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FERMENTATION CHARACTERISTICS AND FEED VALUE OF CORN SILAGE MADE WITH COMMERCIAL MICROBIAL INOCULANTS

By

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ABSTRACT

FERMENTATION CHARACTERISTICS AND FEED VALUE OF CORN SILAGE MADE WITH COMMERCIAL MICROBIAL INOCULANTS

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Fermentation of corn silage was studied in laboratory silos after imposing different environmental manipulation (washing, ammoniation, freezing or autoclaving) followed by inoculation. Ammoniation increased all nitrogen fractions. In trial 2, inoculation increased lactic acid and ammonia-nitrogen but did not alter pH. Environmental manipulation decreased dry matter, ammonia-nitrogen, water soluble nitrogen and lactic acid but increased water soluble carbohydrates. Corn silage in tower silos had greater concentrations of lactic acid, ammonia-nitrogen, water soluble carbohydrates and nitrogen than when ensiled in laboratory silos. Silage characteristics and animal intake improved from top to bottom of tower silos. Inoculation of forage into tower silos did not change silage characteristics except for increasing water soluble nitrogen. Inoculation did not alter intake by milking cows, their milk production or its composition except that low producing cows fed inoculated silage yielded more milk and consumed more feed than similar cows fed control silage. Steers fed control silage had greater intake and gain than did those fed inoculated silage.

DEDICATION

For her inspiration, extreme patience,

and steadfast loyalty,

To the nicest, finest creature,

and my guardian angel

Souhair El-Santawy.

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INTRODUCTION

Corn silage has become an important source of forage for dairy cattle because of its high energy content, ease of mechanization in a feeding system, and high yield. The increasing amounts of this valuable forage, harvested by farmers, is a clear recognition of its profitability and of its nutritional importance for beef and dairy cattle.

Corn is grown extensively in the United States of America and other countries. In Michigan, the production of corn silage was 4,940,000 metric tons in 1983. The acreage was 380,000 and the yield per acre was 13 tons. Corn is still to be Michigan's leading crop in both acreage and value (MI Ag. Stat., 1984).

A brief historical account about corn silage and ensiling process is worthwhile. In my country, Egypt, we have known silage for centuries. Our great ancestors, the Pharaos, were very familiar with the ensiling process as a means of preserving crops, especially corn, wheat and faba beans. Peter McDonald (1981) in his book, The Biochemistry of Silage, praised the ancient Egyptians for that. The first upright silo in the United States was built in 1875 and by 1890 there were 2,500 silos in Wisconsin alone (Bunting et al., 1978).

A preferred aim in conserving or preserving any crop is to harvest it at the optimal stage of growth with minimum loss of nutrients so that it can be used to maximum advantage by animals during periods when the growing crop is unavailable for feed. In terms of ensiling characteristics, corn silage could be considered an ideal crop since it is relatively high in dry matter content, is of low buffering capacity, and contains adequate amounts of water soluble carbohydrates for satisfactory formation of lactic acid. A major disadvantage of corn forage, however, which is commonly used as a silage, is its relatively low content of crude protein. This low content of crude protein could be overcome by proper supplementation with protein or by addition of urea or ammonia at the time of ensiling. Ammonia treatment results in a silage containing a higher crude protein content than the untreated silage and the non-protein nitrogen is utilized by ruminant microbes.

Microbial additives have been explored as a means to improve fermentation of corn silage. Different strains of lactic acid-producing bacteria have been used. The most attractive are those called "homofermentative lactic acid bacteria." Several studies are being conducted to study the effects of microbial additives on fermentation processes. Very few trials have been conducted in field-scale silos. Even fewer trials have been conducted to study the effects of microbial treated corn silage on animal performance.

This study was conducted to compare laboratory-scale fermentation and field-scale fermentation. Another objective was to study the effects of different environmental factors on the fermentation of corn forage. Another objective was to compare animal performances when fed microbial treated corn silage to that when fed control silage.

LITERATURE REVIEW

Fermentation of Corn Silage

The objective of ensiling whole plant maize is to preserve it with minimal loss of nutrients so that it can be used to feed ruminant livestock (Bunting et al., 1978). The major compositional changes during ensiling are fermentation of water soluble carbohydrates (WSC) to short chain organic acids and alcohols. The principle fermentation product is lactic acid produced by bacterial activity (Bunting et al., 1978).

