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EFFECT OF MILES LABORATORY COMMERCIAL INOCULANTS, ON ALFALFA, WHOLE PLANT CORN AND HIGH MOISTURE GRAIN SORGHUM SILAGES

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INTRODUCTION

The main objective in the conservation of any crop is to preserve it at the optimum stage of growth for use during those seasons when the crop is unavailable.

Haymaking has long been the traditional technique of preserving forage, but the need to delay harvesting until a mature stage of growth of relatively high dry matter content was reached, meant that the crop was of low digestibility before it was dried.(McDonald , 1981). This fact together with weather changes, frequently resulting in high losses during the drying process, led to a product of variable composition and also of low nutritional value. Early interest by farmers in conserving crops by natural fermentation as silage stemmed from the realization that this process was less dependent of the weather and the need to harvest crops at an advanced stage of growth.(McDonald , 1981).

Silage is the material produced by the controlled fermentation of a crop of high moisture content. Studies performed by McDonald (1981) indicate that the first essential objective in preserving crops by natural fermentation is the achievement of anaerobic conditions. The most efficient way to accomplish this is to store the material in a hermetically sealed container and under these conditions, the oxygen, trapped by the herbage, would rapidly be removed by respiratory enzymes in the plant.

The second main objective as indicated by McDonald (1981)

is to discourage the activity of the clostridia. These bacteria are usually present on harvested forage in the form of spores, but start to multiply as soon as conditions in the silo become anaerobic. The growth of these organisms is undesirable as they produce butyric acid and degrade amino-acids to a variety of products which are of poor nutritional value. The more common way of inhibiting clostridial growth is to promote lactic acid fermentation to produce an environment which is sufficiently acid (McCullough, 1978). Clostridia is also very sensitive to water availability and requires very wet conditions for active growth.

Stirling and Whittenbury (1963) examined the lactic acid bacteria occurring on growing plants and found members were usually less than 100 colonies/g in fresh grass and were limited to decayed, dead or bruised material. Numbers increased rapidly after cutting.

McCullough (1978) points out that the preservation of crops by high moisture content present considerable problems. If such materials are exposed to the air, microbial activity involving yeasts, fungi and bacteria takes place and subsequently results in high gaseous losses of dry matter (up to 75% in extreme cases). The product is usually inedible and may be toxic. Even when anaerobiosis is achieved, the stored material is still vulnerable to spoilage by the anaerobic bacteria.

The purpose of this study was to evaluate a commercial test culture provided by Miles Laboratories Inc. in wet and dry forms on the silage performance of alfalfa, whole plant corn and high moisture sorghum grain.

LITERATURE REVIEW

The principal objective in the conservation of a particular crop is to preserve it at the best stage of growth for use during the time when the crop is not available.

According to McDonald (1981), haymaking (cutting and drying of grass and herbage for livestock feeding) has been the traditional technique of crop preservation. Weather changes resulting in high moisture losses during the drying process causes a product of low nutritional value and variable composition. People became interested in preserving crops by natural fermentation as silage, realizing that this process was not as dependent upon the need to harvest crops at an accelerated stage of growth and weather.

McDonald (1981) defined silage as the material produced by the controlled fermentation of a crop of high moisture content. The term silo is of ancient origin and historically means a grain pit (McCullough, 1978), or hole sunk in the ground for storing corn (McDonald, 1981).

Barnett (1954) defined the objective of silage making as "to achieve within the ensiled mass a sufficient concentration of lactic acid, produced as a result of the presence of microorganisms within the cut crop, to inhibit other forms of microbial activity and thus preserve the material until such time as it is required".

An ideal silage fermentation should occur when the forage has 28 to 34% dry matter; a minimum buffer capacity; a large population of lactic acid producing bacteria; a temperature and degree of compaction suitable for an immediate bacteria population;

adequate level of water soluble carbohydrates (WSC) (Bolsen, 1978). Under these conditions the main use of carbohydrates is for lactic acid production.Techniques as prewilting, chopping and excluding air are also very important in silage fermentation (Zimmer, 1979).

Advantages in the silage making process as a means of preserving crops are 1) Reduced weather damage at harvest--Zimmer (1977) observed that as dry matter content increased because of method of harvesting, the amount of dry matter lost in the field increased also, 2) Improved cropping practices--silage doubles and sometimes triples capital and land use demands (Hinds, 1983), 3) Improved feeding programs--silage is chopped for convenient mixing, supplies a source of feed of steady composition and its moisture content provides a perfect medium to maintain dry grain in the mixed feed (McCullough, 1978), 4) Total mechanization--the facility to mechanize the whole harvesting, storing and feeding operation over the haymaking process, and 5) Harvesting maximum nutrients per acre--silage production increases the percentage of nutrients that are harvested.

The silage making process also has a few disadvantages: 1) Silage demands efficient management--10 to 30% of the gross energy in silage can be lost before it is ingested by animals, (Zimmer, 1979), 2) Silage does not move in the regular market routes, therefore it is difficult to assign a value for it, 3) Silage holds high amounts of moisture compared to haylage, thus its cost of transportation and handling is high, and 4) Silage has a short shelf-life once removed from the silo.

I. Microbiology of Silage

A. Role of lactic acid bacteria

The first attempt of a classification of the lactic acid bacteria was performed by Orla-Jensen (1919) who defined them as micro-aerophilic, gram-positive, non-spore-forming organisms which ferment sugars to lactic acid. He separated them in two classes, the homofermentative lactic acid bacteria (homolactics) which ferment hexoses to lactic acid, and the heterofermentative lactic acid bacteria (heterolactics) which ferment hexoses to lactic acid, ethanol and acetic acid. The microorganisms associated with silage are listed below:

Classification of lactic acid bacteria important in silage. (McDonald, 1981)

A. Homofermentative

Rod

Lactobacillus casei
Lactobacillus coryniformis subsp. *coryniformis*
Lactobacillus curvatus
Lactobacillus plantarum

Coccus

Pediococcus acidilactici
Pediococcus cerevisiae
Pediococcus pentosaceus
Streptococcus faecalis
Streptococcus faecium

B. Heterofermentative

Rod

Lactobacillus brevis
Lactobacillus buchneri
Lactobacillus fermentum
Lactobacillus viridescens

Coccus

Leuconostoc cremoris
Leuconostoc dextranicum
Leuconostoc mesenteroides

Bucker (1970) reported that the majority of 2,124 strains of lactic acid bacteria isolated from corn silage at different stages of maturity were members of the subgenera Streptobacterium and Betabacterium. Only 3 and 4% respectively of the total number belonged to the genera Leuconostoc and Pediococcus. Beck (1972) studied 1,600 strains of lactic acid bacteria isolated from grass and alfalfa silage and also found low percentages of Pediococcus (7.5%) and Leuconostoc (4.5%).

The biochemical mechanisms and the pathways of homo and heterofermentative lactic acid production differ distinctly. The pathway of homofermentation of hexoses, according to the Embden-Meyerhof scheme, leads to pyruvate in its initial stages. After it, pyruvate is hydrogenated producing lactic acid (Franke and Buchta, 1960). Fred et al., (1921) reported that the accumulation of acetic acid in silage is due to the activity of heterofermentative lactobacilli. Heterofermentation according to the Dickens-Horecker-mechanism is initiated by an asymmetric split of the glucose into 1 molecule of CO_2, 1 molecule of ethanol and 1 molecule of lactate.

Lactobacilli ferment a number of carbohydrates: the main product of fermentation, lactic acid can be in the D(-), L(+) or DL(-) isomeric form (McDonald, 1981). Most lactobacilli (L. curvatus, L. plantarum, L. brevis, L. buchneri, L. fermentum, and L. viridescens) produce mainly DL (-) lactic acid. Wilson and Miles (1975) reported that lactobacilli have complex nutritive requirements for peptides, amino acids, vitamins, fatty acids, nucleic acid derivatives, salts and fermentable carbohydrates. They also pointed out

that lactobacilli have little effect on proteins and if any proteolytic action occurs, it is very slight.

B. Sources of lactic acid bacteria

Plant materials carry a variety of bacteria depending on the crop and environmental conditions. Kroulik et al. (1955a) showed that fresh green alfalfa contained a variable number of microorganisms in the same field during the same day. These variations did not correlate with the time of day. Different numbers of microorganisms were found on different parts of the same plant at a given time. Corn tassel had more bacteria than corn leaves. Orchard grass heads were lower in counts of microorganisms than orchard grass leaves.

Kroulik et al. (1955b) found variations in the numbers of microorganisms on different kinds of plants as follows:

Numbers of microorganisms on different kinds of plants. (Kroulik et al., 1955b)

Plant	Total Count/g	Coliform Count/g	Molds & Yeasts/g
Horse tail	494	1,000	228
Oats	89	1,000	280
Orchard grass	340	<10	4,400
Thistle	2,600	1,000	1,000
White clover	11,780	10	13,200
Wild mustard	21,400	1,000	3,652
Wild onion	120	<10	500

Although there were many variations in the microbial populations on green plants, there was a tendency for the microorganisms to increase with the maturity of the plants and advancement of the season as shown below:

Number of microorganisms with advance in growth period or increase in maturity of plant (Kroulik et al., 1955b).

Plant	Date	No. of Samples	Total Count/g	Coliform Count/g	Mold & Yeasts/g
Alfalfa	4/17-30/51	5	1,804	1,000[a]	580[a]
Alfalfa	5/4-24/51	7	26,383	1,000[a]	763[a]
Alfalfa	6/1-27/51	5	62,116	1,000[a]	14,560[a]
Oats	6/2/53	1	2,600	10,000	1,000
Oats	6/9/53	1	3,350	10,000	15,300
Oats	6/29/53	1	23,100	10,000	26,000
Sour dock	5/25/51	1	28	<10	100
Sour dock	5/29/51	1	1,020	<10	2,848
Sour dock	6/14/51	1	6,960	1,000	680
Sour dock	6/19/51	1	4,840	1,000	320

a = Average of samples studied

A definite increase in numbers of microorganisms was attendant with wilting and chopping of plant material in preparation for ensiling. The predominating microorganisms on fresh green plants were pigmented, aerobic, non-sporeforming, rod-shaped bacteria. The authors reported that relatively few of the microorganisms isolated from fresh green plants were similar to bacteria present in silage, and none were typical of lactobacilli found in silage. The microorganisms found to be similar to the silage bacteria appeared to increase in chopped forage previous to ensiling.

Stirling (1951) reported that the lactic acid bacteria are not usually part of the normal microflora of the growing plant. Nilsson (1956) found very few lactic acid bacteria on aseptically harvested plants suggesting that either an antibiotic action of other microorganisms on the plant surface restrained the growth of the lactic acid bacteria, or that the introduction of these bacteria during harvesting

of the crop accounted for their rapid appearance in silage.

Stirling (1951) stated that when the crop is gathered, the numbers of lactic acid bacteria are influenced by the amount of partially decayed material cut near ground level. Once the crop is cut these bacteria can be spread by the subsequent handling, especially if the plant sap is released by chopping, bruising or lacerating the material. The numbers of organisms found on the harvesting equipment indicate the ability of lactic acid bacteria to multiply where plant sap collects, and the equipment, therefore, seems the means through which the microorganisms can be spread.

C. Role of other microorganisms in silage

1. Fungi

Fungi are eukaryotic microorganisms which grow either as single cells, the yeasts, or as multicellular filamentous colonies, the molds (McDonald, 1981). They are in large numbers in ponds, streams, or vegetation soil where they live mainly on decomposed wood and leaves. Fungi obtain nutrients for their growth by the production of extra cellular enzymes as amylases, proteases, lipases and cellulases which breakdown complex organic molecules to simple monomers that can be absorbed through their cell membranes.

Molds are associated with areas of silage as the sides and surface which are exposed to air. Their presence in silage is undesirable because they not only breakdown sugars and lactic acid through normal respiratory pathways,

but also metabolize and hydrolyze cellulose and other cell wall components (McDonald, 1981). In addition, some molds produce substances called mycotoxins which are harmful to animals.

Ohyama and Hara (1975) demonstrated molds as being mainly responsible for the deterioration of grass silages exposed to air. Some of the important molds isolated in silage are: Fusarium spp., Geotrichum candidum, Monascus purpureus, Aspergillus fumigatus, Penicillium roqueforti and Byssochlamys nivea.

Ruschmann and Graf (1932) first reported that yeasts are components of silage microflora. They isolated Mycoderma and Torula from grass and potato silages at different stages of fermentation. The authors believed that yeasts are useful microorganisms in silage in two respects: a) improve silage quality by the production of aromatic substances and b) positively influence and promote the activity of lactobacilli. On the other hand, Beck and Gross (1964) showed that yeasts are most common in silages of poor quality. They found yeasts to be the main reason for the aerobic deterioration of silage after opening the silo.

There are two physiological groups of yeasts occurring in silage: a) ground-growing yeast (sediment yeasts) which ferment sugars, for example Torulopsis, b) top-growing yeasts (pellicle yeasts) with weak fermentation capacity but a high respiration capacity for lactic acid, for example Hansenula, Pichia, and Candida (McCullough, 1978). Burmeister et al. (1965) and Ohyama and Hara (1975) studying high-moisture corn

silage and grass silages, respectively, also confirmed these findings. Coneva and Kirov (1972) found that the predominant yeasts in corn are Pichia and Saccharomyces.

2. Clostridia

Clostridia are Gram-positive, spore-forming, rod shaped, often motile bacteria that grow under anaerobic conditions and ferment sugars, proteins or organic acids (Smith and Hobbs, 1974). The most widely known name for clostridia is butyric acid bacilli.

Activity of clostridia in silage has a definite influence in the end result of silage fermentation. In general, activity of clostridia should be minimized (McCullough, 1978).

The first studies regarding the relationship between activity of clostridia and its action of counteracidification in silage were reported by Kurstein (1918, 1926) and Burri et al. (1919). Early studies of Ruschmann and Harder (1931), Beynum and Pette (1935, 1936, 1939) and Allen and Harrison (1937) indicated two physiological groups of clostridia were found in silage: a) saccharolytic clostridia, such as Clostridium butyricum which ferments carbohydrates and organic acids, having a weak proteolytic activity and b) proteolytic (putrefying) clostridia, such as Clostridium sporogenes which ferments mainly proteins, having a weak or no activity for carbohydrates.

According to Bhat and Barker (1947), Bryant and Burkey (1956), Winkler (1957), Kutzner (1963), Gibson (1965) and Azeezullah et al. (1973) the most common butyric acid

producing organisms in silage are C. tyrobutyricum, C. butyricum, C. sphenoides, and C. sporogenes and very seldom C. perfringens, C. bifermentans and C. tetanomorphum.

3. The Enterobacteriaceae

Enterics are Gram-negative, non-spore-forming, facultative anaerobic, rod-shaped, typically motile bacteria that ferment carbohydrates (McCullough, 1978). The enterics occurring in silage usually belong to the Coli-Aerogenes group of bacteria. The main characteristic of the non-faecal bacteria of the coliform group is the fermentation of lactose, producing larger amounts of CO_2.

Enterics in silage were first studied by Ruschmann and Harder (1931). They determined that the production of acetic acid in the first fermentation step was caused by this group of microorganisms and thus, named them "acetic acid bacteria". This discovery was supported by Allen and Harrison (1936) but they named the group "coliform" bacteria. This group of microorganisms are normally active only in the first step of fermentation.

Studies performed by Stirling (1951), Langston et al. (1962), Weise (1963), Gouet and Chevalier (1966) established that coliform bacteria are undesirable microorganisms in silage because they compete with lactic acid bacteria for fermentable sugars. Fermentation products were found to be acetic acid, CO_2, lactic acid and to a less degree ethanol, 2,3-butanediol and H_2, depending upon temperature and pH. Generally they do not attack complex proteins and have weak

proteolytic activity. On the other hand, bacteria on the coliform group are capable to decarboxylize and desaminize amino acids.

II. Aerobic Deterioration and Heating of Silage

A good silage fermentation and preservation is the result of anaerobic processes. There is an unavoidable entrapment of air in the crop during silo filling. Mostly, aerobic organisms (fungi and bacteria) jointed with plant respiration produce carbon dioxide that replaces free oxygen in the silo (McDonald, 1973; Watson and Nash, 1960).

Generally the water soluble carbohydrates (WSC) are oxidized during this aerobic phase. The more oxygen entrapped in the ensiled crop or the longer sealing is delayed, the more WSC oxidized; thus less is available to initiate and sustain an active lactic acid fermentation (McCullough, 1978). Kearney and Kennedy (1962) simulated conditions of delayed silo sealing to demonstrate this. They accounted that 50% of the water soluble carbohydrates content of chopped forage were lost in a 30 hour period.

