for lactic acid fermentation of silage is 30 C (65).

Small grain silages may be less dense and have lower water soluble carbohydrates available to ensure proper fermentation. Barley, wheat, and oat silages ensiled at 37-39% DM in experimental silos (1.1m x 1.8m), reached a maximum temperature between 4-7 days of 26.7 C. Bunker silos took longer to reach maximum temperatures which were 5-15 degrees higher than the experimental silage. One end of a bunker of Wascana wheat reached a fermentation temperature of 56 C. This silage had a lower protein digestibility of 59.4 as compared with a protein digestibility of 61.6 for the silage stored in experimental silos. Silage density times percent moisture explained 28% of the variation in temperature (24).

Addition of anhydrous ammonia (ANAM) speeds silage temperature rise. Buchanon, Smith and Yao (8), ensiled whole corn plant at 28 and 40% DM with and without ANAM. The silage without ammonia at 28% DM reached 30 C in 54 hours, and at 40% DM reached 30 C in 70 hours. With ANAM the two silages reached 30 C in 30 and 27 hours respectively.

At temperatures above 60 C the Maillard reaction (or non-enzymatic browning reaction) may occur, with significant reductions in protein and energy value (43, 44, 60, 61). Carbohydrates are subject to thermal degradation in the presence of amines or amino acids and water. Sugars condense with residues of amino acids followed by polymerization to form a compound with 11% nitrogen (N) and the physical properties of lignin (62).

High temperatures may not always occur with the Maillard reaction, as was the case with an alfalfa forage ensiled at the bud stage of maturity and 38.8, 52.1, 57.8 and 65.8% DM. The 57.8 and 65.8% DM silages contained low lactic and acetic acid levels, high isobutyric acid concentrations, elevated pH, and high acid detergent insoluble nitrogen (ADIN). Formation of ADIN occurred during the first 10 days of ensiling. Silage was dark indicating the Maillard reaction took place despite low fermentation temperatures, which peaked at approximately 27 degrees Celcius, thus suggesting that DM plays a significant role in the ensiling process irrespective of the temperature of fermentation. There was no significant effect of DM on the temperature of fermentation, (all silages fermented at approximately 27 C); however, the two highest DM silages contained high percentages of insoluble nitrogen and were dark in color. This may be due to the Maillard reaction proceeding at lower temperatures if the pH remains high (6). The 38.8% DM silage pH was 4.68 at 30 days versus 5.72 for the 65.8% DM silage.

High temperatures increase DM and digestible protein losses. Losses in alfalfa ensiled in lab silos and held at 68 C or 38 C were 2.04 and .76% of DM by day 21 (P<.02) (21). Loss of digestible protein is the result of carbohydrate polymerization with free nucleophilic groups like lysine, methionine and possibly other aromatic amino acids (62). Some heat coagulation of protein may be beneficial in feeding silage to dairy cattle (62). It is recommended that moisture levels be kept greater than 45% to limit heating and the Maillard reaction (62).

#### Moisture Content

Forages for silage are described as one of three groups based on moisture level: high-moisture or direct-cut, with 70% or more moisture; wilted with 60-70% moisture; and low-moisture at a 40-60% moisture content. For crops greater than 55% DM fermentation by lactic acid bacteria plays a minor role in making high quality silage; a pH of 4.5 is considered adequate (21, 49). In these silages acidity is inhibited by the availability of water and the high osmotic pressure; therefore, pH of high DM (<65% water) is inversely related to water content (62). For higher moisture silages a rapid pH decline is important. Well-fermented silage will achieve a pH of 4.2 or less in two or three weeks. The low pH inhibits further microbial growth and enzyme action; silage may be kept for years if air is excluded.

In direct-cut silage moisture levels of 70% or more considerable effluent losses can occur (41). Wilting to decrease moisture reduces seepage losses. Less fermentation occurs in wilted material; a pH of 4.5 or less is indicative of good quality fermentation. With low-moisture silages particular attention must be given to air exclusion, fine chopping, rapid filling of the silo and a good seal. Silage between 40-60% moisture will have limited bacterial growth and fermentation. Improper exclusion of air can result in yeast and mold damage, Listeriosis bacteria growth, and spontaneous heating (19). Spontaneous heating results in the formation of indigestible products which lower protein and energy values. Reductions of 50% in milk production have been noted by the author in cattle fed severely heat damaged alfalfa haylage. With 40% or less moisture in at least part of the ensiled contents, fires may occur within a critical range of conditions involving moisture level, availability of oxygen and heat dissipation (19).

In another study Archibald et al. (2) used data from 102 lots of silage and their green crops and analyzed them for correlations between pH, butyric acid, lactic acid, water content, and volatile bases. The component of the green crop having the highest relationship to quality measures was water which had a negative correlation with lactic acid and a positive correlation with butyric acid. The carbohydrate content was also of some

significance, with variations in green crop sugar content apparently unrelated to variations in maturity or species but influenced by weather, accumulating on dry cool days. This may partially explain why sometimes good silage is achieved without an additive and other times not. The carbon to nitrogen ratio has been reported to be influenced by weather elsewhere (61).

In working with 57 direct-cut silages made from alfalfa, millet and oat crops, with varying levels of added ground corn, urea, and calcium carbonate, the factor McCullough (35) most highly associated with final pH was crude protein of the forage. Absence of a significant correlation between carbohydrates and final pH indicated that the level of carbohydrates required depends upon the course taken by fermentation. Playne and McDonald (48) determined that organic acids were the major buffers and that plant proteins contributed to only 10-20% of the total plant buffering capacity. They also found a high correlation between pH at 4 days and final pH.

Within a crop species and maturity there are wider variations in silage intake than in nutritional content and digestibility; thus determination of factors which decrease or increase intake are important for maximum productivity. The influence of moisture content has been studied extensively but does not appear to be directly responsible for intake fluctuations. Buchanon-Smith (8) however, found that while steers consumed more of a higher DM corn silage (40%), rate of weight gain for those fed 28% DM was the same.

It is probable that some interrelationship of factors affecting fermentation and its end products are involved, and moisture content does have a significant influence on the nature of fermentation which occurs (2, 35, 47). Thomas et al. (58) found that treatment of alfalfa forage with a mechanical press to decrease water content, or with formic acid did not significantly affect silage composition, digestibility or performance over direct-cut silage.

#### **Clostridial Fermentation**

Generally a moisture level between 60 and 70% has resulted in good quality silage as indicated by high lactic acid content and a final pH of less than 4.2. Above 70%, seepage losses occur, the extent varying according to the vertical pressure to which the ensiled mass is subjected (19). With this level of moisture undesireable clostridial fermentations may develop, producing poor quality silage containing a low lactic acid and high butyric acid levels, high pH and large amounts of the products from protein degradation (38).

Saccharolytic clostridia ferment carbohydrates and organic acids producing butyric acid, carbon dioxide and water with considerable loss of energy. The conversion of 2 moles of lactate to 1 mole of butyric by clostridia results in 22.1% loss of the original

energy. Proteolytic clostridia ferment amino acids resulting in a variety of organic acids, carbon dioxide, ammonia and amines such as cadaveríne, histamine, putrescine, tryptamine and tyramine (41). Butyric acid, ammonia and amine production are associated with reduced feed intake (41).

In a clostridia fermentation growth inhibition is a function of pH and DM (36, 38). Growth is restricted with a pH of less than four in unwilted silages and a DM of 40-50% in wilted forages (41). Prevention of a clostridial fermentation may be achieved by reducing moisture in the forage through pressing, wilting, adding DM to less that 70% moisture content or through addition of preservatives such as propionic acid to the forage at ensiling (35, 43).

When forages with DM contents of 40% or greater are ensiled, consolidation becomes more difficult. Proper exclusion of air becomes increasingly important to prevent yeast and mold growth, which compete with lactic acid bacteria for available carbohydrates preventing a rapid lowering of pH (39, 50), and to prevent the Maillard reaction (6). Intakes and productivity of animals fed high quality, low-moisture silage or companion crop hay have been similar, with silage sometimes consumed at slightly lower levels, and other times slightly higher levels than hay (23, 43, 58).

#### Buffering Capacity and pH

High protein, mineral or organic acid levels in plant material or in silage additives increase the silage buffering capacity. An increase in production of lactic acid is required in order to substantially alter the pH (19, 51, 54). The final pH of high protein and highly buffered alfalfa silage has been found to be negatively and linearly correlated to the ratio of sugar to buffering capacity (41). Fisher and Burns (14) found that corn has a lower fresh forage pH and buffering capacity than grain sorghum, forage sorghum, and millet. However, a higher forage pH was associated with a lower conversion and increased retention of total nonstructural carbohydrates after ensiling. The authors hypothesized that the higher pH in fresh forage may increase use of hemicellulose as a substrate.

The length, extent, and course of fermentation depends on the buffering capacity and DM of the forage ensiled (41). Buffering ability has been attributed to inorganic ions, specifically the alkaline minerals K, Ca, Mg which form salts with organic acids during the ensiling process and slow pH decline, and to plant protein, protein degradation products, and organic acids (49, 52, 57). Buffering capacity may also be increased through addition of silage additives such as urea, anhydrous ammonia, or minerals such as limestone (19, 25, 32, 34, 54). As acids are formed, neutralization by ammonia or mineral cations delays lowering of silage pH. Subsequently, more water soluble carbohydrates are required for the achievement of a low, stable pH which will prevent further microbial growth (34).