The decrease in pH resulting from the production of lactic acid and acetic acid effectively preserves the crop (Bunting et al., 1978). The absence of butyric acid and the relatively sweet, yet acidic, smell of the ensiled product is indicative that it has been well preserved with relatively little loss of energy.

Bergen et al. (1974) mentioned that ensiling causes the protein to be degraded because of plant enzymes. According to Bunting et al. (1978) accumulation of acids and reduction of oxygen, eventually inactivates the plant enzymes. Further degradation of protein can occur due to bacterial enzymic activity.

There is generally a small increase in the ash concentration of forage maize associated with the ensiling process, as a result of losses of organic components during the fermentation. However, actual mineral loss may occur with wet crops in the effluent from the silo.

Loss of Nutrients

There is a general recognition that both of the changes in nutritional value of the ensiled crop and consequent nutrient losses in silage making are relatively low (Bolsen et al., 1984; Wilkinson, M, 1978 and Zimmer, E., 1980). The percentage of the "unavoidable" losses due to residual respiration is 1-4% and that caused by fermentation is 3-8% (Wilkinson, 1978 and Zimmer, 1980) under good storage conditions.

Influence of Air Trapped and Aerobic Deterioration

Assuming that ensiling is properly done and silos are adequately sealed, the air trapped in the crop or at the time of ensiling, had an insignificant influence on nutrient loss (Woolford, 1984a). Indeed, in an adequately sealed silo, oxygen is soon exhausted from the silo atmosphere. However, in an inadequately sealed silo, air that enters can exert its influence. This influence is on the process and the final product from the time the filling operation has been completed until the feed is finally consumed by livestock. Air trapped or that entering the inadequately sealed silos, results in breakdown of nutrients to carbon dioxide and water and can lead to considerable dry matter loss (Woolford, 1984).

Irrespective of the efficiency with which air is excluded from the silo, there comes a point when the silage has to be fed out and thus, exposure to air is inevitable at this time.

Definition

Aerobic deterioration is the spoilage that occurs to the silage when it is exposed to air when removed from the silos to be fed out to animals, or at the silage surface in the silo when silage is being removed periodically from the silo.

Relevant factors can be grouped into two areas (Barry et al., 1980). The first concerns the chemical characteristics of the silage at the end of anaerobic fermentation, particularly if these predispose the silage to either stability or to heating and deterioration when it is subsequently exposed to air (Barry et al., 1980). The second factor is the activity of aerobic micro-organisms under aerobic conditions.

Woolford (1984) has concluded that a large increase in aerobic bacterial numbers preceeds the visible increase in fungi in aerobic deterioration of silages.

Feeding Value of Corn Silage

Corn silage is a major ingredient in winter rations for ruminants in the United States due to its ease of production and relatively high energy content, but the low protein and mineral content make it poorly balanced for milk producing cows.

Owen et al. (1957) conducted three experiments over a period of three years to determine the feeding value of corn and sorghum silages. In each feeding trial, the difference in silage consumption, production of 4% fat corrected milk (FCM), and change in body weight were highly significant for the corn silage over a variety of sorghum silages.

Yet sorghum is the principle feed crop grown in some areas of the United States where rainfall is limited.

Earlier investigators (Wheeler, 1895 and Williams, 1904) found increases in milk yield when corn silage replaced hay or some of the grain in dairy cows ration. This increase was due to increased consumption of corn silage (Williams, 1904) or total digestible nutrients (TDN) (Fairchild, 1923). Similar results were obtained by Converse (1928).

Belyea et al. (1975) assigned the dairy cows uniformally to three forage treatments to compare production and body weight over three lactations. Treatments were corn silage ad libitum, corn silage restricted plus hay-crop silage, or hay ad libitum. Milk production did not differ significantly among forage treatments. Changes of body weight were similar, so were feed intakes. They concluded that there was no advantage for hay included with corn silage. Cows fed a high proportion of corn silage had increased incidence of health problems and removal from the herd which can be related to the extraordinarily high energy density of that particular ration.

Holter et al. (1975) used a 4 x 4 Latin square trial to evaluate possible associative feeding effects between corn and hay-crop silages. Ingredients fed were concentrate mixture, urea treated corn silage and early-cut, wilted hay-crop silage in dry matter ratios of 55:45:0, 55:30:15, 55:15:30, and 55:0:45. Ingredients were fed separately, concurrently, and twice daily. Milk yield (18 to 19 Kg) and dry matter intake (2.61% of body weight) were not significantly different among diets. They concluded that no associative feeding effects were significant. The two silages were equivalent in supporting milk production when fed in equal amounts of dry matter. Similar conclusions were made by Johnson et al. (1975).