Ruxton and McDonald (1975) ensiled ryegrass which had either 0.26 or 6.59 ml of oxygen per g of fresh herbage to simulate entrapment of different levels of oxygen. The high oxygen treatments had greater oxidation of water soluble carbohydrates, higher pH, volatile nitrogen and butyrate levels and remarkably lower lactic acid levels relative to the low oxygen content herbage. It was apparent that oxygen inclusion indirectly led to greater clostridial activity and variation in fermentation patterns, producing an undesirable

high pH silage instead of a stable low pH silage (Ruxton et al.,1975).

Besides achieving anaerobiosis, it is also important to sustain anaerobiosis. An increase in dry matter losses from 10 to 17% has been reported when continuous oxygen is infused into silage. The effect of air is limited to the area immediately surrounding the point of infiltration (Ruxton and McDonald, 1975; Meiering, 1975).

Exposure of silages to air should be diminished when the silo is opened for feeding. Silage can deteriorate with microbial growth and surface heating during this period (Ohyama and McDonald, 1975). Wilkins (1975) reported that there is a loss of soluble carbohydrates (WSC), starch structural carbohydrates, and short-chain organic acids (mainly lactate), while pH and ammonia content increase. Wilkins (1975), Ohyama and Hara (1975), Ruxton et al. (1975), and Zimmer (1971) agreed that mainly yeasts, and molds in a lesser degree, are involved in the secondary fermentations.

Abnormal silages are generally more stable than normal silages to aerobic deterioration. Consistently abnormal silages have a higher content of butyric and propionic acids, and total volatile fatty acids which are good inhibitors of yeasts and molds (Huber and Soejono, 1976; Ohyama et al., 1975; Woolford, 1975). Normal silages with high lactic acid content and low pH are more prone to aerobic deterioration because yeasts absorb lactic acid and can tolerate acid conditions. In most cases losses due to aerobic deterioration can be limited by sufficient pack storage in a gas tight

structure, rapid sealing and rapid removal of material from the silo during feeding.

Aerobic and anaerobic fermentations in silage produce heat. The energy involved from fermentation is transferred to ATP. The ATP produced per mole of glucose or fructose is only 1 to 2 moles in anaerobic systems. Thirty-eight moles of ATP are derived from aerobic fermentation of the same carbohydrates as shown below.

Aerobic and anaerobic fermentation of hexose. (McDonald, 1973)

Aerobic:

$$\text{glucose} + 6\ O_2 \text{ -------> } 6\ CO_2 + 6\ H_2O + 38\ \text{ATP}$$

Anaerobic-homofermentative:

$$\text{glucose} \text{ -------> } 2\ \text{lactate} + 2\ \text{ATP}$$

Anaerobic-heterofermentative:

$$\text{glucose} \text{ -------> } 1\ \text{lactate} + 1\ \text{ethanol} + CO_2 + 1\ \text{ATP}$$

$$\text{fructose} \text{ -------> } 1\ \text{lactate} + 1\ \text{acetate} + CO_2 + 2\ \text{ATP}$$

It is hard to differentiate between the direct effect of temperature of ensiling and the effect of oxygen whose respiration causes rise in temperature (Ruxton et al., 1975). Wieringa (1960) reported that maximum butyric acid production occurred at 35°C when silages were incubated at temperatures from 20 to 60°C. He concluded that a high temperature must be avoided. Ruxton et al. (1975) showed that clostridia

and coliforms show optimal growth at 37°C. De Man (1953) compared silages kept at 35°C and 20°C and found that the silage kept at 35°C had less acid, higher pH, more ammonia and butyric acid; therefore it was always less desirable than the silage kept at 20°C.

The best temperature for growth of the lactic acid bacteria is alike to the optimum for coliforms and clostridia (Gibson et al., 1958; Murdoch, 1960). However, the lactic acid bacteria are more tolerant of temperatures lower than 30°C than clostridia or coliforms (Gibson et al., 1958; Ruxton et al., 1975).

High temperatures of silage indicate that water soluble carbohydrates are oxidized. This oxidation together with high temperatures generates a more desirable environment for clostridia, and a less desirable environment for the lactic acid bacteria.

The digestibility of proteins is reduced in the presence of oxygen at high temperatures (Ruxton et al., 1975; Wieringa, 1960; Wieringa et al., 1961). Darrah et al. (1976), Noller et al. (1966), and Van Soest (1962) agreed that the cause of reduced protein digestibility is the Maillard reaction, also named the non-enzymatic browning reaction. In this reaction, hemicellulose and carbohydrate degradation products condense with protein. Brown color and carmelized odor are physical indicators of protein damage in silages.

Protein damage occurs in the presence of oxygen and heating of silage. The longer the heating the more the protein damage. The rate of damage increases with temperature. Reductions in

protein digestibility are drastic above 50°C (Murdoch, 1960). Wieringa et al. (1961) concluded that temperature of silages should be kept below 30°C to reduce protein damage. Although excessively heated silages are readily consumed by livestock, a good proportion of their food value is lost.

III. Microbial Cultures as Silage Additions

Since the early 1900's when French researchers added Lactobacillus cultures to beet pulp silage, decreasing its butyric acid content and producing a more pleasant aroma, there was the possibility of aiding silage acidification and therefore fermentation (Bolsen, 1978).

Watson and Nash (1960) performed a study using microbial cultures for ensilage and found the results to be variable from one experiment to another. On the other hand, Owen (1971) stated "under certain conditions microbial cultures may reduce pH, increase lactic acid and improve the possibility of success in silage making".

Moon et al. (1981) prepared silages from wheat (early bloom), alfalfa (second cutting 20 to 40% bloom), and corn (early dent). They inoculated 5g/kg with a commercial silage additive (Sila-bac) containing Lactobacillus acidophilus and Candida sp. They found that the addition of inoculum had a beneficial effect on numbers of lactobacilli recovered, no significant effect on pH and a negative effect on fermentation acids. The inoculant apparently had little effect in controlling fermentation, and lactic acid bacteria, although present, was unable to cause rapid pH declines, high lactic acid and silage stability. They

suggested that the lack of beneficial effects in the silages could be caused by several reasons: the inoculum was not viable, did not compete successfully with normal flora or did not have sufficient nutrients available for growth. If one of these factors is limiting, silage fermentation is inferior.

Ely et al. (1981) studied effects of inoculation of alfalfa, corn, sorghum and wheat forages with Lactobacillus plantarum (10^7/g silage). Proximate analysis, pH, neutral detergent fiber, acid detergent fiber, soluble carbohydrate, lactic acid, volatile fatty acid contents, total facultative anaerobes, lactobacilli, lactic acid, cocci, yeasts and mold populations were measured in control and treated silages. They reported that total facultative anaerobes were increased in all silages by addition of L. plantarum, and numbers of lactobacilli were increased in all but corn silage. Yeasts and molds were lower only in inoculated alfalfa and wheat silages. The effect of the inoculum was observed on chemical composition for alfalfa and wheat silages only. These treated silages had a lower pH, higher lactic acid content and greater recovery of dry matter, crude protein, detergent fiber (alfalfa) and nitrogen-free extract (wheat). Inoculation had a positive effect on nutrient recovery in alfalfa and wheat silages. They suggested that it is possible that the beneficial effect of an inoculum on nutrient recoveries would be greater if conditions for fermentation were not controlled as carefully, based on the study performed by Ohyama et al. (1975) in which they observed a greater

positive effect of inoculation when silages were sealed improperly and air was introduced.

Burghardi et al. (1980) conducted a study in which they added two different levels, 4.5×10^7 and 22.5×10^7 live organisms per kilogram wet silage of Lactobacillus bulgaricus, L. acidophilus, L. brevis, Streptococcus lactis and S. cremoris to corn silage. They reported that recoveries of DM and crude protein were not affected by inoculation. Nonprotein nitrogen values for treated silages did not differ significantly from those for controls. All microbial treatments tended to increase NH_3-N levels, with L. bulgaricus, S. lactis and S. cremoris resulting in the highest ones. Lactic acid, acetic acid and ethanol concentrations were not affected by microbial treatment. It appeared that time required to complete 95% of total gas production was decreased by Aspergillus oryzae, L. acidophilus and S. lactis. Cell wall constituents, acid detergent fiber and pH were not significantly affected by level or type of organism. They also studied a commercial line bacterial silage additive that contained Bacillus subtilis, L. acidophilus, A. oryzae and mixed lactic ferment enzymes and found out that it did not improve the feedlot performance of steers or digestibility in lambs fed corn silage. Results from these trials showed that the added organisms increased protein degradation and did not result in higher quality corn silage.

Moon et al. (1980) inoculated fresh alfalfa, corn, sorghum, and wheat with L. plantarum at a population of 1×10^6/g. Samples were evaluated for facultative anaerobic

bacteria, lactobacilli, streptococci, lactate utilizers, yeast and molds. All inoculated silages had higher levels of lactic acid and in alfalfa, and wheat, a more rapid rate of pH decline and a lower final pH was observed. Populations of lactobacilli were higher in inoculated alfalfa, wheat and corn silages. They concluded that inoculation with L. plantarum increased the rate of fermentation early, but few differences were noted in silage quality between inoculated or control silages at the end of the 32 day ensiling period.

Rakshit and Voelker (1981) studied the effects of two different cultures on alfalfa silage, corn silage and corn stillage. Culture 1 contained A. oryzae, B. subtilus, lactic acid starter culture, ethoxyquin and some minerals. Culture 2 contained A. oryzae, L. plantarum, potassium sorbate and ethoxyquin. They reported that the cultures increased lactic acid, reduced pH and dry matter losses in alfalfa and corn silage. The cultures showed no apparent effects on proximate analyses of silages. Stability of treated corn silage was improved by cultures, based on silage weights, pH and temperature changes.

A study performed by Huber et al. (1981) with freshly wilted alfalfa treated with 0.05% B. subtilus, dried enzyme soluble, 0.05 or 0.15% lactic acid inocula showed that microbial inocula hastened and stimulated lactic acid production but did not reduce proteolysis in alfalfa haylage.

Ely et al. (1979) evaluated the effect of a dried Lactobacillus and a freeze-dried Lactobacillus in wheat,

ryegrass, alfalfa, corn and sorghum silages. Lactobacillus addition caused no differences in content of silage and recovery of nutrients.

MATERIALS AND METHODS

The effect of two inoculants on silage made from three different crops were evaluated in this study. The inoculants were provided by Dr. W. T. H. Chang from Miles Laboratories Inc. The dry inoculant was in powder form, kept at refrigeration temperature and then at room temperature before silage application. The wet inoculant was kept at freezing temperature, then at room temperature before its application and mixed with water to a final volume of 1% to the silage.

The crops used were: 1) Alfalfa, variety Kanza, third cutting in the bud to 1/10 bloom maturity; field wilted 4 hours to 68% moisture; harvested on July 24, 1981; 2) Whole-plant corn, variety Ferry-Morse 30-20, in the dent maturity and 63-64% moisture; direct harvested on August 21,1981; and 3) High moisture grain sorghum, variety Dekalb DK-59 with 27-28% moisture; direct harvested on October 23, 1981. The effect of the inoculants was evaluated under anaerobic conditions during the first 28 days post ensiling and under aerobic conditions for 7 days after the silos were opened.

I. Anaerobic Study.

Approximately 275 kg of crop were used for each of the following treatments: a) Control (no inoculant added), b) Dry inoculant, c) Wet inoculant. Each crop was placed in a horizontal mixer and then the inoculant was added (2.3×10^6 g/kg). After it was mixed, the treated crop was tightly packed into laboratory silos. These silos consisted of plastic cylindrical containers, 0.27 m in diameter and 0.35 m in height, which were made airtight by a lid adjusted with a

rubber O-ring seal and Bunsen valve. Silos were packed by means of a hydraulic press which allowed all silages within a crop to have uniform density. The silos were then weighed and stored at ca. 80° F at the Sheep Research Center, Kansas State University, during the 28 day ensiling period. The silos were arranged in a completely randomized design. Samples obtained from each silo (pre and post treatment) were immediately frozen in liquid nitrogen and kept frozed until they were analyzed.

The horizontal mixer was sanitized between treatments with a solution containing 200 ppm of chlorine in water. The amount of time between "pre-harvest" until the silos were sealed was approximately 2 1/2 hours.

Triplicates of the silos for each treatment were opened at 1, 2, 4, 7, and 28 days post ensiling. The silos were weighed, the silage mixed for one minute in an electric mixer and then sampled. All samples were analyzed for dry matter, pH, ammonia-nitrogen, lactic acid, volatile fatty acids (acetic, propionic, butyric), crude protein, hot water insoluble nitrogen, and microbial population.

II. Aerobic Study

A 2 kg sample was obtained from each of the silos opened at the 28th day. The sample was taken from the center of each silo at 0.10 m below the silage surface and was placed in an expanded polystyrene container lined with a plastic film at room temperature. A thermocouple, composed of a wire lead introduced into a vacuutainer brand collection tube filled with mineral oil, was inserted into the center

of the silage. In order to prevent contamination a cheesecloth was placed over each container. Containers were kept at room temperature and the silage temperatures were registered daily for 7 days. After 7 days of air exposure, the containers. were weighed, mixed and sampled to determine dry matter, pH and microbial population.

III. Laboratory Analyses.

A. Bacteria and mold populations

Plate Count Agar (PCA) was used for viable cell count (VCC) of bacteria. One microgram of Cycloheximide/ml antibiotic was added to the agar to inhibit mold growth.

Ten (10) grams of the sample were put into a sterile Stomacher bag together with 90 ml of distilled water and run through the Stomacher (Lab-Blender 400 Model # BA 602) for 1 minute. After this, dilutions of the samples were performed from 10^1 to 10^8, and agar was poured into the petri dishes. The agar was kept in a water bath at $47^{\circ}C$ until it was poured into the petri dishes. The samples in the petri dishes were incubated at $32^{\circ}C$ for 48 hours. Colonies were counted and results recorded.

Potato Dextrose Agar (PDA) was used for mold counting. Tetracycline and Chloramphenicol, 1 ug/ml each was added to the agar to inhibit bacteria growth. The same procedure used for bacteria count was followed in this case, with the exception that the petri dishes were incubated at room temperature for 48 hours. Colonies were counted and results recorded.

B. Chemical Analyses

The chemical analyses were performed in the Nutrition

Laboratory, Weber Hall at Kansas State University.

Dry matter losses during storage and fermentation were determined precisely by weighing and sampling every sample of crop ensiled and later weighing and sampling all silage withdrew from the silo.

Crude Protein was determined by the Kjeldahl method. (AOAC, 1965) as follows:

1. Two grams of sample were weighed and deposited into a 500 ml Kjeldahl flask together with 20 ml of standard acid.

2. A couple of boiling beads were added to prevent bumping.

3. Ten grams of K_2SO_4 and 0.3 grams $CuSO_4$ were added.

4. Twenty-five ml of concentrated H_2SO_4 were added.

5. The sample and reagents were digested until all organic material was consumed and the reaction mixture had a clear green color. Thirty more minutes were allowed as extra digestion time.

6. After the mixture was allowed to cool, 250 ml of distilled water were added.

7. Twenty-five ml of boric acid solution were poured into a 250 ml Erlenmeyer flask. A delivery tube was inserted below the surface of the boric acid solution.

8. Seventy-five ml of 40% NaOH was carefully added down the side of the flask to form a layer under the digestion mixture.

9. A piece of mossy zinc was added.

10. The mixture was distilled for 15 minutes after the solution started to boil.

11. The boric acid solution was titrated to a green end point using standardized 1 $\underline{N}$ H_2SO_4.

12. Blank values obtained by running the same analysis described previously without sample were substracted.

13. The formula used was:

$$\% \text{ N} = \frac{(\text{ml of acid})(\text{N of acid})(14)}{\text{sample wt. in mg}} \times 100$$

$$\% \text{ Crude Protein} = (\% \text{ N})\ (6.25)$$

Hot water insoluble matter and its nitrogen (HWIM + HWIM x N) was determined by Goering and Van Soest (1970) procedure, in the following way:

1. Two grams of the sample were weighed and put into a reflux beaker.

2. Two-hundred ml of distilled water were added and refluxed for 1 hour.

3. The solution was filtered through paper No. 4 and washed four times with hot water.

4. The filtrate was dried for eight hours and weighed.

5. Kjeldahl Method was ran on the dry residue and nitrogen was calculated.

6. The following calculations were performed.

$$\text{HWIM} = \frac{\text{weight loss on reflux}}{\text{sample weight}} \times 100$$

$$\text{HWIM x N} = \frac{\text{gN}}{\text{sample wight}} \times 100$$

$$\%\text{CPxF} = 6.25\ (\text{HWIM x N})$$

The pH was measured in the following way:

1. Twenty-five grams of sample was weighed and put into a 400 ml beaker.

2. The sample was covered with distilled water.

3. pH meter was read after one hour.