McCullough (34) found a buffering capacity in alfalfa silage at pH 5 which required 10 times more acid to reach a pH of 4 as compared to the unfermented forage. This increased buffering action was attributed to the products of the disintegration of plant protein.

Playne and McDonald (48) studied the buffering systems of red clover and Italian ryegrass. They determined that between the pH of 4 and 6 plant proteins are not a major buffering system. The main buffering capacity of fresh forage is due to the organic acids malate, citrate, and glycerate, and in fermented silage buffering is mainly due to lactic and acetic acids. Buffering by plant protein was only 10-20% of the total capacity. High levels of organic acids present in fresh clover gave it approximately twice the buffering capacity of ryegrass. Plant organic acids are present largely as salts. When ensiled, breakdown of these acids releases cations which are neutralized by the lactic and acetic acids that have been formed, thus resisting a change in buffering capacity.

Wilting of forage decreases organic acid content and buffering capacity (37, 48), and enzyme activity (35) of plant material. Playne and McDonald (48) found the buffering capacity of wilted clover to be 18% lower than that of fresh, macerated clover. Smaller quantities of lactic and acetic acids have been found in wilted forage after ensiling (43, 48).

Buffering capacity of alfalfa may be inversely related to level of fertilization with alkaline minerals. In a study with greenhouse grown alfalfa and varying fertility levels, alkaline mineral content was significantly correlated to the rate of pH decline. Increased water soluble forms of K and Ca were negatively correlated with pH decline. The authors thought this possibly was a result of stimulation of microbial activity by soluble forms of K and Ca (57).

Non-protein nitrogen and calcium carbonate additives also increase the buffering capacity of silages (35, 41, 54). McCullough, et al. (35) using a mixture of 90% Ga1123 wheat and annual ryegrasses at a DM of 20% found that addition of 5% ground corn and .25% urea, or 5% ground corn, .25% urea and .5% calcium carbonate resulted in very active and extended fermentations. Lactic acid and butyric acids were 4.6% and a trace for control, 1.8 and .9% for silage with added corn and urea, .9 and 2.1% for added corn, urea and limestone. Losses in protein and NFE and significant declines in digestibility were accompanied by significant decreases in intake.

Shirley et al. (54) added urea at rates of 0, .5, and .75% of DM to 37-40% DM corn silage. Acetic, propionic, butyric, and lactic acids increased with increasing levels of urea, except for a slight depression of acetic acid in the .5% added urea silage. At 25 days

after ensiling the acid levels were 1.93, .03, .17, and 4.89% DM for the 0% level added urea silage; 1.26, .12, .13, and 5.91% DM for the .5% level of added urea. Acetic acid production continued for 25 days after ensiling. High VFA's are indicative of possible plant protein breakdown and loss of DM (31). Butyric acid at 15-30g/kg has anti-fungal properties (65).

#### Lactic Acid

Lactic acid production is associated with high quality silage and may range from 1 to 20% of DM, with 2 to 10% the normal range (2, 33, 43, 51). The anaerobic fermentation of water soluble carbohydrates, carried on in large part by the bacteria Lactobacillus spp., Pediococcus spp., Leuconostoc spp., and Streptococcus spp., produces a racemic mixture of D(-) and L(+) isomers (33, 44). Normally only small amounts are fermented to butyrate, ethanol and acetaldehyde (62). The acid acts to lower the pH of the forage and preserve or ensile it for extended periods of time. Total production of lactic acid varies with the plant chemical composition and conditions of ensilement. Lactic acid generally increases in forage following ensilement for a period of 2 to 3 weeks (43, 54).

Abnormal forage composition at ensiling may alter the production of lactic acid. Bodine et al. (6) found that in 57.8 and 65.8% DM alfalfa silage the content of lactic, acetic and propionic acids increased through 30 days following ensiling. The pH stabilized between days 10 and 20. Acid-detergent insoluble nitrogen did not increase after day 10, indicating that protein damage occurred in the first 10 days. Final concentrations of lactic acid were low, .07 and .52 on a wet basis for the 57.8 and 65.8% DM silage as compared to 1.24 and 1.15% for 38.8 and 52.1% DM alfalfa silage.

Schaadt and Johnson (51) studied lactic acid production during fermentation of corn forages harvested at increasing maturity levels. High levels of lactic acid were produced by day two of fermention, and formation was essentially complete at eight days. Lactic acid production decreased with increasing maturity of corn plant material. Limestone and urea treatments of corn silage increased lactic acid formation in all harvested stages of maturity except the mature stage. Urea alone at .5% of a 30% DM silage was not found to have an effect. Shirley et al. (54) found a slight increase in lactic acid content with .5% urea, and the content almost doubled with .75% added urea, when added to 37 and 40% DM corn plant at ensiling.

Anhydrous ammonia treatment of 28 and 40% DM silage has resulted in an extended fermentation and increase in lactic acid production of 40% (8.32 and 6.64% lactic acid) over untreated control silage (5.7 and 4.48% lactic acid) (8). Lactic acid has been

reported to be maximized at 1% added ammonia-N in 32% DM corn silage, decreasing at higher levels (25). Johnson et al. (28) however, found that lactic acid formation was maximized at .52% added ammonia and decreased at 1.08%. Alanine levels were increased indicating that the lowered lactate levels may be due to amination of pyruvate to alanine rather than being reduced to lactic acid. It has also been suggested that alanine acts as a reservoir for ammonia in the rumen.

Infusions of lactic acid into the rumen or supplementation in the ration has caused a depression in the voluntary consumption of DM by animals fed hay. When silage and hay prepared from the same plant material have been fed, DM consumption of the silage has often been lower than that of hay, but a more efficient utilization of energy or a higher concentration of energy in the silage ration has resulted in equal performance. Feeding of two silages prepared from the same plant material has resulted in greater weight gains for the animals consuming the higher lactate silage (33).

Lomas and Fox (32) found that ammonia plus mineral treatment of corn silage gave similar performances to that of corn silage supplemented with soybean meal when fed to calves. The increased net energy of the treated silage was attributed to the higher lactic acid content as compared to untreated corn silage. Increased digestibility and efficient utilization of the energy in silage suggests efficient metabolism of lactic acid or its ruminal fermentation product. Use of carbon 14-lactate in lactate added to whole rumen fluid *in vitro* showed that the main product of its fermentation was acetic acid (33). Griffiths et al. (20) also found that rumen fluid contained a high molar concentration of acetate on a silage diet.

Lactic acid produced during fermentation occurs as a racemic mixture of D(-) and L(+) isomers (33). The D(-) isomer has been found to predominate in all maturities except the flint stage of corn silage (51). When large accumulations of lactic acid occur in the rumen it is possible that some lactate absorption occurs in the small intestine. It is not until the pH of the rumen contents falls below 5 that significant amounts of free lactic acid would be absorbed through the ruminal lining. Absorption of isomers from washed-out ruminoreticulum of sheep appeared to be equal and occur by simple diffusion of lactate and free lactic acid from the rumen down a concentration gradient. When sheep were fed a sulfur deficient ration, D(-) lactic acid was formed and readily converted to L(+) lactic acid in the rumen. The L(+) lactic acid was readily absorbed from the blood of ruminants and metabolized, probably in a manner similar to monogastric metabolism. The D(-) lactic acid, however, has been shown to be removed slowly from the blood of cattle following intravenous injection, and this together with the lesser efficiency of its metabolism in other

animals implies slow metabolism of the isomer when carried in the bloodstream; it is probable that most is excreted.

#### Nitrogen Fractions

When fresh, approximately 15-25% of the total forage nitrogen (N) will be in the non-protein nitrogen (NPN) form with 70-80% of this fraction present as free amino acids. During ensiling protein is converted into various NPN compounds by plant proteolytic enzymes and microbial action other than lactic acid forming bacteria. Lactic acid bacteria are practically non-proteolytic with limited powers of amino acid synthesis (36). Deamination and decarboxylation of amino acids produces ammonia and amines (62). Wilting of crops does not appear to inhibit plant protease activity (44) but enzymes are sensitive to pH levels. Optimum pH for plant leaf proteases is at pH 5-6 (35). For clover proteases the optimum pH is 5.9-6.3. Temperature increases of 10-40 C increase proteolysis (41). A model of forage changes in the silo indicates that NPN concentration is more affected by temperature than by DM content (9).

Levels of NPN increase in lower DM silages (18, 36, 41). As much as 65-75% of the total N in a direct-cut silage may be in the form of NPN. In alfalfa up to 85% of total N may be in the form of NPN after ensiling (41). Levels of NPN is silage may not be indicative of ADIN levels (6). In a study of 30 samples from farm silos NPN averaged 46.4% of total N. Of the NPN-N 10.1% was ammonia-N, 30.4% was alpha amino-N, 1.2% amide-N and 58.3% was not identified. Variation was considerable in all but the values except ammonia-N. Losses of N vary as selective retention of N may occur in low N forages whereas higher N forages with a higher pH are subject to ammonia losses (62). Microbial breakdown of protein is inhibited by a pH below 4.2 or by increasing osmotic pressure of the forage liquid phase through removal of water. Levels of ADIN increase with increasing DM of silages (6, 18).