Greive and co-workers (1980) conducted an experiment to determine the long-term effects of feeding corn silage as the only forage from birth to completion of three lactations in comparison to corn silage plus hay-crop silage or chopped hay. There was no difference of body weight and body condition across treatments. Greater dry matter intake was found for cows fed silage-hay combination than those fed corn silage alone in each period except second lactation. Milk and solid-corrected milk (SCM) yields were greater for animals fed corn silage and hay than corn silage alone in the first lactation. They also concluded that addition of hay-crop silage or chopped hay to corn silage did not influence milk composition or feed efficiency expressed as the ratio of solid-corrected milk production to dry matter intake. Similar results were obtained by Thomas et al. (1970).

An all corn silage program was evaluated (Hemken et al., 1967; Brown et al., 1965; and Rumsey et al., 1963). Corn silage alone or in combination with hay was fed. They concluded that with a high-energy forage, such as corn silage, there should be a little problem in obtaining a good energy intake and maintaining high levels of production. Results obtained by Byers et al. (1967) indicated that milk production results were about equal for both 55% and 32% dry matter harvested corn silage when fed in a combination with hay. Results, however, with all corn silage program were variable. Dry matter intake was low in one experiment (Converse et al., 1952) whereas

Greive et al. (1980) found no difference for all treatments. Others reported low dry matter intake but equal milk production (Brown et al., 1965).

Thomas et al. (1970) and Hemken et al. (1967) referred to the problems that would appear when using corn silage as sole forage. Problems were retained placentae, goiter, low protein intake, low vitamin A intake, and Ketosis. They eventually concluded that corn silage could be used for milking cows as the only forage when the rations were properly supplemented and energy concentration was not excessive.

Corn Silage Additives

Silage additives or aids are used to change fermentation characteristics and decrease dry matter losses during ensiling. The fact that there are large variations in "quality" of crop material emerging from storage facilities (silos) has led to the use of additives. These are often termed "preservatives" and proponents claim that they improve silage quality, reduce losses, and prevent deterioration of the ensiled product.

There are several types of additives and they can influence the process by different means (Thomas, J.W., 1978). Silage fermentation aids include those products that include lactic acid producing microorganisms, nutrients required by these organisms, enzymes and/or microorganisms that increase the availability of fermentable carbohydrates (Bolsen et al., 1984).

Non-Protein Nitrogen Additives

Bunting et al. (1978) mentioned that the relatively low crude protein content of corn silage is widely recognized. Various approaches have been made to overcome this nutritional limitation. Attempts at improving the protein content by breeding have met with little success as dry matter yield appears to be inversely related to the protein content.

Nitrogen fertilizers increase the protein content but results have been small and variable, and its present cost makes this approach uneconomic. A nitrogen source such as urea or ammonia can be added to corn forage as ensiled to increase the nitrogen content of corn silage and this practice is used by many farmers and has become a recommended practice.

Milk production and gains of body weight of cattle fed corn silage treated with ammonia at the blower were equal or superior to those fed control (Boman, 1979; Huber et al., 1979; Huber et al., 1973; Huber et al., 1972).

Mineral Additives

Corn silage is comparatively low in its mineral content and this has implications in the fermentation of silage and in ration balancing. The production of organic acids during the ensiling process decreases pH. The decrease may be sufficient to inhibit or kill the lactic acid-producing micro-organisms. The addition of minerals such as ($CaCO_3$) neutralizes the great decrease in pH so that the bacteria can grow for a longer period. Also, limestone corrects the deficient mineral (calcium) content of the ensiled material.

Acid Additives

Long et al. (1971) stated that direct acidification of forages ranks next to wilting as the method most commonly used on the farms of the world for improving silages. The theoretical principles of this process was developed by A. I. Virtanen about 1925. On the basis of this fundamental work, Virtanen started the use of mineral acids on farms. This use was subsequently known as the A.I.V. process. Later on, organic acids were used. The present goal of direct acidification of silage is to immediately reduce the pH of the ensiled material to about 4. The use of acids includes formic, phospheric, sulfuric, and lactic acids or mixtures of these acids. Formic acid, formaldehyde, and a mixture of both have been used for hay-crop silages in Europe.