The volatile fatty acids (VFA) were calculated by:

1. Twenty-five grams of sample were weighed and placed into a 600 ml Berzilius beaker.

2. The sample was covered with 200 ml of 0.2 N H_2SO_4.

3. The beaker was covered with parafilm and allowed to stand at refrigeration temperature for 2 days before it was ran through Gas Chromatograph.

Lactic acid was measured by Barker and Summerson (1941) procedure:

1. The sample was added in a 20 ml test tube

silage	R.F. (rumen fluid)
0.12 ml	0.25 ml

Fifty % Trichloro HAC

silage	R.F.
4.88 ml	4.75 ml

and stored.

2. The sample was diluted with 4 ml of 0.5% $CuSO_4$ and stirred.

3. One ml sample was pipetted into duplicate test tubes.

4. Four ml of H_2SO_4 were added and the mixture was stirred.

5. Two drops of 4% $CuSO_4$ were added and again the mixture was stirred.

6. The sample was put in a hot water bath for five minutes and then placed in cool water until it reached room temperature.

7. Two drops of color reagent were added to each test tube and mixed well. The color reagent was prepared in the following way:

a) Six pellets of NaOH were dissolved into 25 ml of deionized water .

b) While the solution was stirred on a hot plate,

1.5 g of diphenylphenol was added in small increments until it was dissolved.

c) A 20% $CuSO_4$ solution was prepared adding 500 g $CuSO_4$ .5 H_2O to 1,408 ml deionized water.

9. After 30 minutes the samples were put in hot water for 90 seconds and then in cold water for 15 minutes before transferring it to a cuvette.

10. Absorbance was read on spectrophotometer at 560 nm.

For the ammonia determination, Conway, 1957 microdiffusion method was used. The following reagents were prepared:

a) Indicator stock - Sixty-six milligrams of methyl red together with 33 mg of bromocresol green were dissolved in 70 ml ethanol and brought to 100 ml with deionized water.

b) Boric acid solution - Five grams of boric acid were dissolved in 700 ml deionized water and 200 ml ethanol. Ten ml of indicator stock and 0.25 g Tergitol NPX were added to make the volume up to 1 liter.

c) Potassium carbonate - Four hundred fifty grams of K_2CO_3 and 0.25 g of Tergitol were brought up to 1 liter with deonized water and boiled 15 minutes to drive off ammonia.

d) Titration acid 0.1 N HCl was used.

e) Ammonia standards - Three hundred eighty-two milligrams of ammonium chloride (previously dried to constant weight at 102°C was diluted to 100 ml with 0.2 N HCl. Working standards of 50, 100 and 200 mg of ammonia nitrogen per ml were prepared.

The procedure used was as follows:

1. One ml of boric acid solution was added to the center well of the microdiffusion dish and spread with pipette tip covering the entire surface.

2. One ml of potassium carbonate solution was put in one side of the sample ring and 1.5 ml in the sealing ring.

3. The dish was gently rotated to mix the sample with the carbonate and was incubated at room temperature for one hour.

4. The center well was titrated until a pink endpoint color was obtained with standard titration acid. Microburet units were then read.

Analyses of variance were performed on all variables measured in each sampling date for each crop. The treatment means were compared using Fisher's LSD at the 0.05 level of significance. Linear correlation was used to analyze relationships between variables measured in the end product silage and those measured after 7 days of air exposure. Linear correlation was also used between variables measured in day 7 of the aerobic study. All the statistical analyses were performed using Statistical Analyses System (SAS) available at the computing center at Kansas State University.

RESULTS

I. Anaerobic Phase in Silage Fermentation

A. Bacteria and mold populations

1. Alfalfa Trial.

The initial population of bacteria in the pre-ensiled control alfalfa was ca. 3 X 10^6/g (Table 1) (Fig. 1). The population increased to 3.3 X 10^8/g in the first day and peaked at 3.3 X 10^9/g in the second day, then stabilized at around 2.2 X 10^7/g at the 7th day. On the 28th day the population was 10^5/g (Fig. 1). Alfalfa silage inoculated with wet or dry inoculants had ca. 1.4 X 10^7/g at day 0. The population increased to about 10^8/g in the first seven days and the 28th day population was around 10^5/g. These data indicated that there was a high initial bacterial count in alfalfa with or without the starter culture inoculation and that in the first 7 days of ensiling the population increased one log unit. After 28 days of storage the population stabilized at about 10^5/g and there was no statistical difference between treatments in the bacterial counts. Dry and wet treatments produced significantly higher bacterial counts than the control, on days 4 and 7, respectively.

Mold was detected in the pre-ensiled alfalfa at a level of ca. 2.0 X 10^4/g for control 1.8 X 10^5/g for dry inoculant and 1.3 X 10^4/g for wet inoculant (Table 2). No mold was detected at any of the other sampling days, except at day 4 in the dry and wet inoculated silages (Fig. 2). These data indicate that although molds were present on the pre-ensiled alfalfa they were quickly suppressed by bacterial

Table 1. Bacteria counts from the control and inoculated (dry and wet treatments) alfalfa, whole plant corn and high moisture grain sorghum silages from initial day to 28 days post-ensiling.

	Alfalfa	Corn	Grain Sorghum
Initial Day			
Control	3.5×10^{6} a	5.5×10^{8} b	2.9×10^{7} b
Dry Inoculant	1.1×10^{7} a	9.0×10^{7} a	6.2×10^{7} b
Wet Inoculant	1.8×10^{7} a	9.7×10^{7} a	2.8×10^{8} a
Day 1			
Control	3.3×10^{8} a	---	5.7×10^{7} ab
Dry Inoculant	1.9×10^{8} a	---	8.5×10^{7} b
Wet Inoculant	4.3×10^{8} a	---	2.7×10^{7} a
Day 2			
Control	3.4×10^{9} a	3.3×10^{7} a	3.1×10^{8} a
Dry Inoculant	4.4×10^{7} a	1.4×10^{7} a	6.8×10^{9} a
Wet Inoculant	1.7×10^{8} a	3.0×10^{7} a	3.4×10^{9} a
Day 4			
Control	1.1×10^{8} a	5.6×10^{5} a	2.1×10^{8} a
Dry Inoculant	2.4×10^{8} b	2.9×10^{5} a	3.4×10^{9} a
Wet Inoculant	2.0×10^{8} ab	1.0×10^{6} a	7.9×10^{7} a
Day 7			
Control	2.2×10^{7} a	3.0×10^{5} a	3.4×10^{9} a
Dry Inoculant	5.9×10^{7} ab	4.1×10^{5} a	2.9×10^{8} a
Wet Inoculant	1.0×10^{8} b	3.5×10^{6} a	3.8×10^{8} a
Day 28			
Control	1.0×10^{5} a	1.2×10^{6} a	1.0×10^{10} b
Dry Inoculant	3.0×10^{5} a	3.4×10^{5} a	1.2×10^{6} a
Wet Inoculant	4.2×10^{5} a	4.1×10^{5} a	3.5×10^{9} a

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

All values averages of three silos.

--- = Not determined.

Table 2. Mold counts from the control and inoculated (dry and wet treatments) alfalfa, whole plant corn and high moisture grain sorghum silages from initial day to 28 days post-ensiling.

	Alfalfa	Corn	Grain Sorghum
Initial Day			
Control	2.4×10^4 a	2.7×10^5 a	6.8×10^4 a
Dry Inoculant	1.8×10^5 a	3.7×10^5 a	4.7×10^4 a
Wet Inoculant	1.3×10^4 a	3.3×10^6 a	5.9×10^4 a
Day 1			
Control	ND	---	2.4×10^4 a
Dry Inoculant	ND	---	2.9×10^4 a
Wet Inoculant	ND	---	3.0×10^4 a
Day 2			
Control	ND	ND	1.0×10^3 a
Dry Inoculant	ND	ND	3.7×10^3 a
Wet Inoculant	ND	ND	1.0×10^2 a
Day 4			
Control	ND[a]	ND	ND[a]
Dry Inoculant	1.0×10^3 a	ND	1.1×10^2 a
Wet Inoculant	1.0×10^5 a	ND	ND[a]
Day 7			
Control	ND	ND	ND
Dry Inoculant	ND	ND	ND
Wet Inoculant	ND	ND	ND
Day 28			
Control	ND	ND	2.3×10^4 a
Dry Inoculant	ND	ND	ND[a]
Wet Inoculant	ND	ND	3.7×10^4 a

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

All values averages of three silos.

ND = Not detected.

--- = Not determined.

THIS BOOK CONTAINS NUMEROUS PAGES THAT WERE BOUND WITHOUT PAGE NUMBERS.

THIS IS AS RECEIVED FROM CUSTOMER.

Fig. 1. Bacterial plate count vs. time for the control, and dry and wet inoculant treatments in alfalfa silage.

THIS BOOK CONTAINS NUMEROUS PAGES WITH DIAGRAMS THAT ARE CROOKED COMPARED TO THE REST OF THE INFORMATION ON THE PAGE.

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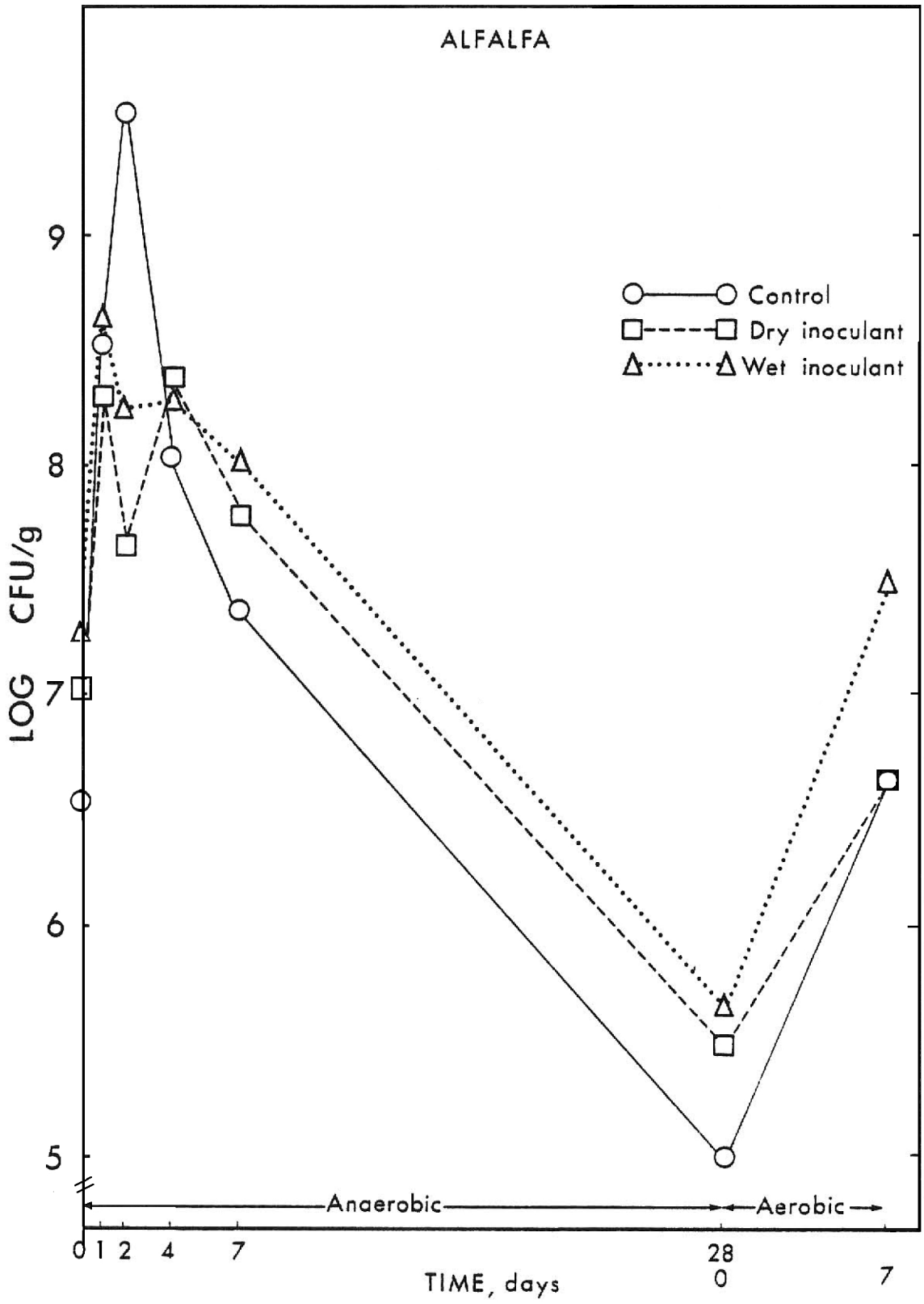
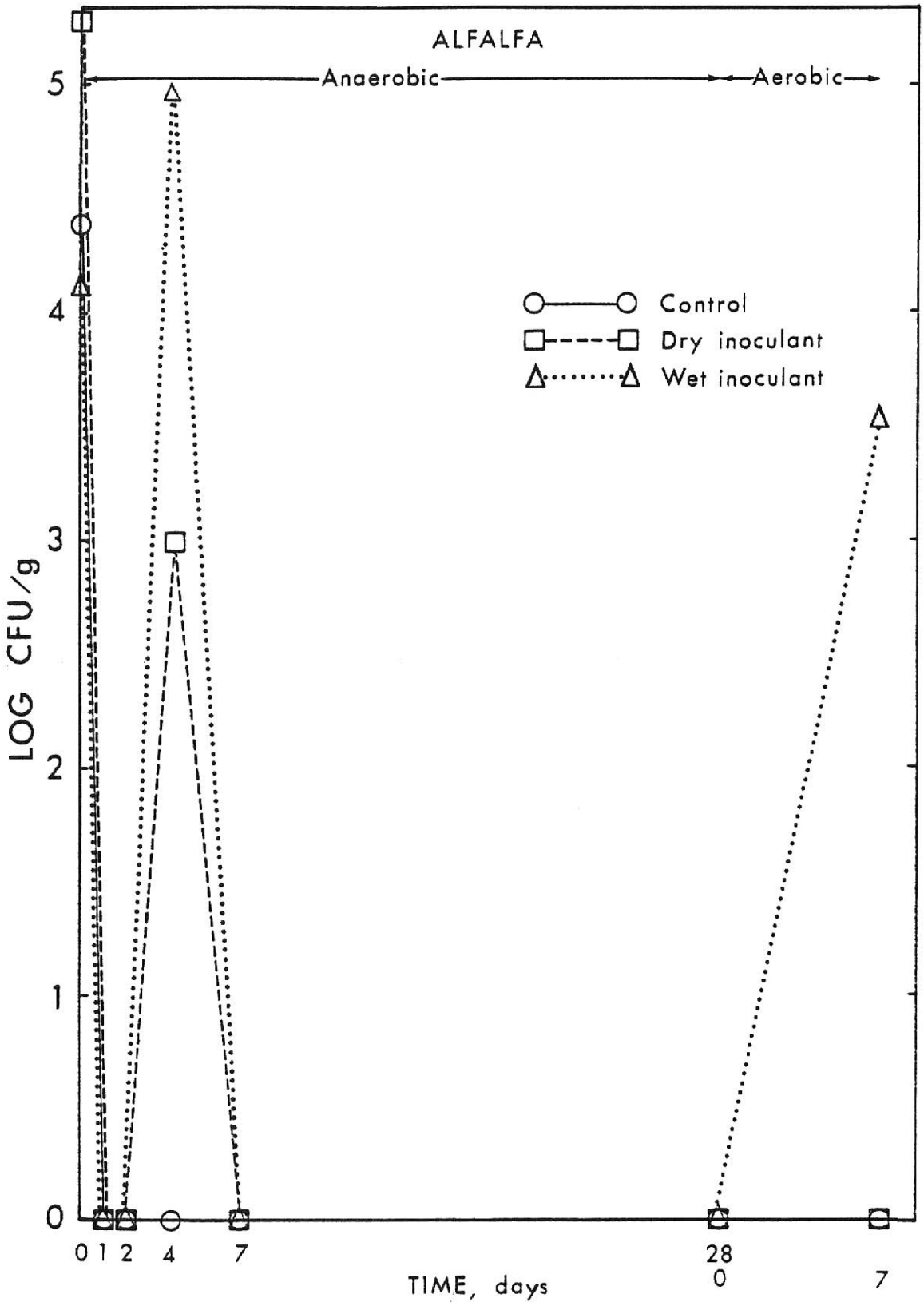

ALFALFA
LOG CFU/g
9
8
7
6
5
Control
Dry inoculant
Wet inoculant
Anaerobic
Aerobic
0 1 2 4 7 28
0 7
TIME, days

Fig. 2. Mold plate count vs. time for the control, and dry and wet inoculant treatments in alfalfa silage.


ALFALFA
Anaerobic
Aerobic
Control
Dry inoculant
Wet inoculant
LOG CFU/g
0
1
2
3
4
5
0 1 2 4 7
28
0 7
TIME, days

populations. This was a good sign since the presence of mold is not desirable in the silage fermentation process.