Prolonged fermentation of silage for McCullough and Sisk et al. (34) resulted in losses of protein, NFE, and digestibility. They found that digestibility of wheat/ryegrass silages affected intake - silage digestibilities were as follows: silage I, 69%; silage II, 70%; and silage III, 63%. Intakes were 2.27, 2.14, and 1.82 respectively when fed to Holstein heifers. The more rapid the lowering of pH the less protein catabolism occurs.

#### Palatability/Intake Factors

Many studies have dealt with the relationship of the chemical nature of silage to intake and utilization by ruminants. Van Soest (62) lists these three theories for low intakes of poor quality silage: toxic production of fermentation products; high acidity decreasing

palatability; or depleted fermentation substrate which deprives the rumen organisms of critical energy substrate making it difficult to ruminate (62).

Palatability has been possitively correlated with acetic acid, propionic acid and total sugars (18) and negatively correlated with butyrate, ammonia and amines (41). Cummins and McCullough (12) investigated intake as related to corn plant maturity. Maximum intake occurred when harvested at the dent stage, which was characterized by 35% DM content, maximum DM per hectare yield and maximum DM disappearance in rumen fluid. Cell wall content and nitrogen accounted for 16.4 and 8.4% of the variation in intake when fed to the African Zebu cattle (30). Reduction of cell wall content from 70 to 50% was associated with significant increases in DM intake, with maximum intake occurring at 45-60% CWC (30). In pearlmillet lignin content was highly correlated with DMD (21).

Both the acidity and digestibility and energy substrate theories are supported by the work of Brown and Radcliffe (7). They examined the relationship of total nitrogen, ammonia nitrogen, titratable acids, acetic, propionic and butyric acids, DM content and acid pepsin DM disappearance. The pH was significantly correlated to DM intake, and a significant correlation (P less than .05) existed between *ad libitum* intake and *in vitro* digestibility of DM and rate of digestion. Silage intake was negatively related to the degree of fermentation. There was no significant relationship between chemical composition and DM intake, organic matter intake or energy intake, and no individual silage characteristic accounted for greater than 34% of variation in forage intake. The coefficient of variation between 20 silages was 10 times greater for *ad libitum* intake than for *in vivo* digestion; therefore, intake was indicated to be the more important component when determining differences in nutritional value between silages. The authors hypothesized that the rate of digestion in the reticulorumen has an effect on intake of silage.

Bergen et al. (3) also found that intake by mature crossbred whethers was not influenced by the extent of fermentation or chemical composition of whole corn plant silage varying in organic acid content from 12.2 to .3%. Their data indicated that in corn silage, nitrogen availability may be limiting and that both protein status of the animal and lowered rumen function may depress voluntary forage consumption. Several studies (24) have shown the value of supplementing rations containing these forages with protein.

McCullough and Sisk et al. (34) also found that digestibility of wheat/ryegrass silages affected intake - silage digestibilities were as follows: silage I, 69%; silage II, 70%; and silage III, 63%. Intakes were 2.27, 2.14, and 1.82 respectively when fed to Holstein heifers. Prolonged fermentation of silage III resulted in losses of protein, NFE, and digestibility.

Addition of urea and anhydrous ammonia have increased DM intake and gain over untreated corn silage (8). Feeding of hay at 1 pound per 100 pounds of body weight has tended to negate silage intake effects (34).

#### Addition of Anhydrous Ammonia

Chopped whole corn plant for silage is inherently deficient in protein. Anhydrous ammonia (ANAM) may be added at ensiling as an economical source of nitrogen (19). Several feeding trials have demonstrated that ANAM can be beneficial as an additive to corn silage.

Buchanon-Smith (8) added ANAM to 28 and 40% DM corn silage at 1% of DM. When fed to 300 kg Charolais steers, intake and gain were increased 5 and 6% respectively as compared to untreated silage supplemented at feeding with urea. Retention of added nitrogen in this trial was low, 60% in both corn silages. Lomas and Fox (32) fed 33% DM corn silage treated with 15.6 g ANAM per 1 kg silage DM and a complete mineral mix. When compared with untreated corn silage supplemented at feeding with soybean meal, performances were similar.

Degradation of plant protein appears to be inhibited by addition of ANAM (3, 8, 25, 26, 28, 32). Lomas and Fox (32) found that the magnitude of the plant protein sparing effect can vary. Both ANAM and mineral treatment increased silage insoluble protein and net energy values over untreated silage. When added at recommended levels ANAM treatment resulted in at least 57% of plant protein being in the insoluble form with a range of 57 to 62%. Untreated silage; however, had from 38 to 57% of its protein in the insoluble form. Thus ANAM treatment provides a safety factor by preventing excess degradation under certain conditions. Huber et al. (21) determined that ANAM accounted for 59% of the increase in insoluble nitrogen in fermented silage, the remaining 41% was attributed to a decreased breakdown of plant protein. Limited amounts of ammonia, 5-10%, were indicated to have been incorporated by microbes. In fermented silage (33% DM) recovery of added nitrogen averaged 95%. Five to 10% of the added nitrogen was incorporated into microbial protein. Lomas and Fox (32) studied the performances of calves fed silage with added ANAM. At lighter weights, their performance corresponded closely to diet insoluble protein levels. Buchanon-Smith (8) reported that results from feeding frozen corn forage and corn silage to yearling steers have not indicated an advantage in efficiency of nitrogen utilization due to the decreased proteolysis of plant protein.

Other effects on fermentation include a delay in lactic acid production and higher final pH with increasing ammonia levels. Fermentation is prolonged and the decrease in

pH delayed resulting in an overall increase in lactic acid content with additions of ammonia nitrogen (AN) up to 1-1.5% of DM (8, 19, 25, 26, 32). The high energy content of corn silage is of benefit in production of sufficient lactic acid to overcome the buffering ability of the ANAM and acheive a desireable pH, and is beneficial in the utilization of non-protein nitrogen in the rumen of cattle (19). At the higher levels of ANAM application to corn silage a reduction in lactic acid production may occur (25, 28).

For Buchanon-Smith (8) the longer fermentation, in which the silage took 50% longer to reach 30 degrees C was not associated with an increase in nutrient loss. Yeast counts were reduced to less than one-fifth that of untreated silage. An added benefit noted in the use of ANAM as a non-protein nitrogen additive has been an increased aerobic stability at feeding (8), whereas urea treated silage has been observed to heat readily upon removal from storage (19).

After fermentation is essentially complete continued losses in DM and quality occur due to oxygen infiltration (9, 65). Computer models for losses from corn and alfalfa silage indicates that oxygen infiltration through the silo wall or cover is responsible for most of the DM loss with the rate primarily influenced by silo size, wall/cover permeability and the length of storage (9). Pitt (46) predicted that long-term storage losses in concrete tower silos range from 1-3% with total DM storage losses typically at 3-25%. USDA studies were reported to estimate losses in conventional silos at an average of 10-20% and in sealed silos at 5-10%. Decreasing container permeability and increasing forage density was predicted by the model to have the greatest effect in reducing losses, most of which occurs in the outer 10% of a tower silo (46). Additional losses occur during the fourth phase, silage feed-out, but are not relevant to the data obtained in this study.

#### Forages for Silage

Whole plant corn forage is a standard crop for ensiling due to its high yields, high energy content, and quality preservation. To maximize production, extend the harvest season, and for flexibility in harvest method interest has been stimulated in ensilement of alternative crops. These crops are often used when double-cropping or for late plantings and drought is often a problem. Crops with good drought tolerance have the potential to out-produce late planted corn. Sorghums, millets, soybeans and small grain crops are valuable in the development of extended grazing and harvest periods with flexibility as to means of harvest. Sorghums and millets have an added advantage in increased resistance and recovery from summer droughts (16). Soybeans are interplanted with sorghum and millets in an attempt to offset low protein yields of high-energy crops and to produce a silage that more nearly meets requirements of the ruminant diet.

#### Corn Silage

Ensiled whole plant corn produces a highly palatable feed. Corn silage contains, on a dry matter basis, 8.9% crude protein (CP), 68.5% total digestible nutrients (TDN), .33% calcium (Ca) and .21% phosphorus (P). Corn silages are relatively low in protein content and buffering capacity and high in energy content which makes it an easily ensiled feed (17).

Maximum yields of corn silage DM occur when harvested between the dent and glaze stages of maturity (12). Maximum dry matter digestibility (DMD) has been obtained with corn plants harvested at the milk-early dough to dough-dent stages of maturity. Dry matter digestibility changes with maturity were small but significant. The DMD averaged 71.9% for the two years at the dough-dent stage of maturity and decreased to 69% at the postfrost stage. Crude protein digestibility decreased from 77.5% at the milk-early dough stage to 67% at the flint stage of kernel maturity. Maximum intake occurred at the blister to glaze stage, decreasing at the flint and postfrost stages. With maximum yields occurring at the dent to glaze stages, and since DMD and intake are at or near maximum at these stages, it appeared that corn should be harvested for silage at the dent to glaze stages. At this time the forage contained 27.5-33.9% DM. Others (12, 42) have suggested that corn silage should be made when the corn plants contain about 35% DM.