Microbial Additives

Woolford (1984) mentioned that inoculum treatments are, in theory at least, intended to promote a desirable fermentation and yield a good quality material. Actually, French investigators applied Lactobacillus to silages in the early 1900's.

Microbial cultures (Kiros, 1962 and Papendick et al., 1970) have been shown to affect chemical composition of silages. Gross (1969) ensiled whole-plant corn (43% dry matter) with several levels of Lactobacillus cultures. He found Lactobacillus treated silages had a lower pH, higher lactic acid, and lower butyric acid concentrations than untreated silage.

Buchanan-Smith et al. (1981) found that an additive containing lactic acid bacteria did not affect final pH, or concentrations of lactic or acetic acids, crude protein or ammonia-nitrogen in the resulting corn silage of 35-40% dry matter.

In an earlier experiment, Burghardi et al. (1980) evaluated the fermentation of microbial treated corn silage in laboratory silos. They inoculated Lactobacillus Bulgaris, Lactobacillus Acidophilus, Lactobacillus Bervis, Streptococcus Lactic, and Streptococcus Cremoris at two levels [(4.5 x 10^7) and (22.5 x 10^7)] of live organisms per kilogram wet forage. They found that recoveries of dry matter and crude protein were not affected by inoculations. Non-protein-nitrogen proportions tended to be increased by the higher level of added organisms. However, values for treated silages did not differ significantly from those for control silages. All microbial treatments tended to increase ammonia-nitrogen levels. With three species of the bacterial cultures (L. Bulgaris, S. Lactic, and S. Cremoris), there was no effect of the treatment on lactic acid, acetic acid, and ethanol concentrations. Acid detergent fiber, cell wall constituents and pH were not significantly affected by level or type of organisms. With commercial live bacterial silage additives, they found no effect on feedlot performance of steers. They concluded that the added organisms increased protein degradation and did not result in higher quality corn silage. However, it is known that field situations may differ from laboratory conditions. The same conclusion may also apply to experimental silos (usually termed laboratory silos) and field silos (upright concrete silos, or other types). Concerning this matter, Thomas (1978) quoted from McDonald and Whittenbury (1973) the following: "In laboratory experiments, the use of a mixed inoculum of L. Plantarium and S.

Faecolis at a rate of 10^7 live organisms per gram fresh forage caused inoculated material to preserve while uninoculated spoiled; however, in the field, results have been disappointing; even though pH of inoculated silage decreased more rapidly, there was no discernible benefit from inoculation." Thomas (1978) also mentioned that similar statements were made by Japanese investigators (Ohyama et al., 1973).

In many trials with organisms, the number of live organisms added has not been known or measured. Lately, some microbial cultures have been used under field and laboratory conditions in which the organisms are known to be viable and have increased microbial numbers in the resulting silage (Thomas, J. W., Personal Communications). Claims for rapid decrease in pH of this inoculated silage need verification.

Other Additives

Other additives for corn silage have been used but experimental evidences for their improving silage quality are inconclusive. Other additives like enzymes, sodium hydroxide, and hydrolytic enzymes with an antioxidant have been used.

However, attempts to improve the nutritive values of corn silage are too numerous for this review. One of the ways to improve the nutritive values of corn silage would be to increase cellulose digestion in the rumen or increase the predigestion of cellulose in the silo. Thomas et al. (1968) continued their introduction saying that predigestion or cellulose hydrolysis while the forage is stored in the silo offers one plausible way of upgrading forages. They mentioned that one investigator reported success with this approach (McCullough et al., 1966).

One experiment was designed to obtain information on composition, digestibility, and animal performance of corn silage to compare with previously published values (Klopfenstein et al., 1967).

Corn silage was ensiled with 35% glycerol-cellulase preparation and fed ad libitum to fourteen Holstein cows (Thomas et al., 1968). They found that cellulase treatment did not decrease the cellulose or crude fiber content of the resulting silage. The acid detergent fiber, cell walls, and hemicellulose (cell wall minus acid detergent fiber) were less for the cellulase treated silage. When fed as the only forage to milking cows, silage consumption was essentially the same (91.7 vs. 91.0 pounds/cow/day) for the two silages. Milk and fat yields, as well as the fat content of the milk, were practically identical for cows fed either silage. None of the above differences even approached statistical significance.