2. Corn trial.

The initial population of bacteria in corn forage was approximately 5.5 X 10^8/g in the control (Table 1). The population decreased to 3.3 X10^7/g in the second day and stabilized at the 7th day. On the 28th day the population was 10^6/g (Fig. 3). Corn forage inoculated with dry or wet inoculants had approximately 9.4 X 10^7/g initially. The population decreased to 2.9 X 10^5/g and 1.0 X 10^6/g for the dry and wet inoculant respectively, at day 4. On the 28th day both dry and wet inoculant silages had a population of ca 3.8 X 10^5/g. These results indicated that there was a high initial bacterial count in corn forage with and without inoculation and that in the first 7 days of ensiling the population decreased one log unit. There were no statistical differences in bacterial count at any of the sampling days, with the exception of the initial day, when control silages had significantly higher bacterial counts than inoculated silages (Table 1).

Mold was detected in the pre-ensiled corn forage at approximately 3.2 X 10^5/g for the control and dry treatment and at 3.3 X 10^6/g for the wet treatment (Table 2). No mold count was detected at any of the other days of sampling (Fig. 4). These results indicate that the bacterial population quickly supressed the mold count present at the beginning of the ensiling process.

Fig. 3. Bacterial plate count vs. time for the control, and dry and wet inoculant treatments in whole plant corn silage.

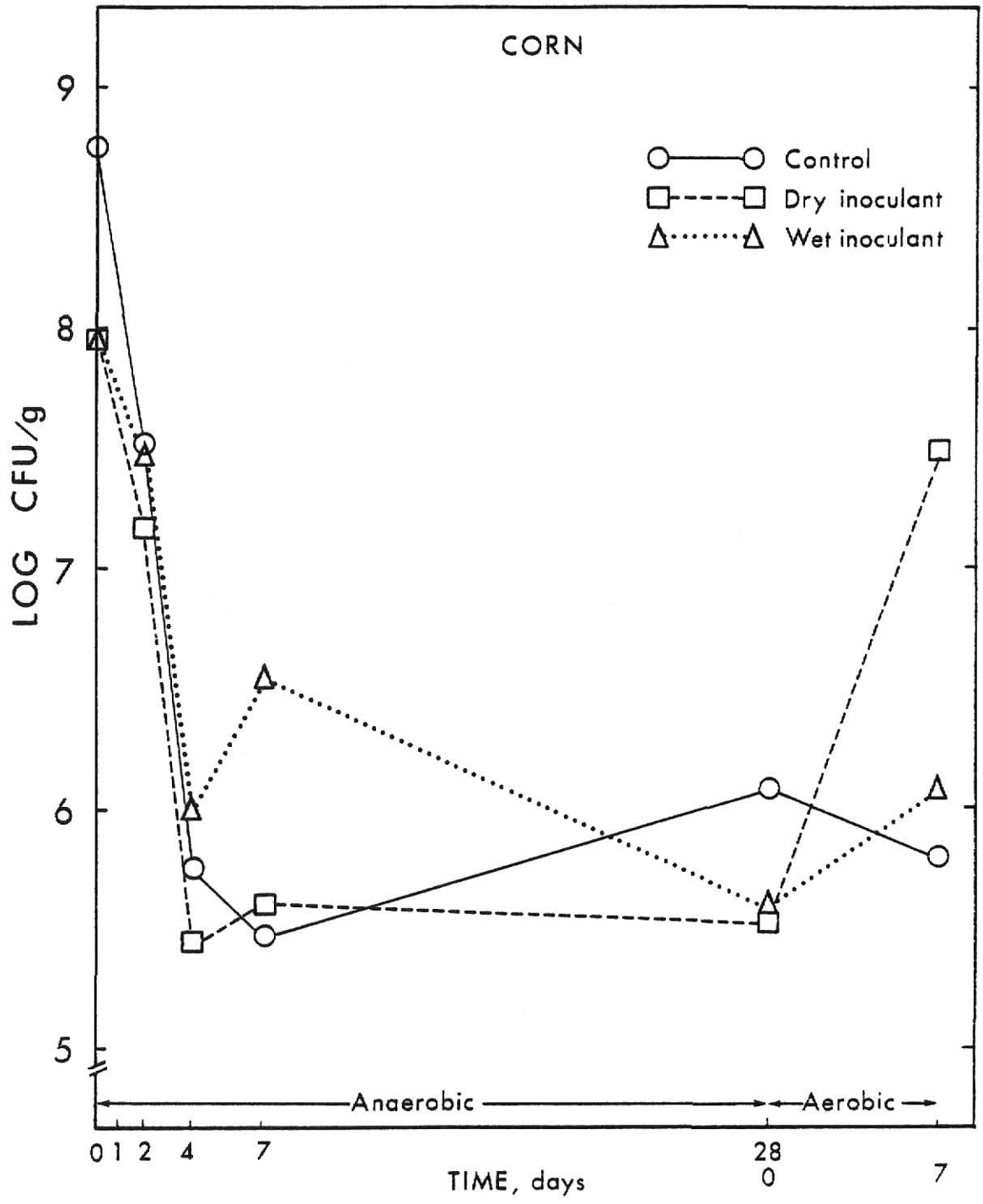
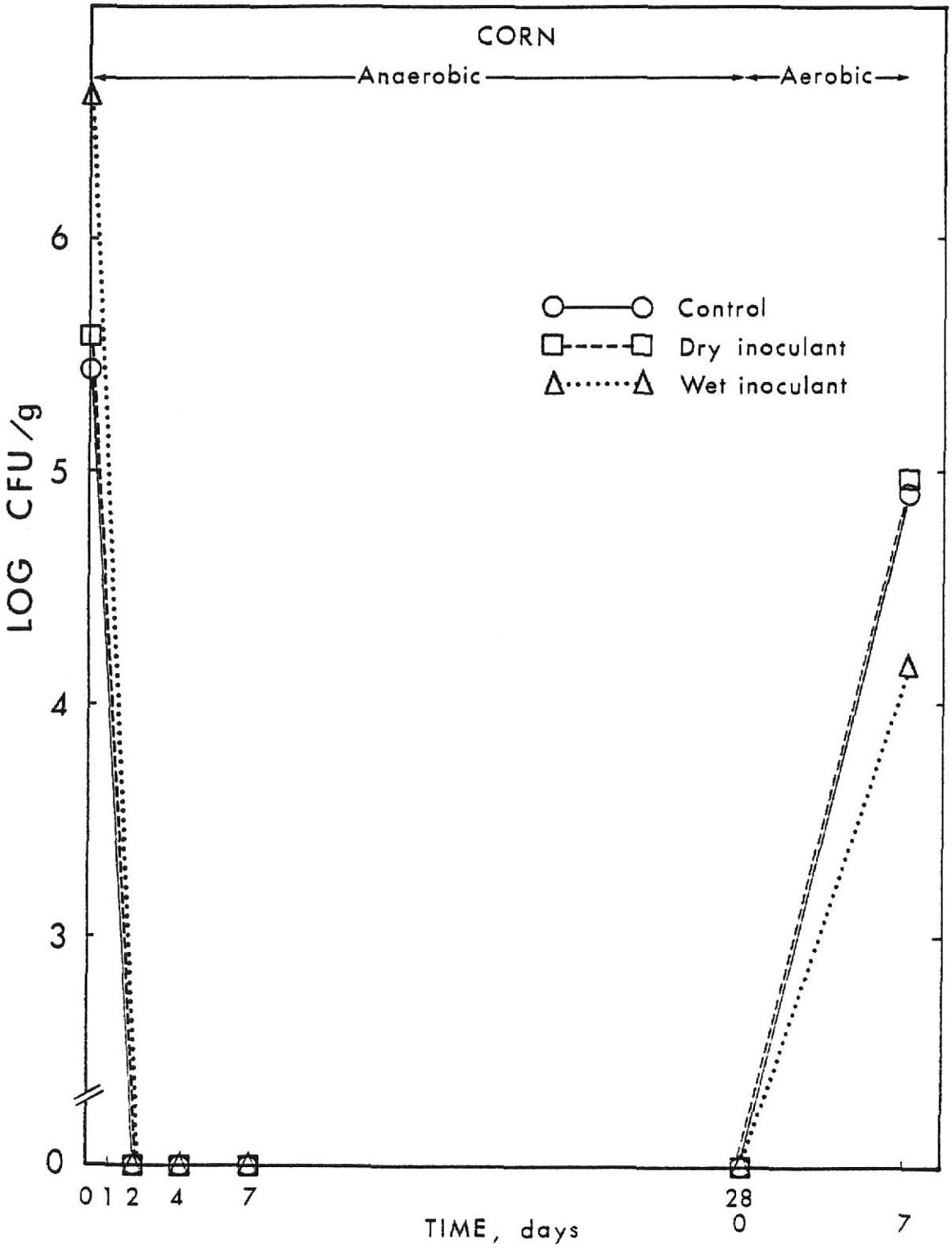

CORN
Control
Dry inoculant
Wet inoculant
LOG CFU/g
9
8
7
6
5
Anaerobic
Aerobic
0 1 2 4 7 28 0 7
TIME, days

Fig. 4. Mold plate count vs. time for the control, and dry and wet inoculant treatments in whole plant corn silage.


CORN
Anaerobic
Aerobic
Control
Dry inoculant
Wet inoculant
LOG CFU/g
6
5
4
3
0
0 1 2 4 7
28
0
7
TIME, days

3. Grain sorghum trial.

The initial population of bacteria in control grain sorghum samples was approximately 3.0 X 10^7/g (Table 1). The population increased to 10^8/g in the second day and by the seventh day it had increased to 3.4 X 10^9/g. It reached its peak on day 28 in which the bacterial count was 10^{10}/g (Fig. 5). Grain sorghum silage inoculated with the dry inoculant had an initial population of approximately 6.2 X 10^7/g. By the second day it had reached 6.8 X 10^9/g where it stabilized. The bacterial population had decreased one log unit by day 7 and by the 28th day it had reduced two log units. The wet inoculant grain sorghum started with a population of 2.8 X 10^8/g, increased to 3.4 X 10^9/g on day 2, decreased to 7.9 X 10^7/g on day 4 and again increased to 3.5 X 10^9/g by the 28th day. Although the grain sorghum data did not follow the same pattern of the other two crops, it indicated that the bacterial population in the grain sorghum was high with and without the inoculants. On the first day, the dry inoculant produced a significantly higher bacterial population than the wet inoculant. On the other hand, on day 28 the control grain sorghum silage showed a significantly higher bacterial population than the dry and wet inoculant silages.

Mold population was of 6.8 X 10^4/g for the control sorghum grain on the initial day, decreased to a nondetectable level by day 4 and again increased to 2.3 X 10^4/g on day 28 (Table 2, Fig. 6). There were no statistically significant differences between treatments at any of the other days of sampling.

Fig. 5. Bacterial plate count vs. time for the control, and dry and wet inoculant treatments in high moisture grain sorghum silage.

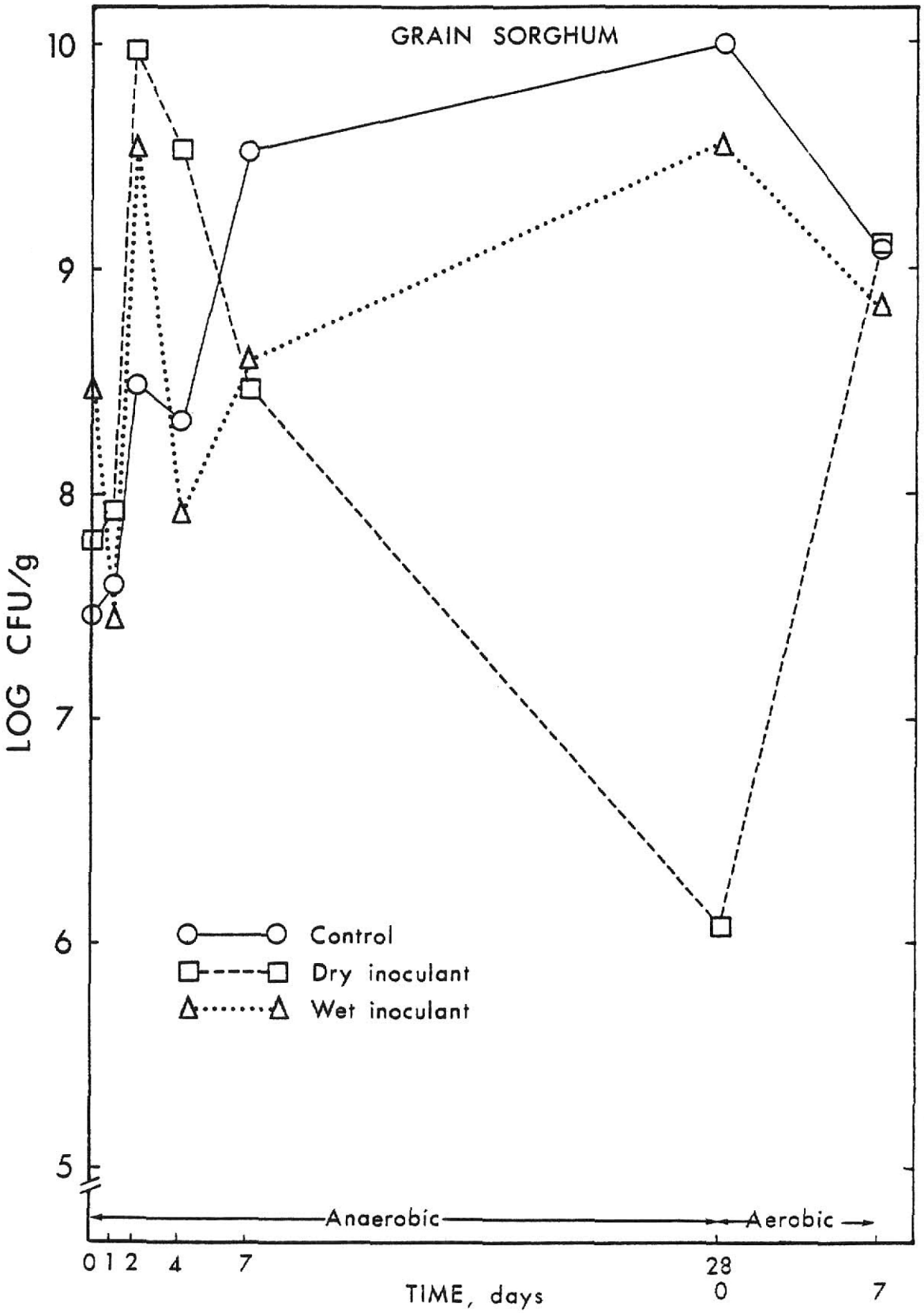
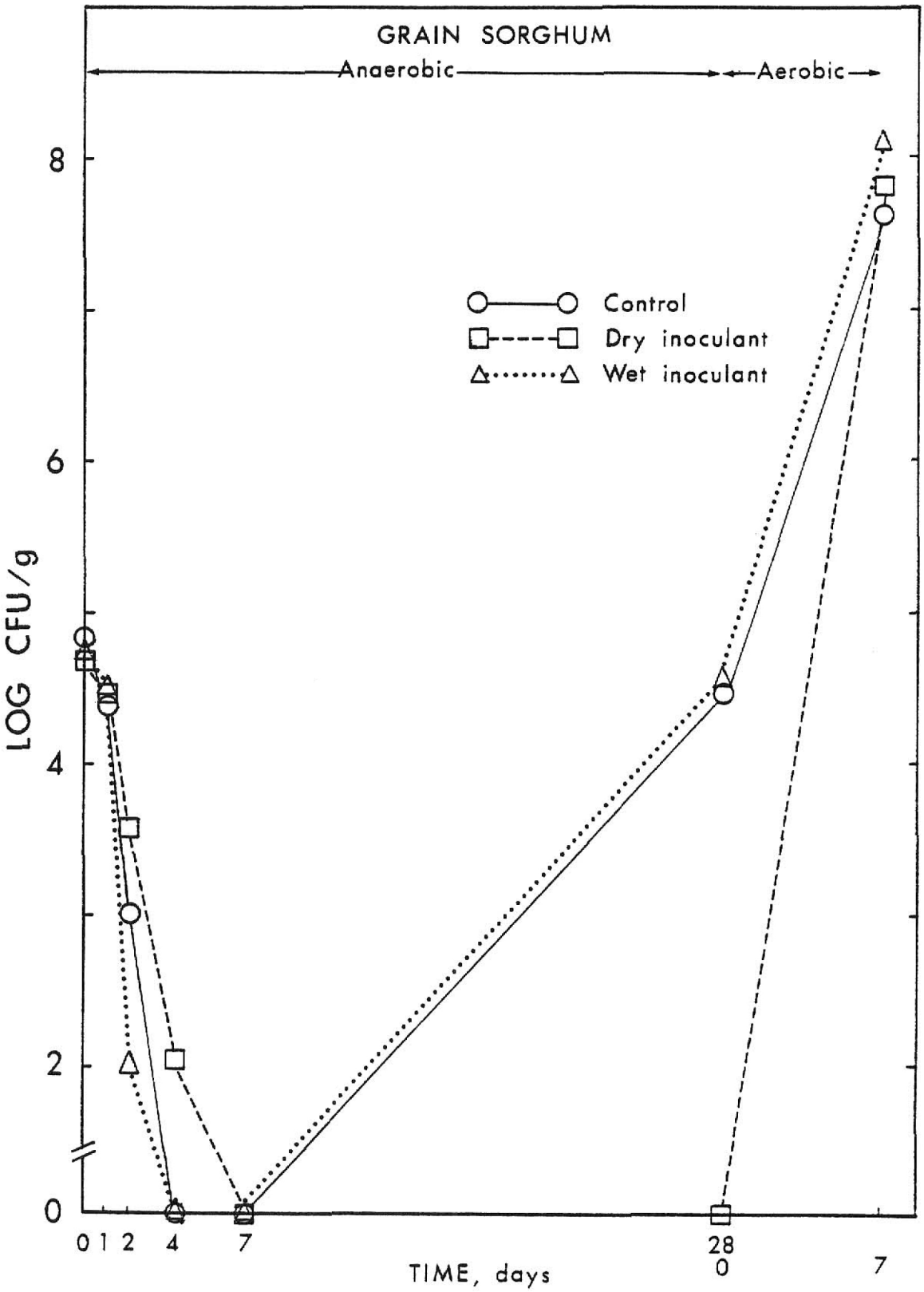

GRAIN SORGHUM
LOG CFU/g
10
9
8
7
6
5
Control
Dry inoculant
Wet inoculant
Anaerobic
Aerobic
0 1 2 4 7 28 0 7
TIME, days

Fig. 6. Mold plate count vs. time for the control, and dry and wet inoculant treatments in high moisture grain sorghum silage.