### Sorghum

Sorghum is considered a relatively drought tolerant crop and is grown extensively in the southwest where despite its lower energy content, high DM yields make it a more economical crop than corn (5). Fribourg et al. (16) however, found that in Tennessee lowering of soil moisture supplying capacity had a similar adverse effect on both corn and grain sorghum productivity for silage. Gahi-1 pearlmillet was best able to compensate for late planting when compared to corn, and forage and grain sorghums, and was benefitted less by irrigation than were the other crops.

Reported harvests from studies in Tennessee have favored corn with yields of 12,330 kg/ha DM and 8,030 kg/ha TDN for sorghum and 13,450 kg/ha DM and 9,220 kg/ha TDN for corn (16). Performances of cattle fed sorghum silage vary with plant type and maturity (5) but have achieved 97% of the rate of gain of animals fed corn silage (21). Rates of gain and feed efficiency have been significantly improved by feeding rolled sorghum silage (19). Morgan et al. (40) fed 11 combinations of 4 silages, corn (A), grain sorghum (B), and forage sorghum (C), and (D) with three concentrate rations varying in crude protein content. Responses to concentrate rations were not significant. For silages

A, B, C, and D the DM consumptions were 12.8, 10.8, 11.4, and 14.2 kg/cow/day with digestible dry matters of 65, 52.2, 51.6, and 56.4%. Lactation trial results for 4% fat corrected milk were 20.9, 18.1, 18.0, and 19.6 kg/cow/day with mean butterfat percentages of 4.00, 3.66, 3.82, and 4.09.

Highest yields of gross and digestible energy occur at late milk to early dough stage after which energy content rapidly declines. Total digestibility is highest at the early bloom stage. Percentage of grain heads increases from 5% at early bloom to 36% at hard dough. The percentage leaves decreases with maturity from 31 to 18%. Dry matter, pH, lactic acid, acetic acid, propionic acid and butyric acid at the early bloom stage of maturity were 19.8, 3.75, 19.4, 6.6, 0, and 0 mg/g. At late milk/early dough stage they were 27.4, 3.53, 20.7, 7.5, 0 and 0 mg/g. At the dough stage energy levels dropped with the resulting values of 27.6, 3.95, 6.2, 8.7, .22, .04 and 0 mg/g (5).

In a study by Sprague et al. (57), sorghum harvested at the hard dough stage and ensiled at 28.3% DM experienced an unexplained 11% loss of protein although no seepage losses were noted. Corn silage harvested at similarly late maturity did not experience significant loss suggesting that proteins or proteolytic enzymes may differ between crops.

#### Soybeans

Soybeans and other leguminous plants are of interest of forage crops for silage due to their high protein content and ability to increase soil nitrogen fertility through decomposition of nitrogen-fixing nodules. They are adaptable to a wide range of planting dates and adapted to most climates.

Soybeans can be of significant nutritional value when grown for forage. Johnson (28) harvested a June 6 planting of Bragg soybeans at 75 days and in full bloom stage with a yield of 4,102 kg/ha, 18% crude protein, 2% crude fat, and 59% digestible DM. Plants harvested at 117 days with seed pods well matured and leaves yellowed and dropping noticeably yielded 7,958 kg/ha, 21% crude protein, and 7% crude fat with a digestible DM of 61%. Moisture content of the late harvest of soybeans was high, 71%, and the resultant silage had an undesireable odor. Replacement of one-half of the corn silage for lactating dairy cattle with this soybean silage allowed a 50% reduction in cottonseed meal from concentrates.

#### Intercropped Soybeans and Grain Sorghum

Reasons cited for use of intercropped legumes and a companion crop have included provision of a more balanced nutritional supply of energy and protein, profit and resource maximization, efficient water utilization, inexpensive weed control, minimization of agricultural risks, and improved soil fertility (49).

Yields of soybean-sorghum crops can be competitive with whole plant corn crops, yields of 31384.7 kg/ha testing 12% crude protein, and 62% TDN have been reported<sup>a</sup>. Cummins (11) reported research data covering a three year period in which DeKalb FS-16 sorghum and Bragg soybeans were grown with seeding rates of 4.5, 9, and 13.5 kg/ha for sorghum and 33 seeds/meter for soybeans. At harvest, sorghum was in the dough stage of maturity and soybeans in full to late bloom. Maximum yields were achieved at a seeding rate of 9 kg/ha for sorghum, increased seeding rates did not significantly increase yields. Soybeans did not significantly effect head, leaf or stalk content except in non-irrigated 13.5 kg/ha treatments, although a trend for an increase in protein content did exist with 1.6% protein in the 4.5 kg/ha sorghum seeding rate as compared to .8% protein for the 12#/A seeding rate..

Shirley and Evans (55) studied the effect of stage-of-maturity and the ratio of plants on total protein and its solubility. Interplanted soybeans (100 to 135 kg/ha) and grain sorghum (17 to 23 kg/ha) were harvested at 9 weeks post planting and four additional harvests were made during the following 9 weeks. Dry matter yield increased from 6,300 kg/ha at 9 weeks to 15,000 kg/ha with 52% vegetation at 18 weeks. A higher protein quality was indicated for grain sorghum with sodium chloride insoluble nitrogen values of 79% for grain sorghum head, and 67% for grain sorghum vegetation in comparison with 58% for both portions of the soybean plant.

Light has been indicated as a critical factor in soybean-sorghum competition with genotypic tolerance dependent upon plant height (64). Varietal combinations affect percentages of vegetation and seed parts. Shirley et al. (55) found that the vegetable parts of a soybean/grain sorghum mix at 123 days averaged 56% TDN, 46% ADF, and 9% protein. Seed parts were 75% TDN and 9% protein for the grain sorghum and 29% protein for the soybeans. Wahua and Miller (63) found that intercropping Calland soybeans with tall sorghum reduced yields by 75 and 14%, and soybeans with semi-dwarf sorghum by 17 and 74% respectively indicating that light is a critical competition factor and that genotypic tolerance to intercropping may depend upon plant height. Yields of DM were 4,399.2 kg/ha for the tall sorghum/soybean crop and 3,675.3 kg/ha for the dwarf sorghum/soybean crop. Relative yield totals, or the sum of the relative yields for each of the mixture components were 111 and 108% respectively (64).

Several varietal combinations have been recommended for maximum yields of DM and nutrition including Northrup King Silo Milo with Larado, Wilson, or Biloxi soybeans, and DeKalb C42A or C42Y with Larado or Lee soybeans at approximately 134 kg/ha for soybeans and 22.4 kg/ha for grain sorghum. Further general recommendations for growing a soybean/grain sorghum crop include seeding between May 1 through July 10 when soil temperatures have reached 60 degrees; using fertilization rates of 33.6 to 44.8 kg actual nitrogen, 67.3 to 100.8 kg phosphate, and 100.8 to 134.5 kg of potash per hectare preplant; and harvest at 60 to 100 days after seeding when soybeans are in the early bloom stage and grain sorghum in the soft dough stage of maturity<sup>1</sup>

Nitrogen fixation by soybeans is reduced by intercropping (59, 63). Trang and Gidddens (59) found highest nitrogen fixation for soybeans grown in monoculture at 18% shading. It has also been suggested that companion crops may gain nitrogen from intercropped legumes through movement of nitrogen from the leguminous plant root nodules to companion crop roots (51); however, studies have not shown any evidence of a direct transfer prior to nodule decomposition (63).

#### Pearlmillet

Pearlmillet (Pennisetum americanum (L.)) seeded alone or in combination with soybeans produces good quality hay or silage for ruminants. Dry matter yields of 15,020 to 20,176 kg/ha have been achieved with pearlmillet under good management practices (11) with most of its growth occurring from late June through late August (11). Favorable temperatures for growth are between 25-30 C with a minimum temperature of 15 C, millets are sensitive to low temperatures but tolerant of pathogens and high humidity (16). Yields superior to sorghum-sudangrass hybrid yields have been achieved on upland sandy soils of southeast coastal plains, but on clay loams there were no notable differences in DM yields (10).

An advantage to use of pearlmillet is it's drought tolerance, growing where precipitation is 400-650 mm or less and it can resume vegetative growth after a dormant period (16). Fribourg et al. (17) found that early planted Gahi-1 pearlmillet outproduced late-planted pearlmillet in only 5 of 12 comparisons. The percent of total production occurring after August 1, for greenchop, was essentially the same for both plantings, indicating an ability of Gahi-1 to compensate for late planting. Irrigation benefited grain sorghum and Sudax SX-11 in 6 out of 10 situations, and forage sorghum 7 out of 10 situations, with production increases of 5% or more. Gahi-1, on the other hand, benefited from irrigation in only one-half of the growing situations. Gahi-1 compensated for late

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<sup>&</sup>lt;sup>1</sup>Personal communication with Dr. John E. Shirley, current address Kansas State University, Manhatten, KS.

planting and drought conditions better than did corn, grain and forage sorghum, and sorghum-sudangrass hybrid.