Corn silage was satisfactory when used as the sole forage or in combination with hay for dairy cows. Additives have been used to "preserve" corn silage. Some of these additives did not improve corn silage as compared to a control. Corn forage with microbial additives performed similar to control. More research is needed to evaluate the form of some additives such as ammonia. Use of viable and defined homo-fermentative microbial cultures offers a very plausible approach to improving the resulting silage and nutrient recovery. Well conducted and planned studies in this area could provide basic and practical information that could lead to improved silage under field conditions.

MATERIALS AND METHODS

Forage, Silage, and Chemical Analysis

In experiment 1, corn forage was harvested with a field chopper at about 39% dry matter and portions were then treated with the following:

a. Nothing (control)

b. 500 grams/ton (low level) of commercial (Furst-McNess Co., Freeport, Illinois) inoculum *Lactobacilus plantarium*, *L. brevis*, and *Pediococcus acidilactici*

c. 1,000 gram/ton (high level) of the same inoculum as above

d. 500 gram/ton of the same inoculum as above plus 1% of the dry matter as ammonia (NH_4OH)

e. 1% ammonia (NH_4OH)

The addition of microbes was applied to the forage as a suspension at the rate of 1 gallon of water per ton of forage. A measured amount of ammonia was sprayed on a weighed portion of the forage and mixed by hand. Then portions of the treated forage was compacted into experimental silos (large plastic, thick walled bags of 17 Kg each), evacuated with a vacuum cleaner, sealed, and allowed to ferment for varying times. Each treatment was in triplicate. The silos were opened at 0, 3, 4, 14, and 50 days of fermentation. Samples were taken and frozen at -5°C until time of analysis. Chemical analysis included dry matter, nitrogen fractionations (total nitrogen, ammonia nitrogen, and water soluble nitrogen), and fermentation parameters (lactic acid, pH, water soluble carbohydrates). Inocula was added as a dry powder at the rate of 1 lb/ton.

Chemical Analysis

Frozen silages were allowed to thaw and then ground for 5 minutes to break the long fibers using a Hobart chopper (the Hobart Manufacturing Co., Ohio, USA). Dry matter was calculated by difference after drying 5-6 gram sample in a dry oven at 65°C for 48 hours. Another sample was placed on a tray in a thin layer under a hood that has a fan and the sample air dried. When the sample was dry, it was ground in a Wiley mill to pass a 1 mm screen. Portions were then taken and analyzed for DM, ADF, and ash by procedures described by Goering and Van Soest (1970). Samples were ashed at 600° for 4 hours in a miffle furnace.

Analysis for the nitrogen portions except ammonia nitrogen, which was determined according to procedures described by Chaney and Marbach (1962), was done by using the conventional Kjeldahl procedure. A three gram sample of fresh silage for total nitrogen determination was placed into a pyrex flask, then 25 ml of 98% H_2SO_4 was added with Kjeldahl mixture and boiling beads. Digestion was carried out for 45-60 minutes or until the sample color was bluish-green. Flasks were left to cool and 25 ml of distilled water was added plus 60 ml of sodium hydroxide solution. Distillation was carried out for about 25 minutes and the distillant collected in boric acid solution. For titration, we used HCl (0.1 N). Crude protein was determined by multiplying nitrogen concentration by 6.25. A 10 gram sample plus 100 ml distilled water was homogenized in a sevrall omni mixture. Afterward, the entire mass was filtered through cheesecloth to obtain a filtrate (water soluble extract). The pH was determined immediately using a glass electrode with a digital pH meter (digital ionalyzer model 801, Oiron Research, Cambridge, Mass.). The filtrate was then centrifuged for 20 minutes at 19,000 rpm and the pellet discarded. Aliquotes of this clear homogenate were used to determine water soluble nitrogen, ammonia-nitrogen, water soluble carbohydrates, and lactic acid. The clear homogenate was stored frozen for future analysis. Lactic acid concentrations were determined according to procedures described by Baker and Summerson (1941). Water soluble carbohydrate content was determined using the method described by Dubois et al. (1951) and the color readings were made using Colorimeter (Spectronical, Bausch and Lomb, USA) as in lactic acid analysis.