GRAIN SORGHUM
Anaerobic
Aerobic
Control
Dry inoculant
Wet inoculant
LOG CFU/g
8
6
4
2
0
0 1 2 4 7
28
0 7
TIME, days

B. Biochemical changes in silage fermentation

1. Alfalfa trial

The percentage of dry matter in the pre-ensiled alfalfa ranged from 30.7 to 32.0 (Table 3). In all cases, except for data obtained in day 1, the control silage had a significantly lower dry matter content than those silages treated with dry or wet inoculant. The percentage recovery of dry matter tended to be higher in the wet inoculant treated silage when compared to the control, although these differences only reached significance on days 2 and 4. In general, the recovery was very high (90 to 97%) which indicated good ensiling conditions in the entire experiment.

The pH of the alfalfa started at around 6.8 and dropped one log unit as the fermentation progressed (Table 4). This indicated that a typical fermentation occurred.

The total acids were around 6 to 7% of the silage dry matter for the first 7 days and on day 28 the percentage reached nearly 10% (Table 4). No statistical differences were observed except on day 1 when the wet inoculant silage was significantly lower than the control and the dry inoculant silages.

The lactic acid concentration was about 3 to 4% during the first 7 days of fermentation (Table 4). On day 28 the level dropped to 1-2%. There were statistical differences between treatments on day 1, when the control silage had a significantly higher lactic acid concentration than the two inoculant silages, and on day 2, when the wet inoculant silage had a significantly lower lactic acid concentration than the other two treatments.

Table 3. Dry matter (DM) content and dry matter recovery (DM recovery) for the alfalfa, whole plant corn, and high moisture grain sorghum silages from initial to 28 days post-ensiling.

Day and treatment	Alfalfa		Corn		Grain Sorghum	
	Dry matter	DM recovery[1]	Dry matter	DM recovery[1]	Dry matter	DM recovery[1]
	%		%		%	
Initial Day						
Control	30.71[a]	---	35.76[a]	---	72.38[a]	---
Dry Inoculant	31.64[b]	---	37.09[a]	---	72.38[a]	---
Wet Inoculant	31.99[b]	---	37.06[a]	---	72.38[a]	---
Day 1						
Control	30.72[a]	96.44[a]	35.32[a]	98.16[a]	72.05[ab]	99.45[a]
Dry Inoculant	31.63[a]	96.92[a]	36.55[b]	98.37[a]	71.88[a]	99.54[a]
Wet Inoculant	31.93[a]	97.58[a]	36.58[b]	98.48[a]	72.28[b]	99.42[a]
Day 2						
Control	30.58[a]	96.17[a]	34.84[a]	96.71[a]	72.05[a]	99.20[b]
Dry Inoculant	31.56[b]	96.50[a]	36.18[b]	96.95[a]	71.57[a]	98.91[a]
Wet Inoculant	31.76[b]	97.23[b]	36.21[b]	97.39[a]	72.32[a]	99.34[b]
Day 4						
Control	30.58[a]	95.64[a]	34.19[a]	95.10[a]	72.01[a]	99.05[a]
Dry Inoculant	31.56[b]	96.13[ab]	35.96[b]	96.34[a]	71.54[a]	98.61[a]
Wet Inoculant	31.76[b]	96.52[b]	35.90[b]	96.45[a]	72.28[a]	99.49[a]
Day 7						
Control	30.23[a]	94.46[a]	34.30[a]	95.34[a]	71.98[b]	98.91[ab]
Dry Inoculant	31.42[b]	95.47[a]	35.91[b]	96.24[a]	71.52[a]	98.62[a]
Wet Inoculant	31.70[b]	96.15[a]	35.98[b]	96.50[a]	72.24[b]	99.14[b]
Day 28						
Control	29.38[a]	90.99[a]	33.51[a]	93.03[a]	71.64[a]	98.32[a]
Dry Inoculant	30.97[b]	93.61[a]	34.86[a]	93.20[a]	71.41[a]	98.37[a]
Wet Inoculant	30.84[b]	92.74[a]	35.44[a]	94.40[a]	71.99[a]	98.71[b]

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

[1] As % of the dry matter ensiled.

--- = Not determined.

Table 4. pH, lactic acid, volatile fatty acids, and total fermentation acids for the alfalfa silage from initial to 28 days post-ensiling.

	pH	Lactic	Acetic	Propionic	Butyric	Total
		as % of the silages DM				
Initial Day						
Control	6.80[a]	---	---	---	---	---
DryInoculant	6.84[a]	---	---	---	---	---
Wet Inoculant	6.82[a]	---	---	---	---	---
Day 1						
Control	5.32[a]	4.37[b]	3.02[a]	0.06[a]	ND	7.45[b]
Dry Inoculant	5.47[b]	3.68[a]	3.47[b]	0.06[a]	Trace	7.21[b]
Wet Inoculant	5.39[ab]	3.14[a]	3.26[ab]	0.07[a]	Trace	6.48[a]
Day 2						
Control	5.56[b]	3.61[b]	3.23[a]	0.06[a]	ND	6.91[a]
Dry Inoculant	5.55[b]	3.50[b]	3.92[a]	0.16[a]	ND	7.58[a]
Wet Inoculant	5.52[a]	3.25[a]	3.69[a]	0.08[a]	ND	7.02[a]
Day 4						
Control	5.64[b]	3.16[a]	4.00[a]	0.02[a]	ND	7.18[a]
Dry Inoculant	5.60[a]	3.55[a]	3.93[a]	0.04[a]	ND	7.52[a]
Wet Inoculant	5.57[a]	3.33[a]	3.94[a]	0.08[a]	ND	7.40[a]
Day 7						
Control	5.84[b]	3.10[a]	4.55[b]	0.06[b]	ND	7.70[a]
Dry Inoculant	5.71[a]	3.00[a]	4.33[ab]	0.03[ab]	ND	7.36[a]
Wet Inoculant	5.66[a]	2.85[a]	4.13[a]	ND[a]	ND	6.98[a]
Day 28						
Control	5.77[a]	0.91[a]	7.38[a]	1.47[b]	0.21[b]	9.98[a]
Dry Inoculant	5.76[a]	1.72[a]	7.38[a]	0.79[a]	0.05[a]	9.94[a]
Wet Inoculant	5.81[a]	1.43[a]	7.15[a]	0.91[a]	0.21[b]	9.70[a]

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

ND = Not detected.

--- = Not determined.

Acetic acid concentration was around 3-4% during the first 7 days, but increased to over 7% at day 28 (Table 4). Significant differences were observed on day 1 when the dry inoculant silage had a higher acetic acid concentration than the control silage. On day 7 the control had a significantly higher acetic acid than the wet inoculant silage.

Propionic and butyric acids (Table 4) were very low (0.02 - 0.16 to nondetectable) for the entire ensiling period which indicated that the undesirable Clostridial activity in these silages either did not occur or occurred at a very low rate.

The crude protein level of alfalfa was about 24% of the dry matter at the ensiling process, and dropped slightly to about 23% throughout the experiment (Table 5). The hot water insoluble nitrogen dropped from around 42% on day 1 to approximately 29% on day 28.

The ammonia concentration had approximately a threefold increase from day 1 to the 28th day for all treatments. The control silage had a significantly lower ammonia concentration than the other two treatments on day 1, but not on the following days (Table 5).

2. Corn trial

The percentage of dry matter in the pre-ensiled corn forage ranged from 35.8 to 37%. The control silage had significantly lower dry matter content than those silages treated with dry and wet inoculant at all days post-ensiling, with the exception of the 28th day in which there were no significant differences (Table 3). There was a trend for

Table 5. Crude protein (CP), ammonia nitrogen (NH_3) and hot water insoluble nitrogen (HWIN) for alfalfa silages from initial day to 28 days post-ensiling.

	CP[x]	NH_3[x]	HWIN[y]
Initial Day			
Control	24.3	---	---
Dry Inoculant	24.3	---	---
Wet Inoculant	24.3	---	---
Day 1			
Control	23.81	0.41[a]	39.30
Dry Inoculant	24.55	0.65[b]	44.50
Wet Inoculant	25.10	0.70[b]	45.70
Day 2			
Control	24.41	0.68[ab]	41.40
Dry Inoculant	24.09	0.97[b]	45.80
Wet Inoculant	24.08	0.52[a]	44.30
Day 4			
Control	23.75	0.79[a]	38.90
Dry Inoculant	23.81	0.61[a]	42.90
Wet Inoculant	23.99	0.57[a]	40.10
Day 7			
Control	23.39	0.78[a]	37.60
Dry Inoculant	22.95	0.76[a]	39.50
Wet Inoculant	23.07	1.10[a]	38.80
Day 28			
Control	23.13	1.69[a]	27.90
Dry Inoculant	23.42	1.67[a]	27.60
Wet Inoculant	23.03	1.50[a]	29.90

CP and HWIN were analyzes of composited samples per treatment per day, thus no statistical analysis were performed.

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

[x]As % of the dry matter.

[y]As % of total nitrogen.

the percentage recovery of dry matter to be higher in the wet inoculant silage, although the differences were not statistically significant. The recovery was quite high (93 to 98%) indicating good ensiling conditions.

The pH of the corn forage was around 6.80 on the initial day and by the first day it had already decreased to nearly 4.0 (Table 6). The control silage showed significantly lower pH than the treated silages on days 1, 2, 4 and 7. This suggested that a rapid fermentation occurred even without the addition of inoculants.

The concentration of total acids varied between 3.57 and 3.69% on the first day and increased as fermentation progressed (Table 6). There were no statistical differences between treatments at any of the days post-ensiling, with the exception of day 28 in which the control silage had a significantly higher concentration of total acids than the treated silages.

The lactic acid concentration was between 2 and 3% on the first day and increased to 5% by the second day. No statistical differences were observed between treatments, although the control silage showed a trend of higher amounts of lactic acid than the other treatments on days 2, 7, and 28.

Acetic acid concentrations started at 1% on the first day and decreased to 0.9% on day 7. On day 4, the corn silage treated with the wet inoculant had a higher acetic acid concentration than the dry silage, but it did not show significant difference compared to the control silage.

Propionic and butyric acids were present in very low

Table 6. pH, lactic acid, volatile fatty acids, and total fermentation acids for the whole plant corn silages from initial to 28 days post-ensiling.

	pH	Lactic	Acetic	Propionic	Butyric	Total
		as a % of the silage DM				
Initial Day						
Control	6.80[a]	---	---	---	---	---
Dry Inoculant	6.78[a]	---	---	---	---	---
Wet Inoculant	6.79[a]	---	---	---	---	---
Day 1						
Control	4.09[a]	2.48[a]	1.09[a]	Trace	Trace	3.57[a]
Dry Inoculant	4.14[b]	3.24[a]	1.09[a]	Trace	Trace	4.33[a]
Wet Inoculant	4.15[b]	2.68[a]	1.01[a]	Trace	Trace	3.69[a]
Day 2						
Control	3.72[a]	5.20[a]	1.22[a]	Trace	Trace	6.42[a]
Dry Inoculant	3.76[b]	5.05[a]	0.58[a]	Trace	Trace	5.63[a]
Wet Inoculant	3.77[b]	4.81[a]	1.13[a]	Trace	Trace	5.94[a]
Day 4						
Control	3.65[a]	5.30[a]	0.94[ab]	0.04[a]	ND	6.28[a]
Dry Inoculant	3.69[b]	4.82[a]	0.82[a]	Trace	ND	5.64[a]
Wet Inoculant	3.70[b]	5.36[a]	1.00[b]	Trace	ND	6.36[a]
Day 7						
Control	3.69[a]	5.47[a]	0.95[a]	Trace[a]	ND	6.42[a]
Dry Inoculant	3.72[b]	5.04[a]	0.94[a]	0.02[a]	ND	5.99[a]
Wet Inoculant	3.73[b]	4.87[a]	0.93[a]	0.04[a]	ND	5.83[a]
Day 28						
Control	3.53[a]	5.82[a]	0.93[a]	0.01	ND	6.77[b]
Dry Inoculant	3.58[a]	4.17[a]	0.98[a]	Trace	Trace	5.15[a]
Wet Inoculant	3.63[a]	3.62[a]	1.04[a]	Trace	ND	4.67[a]

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

ND = Not detected.

--- = Not determined.

amounts throughout the entire ensiling time. Traces of butyric acid were detected on days 1 and 2 with none detected after the second day. The low amounts of butyric acid in the corn silages indicated that there was no clostridial growth.

The crude protein level of corn was about 6% on the day 1 and increased to over 7% during the fermentation process (Table 7). The control silage showed a slightly higher amount for crude protein on days 1 and 4.

The ammonia content was approximately 0.24% on the first day with the wet inoculant silage having a significantly lower percentage than the control silage. Day 7 also showed a significantly lower percentage of the wet inoculant over the control silage.

The HWIN was approximately 60% on day 1, decreased to 47% on day 4, and increased to 51% on the 28th day for the control silage. The dry inoculant silage seemed to have higher HWIN than the wet inoculant and control silages, beginning with 62% on the first day but dropping to only 52% on day 28. The wet inoculant silage HWIN dropped consistently from 62% to 46%.

3. Grain sorghum trial

The percentage of dry matter in the pre-ensiled grain sorghum was 72.4% (Table 3). The wet inoculant silage consistently had a higher percentage of dry matter than the dry inoculant silage, although the differences reached significance at days 1 and 7 only. The wet inoculant silages had a higher dry matter recovery than the dry

Table 7. Crude protein (CP), ammonia nitrogen (NH_3), and hot water insoluble nitrogen (HWIN) for whole plant corn silages from initial day to 28 days post-ensiling.

	CP[x]	NH_3[x]	HWIN[y]
Initial Day			
Control	6.69	---	---
Dry Inoculant	6.62	---	---
Wet Inoculant	6.70	---	---
Day 1			
Control	7.47	0.24[b]	59.90
Dry Inoculant	7.28	0.24[ab]	61.90
Wet Inoculant	7.31	0.23[a]	61.20
Day 2			
Control	7.66	0.25[a]	55.60
Dry Inoculant	---	0.18[a]	64.50
Wet Inoculant	7.53	0.22[a]	56.60
Day 4			
Control	7.73	0.24[a]	47.30
Dry Inoculant	7.43	0.23[a]	53.20
Wet Inoculant	7.70	0.23[a]	48.90
Day 7			
Control	7.37	0.25[b]	---
Dry Inoculant	7.24	0.24[ab]	---
Wet Inoculant	7.56	0.23[a]	---
Day 28			
Control	---	0.20[a]	50.83
Dry Inoculant	---	0.20[a]	52.23
Wet Inoculant	---	0.18[a]	46.37

CP and HWIN were analyses of composited samples per treatment per day, thus no statistical analyses were performed.

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

[x]As % of the dry matter.

[y]As % of total nitrogen.