Pearlmillets vary widely in DMD and leafiness with maturity. Hart (21) reported correlations between leafiness and DMD were significant between the boot stage and maturity but not significant during earlier growth stages. Dry matter digestibility (DMD) declined with age but at different rates for different genotypes. Leafiness at the earliest harvest dates were nearly 100% for all genotypes. At last harvest, however, leafiness varied from 28 to 73% in 1961 and 31 to 76% in 1964. Leafiness was not significantly correlated with digestibility at late stages of maturity; however, stem digestibility was significantly and positively correlated with leafiness. Lignin content was highly correlated with DMD. Digestibility of stems decreased 5 percentage units for each 1% increase in lignin content, and digestibility of leaves decreased 3.9 percentage units.

#### Small Cereal Grain Ensilages: Wheat

Small cereal grain crops yield 6,000 to 7,000 kg/ha for oats, barley, wheat, and rye (45, 24). Markets are generally better for wheat than for other small cereal crops allowing for greater flexibility in harvesting as a grain or forage crop. Wheat can be planted later in the fall, following removal of a summer crop, than can oats or barley because of it's greater winter hardiness. The use of moderate fall and winter grazing has been reported to have little deleterious effect on subsequent grain yields and may even decrease problems with lodging and increase tillering. Harvest for silage in the spring allows for earlier planting of a second crop and optimum use of soil moisture in the humid South. When cut at the soft dough stage, wheat contains 65-70% moisture and can be ensiled directly without seepage losses (20).

Bishnoi et al. (4) ensiled small grain crops at the dough stage. The resultant silages were of good quality with pH values between 4.0 and 4.3. They found triticale and rye contained similar crude protein, ash, and ether extract levels. Wheat and oats were higher in these constituents and barley significantly lower. No significant differences in pH, phosphorus, calcium, or gross energy were found among cultivars except for in the case of rye and barley which were higher in calcium at the dough stage. Protein content of all silages were significantly higher at bloom than at the dough stage. Crude fiber and ash content were higher at bloom in all silages except wheat which was lower in fiber at the bloom stage. Hingston et al. (24) however, found barley and wheat silages to be similar in energy and protein digestibility and higher in these constituents than were oats. The lower digestibility of the oat silage was offset by an increased dry matter intake (DMI), resulting in essentially the same digestible energy intake from all silages for growing Hereford

steers. Lassiter et al. (31) fed oat silage to lactating Holstein cows and heifers. For lactating cows oat silage fedat 63% of total roughage was superior to corn silage, but at 77% cows produced less than animals fed corn silage or alfalfa hay and produced significantly less average daily gain.

Fisher and Lessard (14) compared wheat ensiled at the milk, soft dough, and firm dough stages of maturity to 30% DM corn silage. Protein contents of the four silages ranged from 7 to 8.3%. Crude fiber content of corn was 22%, and that of wheat ranged from 27.4 at firm dough stage to 34.5% at the milk stage. Dry matter intakes were lowest for corn silage and highest for firm dough wheat silage. There were no significant differences in milk yield or composition. Lower DMI of corn silage was offset by a higher dry matter digestibility (DMD). Crude protein digestibilities were essentially equal for the four silages. These results supported previous work which indicated that wheat peaks at the boot stage and again at firm dough. Cow response in terms of body weight change, milk yield, DMI, and DMD indicate whole plant wheat silage should be harvested at the firm dough stage of maturity (14).

#### EXPERIMENTAL PROCEDURES

### Trial I

Trial I consisted of three forage crops which were chopped and ensiled in 4.2 by 10.7 m metal tower silos between October 7 and 13 of 1981. The ensiled crops included interseeded Lee soybeans and DeKalb A28 grain sorghum (SBGS) planted on July 3 with a 17.8 cm grain drill at 134 kg/ha for soybeans and 22.4 kg/ha for grain sorghum and harvested after 95 days of growth. The second forage was O's Gold corn (CSA) planted June 11 in 76.2 cm rows, conventional seeded for a population of 51,893 plants/ha, and harvested for silage on October 9 with application of anhydrous ammonia (ANAM) at a rate of 2.9 g/kg of fresh chopped forage to attain a level of 1% added ANAM on a dry basis. The third crop ensiled was Southern States 710 corn (CSC) planted on June 18 in 76.2 cm rows for a population of 51,893 plants/ha and harvested for silage at the dent stage on October 12. Due to extensive insect damage to the corn in this field and subsequent loss of grain production, shelled corn was added to the forage material as ensiled at a rate of 15% of fresh forage. Fertilizer and production costs are listed in Table 1.

#### Trial II

Two forages were ensiled in Trial II. Hart wheat (WH) was ensiled on May 11 of 1982 at the boot stage of maturity and Ice soybeans and DeKalb pearlmillet (SBPM), interseeded at a rate of 134 kg/ha and 22.4 kg/ha, was ensiled on August 15. Silos were the same as used in the previous trial.

#### Sampling Procedures

Composite samples were obtained from samples taken from each wagon load as the forages were placed in the silos. Samples were placed in plastic bags during the course of silo filling (one day); it is expected that respiration and bacterial growth occurred during this time. Thereafter samples were collected at 5 day intervals for a period of 25 days via 4 cm holes bored into silo doors approximately midway up the silo, and fitted with rubber stoppers to prevent oxygen seepage and silage decomposition. Samples were obtained by drilling into the silage mass with a Penn State bale core sampler (3 cm diameter, stainless

steel tube with cutting teeth on one end) inserted to a depth of 30 to 45 cm. The composite and core samples obtained were placed in plastic bags and frozen until analyzed.

#### Temperature Data Collection

Temperature data were obtained from all silages except the wheat by placement of thermocouple wire midway up the silo and in the center of the forage mass as the silo was filled. Pyrometer readings were taken at intervals of 2 hours initially, decreasing to every 24 hours over a 60 day period. Type T, copper-constantine wire was used in Trial I and Type K, Chromel-alumel wire in Trial II.

#### Proximate Analysis

Proximate analyses for ash, ether extract, crude fiber, and macro-kjeldahl crude protein, followed 1980 standards of procedure as prescribed by the Association of Official Agricultural Chemists (1). Dry matter content of samples were determined by placement of forage material in a forced-air oven, at 100 C for 24 hours.

#### pH Analysis

Measurements of silage pH were obtained by hydrogen ion readings of silage filtrate using a Beckman pH meter equipped with a glass electrode. The filtrate was prepared by combining 20 g of water with 10 g of frozen sample, and grinding intermittantly for thirty minutes. The sample was then strained through a double layer of cheesecloth prior to analysis.

#### **Buffering Capacity**

Buffering capacities were determined by titration as described by Playne and McDonald (40). Ten grams of silage were macerated in 250 mls of distilled water. The pH values were recorded for the macerate which was then titrated to a pH of 3 with .1N hydrochloric acid to release bicarbonate as carbon dioxide. Samples were then back titrated to a pH of 6 with .1N sodium hydroxide. Buffering capacity was expressed as the milliequivalents of alkali required to change the pH from 4 to 6 per 100 grams of dry matter. Titration values were corrected on a 250 ml blank.

#### Silage Extract Preparation

An extract was prepared for use in obtaining values for total lactic acid, L(+) lactic acid, and volatile fatty acids. Frozen samples were ground in a Wiley Mill. Twenty grams of each sample were weighed into beakers, combined with 50 ml of .4 N sulfuric acid,

covered with parafilm and refrigerated at 3 C for 72 hours. Samples were then filtered through a double layer of cheesecloth and centrifuged for 10 minutes at 5,000 rpm. The supernatants were collected and stored at 3 C until analyzed.

#### Total Lactic Acid

Total lactic acid determinations were made following the procedures outlined by Savory and Kaplan (47). One milliliter of silage extract was mixed with .1 ml of ceric sulfate and incubated at 37 C for ten minutes to allow oxidation of lactic acid contained in the solution to acetaldehyde. Ten microliters of the oxidized solution was injected through a septum covered test tube onto .15 g of copper sulfate. The test tube was connected to a trap for collection of acetaldehyde for 5 minutes, and the trap was then flushed into a Varian model 3700 gas chromatograph. The column temperature setting was 300 C, run time at 10 minutes, and the FID attenuation was set at either 32 or 64. Lactic acid and acetaldehyde standards were prepared, and distilled water blanks run at intervals.

#### L(+) Lactic Acid

Enzyme analysis for L(+) lactic acid used preparations and directions obtained from Sigma Chemical Corporation, St. Louis, Missouri. Two milliliters of silage extract were added to 4 ml of 8% perchloric acid solution, swirled and refrigerated to allow protein precipitation. Two millimeters of glycine buffer, 4 mls of water and .1 ml lactic acid dehydrogenase were pipetted into vials containing nicotinamide adenine dinucleotide (NAD). These were inverted to dissolve the NAD. Two and eight-tenths milliliters of the NAD solution were pipetted into labelled test tubes containing 20 mls of the protein-free silage extract, inverted to mix and allowed to stand for 1 hour at room temperature. Samples were read on a Kary 14 Spectrophotometer using 1 cm cuvets. Distilled water was used for adjustment to zero absorbancy at 340 nm, visual region light. Blank samples were run with results adjusted accordingly.

#### D(-) Lactic Acid

Determination of D(-) lactic acid was by subtraction of the L(+) isomer from the total lactic acid values.