In experiment 2, corn forage was harvested by a field chopper at 35% dry matter and divided into portions in preparation for placement into experimental silos. The silos were double plastic bags each containing 5 Kg of corn forage. The treatments were:

a. Control (with or without inocula; at the rate of 1,000 g/ton).
b. The standing corn plant was drenched with water from a hose in the field, then chopped and ensiled with or without inocula (as above).
c. After field chopping, corn forage was frozen at -20°C for 2-3 days, then thawed and ensiled with or without inocula (as above).

d. After field chopping, corn forage was autoclaved for 1 hour, then ensiled with or without inocula.

The forage was placed in the plastic bags, then evacuated and sealed and allowed to ferment. A sufficient number of bags were prepared for each experimental lot for duplicates to be opened at 0, 2, 4, 8, and 30 days after ensiling. After the designed date, the silages were frozen until thawed for chemical analysis which included dry matter, pH, lactic acid, water soluble carbohydrates, and nitrogen fractionation as described above.

In experiment 3, corn forage was cut and chopped with a field chopper at 35% dry matter, then 454 grams inocula per ton were added as the forage entered the blower in a 16 x 50 feet concrete silo. A similar silo was filled with untreated silage to serve as a control. Forages were sampled as the loads were unloaded and these samples combined into 3 composites and then frozen for later analysis. When fed, the silages were sampled three times a week and frozen. Prior to chemical analysis, samples were thawed and then composited by two week periods. Analyses were performed as mentioned previously.

Animals

Corn silages from the concrete silos were fed to lactating dairy cows. Twenty-two lactating dairy cows were divided into two groups, one fed treated silage and the other fed control silage. The cows were paired based on stage of lactation and average milk production of two weeks prior to initiation of the experiment and one of each pair randomly assigned to receive one of the silages. Average milk production of those two weeks were 74.2 and 73 lb for the two groups on treated and control silages, respectively (Table 1). Average number of lactations was 2.9 and 2.6 and average number of days in lactation were 88.8 and 78.6 days for treated and control groups, respectively (Table 1). The two groups' average age was 3.7 and 3.5 years for treatment and control, respectively.

Cows were fed a complete balanced ration of either treated or untreated corn silage twice daily at 7:00 a.m. and 1:00 p.m. for 70 days (ten weeks divided into five periods). Rations were supplemented with protein and mineral supplements. Ration ingredients are given in Table 2. Rations were composed of 50% corn silage, 25% high moisture corn, 10% chopped hay, 14.5% protein supplement (Table 3), and 0.5% trace mineralized salt. Cows were offered the feed ad libitum in sufficient amounts to allow 10% refusal. Refusals were collected and weighed daily and feed intakes were calculated by subtracting refusal from amount offered. Animals were kept in individual tie stalls and water was offered on free choice basis. Cows were milked twice daily at 3:00 p.m. and 4:00 a.m. Milk production was recorded also daily. Biweekly milk samples were taken for chemical analysis performed by the Michigan DHIA for protein, fat with total solids calculated. Animals were weighed one week after the initiation and at the end of the experiment for two consecutive days. During the experiment, one of the cows was sick and declined in terms of health and production. The cow was eliminated from the herd before the end of the experiment. All data from this cow and her pair-mate were eliminated.

Table 1

Characteristics of 20 cows listed in two groups (basis on which cows were paired)

Group A (Treatment) Cow #	Age at the Beginning Years	Days	# of Lactations	Days in Current Lactation	Average Milk Production Two Week Prior to Experiment	Pair #	Remarks
1669	4	75	5	50	92.5	1	
1796	4	52	3	82	92.4	2	1st five
1801	4	50	3	101	81.0	3	high producers
1738	5	61	4	75	76.7	4	(treatment)
1873	3	39	2	87	75.7	5	$\bar{x}$ = 83.66 Lb
1829	4	45	3	117	73.6	6	
1687	6	71	5	116	69.2	7	1st five
1063	2	25	1	79	66.0	8	low producers
1877	3	38	2	86	60.6	9	(treatment)
1953	2	26	1	95	54.0	10	$\bar{x}$ = 64.68 Lb
Group B (Control)							
1871	3	41	2	62	94.6	1	
1857	3	42	2	95	82.8	2	1st five
1883	3	39	2	38	81.7	3	high producers
1744	5	60	4	93	79.2	4	(control)
1752	5	61	4	43	76.0	5	$\bar{x}$ = 82.86 Lb
1809	4	49	3	104	74.5	6	
1805	4	49	3	82	64.2	7	1st five
1874	3	39	2	88	62.6	8	low producers
1889	3	37	2	82	60.3	9	(control)
1944	2	28	1	99	54.1	10	$\bar{x}$ = 63.14 Lb
ave. A	3.7	48	2.9	88.8	74.2	A	
ave. B	3.5	45	2.6	78.6	73	B	