--- Not determined.

inoculant silages, with days 2, 7 and 28 showing statistically significant differences. The dry matter recovery for all grain sorghum silages was very high (98 and 99%), suggesting an excellent conservation process.

The pH of grain sorghum silage started at 6.80 on the initial day, decreased to approximately 5.5 in the second day and by the fourth day it stabilized around 4.1 (Table 8). Statistically significant differences were observed on days 2, 4, and 7, where the wet inoculant exhibited the fastest drop over the other two silages. The rapid decrease in pH is further indication that an efficient fermentation occurred in the experimental silos.

The percentage of total acids ranged between 0.20 to 2.04 with significant differences among silage treatments observed for days 4, 7 and 28. The control silage had a lower total acid concentration than the wet inoculant silage on day 4, while the converse was true on day 28. The wet inoculant silage had a higher total acids than the dry inoculant silage for days 4 and 7.

The lactic acid concentration was about 0.15% on the first day for the control silage, increased to 0.41 by day 2 and, stabilized at approximately 1% by the fourth day. Lactic acid reached its peak of 1.74% on the 28th day. The lactic acid content of the wet inoculant silage was significantly higher than that of the dry inoculant silage on day 7 and it was significantly higher than that of the control silage on day 28.

Table 8. pH, lactic acid, volatile fatty acids, and total fermentation acids for the high moisture grain sorghum silage from initial to 28 days post-ensiling.

	pH	Lactic	Acetic	Propionic	Butyric	Total
		as % of the silage DM				
Initial Day						
Control	6.80[a]	---	---	---	---	---
Dry Inoculant	6.80[a]	---	---	---	---	---
Wet Inoculant	6.80[a]	---	---	---	---	---
Day 1						
Control	6.46[a]	0.15[a]	0.05[a]	ND	ND	0.20[a]
Dry Inoculant	6.53[a]	0.14[a]	0.04[a]	ND	ND	0.18[a]
Wet Inoculant	6.48[a]	0.16[a]	0.02[a]	ND	ND	0.19[a]
Day 2						
Control	5.54[b]	0.41[a]	0.08[a]	ND	ND	0.50[a]
Dry Inoculant	5.67[b]	0.41[a]	0.11[a]	Trace	ND	0.52[a]
Wet Inoculant	5.12[a]	0.56[a]	0.08[a]	ND	ND	0.63[a]
Day 4						
Control	4.81[b]	1.10[a]	0.30[a]	Trace	Trace[a]	1.40[a]
Dry Inoculant	4.84[b]	1.03[a]	0.30[a]	Trace	0.02[a]	1.34[a]
Wet Inoculant	4.38[a]	1.57[a]	0.27[a]	ND	ND[a]	1.83[b]
Day 7						
Control	4.37[b]	0.94[ab]	0.20[a]	Trace	ND	1.14[ab]
Dry Inoculant	4.37[b]	0.78[a]	0.16[a]	ND	ND	0.94[a]
Wet Inoculant	4.13[a]	1.39[b]	0.19[a]	ND	ND	1.58[b]
Day 28						
Control	4.12[ab]	1.74[b]	0.29[a]	0.01	ND	2.04[b]
Dry Inoculant	4.15[b]	1.51[ab]	0.24[a]	ND	ND	1.75[ab]
Wet Inoculant	4.10[a]	1.18[a]	0.16[a]	ND	ND	1.35[a]

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

ND = Not detected.

--- = Not determined.

Acetic acid percentage of the grain sorghum silage was around 0.05 in the first day for the control silage, increased to 0.30 on the fourth day, and decreased to 0.29 by day 28. No statistically significant differences were observed for any day of the ensiling period, however, the control silages consistently showed a slightly higher percentage than the inoculant silages.

Propionic and butyric acids were found in very small amounts and this indicated that there was no undesirable clostridial activity in the grain sorghum silages.

The crude protein level of grain sorghum was about 11.80% for the initial day (Table 9). The value dropped slightly on the first day, increased to 12% on the second day, again showed a slight increase by the seventh day but decreased to near 11% at the 28th day. The wet inoculant silage had a slightly higher crude protein over the control and dry inoculant silages for days 1, 2, 4, and 28, although no statistically significant differences were observed.

In general, the HWIN percentage decreased throughout the ensiling period. The only significant differences were for day 1, where the wet inoculant silage had a larger percentage than the control and dry inoculant silages.

Ammonia percentage started at 0.08 on day 1, increased to 0.12% by day 4 and stabilized at 0.13% at day 28 for the control silage. Statistical significant differences were only shown on the 28th day where the control silage exhibited the lowest amount of ammonia. Dry and wet inoculant silages

Table 9. Crude protein (CP), ammonia nitrogen (NH_3), and hot water insoluble nitrogen (HWIN) for the high moisture grain sorghum silages from initial to 28 days post-ensiling.

	CP[x]	NH_3[x]	HWIN[y]
Initial Day			
Control	11.80[a]	---	---
Dry Inoculant	11.80[a]	---	---
Wet Inoculant	11.80[a]	---	---
Day 1			
Control	11.24[a]	0.08[a]	86.83[a]
Dry Inoculant	11.44[a]	0.08[a]	87.32[b]
Wet Inoculant	11.45[a]	0.08[a]	93.36[b]
Day 2			
Control	12.11[a]	0.07[a]	83.99[a]
Dry Inoculant	12.03[a]	0.06[a]	82.41[a]
Wet Inoculant	12.29[a]	0.07[a]	85.39[a]
Day 4			
Control	11.77[a]	0.12[a]	77.06[a]
Dry Inoculant	11.62[a]	0.13[a]	85.82[a]
Wet Inoculant	11.68[a]	0.11[a]	86.09[a]
Day 7			
Control	12.02[a]	0.13[a]	79.69[a]
Dry Inoculant	12.48[a]	0.12[a]	73.77[a]
Wet Inoculant	12.42[a]	0.12[a]	70.48[a]
Day 28			
Control	---	0.13[a]	---
Dry Inoculant	11.11[a]	0.21[b]	77.09[a]
Wet Inoculant	11.41[a]	0.25[b]	78.02[a]

Means in each column within days followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

--- = Not determined.

[x]As % of the dry matter.

[y]As % of total nitrogen.

had ammonia percentages of 0.08%, increased to 0.13 and 0.12%, respectively, and reached their peaks on day 28 when the dry inoculant silage had 0.21% and the wet inoculant silage had 0.25%.

II Aerobic Phase in Silage Fermentation

1. Alfalfa trial

The percent of dry matter loss and pH did not show statistically significant differences among the silages after 7 days of air exposure (Table 10). The wet inoculant silage had a significant lower maximum and cumulative temperature than the control and dry inoculant silages, 20.52°C and 3.96°C respectively. There were no statistically significant differences between treatments either in bacterial or mold population. All three alfalfa silage treatments were stable, MPC went up in wet treatment only.

Correlations performed between aerobic stability measurements after 7 days of air exposure for the control silage showed a high correlation (0.93, $P \leq 0.01$) for maximum temperature rise based on actual temperature and cumulative temperature rise above room temperature over 7 days (Table 11).

Data summarized in Table 12, showing correlations for the dry inoculant after 7 days of air exposure pointed out a high correlations between pH and maximum temperature (0.81, $P \leq 0.05$).

Bacterial plate count (BPC) had a high correlation coefficient (0.88, $P \leq 0.01$) with mold plate count for

Table 10. Aerobic stability of the inoculated and uninoculated alfalfa silage after 0 and 7 days of air exposure.

			Inoculant	
	Day	Control	Dry	Wet
DM Loss (% of DM exposed)	7	1.61[a]	6.66[a]	4.50[a]
pH	0	5.77[a]	5.76[a]	5.81[a]
	7	5.78[a]	6.66[a]	6.35[a]
	T changes above room T; °C			
Max T	7	20.52[b]	20.39[b]	20.01[a]
Cum DD	7	4.04[b]	3.96[b]	2.85[a]
	Microorganisms/g			
BPC	0	1.0X10[5a]	3.0X10[5a]	4.2X10[5a]
	7	4.4X10[6a]	4.4X10[6a]	3.1X10[7a]
MPC	0	ND[a]	ND[a]	ND
	7	ND[a]	ND[a]	3.3X10[3a]

Means in the same line followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count expressed as number of bacteria per gram.

MPC - Mold plate count expressed as number of mold per gram.

ND - Not detected

Table 11. Alfalfa silage correlations between aerobic stability measurements in the control silages after 7 days of air exposure.

	DM Loss			
pH	0.65	pH		
Max T	0.35	0.60	Max T	
Cum DD	0.42	0.69	0.93**	Cum DD
BPC	-0.58	-0.45	-0.50	-0.51

*P $\leq$ 0.05 **P $\leq$ 0.01

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

Table 12. Alfalfa silage correlations between aerobic stability measurements in the dry inoculant silages after 7 days of air exposure.

	DM Loss			
pH	0.47	pH		
Max T	0.65	0.81*	Max T	
Cum DD	0.71	0.78	0.93**	Cum DD
BPC	-0.08	-0.50	-0.65	-0.62

*P $\leq$ 0.05 **P $\leq$ 0.01

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

the wet inoculant sample (Table 13).

Correlations between end-product compositions and aerobic stability measurements performed on the control sample showed a high correlation coefficient (0.82, $P \leq 0.05$) between lactic acid and DM loss. Butyric acid had a very high correlation (0.99, $P \leq 0.01$) with pH. Cumulative temperature was significantly negative correlated to both dry matter and dry matter recovery (-0.73, $P \leq 0.05$) (Table 14).

There were high negative significant correlations between maximum temperature, dry matter and dry matter recovery (-0.73 and -0.69 respectively, $P \leq 0.05$) in the dry inoculant sample. Bacterial plate count was highly correlated with dry matter and dry matter recovery (correlation coefficient 0.98, $P \leq 0.01$) (Table 15).

Correlations data for the wet sample summarized in Table 16 indicated that end-product pH and aerobic stability pH were highly correlated. Besides, the aerobic stability pH was significantly correlated to the butyric acid content of the end-product, correlation coefficient was 0.98.

2. Corn trial

There were not statistically significant differences among control and the two treated silages after 7 days of air exposure for dry matter loss or pH as shown in Table 17.

The control silage maximum temperature was significantly higher than the wet inoculant silage. The wet inoculant silage had a significantly lower cumulative temperature rise above room temperature over 7 days than control and dry

Table 13. Alfalfa silage correlations between aerobic stability measurements in the wet inoculant silages after 7 days of air exposure.

	DM Loss				
pH	-0.53	pH			
Max T	0.11	0.56	Max T		
Cum DD	-0.63	0.38	0.76*	Cum DD	
BPC	-0.18	0.30	0.16	0.06	BPC
MPC	-0.17	0.05	0.20	0.35	0.88*

*P $\leq$ 0.05 **P $\leq$ 0.01

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

MPC - Mold plate count.

Table 14. Alfalfa silage data correlations between end-product composition and aerobic stability measurements for the control silages.

	Aerobic Stability				
End Product	DM Loss	pH	Max T	Cum DD	BPC
DM	-0.14	-0.78	-0.51	-0.73*	0.11
DM Rec	-0.14	-0.78	-0.51	-0.73*	0.11
pH	0.25	-0.40	-0.38	-0.60	-0.18
Lactic	0.82*	0.84*	0.33	0.32	-0.59*
Acetic	-0.75	-0.40	-0.03	0.10	0.54
Prop	-0.44	-0.96**	-0.54	-0.71*	0.32
Buty	0.55	0.99**	0.53	0.66*	-0.40
CP	0.00	0.00	0.00	0.00	0.00
NH_3	-0.81*	-0.87*	-0.35	-0.35	0.58
HWIN	---	---	0.00	0.00	---
BPC	0.09	-0.57	-0.44	-0.68*	-0.06

*$P \leq 0.05$ **$P \leq 0.01$

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

Prop - Propionic acid.

Buty - Butyric acid.

CP - Crude protein as a % of the dry matter.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of total nitrogen.

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

Table 15. Alfalfa silage data correlations between end-product composition and aerobic stability measurements for the dry inoculant silages.

	Aerobic Stability				
End Product	DM Loss	pH	Max T	Cum DD	BPC
DM	0.01	-0.53	-0.73*	-0.62	0.98**
DM Rec	0.08	-0.48	-0.69*	-0.56	0.98**
pH	0.70	0.66	0.70*	0.84**	-0.52
Lactic	0.52	0.69	0.81**	0.87**	-0.77
Acetic	-0.76	-0.60	-0.61	-0.78*	0.35
Prop	-0.73	-0.63	-0.66**	-0.81**	0.44
Buty	0.61	0.69	0.77*	0.86**	-0.66
BPC	0.00	0.53	0.73*	0.62	-0.99**
NH_3	-0.20	0.40	0.61	0.46	-0.95**
HWIN	---	---	0.00	0.00	---

*P $\leq$ 0.05 **P $\leq$ 0.01

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

Prop - Propionic acid.

Buty - Butyric acid.

BPC - Bacterial plate count.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of total nitrogen.

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

Table 16. Alfalfa silage data correlations between end-product composition and aerobic stability measurements for the wet inoculant silages.

	Aerobic Stability					
End Product	DM Loss	pH	Max T	Cum DD	BPC	MPC
DM	-0.19	0.70	0.46	0.42	-0.12	-0.40
DM Rec	-0.23	0.75	0.48	0.44	-0.09	-0.37
pH	-0.60	0.98**	0.57	0.47	0.24	0.07
Lactic	-0.67	0.79	0.42	0.31	0.42	0.41
Acetic	0.46	-0.23	-0.06	0.01	-0.45	-0.62
Prop	0.13	0.32	0.26	0.27	-0.31	-0.58
Buty	-0.62	0.98**	0.56	0.46	0.26	0.11
NH_3	-0.20	0.72	0.47	0.43	-0.11	-0.39
BPC	0.68	-0.90*	-0.49	-0.38	-0.37	-0.30

*$P \leq 0.05$ **$P \leq 0.01$

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

Prop - Propionic acid.

Buty - Butyric acid.

BPC - Bacterial plate count.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of total nitrogen.

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

Table 17. Aerobic stability of the inoculated and uninoculated whole plant corn silages after 0 and 7 days of air exposure.

			Inoculant	
	Day	Control	Dry	Wet
DM Loss (% of DM exposed)	7	3.38[a]	2.60[a]	3.35[a]
pH	0	3.53[a]	3.58[a]	3.63[a]
	7	4.52[a]	4.93[a]	4.25[a]
	T changes above room T; °C			
Max T	7	47.22[b]	45.22[ab]	40.11[a]
Cum DD	7	46.96[b]	44.96[b]	31.56[a]
	Days			
Rot	7	5.56[a]	5.44[a]	6.00[a]
DD Heat	7	6.67[a]	6.44[a]	6.78[a]
	Microorganisms/g			
BPC	0	$1.2X10^{6a}$	$3.4X10^{5a}$	$4.1X10^{5a}$
	7	$6.1X10^{5b}$	$3.1X10^{7a}$	$1.2X10^{6b}$
MPC	0	ND	ND	ND
	7	$8.0X10^{4a}$	$9.2X10^{4a}$	$1.4X10^{4a}$

Means in the same line followed by the letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

Max T - Maximum temperature rise based on actual temperatures.

Cum DD - Cumulative temperature rise above room temperature over 7 days (degree days).

Rot - Day that rise above room temperature exceeded 1.7°C.

DD Heat - Day of maximum temperature rise.

BPC - Bacterial plate count expressed as number of bacteria per gram.

MPC - Mold plate count expressed as number of mold per gram.

ND - Not detected.

inoculant silages. The dry inoculant sample showed a significant higher bacterial population than the control and wet inoculant silages.

Data summarized in Table 18 indicated that pH and day that rise above room temperature exceeded 1.7°C were negatively high correlated (-0.79, $P \leq 0.01$) as well as day of maximum temperature rise and mold population.

Correlations between aerobic stability measurements for the dry inoculant sample indicated a high correlation (0.80, $P \leq 0.01$) between pH and day to reach maximum temperature also pH and rot (day of initial temperature rise). There was a significant negative correlation between day that rise above room temperature exceeded 1.7°C and bacterial population (-0.72, $P \leq 0.05$) (Table 19).

Dry matter loss showed a significant correlation with maximum temperature and cumulative temperature rise above room temperature over 7 days (0.89 and 0.87, $P \leq 0.01$ respectively) for the wet inoculant sample as shown in Table 20. The pH was negatively highly correlated with day that rise above room temperature exceeded 1.7°C. The three treatments showed a negative high correlation between pH and day of initial temperature rise as expected, indicating that the earlier day of initial temperature rise occurred, the higher the pH.

Correlations between end-product composition and aerobic stability measurements performed in the control silage showed a significant correlation (0.68, $P \leq 0.05$) between end product pH and aerobic stability pH (Table 21).