#### Volatile Organic Acids

Procedures of the Association of Official Agricultural Chemists (1) were followed for determination by gas chromatograph of the volatile acids, acetic, propionic, and butyric. Three microliters of silage extract were injected into a Varian model 3700 gas chromatograph with hydrogen flame ionization detector. Column packing consisted of 10% ethylene glycol adipate, and 2% phosphoric acid on Supelcoport 100/120 mesh. Column temperature was 110 C, FID attenuation 4, and run time was set at 10 minutes. Appropriate standards and blanks were run and used in adjusting final values.

#### Ammonia Nitrogen

The ammonia nitrogen concentrations in the silage samples were determined using an enzymatic procedure from Sigma Chemical Company, St. Louis, Missouri. The method is based on reductive amination of 2-oxyglutarate using glutamete dehydrogenase and reduced nicotinamide adenine dinucleotide (NADH) to produce glutamate and oxidized NAD. The decrease in absorbance at 340 nm, which results from oxidation of NADH, is proportional to the ammonia concentration. Blank samples contained 3.0 ml ammonia assay solution and .2 mls water. The control was 3.0 ml ammonia assay solution and 12 ml of ammonia control solution. Test cuvets contained 3.0 ml ammonia assay solution and .03 ml silage extract. After 5 minutes during which samples were allowed to reach equilibrium, the absorbancy of the sample was read at 340 nm using distilled water as a reference. Then .02 ml of L-glutatmate dehydrogenase was added to all cuvets, mixed by gentle inversion and readings taken following a 5 minute equilibrium period.

#### **RESULTS AND DISCUSSION**

#### Yield Data Trial I

Yields for whole plant corn forage (CSC) and soybeans and grain sorghum forage (SBGS) along with corresponding seed and fertilizer costs are as listed in Table 1. The return over expenses, as determined by subtraction of variable expenses from forage value, indicates a significant advantage to the corn silage crop. Variable production expenses included seed, fertilizer and sprays. Forage values were determined by use of Peterson Feed Values. Determinations are based on the energy value of shelled corn and protein value of 44% soybean meal with average alfalfa hay at \$50/t, and the digestibility of the crop. The return over variable for SBGS forage is \$542.77, and for CSC forage it is \$762.59 leaving an advantage of \$219.82 to whole plant corn forage. The corn crop had a potential yield advantage having been planted two weeks to one month earlier than was the SBGS crop. Different fields were utilized for production without replications within the fields for the two forage crops; hence, statistical analyses of yield data are not possible.

In terms of plant nutrient yield there was an advantage for SBGS in protein content. Protein yield for SBGS was 3643 kg/ha and for CS was 2,789 kg/ha. At \$0.31/kg of 44% protein soybean oil meal, protein would have a value of \$0.70/kg. The 854 kg/ha of extra protein yield for SBGS forage would amount to \$597.80/ha reduced purchased protein costs. Some advantage, however, would potentially be lost due to the lower digestibility of sorghum plant as compared to corn silage (19).

#### Forage Analyses

Samples were collected from five holes drilled into silo doors, one for each sample collection date. Possible alteration of silage composition may have occurred due to oxygen infiltration when samples were obtained. Rubber stoppers were used to plug the holes when the silo was filled. Some of them were forced out by the silage pressure and had to be replaced. Oxygen infiltration has been suggested to occur 25 cm into silage dry matter but is dependent on density of material and moisture content (45).

Testing errors may also have risen due to placement of silage samples in plastic bags which may have been somewhat permeable to air. It is possible that some volatile gasses were lost before grinding and placement in glass jars (62).

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		SBO	35	C	s
U	nit value	kg/ha	Value	kg/ha	Value
Crop yield/value1		28022	\$773.44	26677	\$1051.85
Variable expenses:					
Seed (soybean)	\$0.53/kg	134.5	\$71.29		
(sorghum)	\$1.54/kg	22.4	\$34.52		
(corn)	-				\$44.48
Fertilizer					
33% Am nitrate	\$0.171/kg	112	\$19.15		
Anhydrous am	\$0.154/kg			140	\$21.56
18-46-0	\$0.309/kg			112	\$34.61
Phosphate	\$0.259/kg	112	\$29.01		
Potash	\$0.171/kg	224	\$38.30	224	\$38.30
SMgO	\$0.143/kg			34	\$4.86
Zinc	\$0.231/kg	11	\$2.54		
Lime	\$0.008/kg	4483	\$35.86	4483	\$35.86
Spray <sup>2</sup>					\$109.59
Total variable			\$230.67		\$289.26
Return over variable			\$542.77		\$762.59

TABLE 1. Trial I. Crop yield, variable expenses and return over expenses for soybean/grain sorghum silage and corn silage.

<sup>1</sup>Forage values used are based on Petersons Feed Values to compare the relative feed values of feeds determined the prices of a base protein and base energy feed. The base feeds here are shelled corn and 44% soybean oil meal, with whole shell corn at \$0.16/kg, soybean oil meal price at \$0.31/kg, and average alfalfa hay at \$50.00/t. Hay values thus generated were \$0.039/kg for corn silage and \$.028/kg for sorghum/small grain silage.

<sup>2</sup>Spray costs include \$5.27 for machinery costs per hectare.

Fresh forage dry matter percentages were as follows: soybeans and grain sorghum (SBGS), 36.6; corn silage with anhydrous ammonia (CSA), 38.8; corn silage control (CSC), 38.2; wheat (WH), 45.1; and soybeans and pearlmillet (SBPM), 44.0. These wilted dry matter levels increase the need for rapid fill and good oxygen exclusion. Dry matter values may have been inflated as volatile substances are lost upon drying (62),

#### Temperature Data Results

Temperature data were collected on four of the ensiled forages as the fermentation process took place: CSC, CSA, SBGS and SBPM. Data was not obtained from the wheat silage.

Temperature data collected on the forages in Trial I (CSC, CSA, SBGS) covered 57 days from October 8 through December 4. The maximum recorded temperatures for each of the forages were 37, 37, and 24 degrees Celcius, achieved by day 11, 4, and 11 of ensilement respectively. Aerobic activity is perceived to be the primary source of heat production (36, 38, 65) with oxygen, pH, and moisture content the three major limiting factors for plant respiration and aerobic microbial growth (41). The rapid temperature increase for material treated with ANAM has been noted previously (8) and in this study treated silage temperature increased more rapidly than untreated corn silage, and sustained peak temperature for an extended period of time.

Final temperatures readings were 22, 25, and 17 C. Ambient temperature was not reached in the ensiled mass during the period in which temperature data were recorded. Ambient temperature ranged from a daily maximum of 7.5 to 25 C with two days close to 0. This was in the range of 10 C of the silage temperature for most of the period in which data was recorded; therefore, there was no net loss of heat. Thermocouples were placed in the center of the silage mass when approximately one-third of the silo was filled, thus were well insulated from ambient temperature change (38).

On October 16 all three silages in Trial I (Figure 1) decreased suddenly in temperature; the SBGS dropped three degrees, and the corn silages each fell seven degrees but regained their previous temperature within three days. The sudden drop in temperature on October 16 did not appear to be related to the fermentation and is not readily explainable.

The SBPM ensiled on August 22 of the following year (Figure 2) quickly reached a maximum temperature of 44 C at 4 days of ensilement, gradually decreasing over a 58 day period to 35 degrees on October 19. Optimum temperatures for lactic acid producing bacteria are between 27-38 C (35). At 40 C a selective effect has been noted to prevent growth of many lactic acid producing bacteria and to favor clostridia growth (24, 36, 38).



Figure 1. Trial I. Temperature data collected on soybean/grain sorghum (SBGS), corn with anhydrous ammonia (CSA) and corn control (CSC) forages during 55 days of ensilement.



Figure 2. Trial II. Temperature data collected on soybean/pearlmillet silage during 56 days of ensilement.



Figure 3. Daily maximum ambient temperatures and silage temperatures during 56 days of ensilement.

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Further evidence of an improper fermentation in the SBPM silage is noted with the results of chemical analyses.

Although various peaks and valleys of one to three degrees occurred throughout the collection period when silage temperatures were graphed with the average ambient temperatures for the same period there was no significant correlation (Figure 3).

#### The pH and Buffering Results

A pH of 4.2 is considered adequate to properly preserve high moisture forage material (11, 43, 62). Four of the five silages were adequately preserved by day 25 (Figure 4). The pH of the CSC silage reached 3.7 by day five and did not alter significantly in the remaining samples taken at later dates, indicating that the majority of fermentation occurred during the first five days. The SBGS achieved a stable pH of 4.2 by the tenth day of fermentation. Readings for WH were 4.1 on day 10 and on day 25 the final pH was 4.0. Readings from sample dates 15 and 20 were higher, 4.6 and 4.5, indicating a possible fermentation problem due to oxygen infiltration from the bore holes.

For CSA a delay in pH drop as compared to the control was expected due to the increase in buffering capacity associated with addition of ANAM (8, 19, 25, 32). The pH of samples obtained on days 15 and 20 were 4.2, while those from days 10 and 25 were 5.3 and 4.7. Protein and nitrogen analyses indicate that ANAM distribution was uneven, with high levels applied to these samples. Ammonia reacts with lactate preventing pH decline (36, 62).