Table 18. Whole plant corn silage correlations between aerobic stability measurements in the control silages after 7 days of air exposure.

	DM Loss						
pH	-0.34	pH					
Rot	0.42	-0.79*	Rot				
Max T	-0.05	0.43	-0.03	Max T			
DD Heat	-0.56	0.63	-0.63	0.43	DD Heat		
Cum DD	-0.31	0.39	-0.03	0.72*	0.48	Cum DD	
BPC	-0.21	0.34	-0.34	0.19	0.06	-0.27	BPC
MPC	0.25	-0.60	0.62	-0.44	-0.88**	-0.28	-0.04

*P $\leq$ 0.05 **P $\leq$ 0.01

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise above room temperature over 7 days.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

MPC - Mold plate count.

DD Heat - Day of maximum temperature rise.

Table 19. Whole plant corn silage correlations between aerobic stability measurements in the dry inoculant silages after 7 days of air exposure.

	DM Loss						
pH	0.26	pH					
Rot	-0.17	-0.71*	Rot				
Max T	-0.44	-0.29	0.01	Max T			
DD Heat	0.14	0.80**	-0.80**	0.26	DD Heat		
Cum DD	0.22	0.41	-0.49	0.45	0.66*	Cum DD	
BPC	0.46	0.29	-0.71*	-0.08	0.47	0.26	BPC
MPC	-0.24	-0.39	0.42	0.31	-0.34	-0.42	-0.31

*P $\leq$ 0.05 **P $\leq$ 0.01

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise above room temperature over 7 days.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

MPC - Mold plate count.

DD Heat - Day of maximum temperature rise.

Table 20. Whole plant corn silage correlations between aerobic stability measurements in the wet inoculant silages after 7 days of air exposure.

	DM Loss						
pH	0.75*	pH					
Rot	-0.93**	-0.88**	Rot				
Max T	0.89**	0.74*	-0.87**	Max T			
DD Heat	-0.04	0.09	0.00	-0.40	DD Heat		
Cum DD	0.87**	0.74*	-0.87**	1.00**	-0.44	Cum DD	
BPC	0.48	0.86*	-0.62	0.39	0.24	0.40	BPC
MPC	0.38	0.45	-0.36	0.45	-0.43	0.50	0.41

*P $\leq$ 0.05 **P $\leq$ 0.01

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise above room temperature over 7 days.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

MPC - Mold plate count.

DD Heat - Day of maximum temperature rise.

Table 21. Whole plant corn silage data correlations between end-product composition and aerobic stability measurements for the control silage.

End Product	Aerobic Stability							
	DM Loss	pH	Rot	Max T	DD Heat	Cum DD	BPC	MPC
DM	-0.38	0.81**	-0.69*	0.25	0.25	0.12	0.45	-0.28
DM Rec	-0.33	0.77*	-0.64	0.21	0.16	0.08	0.46	-0.21
pH	0.25	0.68*	0.55	-0.15	0.00	0.00	-0.45	0.07
Lactic	0.06	0.28	-0.16	-0.08	-0.49	-0.24	0.37	0.37
Acetic	0.45	-0.87**	0.75*	-0.30	-0.39	-0.19	-0.44	0.41
HWIN	0.50	-0.90**	0.79*	-0.34	-0.50	-0.24	-0.42	0.50
BPC	-0.50	0.90**	-0.79*	0.34	0.51	0.24	0.42	-0.50
CP	0.20	-0.62	0.49	-0.11	0.09	0.04	-0.45	-0.01
NH_3	0.58	-0.92**	0.84**	-0.41	-0.73*	-0.35	-0.35	0.69*

*$P \leq 0.05$ **$P \leq 0.01$

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

BPC - Bacterial plate count.

MPC - Mold plate count.

CP - Crude protein as a % of the dry matter.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of total nitrogen.

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise based on actual temperature .

DD Heat - Day of maximum temperature rise.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

Hot water insoluble nitrogen was negatively high correlated to pH (-0.90, 0.01).

The end product pH was negatively correlated with (-0.82, $P \leq 0.01$) aerobic stability pH in the dry inoculant sample as indicated in Table 22. The bacterial population was highly correlated to dry matter, dry matter recovery, lactic and acetic acid (0.90, 0.97, 0.84, 0.97, respectively, $P \leq 0.01$). End product bacterial population was negatively correlated (-0.98, $P \leq 0.01$) to aerobic stability bacterial population.

Table 23 indicated a high negative correlation in the wet inoculant sample between pH, lactic acid and bacterial plate count (-0.71, -0.88, $P \leq 0.05$, ≤ 0.01 respectively). The bacterial population was negatively correlated (-0.92, $P \leq 0.01$) with maximum temperature reached.

3. Grain sorghum trial

There were not statistically significant differences for dry matter loss among control and the two inoculant treated samples (Table 24). The wet inoculant sample had a significantly higher pH than the control and dry inoculant samples (7.04, 5.58, 5.75 respectively).

The wet inoculant sample had a significantly higher maximum temperature than the control sample but it was not statistically different to the dry treated sample. The wet inoculant sample had also a significantly higher cumulative temperature than the control and dry inoculum treated sample.

There were statistically significant differences for

Table 22. Whole plant corn silage data correlations between end-product composition and aerobic stability measurements for the dry inoculant silage.

End Product	Aerobic Stability							
	DM Loss	pH	Rot	Max T	DD Heat	Cum DD	BPC	MPC
DM	0.30	0.53	-0.82**	-0.30	0.55	0.12	0.90**	-0.44
DM Rec	0.35	0.36	-0.76*	-0.21	0.46	0.14	0.97**	-0.37
pH	0.06	-0.82**	0.55	0.45	-0.55	0.04	-0.10	0.41
Lactic	0.26	0.61	-0.84**	-0.34	0.59	0.10	0.84**	-0.46
Acetic	0.35	0.37	-0.76**	-0.21	0.46	0.14	0.97**	-0.37
BPC	-0.38	-0.14	0.63	0.09	-0.32	-0.16	-0.98**	0.27
Buty	0.38	0.14	-0.63	-0.09	0.32	0.16	0.98**	-0.27
CP	0.05	-0.82**	0.57	0.45	-0.56	0.04	-0.13	0.42
NH_3	-0.35	0.37	0.19	-0.19	0.06	-0.15	-0.74*	-0.02
HWIN	-0.27	0.59	-0.08	-0.31	0.26	-0.13	-0.49	-0.18

*P $\leq$ 0.05 **P $\leq$ 0.01

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

MPC - Mold plate count.

Buty - Butyric acid.

CP - Crude protein as a % of the dry matter.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of total nitrogen.

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise based on actual temperature.

DD Heat - Day of maximum temperature rise.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

Table 23. Whole plant corn silage data correlations between end-product composition and aerobic stability measurements for the wet inoculant silage.

End Product	Aerobic Stability							
	DM Loss	pH	Rot	Max T	DD Heat	Cum DD	BPC	MPC
DM	0.71*	0.32	-0.61	0.84**	-0.69*	0.85**	-0.01	0.48
DM Rec	0.84**	0.60	-0.80*	0.97**	-0.53	0.98**	0.22	0.47
pH	-0.44	0.06	0.29	-0.54	0.76*	-0.56	0.28	-0.41
Lactic	-0.28	-0.71*	0.44	-0.27	-0.50	-0.24	-0.62	0.07
Acetic	-0.81**	-0.50	0.74*	-0.94**	0.60	-0.95**	-0.14	-0.48
HWIN	0.78*	0.45	-0.70*	0.91**	-0.63	0.93**	0.10	0.48
BPC	-0.82**	-0.88*	0.87**	-0.92**	0.12	-0.90**	-0.53	-0.33
CP	-0.46	-0.81**	0.59	-0.48	-0.36	-0.45	-0.65	-0.04
NH_3	0.87**	0.80**	-0.88**	0.98**	-0.29	0.97**	0.43	0.40

*P $\leq$ 0.05 **P $\leq$ 0.01

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

BPC - Bacterial plate count.

MPC - Mold plate count.

CP - Crude protein as a % of the dry matter.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of the total nitrogen.

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise based on actual temperature.

DD Heat - Day of maximum temperature rise.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

Table 24. Aerobic stability of the inoculated and uninoculated high moisture grain sorghum silages after 0 and 7 days of air exposure.

			Inoculant	
	Day	Control	Dry	Wet
DM Loss (% of DM exposed)	7	1.64^{a}	2.07^{a}	2.35^{a}
pH	0	4.12^{ab}	4.15^{b}	4.10^{a}
	7	5.58^{a}	5.75^{a}	7.04^{b}
	T changes above room T; °C			
Max T	7	35.53^{a}	38.59^{ab}	41.56^{b}
Cum DD	7	27.98^{a}	33.77^{a}	56.74^{b}
	Days			
Rot	7	5.33^{b}	5.11^{b}	2.67^{a}
DD Heat	7	6.33^{b}	5.56^{b}	4.11^{a}
	Microorganisms/g			
BPC	0	$1.0X10^{10b}$	$1.2X10^{6a}$	$3.5X10^{9a}$
	7	$1.2X10^{9a}$	$1.3X10^{9a}$	$6.6X10^{8a}$
MPC	0	$2.3X10^{4a}$	ND^{a}	$3.7X10^{4a}$
	7	$4.2X10^{7a}$	$6.6X10^{7a}$	$1.2X10^{8a}$

Means in the same line followed by the same letter do not differ significantly (Fisher's L.S.D., $P < 0.05$).

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

Rot - Day that rise above room temperature exceeded 1.7°C.

DD Heat - Day of maximum temperature rise.

BPC - Bacterial plate count expressed as number of bacteria per gram.

MPC - Mold plate count expressed as number of mold per gram.

ND - Not detected.

day of maximum temperature and day in which temperature exceeded 1.7°C. The wet inoculant sample reached maximum temperature significantly earlier than the control and the dry inoculant sample. Besides, it also reached day that rise above room temperature exceeded 1.7°C significantly earlier than the control and wet inoculant treated silages. The microbiological population did not show statistically significant differences.

Table 25 summarizes correlations between measurements in the control silage after 7 days of air exposure. Dry matter loss was highly correlated to mold population (0.90, $P \leq 0.01$). The dry matter was not highly correlated to pH (0.42), day that rise above room temperature exceeded 1.7°C (-0.14) and cumulative temperature (0.44) for the control high moisture grain sorghum silage suggesting that it was quite stable compared to the other treated silages.

The pH was significantly correlated (0.80, $P \leq 0.01$) to maximum temperature and also to cumulative temperature over 7 days (0.90, $P \leq 0.01$), day of maximum temperature rise (-0.84, $P \leq 0.01$) and day that rise above room temperature exceeded 1.7°C (-0.77, $P \leq 0.05$) as expected in the dry inoculant treatment (Table 26). Day of maximum temperature rise was highly correlated (0.89, $P \leq 0.01$) to cumulative temperature rise over 7 days.

Dry matter loss was significantly correlated to maximum temperature and cumulative temperature over 7 days (0.85, 0.90 respectively, $P \leq 0.01$) in the wet inoculant treatment. The cumulative temperature over 7 days and maximum temperature

Table 25. High moisture grain sorghum silage correlations between aerobic stability measurements in the control silage after 7 days of air exposure.

	DM Loss						
pH	0.42	pH					
Rot	-0.14	0.03	Rot				
Max T	0.20	0.06	-0.33	Max T			
DD Heat	0.07	0.17	0.00	-0.63	DD Heat		
Cum DD	0.44	0.35	-0.67	0.80**	-0.32	Cum DD	
BPC	-0.17	-0.43	-0.21	0.06	-0.19	-0.18	BPC
MPC	0.90**	0.20	-0.21	-0.06	0.29	0.23	-0.09

*P $\leq$ 0.05 **P $\leq$ 0.01

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise based on actual temperature.

DD Heat - Day of maximum temperature rise.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

MPC - Mold plate count.

Table 26. High moisture grain sorghum silage correlations between aerobic stability measurements in the dry inoculant silage after 7 days of air exposure.

	DM Loss						
pH	0.57	pH					
Rot	-0.71*	-0.77*	Rot				
Max T	0.37	0.80**	-0.42	Max T			
DD Heat	-0.59	-0.84**	0.70*	-0.56	DD Heat		
Cum DD	0.49	0.90**	-0.66*	0.89**	-0.65	Cum DD	
BPC	0.50	0.09	-0.08	-0.06	-0.34	0.14	BPC
MPC	0.37	0.54	-0.42	0.60	-0.44	0.51	-0.22

*P $\leq$ 0.05 **P $\leq$ 0.01

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise based on actual temperature.

DD Heat - Day of maximum temperature rise.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

MPC - Mold plate count.

Table 27. High moisture grain sorghum silage correlations between aerobic stability measurements in the wet inoculant silage after 7 days of air exposure.

	DM Loss						
pH	-0.88**	pH					
Rot	-0.02	0.15	Rot				
Max T	0.85**	-0.71*	-0.31	Max T			
DD Heat	0.47	-0.50	0.49	0.33	DD Heat		
Cum DD	0.90**	-0.74*	-0.04	0.91**	0.51	Cum DD	
BPC	0.30	-0.11	-0.10	0.65	0.20	0.52	BPC
MPC	-0.49	0.40	-0.10	-0.25	-0.45	-0.45	0.33

*P $\leq$ 0.05 **P $\leq$ 0.01

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise based on actual temperature.

DD Heat - Day of maximum temperature rise.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

MPC - Mold plate count.

Table 28. High moisture grain sorghum silage data correlations between end-product composition and aerobic stability measurements for the control silage.

End Product	Aerobic Stability						
	DM Loss	pH	Rot	Max T	Cum DD	BPC	MPC
DM	-0.40	-0.40	0.87**	-0.49	0.88*	0.04	-0.31
DM Rec	0.43	0.45	-0.83**	0.50	0.87**	-0.07	0.32
pH	-0.15	0.03	1.00**	-0.33	-0.67*	-0.21	-0.21
Lactic	0.43	0.45	-0.83**	0.50	0.88**	-0.07	0.32
Acetic	0.47	0.52	-0.77*	0.52	0.88**	-0.12	0.33
Prop	0.26	0.15	-0.98**	0.41	0.78*	-0.11	0.26
CP	0.53	0.66*	-0.60	0.53	0.86*	-0.22	0.33
NH_3	-0.16	0.00	1.00**	-0.34	-0.69*	-0.20	-0.22
HWIN	-0.53	-0.85**	-0.16	-0.35	-0.45	0.47	-0.22
MPC	0.56	0.72*	-0.50	0.52	0.83**	-0.28	0.33

*$P \leq 0.05$ **$P \leq 0.01$

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

Prop - Propionic acid.

CP - Crude protein as a % of the dry matter.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of total nitrogen.

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise based on actual temperature.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

MPC - Mold plate count.

were negatively correlated to pH (-0.74, -0.71 respectively, $P \leq 0.05$) (Table 27).

Correlations between end product composition and aerobic stability measurements for the control sample summarized in Table 28 showed a high negative correlation between day that rise above room temperature exceeded 1.7°C and dry matter recovery, lactic, acetic and propionic acid compositions (-0.83, -0.83, -0.77, -0.98, respectively, $P \leq 0.05$).

End product pH was highly correlated to aerobic stability pH (0.91, $P \leq 0.01$) as shown in Table 29 for the dry inoculant treatment. Also it was significantly correlated to maximum temperature (0.86, $P \leq 0.01$).

Table 30 indicates a significant correlation between end product pH and dry matter loss (0.69, $P \leq 0.05$) for the wet inoculant treatment. The mold population was highly correlated to dry matter loss and day of maximum temperature (0.69, $P \leq 0.05$).

Table 29. High moisture grain sorghum silage data correlations between end-product composition and aerobic stability measurements for the dry inoculant silage.

End Product	Aerobic Stability							
	DM Loss	pH	Rot	Max T	DD Heat	Cum DD	BPC	MPC
DM	-0.10	-0.35	0.29	0.03	0.40	-0.35	-0.63	0.18
DM Rec	-0.17	-0.47	0.38	-0.09	0.49	-0.46	-0.60	0.07
pH	0.55	0.91**	-0.65	0.86**	-0.70*	0.78*	-0.15	0.68
Lactic	0.38	0.79*	-0.61	0.44	-0.72*	0.73*	0.43	0.24
Acetic	0.10	0.35	-0.29	-0.03	-0.40	0.35	0.63	-0.18
CP	0.31	0.37	-0.24	0.60	-0.20	0.29	-0.54	0.62
NH_3	0.00	-0.17	0.17	0.18	0.26	-0.20	-0.65	0.32
HWIN	0.10	0.36	-0.30	-0.02	-0.40	0.36	0.60	-0.17
BPC	-0.26	-0.60	0.47	-0.22	0.59	-0.57	-0.55	-0.04

*$P \leq 0.05$ **$P \leq 0.01$

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

CP - Crude protein as a % of the dry matter.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of total nitrogen.