The only silage which did not appear to reach a desireable pH was the SBPM. Initial pH was high, 7.4, dropped rapidly to 5.1 on day 5, but remained constant in succeeding samples. This indicates that a rapid initial fermentation occurred but either insufficient soluble carboydrates were available for fermentation or high oxygen incorporation into the ensiled material prevented a further decrease in pH and proper preservation of plant material (43, 48). Proximate analyses showed a high dry matter content of 43.2%, and an ash content of 10.3%, suggesting both insufficient consolidation of forage material and a high buffering capacity with low carbohydrate availability. As noted previously the high temperature also is thought to inhibit lactic acid producing bacteria (24, 38).

Initial buffering capacity of the soybean mixture forages, shown in Figure 4, were considerably higher than the other forage materials. The increases in buffering capacity for SBGS and SBPM were 69 and 64.2%. This was much lower than the change found in CSC, CSA, and WH of 133.5, 290.5, and 87.4%, respectively. Most of the increase in



Figure 4. The pH of silage samples obtained at 5-day intervals during ensiling.



Figure 5. Buffering capacity of silage samples obtained at 5-day intervals during ensiling.

buffering capacity during ensiling is due to the formation of organic acids which buffer between a pH of 4-6 (52). The small increase in buffering capacity for SBPM corresponds with the insufficient levels of lactic acid production.

### Crude Protein and Ammonia Results

Ensiled corn silage with anhydrous ammonia (CSA) averaged 14.9% crude protein (Table 2). The five samples of corn silage control (CSC) obtained during fermentation averaged 9.9% CP. Protein content of wheat (WH) in this study is higher than those reported by Fisher and Lessard for wheat ensiled at the milk, soft dough and firm dough stages with crude protein values ranged from 7 to 8.3%. Bishnoi (4) found values of 9.9% for Arthur wheat harvested at the bloom stage. The wheat used in this study averaged 13.6% CP but was ensiled at the boot stage, which is the highest plant protein stage for small grain plants (16). The soybean and grain sorghum silage (SBGS) averaged 13.6% CP and the soybean and pearlmillet (SBPM) averaged 11.4% CP.

The day 0 composite sample for CSA was obtained before application of anhydrous ammonia and averaged 9.5% CP. The treated sample at day 5 of fermentation tested 11.7% CP, at day 10 was 18.7% CP, and for days 15 and 20 were 11.8 and 11.8% CP respectively. At day 25 the sample was 20.5% CP. The variability of protein in samples indicates that distribution of the cold-flow anhydrous ammonia (ANAM) was uneven.

Ammonia nitrogen (AN) levels as listed in Table 4, were lower than found by some researchers for corn silage (8, 10, 25) with values ranging from .1% in untreated corn silage to .351% of dry matter in corn silage treated with 1% ANAM. Values were not as high as those found by Buchanon-Smith (8) for ammonia-treated silage in which a value of .49% AN on a dry matter basis was obtained in 40% DM corn silage treated with 1% ANAM and testing 13.13% CP. In this study the high AN samples on days 10 and 25 in the CSA material contained .282 and .351% AN and tested 18.7 and 20.5% CP.

Concentrations in the other silages tested .061 - .131 AN, and were somewhat lower the than values obtained for grasses and small grains by Brown et al. (7), for which most samples were .1-.13% AN.

Ammonia nitrogen levels were high in the composite samples. The reason for this is unknown. Samples had been exposed repeatedly to room temperature for brief periods as AN tests were among the last run. Fermentation samples were lower in CSA than found in silages in some previous studies (7, 26) but were similar to those found by others (8, 25). The use of enzymatic determination of AN concentrations in silage samples was not compared to standard AN analyses for accuracy.

Da	ays	% DM	% Ash	% CP	% EE	% CF	% AN	AN% TN1
					(% of	dry matter) -		
0	1	36.6	8.23	13.0	2.5	20.4	0.116	5.57
5		37.2	7.81	13.2	2.7	19.8	0.127	6.01
10		37.7	7.67	13.4	4.2	20.2	0.121	5.72
15		35.7	6.66	14.0	4.2	24.5	0.126	5.63
20		35.5	7.68	13.4	3.9	24.9	0.131	6.20
25		35.0	8.27	14.2	4.0	26.7	0.117	5.15
CSA <sup>2</sup>								
0	1	38.8	4.49	9.5	1.2	18.1	0.115	6.91
5		36.0	5.41	11.7	4.9	23.6	0.121	7.27
10		36.0	3.96	18.7	5.4	21.0	0.282	9.43
15		38.2	4.81	11.8	4.5	22.1	0.116	6.14
20		37.2	5.35	11.8	3.8	23.5	0.119	6.30
25		35.4	4.30	20.5	5.7	23.2	0.351	17.83
CSC2							0.001	11105
0	1	38.2	4 27	10.0	28	170	0 109	6.81
5		413	3 73	10.1	3.9	16.9	0.106	6.90
10		415	3.99	94	50	18.1	0.100	7.24
15		413	4 26	10.0	60	197	0.105	6.63
20		40.7	4 52	98	5.4	183	0.088	5.61
25		44 5	4 31	9.8	61	20.8	0.0007	6 10
w/u2			4.51	2.0	0.1	20.0	0.077	0.19
	1.1.1.1	45.1	6 75	127	25	225	0.000	2.04
5		43.1	6.79	11.7	3.5	33.3	0.060	3.94
10		39.6	7.09	12.7	5.7	29.0	0.001	5.75
10		38.0	7.98	13.7	5.7	32.4	0.120	5.75
10		44.0	7.13	14.4	5.2	33.1	0.100	4.50
20		43.9	7.81	14.1	5.1	33.8	0.055	2.44
25		36.0	7.92	15.6	5.2	36.5		
SBPM <sup>2</sup>								
0	1	44.0	9.56	12.3	4.8	34.1	0.076	3.86
5		44.3	10.49	12.3	3.5	40.2	0.091	4.62
10		39.4	8.71	12.6	5.2	35.4	0.101	5.01
15		43.6	8.40	15.6	3.7	37.3	0.089	3.57
20		43.9	10.50	14.2	5.3	39.8	0.090	3.96
25		45.0	7.30	14.5	5.0	34.4	0.086	3.71

TABLE 4. Proximate and ammonia-nitrogen (AN) analyses of forage samples obtained at 5 day intervals during ensiling.

<sup>1</sup>Ammonia-nitrogen as a percentage of total N.

<sup>2</sup>Abbreviations for silages are as follows: SBGS, soybeans/grain sorghum; CSA, corn silage with anhydrous ammonia; CSC, corn silage control; WH, wheat; SBPM, soybean/pearl millet.

<sup>3</sup>Values not obtained.

#### Volatile Fatty Acid Results

Forage samples were analyzed for the volatile fatty acids, acetic, propionic, and butyric. Concentrations of acetic acid (Figure 6) tended to be lower than reported in the literature (32, 54), but were consistent in trend as expected by results of other analyses on the samples. For the CSC formation of acetic acid was essentially complete on sample day 5 with only small increases occurring thereafter. This is as expected because of the low buffering capacity and high levels of readily-available soluble carbohydrates of corn silage. In the remaining silage samples significant formation of acetic acid was delayed, to day 15 for SBGS, CSA, and WH and to day 20 for SBPM. The higher levels of acetic acid found in the latter silage are indicative of an abnormal fermentation wherein lactic acid is catabolyzed and acetic acid production increases (36, 65). Acetic acid normally increases in silage until the pH declines to the point that favors lactic acid bacteria (41). If insufficient pH decline occurs then higher levels of acetic acid are produced. Normally it is expected that increased levels of butyric acid would also be produced but in the SBPM this did not occur. Increased concentrations of acetic acid were also noted in samples of CSA on days 10 and 25, due to the high concentration of ANAM applied to these samples, resulting in increased buffering capacity, and a delay in the lowering of pH. The WH contained low levels in all samples.

Concentrations of propionic acid were highest in CSC silage with the exception of high concentrations in the high ANAM sample on days 0 and 5 of CSA and increases on day 25 for both SBGS and WH.

Traces of butyric acid were found in SBGS composite samples, WH silage from days 5 and 15, and in composite samples of SBPM. Butyric acid production is usually the result of a clostridial fermentation (36, 41).

### Total Lactic acid, L(+) and D(-) Lactic Acid Isomers

The WH, SBPM, and SBGS silages were similar in regard to production of lactic acid (Figure 7). The CSA day 10 and 25 samples were well above the other silages in lactic acid content. Values for SBPM were similar to the SBGS, WH and CSC except for the 25 day sample which was rather low in lactic acid. The day 15 and 20 of the CSA were also low in lactate.

Where high concentrations of ammonia occurred in the CSA fermentation was prolonged as indicated by the excessive amounts of lactic and acetic acids and the low pH in these samples. The low buffering capacity of the CSC is indicated by the normal concentrations of lactate and acetate but more acidic pH when compared to the other forages. Values for L(+) lactic acid agree with the data of Schaadt and Johnson (51). They ranged from .2 to .523 in fresh forage to 5.66% of dry matter in the day 25 of the CSA.