Rot - Day that rise above room temperature exceeded 1.7°C.

Max T - Maximum temperature rise based on actual temperature.

DD Heat - Day of maximum temperature rise.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

Table 30. High moisture grain sorghum silage data correlations between end-product composition and aerobic stability measurements for the wet inoculant silage.

End Product	Aerobic Stability						
	DM Loss	pH	Max T	DD Heat	Cum DD	BPC	MPC
DM	-0.39	0.28	-0.41	0.69*	-0.64	-0.34	0.39
DM Rec	-0.31	0.16	-0.37	-0.40	-0.58	-0.37	0.32
pH	0.69*	-0.80**	0.52	0.69*	0.74*	0.03	-0.62
Lactic	-0.12	0.39	0.08	-0.00	0.18	0.44	0.06
Acetic	0.05	0.18	0.20	0.17	0.36	0.43	-0.09
CP	0.62	-0.87**	0.36	0.54	0.47	-0.24	-0.53
NH_3	0.68*	-0.77*	0.52	0.69*	0.75*	0.06	-0.62
HWIN	0.65	-0.70*	0.52	0.67*	0.76*	0.12	-0.60
BPC	0.53	-0.48	0.48	0.59	0.72*	0.26	-0.50
MPC	0.69*	-0.80**	0.52	0.69*	0.74*	0.03	-0.62

*$P \leq 0.05$ **$P \leq 0.01$

DM Rec - Dry matter recovery expressed as a % of the dry matter ensiled.

CP - Crude protein as a % of the dry matter.

NH_3 - Ammonia as a % of the dry matter.

HWIN - Hot water insoluble nitrogen as a % of total nitrogen.

Max T - Maximum temperature rise based on actual temperature.

DD Heat - Day of maximum temperature rise.

Cum DD - Cumulative temperature rise above room temperature over 7 days.

BPC - Bacterial plate count.

MPC - Mold plate count.

DISCUSSION

The inoculants were a Gram-negative non-motile, non-spore forming rod-shaped bacteria. They did not grow aerobically but grew anaerobically after precultured in cook meat medium which suggested that they were anaerobic bacteria. They were catalase negative and did not ferment adonitol, dulcitol, inositol, rhamnose, ribose, and sorbitol. These data suggested that the organisms were related to the genus Fusobacterium. According to Bergey's Manual of Determinative Bacteriology (Moore and Holdeman, 1974) major products of Fusobacterium from carbohydrate or peptone include butyric acid, often with acetic and lactic acids, and lesser amounts of propionic, succinic and formic acids; which makes a logical choice for testing as a silage inoculum. No further tests or identification were made of this organism.

I. Anaerobic Phase in Silage Fermentation

1. Alfalfa trial

The alfalfa silages did not stabilize and their compositions changed extensively from day 1 to day 28. Only the dry inoculant silage appeared to have some beneficial results on the alfalfa silage. Moon et al. (1980) concluded that a commercial silage inoculant had beneficial effect on numbers of bacteria (increase), no significant effect on pH, and a negative effect on fermentation acids (decrease) in wheat, alfalfa and corn. The inoculant had little effect in controlling fermentation,and bacteria, although present, were unable to cause rapid pH declines, high lactic acid,

and silage stability. They suggested that the lack of beneficial effects in the silages could have been due to: 1) the inoculant did not compete successfully with natural flora, 2) it did not have sufficient nutrients available for growth and 3) it was not viable. Data in this thesis were in agreement with those of Ely et al., (1979) who found that a dried starter culture and a freeze-dried starter culture did not improve silages of wheat, ryegrass, alfalfa, corn, and sorghum. The authors reported that the culture additives had no effect on recovery of silage nutrients, and although some differences occurred they were not statistically significant.

2. Corn trial

The dry and wet inoculants did not improve the corn silage as compared with the control silage. Nonprotein nitrogen values for treated silages did not differ significantly from those for the control. The microbial treatment increased NH_3-N levels. Lactic acid, acetic acid and ethanol concentrations were not affected by microbial treatment. The pH was not significantly affected by the microbial treatment. Burghardi et al., (1980) reported that recoveries of corn DM and crude protein were not affected by inoculation. Moon et al., (1980) observed that inoculation with Lactobacillus plantarum increased the rate of fermentation of alfalfa, corn, sorghum, and wheat at early stages but few differences were noted in silage quality between inoculated or control silages at the end of a 32 day ensiling period.

3. Grain sorghum trial

The grain sorghum silages data were the most variable and inconsistent. Both the dry and wet inoculants produced similar results throughout the trial. Ely et al., (1981) reported that total facultative anaerobes were increased in alfalfa, corn, sorghum and wheat silages with addition of L. plantarum, and numbers of lactobacilli were increased in all but corn silage. Yeasts and molds were lower only in inoculated alfalfa and wheat silages. These treated silages had a lower pH, higher lactic acid content and greater recovery of dry matter, crude protein, detergent fiber and nitrogen-free extract. They suggested that it is possible that the beneficial effect of an inoculum of nutrient recoveries would be greater if conditions for fermentation were not controlled as carefully, based on a study done by Ohyama et al. (1975) where they observed a positive efect of inoculation when silages were sealed improperly and air was introduced.

II. Aerobic Phase in Silage Fermentation

1. Alfalfa trial

The dry matter loss of the alfalfa silage after exposed to air was not significantly different between the control silage and the inoculant silages, although the dry inoculant showed the highest dry matter loss (Table 10).

The pH values ranged between 5.78 and 6.66 with no statistical differences but again the dry inoculant showed the highest pH (Table 10).

Maximum temperature values showed statistically significant differences with the control sample having the highest value, 20.52°C (Table 10). However none of the silages heated, which was an indication of stable silages for the three treatments. Also the cumulative temperature rise above room temperature over 7 days was highest and significantly different for the control sample with 4°C above the room temperature.

Correlations between aerobic stability measurements in the control silage after 7 days of air exposure showed a high correlation (0.93, $P \leq 0.01$) between maximum temperature and cumulative temperature (Table 11).

Correlations between aerobic stability measurements in the dry inoculant after 7 days of air exposure showed a high correlation (0.81, $P \leq 0.05$) for pH and maximum temperature suggesting that as temperature increases, pH increases. Also there was a high correlation coefficient (0.93, $P \leq 0.01$) between maximum temperature and cumulative temperature (Table 12).

Correlations of temperatures, days dry matter and pH in the wet inoculant after 7 days of air exposure showed again a high correlation (0.76, $P \leq 0.05$) between maximum temperature and cumulative temperature (Table 13). As bacterial plate count increased, mold plate count increased as indicated by the correlation coefficient (0.88, $P \leq 0.05$).

Correlations between end product compositions and aerobic stability measurements were performed for the

control silage, dry and wet inoculant silages. As the lactic acid composition of the end product silage for the control silage increased, the dry matter loss of the aerobic stability also increased indicating that the lactic acid bacteria have utilized dry matter effectively. As butyric acid content of the end product increased the pH of the aerobic stability study increased suggesting that Clostridium caused an increase in pH. The bacterial plate count (BPC) showed a high negative correlation (-0.68, $P \leq 0.05$) with cumulative temperature suggesting that as more bacterial population was present the smallest the cumulative temperature rise above room temperature over 7 days. Also the higher the dry matter and dry matter recovery; the lower the cumulative temperature indicating that silages with a large amount of DM do not reach high temperatures after exposure to air (Table 14).

The dry inoculant showed similar results to the control silage displaying a negative high correlation coeficient between DM and DM recovery (-0.73, -0.69, $P \leq 0.05$ respectively) with maximum temperature suggesting that silages with high percent of DM do not reach very high temperatures. Another interesting correlation is the one between bacterial plate count and maximum temperature (0.73, $P \leq 0.05$) implying that if the BPC is high, the maximum temperature rise based on actual temperature is high also. As expected, dry matter and dry matter recovery were highly correlated to BPC (Table 15).

The wet inoculant silage showed quite different results compared to the control sample and dry inoculant silages. The pH of the end product silage had a high correlation coefficient (0.98, $P \leq 0.01$) with aerobic stability pH. As butyric acid content increases, pH increases, this is a good environment for Clostridium survival (Table 16).

2. Corn trial

The dry matter loss of the corn silage was not statistically different among control and the two treated silages; however, the control sample exhibited the higher dry matter loss (Table 17). The pH values ranged between 4.25 and 4.93 (Table 12). No significant differences were observed but the dry inoculant showed a slight higher pH than the other two silages. Wilkins (1975) reported that exposure of silages to air caused a loss of water soluble carbohydrates, starch and structural carbohydrates, short-chain organic acids while pH and ammonia content increased.

Maximum temperature values showed statistical significant differences with the wet inoculant displaying the lowest temperature, 40.11°C and the control treatment having the highest temperature 47.22°C. Cumulative temperatures also showed significant differences with the control silage exhibiting the highest temperature of all, 46.96°C (Table 17).

No statistical differences were observed for the day that rise above room temperature exceeded 1.7°C (Rot) or day of maximum temperature rise although the dry inoculant showed the earliest rise above room temperature and also

the earliest day of maximum temperature (Table 17).

The bacterial plate count (BPC) showed a significant difference among treatments with the dry inoculant samples having the highest value (Table 17). This is in agreement with Ohyama and McDonald (1975) observations. The mold plate count (MPC) did not exhibit statistical differences

Correlations between aerobic stability measurements were performed for the control, dry and wet inoculants after 7 days of air exposure. As the maximum temperature increased (Table 18) for the control silage the cumulative temperature rise above room temperature over 7 days increased also (0.72, $P \leq 0.05$). Silage with fastest temperature increase had the lowest mold count indicating that temperatures rise had an inhibitory effect in mold proliferation (-0.88, $P \leq 0.01$).

Dry inoculant silage showed a high correlation coefficient between pH and days to reach maximum temperature (0.80, $P \leq 0.01$) (Table 19). The negative high correlation (-0.72, $P \leq 0.05$) between the day that temperature rise above room temperature exceeded 1.7°C and bacterial population indicates that the sooner the temperature increases the faster bacterial population decreases.

The wet inoculant showed as expected a high negative correlation (-0.93, $P \leq 0.01$) between dry matter loss and day in which rise above room temperature exceeded 1.7°C (Table 20). These results agree with Wieringa et al., (1961). The pH and bacterial population showed a high correlation coefficient (0.86, $P \leq 0.01$) suggesting that

the higher the pH, the larger the bacterial population. These bacteria are probably proteolytic type organisms not related with those in the anaerobic phase.

Correlations between end product compositions and aerobic stability measurements were performed for the control, dry and wet inoculant silages. As the pH of the end product in the control sample of the corn silage aerobic stability increased, aerobic stability pH increased also (correlation coefficient 0.68, $P \leq 0.05$) suggesting that if silage pH is high it will remain high after air exposure. Hot water insoluble nitrogen (HWIN) and pH had a high negative correlation (-0.09, $P \leq 0.01$) indicating that if more "true protein" is available in the silage, more lactic acid bacteria is present and therefore there is a lower pH (Table 21).

The dry inoculant showed a different pattern to the control sample. As the end product pH increased the aerobic stability pH decreased (-0.82, $P \leq 0.01$) suggesting that one did not influence upon the quality of the other. The bacterial population showed interesting correlations with dry matter, dry matter recovery, lactic and acetic acid (0.90, 0.97, 0.84, 0.97, $P \leq 0.01$). This explains clearly that silages with high dry matter content, dry matter recovery and pH support a high bacterial population.

The wet inoculant exhibited a high negative correlation between lactic acid and pH (-0.71, $P \leq 0.05$) indicating that a large lactic acid bacteria population can not tolerate a high pH. This is again stated by the high negative correla-

tion between the bacterial population and pH. The bacterial population also caused a decrease in the cumulative temperature rise above room temperature (-0.90, $P \leq 0.01$) (Table 23).

3. Grain sorghum trial

The dry matter loss of the grain sorghum silage was not significantly different for the control sample and the inoculants (Table 24). The loss of DM in silage reflects the activity of respiring aerobic microorganisms. This activity involves the production of carbon dioxide and water. Lactic acid, acetic acid, proteins and sugars from the silage are used as sources of energy (Woolford, 1978). The carbon dioxide produced constitutes the loss of dry matter (Daniel et al., 1970).

The pH values ranged between 5.58 and 7.04. There were statistical differences with the wet inoculant showing a significantly higher value than the control sample and dry treatment. This increase in pH is probably due to the release of ammonia by the deamination of amino acids and also the breakdown of lactic and acetic acids to form carbon dioxide and water (Daniel et al. 1970).

Maximum temperatures showed statistically significant differences. The wet inoculant sample had the significantly highest temperature compared to the control sample. The rise in temperature of silage exposed to air is due to the activities of the microorganisms breakdown sugars to produce carbon dioxide and water; this reaction is exothermic raising the silage temperature (Woolford, 1978).

The cumulative temperature was significantly higher for

the wet inoculant compared to the control sample and dry inoculant. The wet treatment sample reached an increase of 1.7°C over room temperature earlier than the control and dry samples (Table 24) and also reached the maximum temperature earlier than the other two treatments.

Microbiological population did not show a significant difference, and both bacteria and mold counts were quite high.

Correlations between aerobic stability measurements performed after 7 days of air exposure in the control sample showed that the higher the DM loss, the higher the mold population indicating utilization of dry matter (Table 25).

Correlations conducted in the dry inoculant sample related dry matter loss with day in which the rise above room temperature exceeded 1.7°C suggesting that the larger the DM loss, the sooner an increase in room temperature occurred (Table 26).

The wet inoculant sample showed a different pattern compared to the control and dry inoculant samples. It showed a high negative correlation between pH and DM loss and maximum and cumulative temperature. For unexplainable reasons, the correlation data from Table 27 do not agree with similar data obtained from Table 24.

CONCLUSIONS

High bacteria population was recorded in the pre-ensiled alfalfa with and without inoculant application. The mold population was supressed by bacteria population early in the anaerobic phase of the study. Dry and wet inoculant forms did not affect total acids composition, lactic and acetic acid percentages, crude protein, hot water insoluble nitrogen or ammonia values. Silage dry matter contents were increased by both dry and wet inoculants. The wet inoculant generally decreased pH earlier than control and dry inoculant. Dry matter loss and pH of alfalfa silages were not affected by the dry or wet inoculants during the aerobic phase of the study. However the wet inoculant decreased both maximum and cumulative temperatures.

High bacteria population was detected in the pre-ensiled corn forage with and without inoculant application. The mold population was supressed by bacteria population at early stages during the anaerobic phase of the study. Dry and wet inoculant forms did not affect pH, total acids, lactic, acetic, propionic or butyric acid percentages. Silage dry matter contents were increased by both dry and wet inoculants. The wet inoculant decreased ammonia. Dry matter loss, pH, maximum temperature, day that rise above room temperature exceeded 1.7°C and mold population were not affected by dry or wet inoculants throughout the aerobic phase of the study. The wet inoculant decreased maximum and cumulative temperatures.

High bacteria population was observed in the pre-ensiled grain sorghum silages with and without inoculant application. Mold population was recorded in all three silages during the anaerobic phase of the study although it was at low levels. The inoculants did not affect acetic acid, crude protein, hot water insoluble nitrogen or ammonia. The wet inoculant increased the dry matter content total and lactic acids and decreased the pH value. Dry matter loss and bacteria and mold populations were not affected by either of the inoculants during the aerobic phase of the study. The wet inoculant increased the pH and cumulative temperature and decreased the day of maximum temperature rise and day in which rise above room temperature exceeded 1.7^{o}C.

In general, the dry inoculant did not improve silage quality of alfalfa, whole plant corn and high moisture grain sorghum silages. The wet inoculant seemed to favorably affect silage composition of alfalfa only.

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EFFECT OF MILES LABORATORY COMMERCIAL INOCULANTS, ALFALFA, WHOLE PLANT CORN AND HIGH MOISTURE GRAIN SORGHUM SILAGES

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ABSTRACT

The effect of two inoculants, dry and wet form, on alfalfa, whole plant corn and high moisture sorghum grain silages was examined. Microbiological and biochemical tests were performed. Viable cell count and mold plate count were determined. Crude protein, lactic, acetic, propionic, butyric, and total acids, dry matter, dry matter recovery, pH, ammonia and hot water insoluble nitrogen were monitored during anaerobic stages. Dry matter loss, pH, maximum temperature, cumulative temperature over 7 days, day in which rise above room temperature exceeded 1.7^{o}C and day of maximum temperature rise were determined during aerobic conditions.

The dry inoculant did not improve alfalfa, corn and grain sorghum silages. The wet inoculant seemed to have a beneficial effect in alfalfa silage by decreasing pH (days 2, 4, and 7), The wet inoculant did not improve corn silage compared to control treatment since both had similar stability. The wet inoculant did not improve grain sorghum silage quality.