Total lactic acid was analyzed by gas chromatograph, L(+) lactic acid isomer by an enzymatic method, and the D(-) isomer by subtraction. Results from the total lactic acid tests appeared to be unreliable; therefore, results for these and the concentration of D(-) lactic acid are unavailable. Day 0 samples from total lactic acid analysis were relatively high in lactic acid concentration while results from the L(+) lactic acid determinations were almost insignificant as expected in unfermented forage material. The small amounts of acids found were the result of holding of composite samples in plastic bags as the silo was filled. Total lactic acid values did follow the general pattern as seen in the L(+) results with the majority of lactic acid formation complete by day 10. Overall, levels were lowest in SBPM, indicating a deficiency of carbohydrates needed for desireable microbial growth and production of volatile fatty acids. The SBPM samples were also high in ash and crude fiber. Earlier cutting of plant material would very likely aid in enhancing the fermentation of such silage in the future. The days 15 and 20 of CSA were also low in lactic acid content. However, as pH was low this would indicate that buffering capacity was low and fermentation rapid and complete in these samples. Days 10 and 25 of CSA were very high in comparison, due to the sustained activity of lactic acid producing bacteria, which continues until either carbohydrates are no longer available or silage pH drops to a level at which the bacteria can no longer grow and function (36, 65). High buffering capacity has been noted to increase production of lactic acid (2, 8, 32, 36, 48, 54, 65).



Figure 6. Acetic acid concentrations of silage samples obtained at 5-day intervals during ensiling.



Figure 7. L(+) Lactic acid concentrations of silage samples obtained at 5day intervals during ensiling.

#### SUMMARY

A comparison of the relationship of fermentation to chemical composition was made for forages which were wilted and ensiled at 35 to 45 percent dry matter. Trial I consisted of three forages ensiled in October, 1981: interseeded soybeans and grain sorghum, whole plant corn with added anhydrous ammonia, and whole plant corn with shelled corn added at a rate of 150 kg/t of fresh forage. Trial II consisted of two forages ensiled in 1982: interseeded soybeans and pearlmillet, and wheat. Temperatures of fermentation were collected, and chemical composition during the first 25 days of fermentation analyzed.

Production data were also collected in Trial I. The cash expenses and yields do not indicate a significant advantage to either crop in this study in terms of yields and return over variables. In terms of plant nutrient content there was an advantage for soybean/grain sorgum silage in protein yield of 854 kilograms per hectare as compared to whole plant corn silage.

Temperature data collected on the forages in Trial I covered 57 days from October 8 through December 4. The maximum recorded temperatures for soybean/grain sorghum, corn silage with added anhydrous ammonia and the corn silage control were 37, 37, and 24 degrees Celcius, achieved by day 11, 4, and 11 of ensilement respectively. The rapid temperature increase for material treated with anhydrous ammonia confirmed previous reports. Small fluctuations occurred in silage temperatures but these were not correlated to ambient temperature. Temperatures declined very slowly in all forages, with the lowest reading for silages by day 57 recorded at 19 C for soybean/grain sorghum silage. Ambient temperature was not reached in the ensiled mass during the 57 day period in which data were recorded.

The soybean/pearlmillet ensiled in Trial II quickly reached a high peak temperature of 44 C at 4 days of ensilement, gradually decreasing over a 57 day period to 35 degrees on October 19. The silage did not reach a desireable pH. Initial forage pH was high, 7.4, dropped rapidly to 5.1 on day 5, but did not decline further in succeeding samples. Samples were low in lactic acid and high in acetic acid content.

Initial buffering capacities for both of the soybean mixture forages were considerably higher than the other forage materials. Buffering capacities at day 0 for whole plant corn, whole plant corn treated with anhydrous ammonia, soybean/grain sorghum, soybean/pearlmillet and wheat forages were 19.4, 20.0, 35.3, 35.3. 38.6 and 22.2 meq/100 g of dry matter respectively. Increases in buffering capacities during fermentation were smaller for soybean/grain sorgum and soybean/pearlmillet. Buffering capacity increases for the forages were 133.5, 290.5, 69.1, 64.2 and 87.4%.

High ammonia nitrogen levels were found in samples of whole plant corn silage with added anhydrous ammonia obtained on days 10 and 20 of ensilement. These contained .282 and .351% ammonia-nitrogen and tested 18.0 and 20.1% crude protein. The increase in buffering capacity which occurrs with addition of anhydrous ammonia was confirmed by the higher acetic acid and pH levels, with corresponding decreases in lactic acid from samples taken days 10 and 25. Crude protein levels were 17.9 and 17.4 percent while the other samples contained only 10-12% crude protein. It appeared that application of anhydrous ammonia was not uniform throughout the silage. Where high concentrations of ammonia occurred fermentation was prolonged as indicated by excessive amounts of lactic and acetic acids and a high pH in these samples. Values for L(+) lactic acid ranged from .2 to .523 in fresh forage to 5.66% of dry matter on day 25. Concentrations of ammonia-nitrogen in the other silages ranged from .061 to .131%.

The low buffering capacity of whole plant corn silage was reflected by normal concentrations of lactate and acetate but a more acidic pH when compared to the other forages. Lactic and acetic acid production for soybean/grain sorghum silage was similar to that of the corn silage control. Corn silage pH was lower, however, throughout fermentation and reached a stable pH by day 5 of fermentation. The wheat silage went through a gradual fermentation with low lactic acid production, and an intermediary ending pH of 4.0.

Fermentation was essentially complete by day 10 in all silages as indicated by pH, buffering capacity, and lactic acid production; however, there was a tendency for buffering capacity and acetic acid content to increase in all of the ensiled materials throughout the 25 day collection period.

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#### APPENDIX

Date	CSC1	CSA <sup>1</sup>	SBGS <sup>1</sup>	SBPM <sup>1</sup>	WH1
0	5.0	5.3	6.0	7.4	5.8
5	3.7	4.4	4.7	5.1	5.1
10	3.8	5.3	4.2	5.1	4.1
15	3.7	4.2	4.3	5.1	4.6
20	3.8	4.2	4.3	5.0	4.5
25	3.7	4.7	4.2	5.0	4.0

TABLE 1-A. Changes in pH with time ensiled for silage samples obtained at 5-day intervals during ensiling.

<sup>1</sup>Abbreviations for silages are as follows: SBGS, soybeans/grain sorghum; CSA, corn silage with anhydrous ammonia; CSC, corn silage control; WH, wheat; SBPM, soybean/pearl millet.

Date	CSC1	CSA <sup>1</sup>	SBGS <sup>1</sup>	SBPM <sup>1</sup>	WH1
		(mi	lliequivilants/10	0 g DM)	
0	19.4	20.0	35.3	38.6	22.2
5	39.2	26.6	39.2	49.7	26.4
10	42.4	53.9	47.6	66.0	28.4
15	44.2	27.9	56.8	59.6	25.0
20	42.1	28.5	45.9	63.8	38.7
25	45.3	78.1	59.7	63.4	41.7
% increase <sup>2</sup>	133.5	290.5	69.1	64.2	87.4

TABLE 2-A. Changes in forage buffering capacity at 5-day intervals during ensiling.

<sup>1</sup>Abbreviations for silages are as follows: SBGS, soybeans/grain sorghum; CSA, corn silage with anhydrous ammonia; CSC, corn silage control; WH, wheat; SBPM, soybean/pearl millet.

<sup>2</sup>Increase in buffering capacity from forage day 0 to day 25 of ensilement.

Sample	% L(+)	% Acetic	% Propionic	% Butyric	
			% of DM )		
SBGS <sup>1</sup>			,		
0	0.14	0.523	0.308	0.288	
5	1.91	0.968	0.440	2	
10	3.03	1.036	0.222		
15	2.65	1.480	0.149		
20	2.81	1.481	0.307		
25	3.43	1.284	0.737		
CSA <sup>1</sup>					
0	0.13	0.399	0.611		
5	1.15	0.40	0.622	2	
10	3.76	1.926	0.188		
15	1.17	1.044	0.279		
20	1.31	1.037	0.254		
25	5.66	2.35	0.334		
CSC1					
0	0.21	0.468	0.374		
5	2.28	1.164	0.149		
10	2.94	1.281	0.536		
15	2.51	1.268	0.512		
20	2.54	1.352	0.429		
25	3.00	1.427	0.107		
WH1					
0	0.19	0.221	0.260		
5	0.79	0.396	0.327	0.140	
10	2.76	1.297	0.205	0.133	
15	2.05	0.990	0.259		
20	2.21	1.072	0.116		
25	2.36	2.55	1.028		
SBPM1					
0	0.28	0.941	0.266	0.163	
5	1.76	2.022	0.187		
10	2.18	2.730	0.195		
15	1.45	3.440	3		
20	2.26	2 376	3		
25	1.33	3.337	0.287		
10 15 20 25	2.18 1.45 2.26 1.33	2.730 3.440 2.376 3.337	$ \begin{array}{r} 0.195 \\ 3 \\ -3 \\ 0.287 \end{array} $		

TABLE 3-A. Concentration of lactic acid (LA) and volatile fatty acids (VFA) of silage samples obtained at 5 day intervals during ensiling.

<sup>1</sup>Abbreviations for silages are as follows: SBGS, soybeans/grain sorghum; CSA, corn silage with anhydrous ammonia; CSC, corn silage control; WH, wheat; SBPM, soybean/pearl millet.

<sup>2</sup>Trace amount.

<sup>3</sup>Values not obtained.

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