Table 2a

Microbial treated and control corn silage chemical ingredients.*

									VFA's		
		TN	WSN	NH_3-N	WSC	LA	ADF	Ash	c_1	c_2	c_3
Ingredient	DM%**	% of DM									
TRT	34.13	1.30	0.738	0.152	2.05	5.68	25.17	5.08	1.47	0.06	0.11
CTRL	35.58	1.23	0.682	0.145	1.91	4.88	23.28	4.70	1.53	0.10	0.08

Table 2b

High moisture corn chemical ingredients

M%	TN	WSN	NH_3-N	WSC	LA	ADF
	% of DM					
70.8	1.45	0.79	0.11	1.96	2.37	2.96

Table 2c

Chopped hay chemical ingredients

DM%	TN	ADF	Ash
	% of DM		
91.79	1.67	52.69	6.34

*Values are averages of 3 samples each.

**DM = Dry matter; TN = Total nitrogen; WSN = Water soluble nitrogen; NH_3-N = ammonia-nitrogen; WSC = Water soluble carbohydrates; LA = Lactic acid; ADF = Acid detergent fiber; VFA = Volatile fatty acid (c_1 = acetic; c_2 = butyric; c_3 = propionic).

Table 3

Protein supplement ingredients*

Ingredient	%
Crude protein**	44.0
Crude fat, minimum	1.6
Crude fiber, maximum	8.0
Calcium, minimum	2.9
Calcium, maximum	3.2
Phosphorous, minimum	1.5
Salt (NaCl), minimum	1.8
Salt (NaCl), maximum	2.2
Vitamin A, minimum	15,000†
Vitamin D_3, minimum	3,000†

*Guaranteed analysis (Kent Feeds, Inc., Iowa).

**This includes not more than 4.4% equivalent crude protein from non-protein nitrogen.

†Usp units/lb.

Corn silages from the concrete silos were also fed to steers. Thirty-two growing Holstein steers weighing 200-225 Kg were randomly chosen from the herd and randomly allotted to four pens (eight steers per pen). Treatments were assigned randomly to pens (two pens per treatment). Animals were fed once a day for 56 days (eight weeks divided into four periods). Corn silage was offered ad libitum as the sole forage to allow 10% refusal. The ration was supplemented with protein and mineral supplement (Table 4). The supplement was mixed with silage prior to introduction to the animals. Feeds were delivered to animals using an automatic belt feeder. Feed refusals were collected and weighed every other day and feed intakes were

Table 4

Proteins, vitamins, and minerals in the supplement fed to steers

Ingredient	%
Corn (dry ground shelled)	14.2
Soybeans (protein)	77.2
Limestone	6.4
Trace mineral salt	1.8
Selenium 200	0.34
Vitamin A	0.05
Vitamin D_3	0.06

calculated by subtracting refusal from amount offered. Steers were weighed for two consecutive days at the beginning and at the end of the experiment and once every other week. Body weights were recorded and average daily gains (ADG) were calculated by subtracting the recent body weight from the previous one and the result was divided by 14.

Statistical Analysis

Statistical analysis of fermentation parameters was carried out using repeat measurement design as described by Gill (1978). Animal performance data were analyzed as a repeat measurement design split plot with blocking of subjects (split block) and calculated using least square method. Persistency of milk production and body weight were analyzed according to "paired-data" design as described by Gill (1978) using student "t" test. Further analysis of cow performance was performed using student "t" test. All data from the animal experiments were analyzed using hand calculations. A terminal (silent 700 ASR Texas Instruments) with a CDC 7200 computer was used for the silage data with statistical packages for Social Science (SPSS).

RESULTS AND DISCUSSION

Chemical Constituents of the Silage

a. Silage Dry Matter

Dry matter (DM) content (Table 5) of the corn silages were not affected by addition of inocula or its amount of ammonia or the combination of ammonia and inocula.

In experiment 2, the different environmental manipulations (treatments) altered the DM content ($P < 0.001$) and that was expected. All treated forages had less DM than control and all treated silages (except one) had less DM than control ($P < 0.001$). However, the DM content of the watered silage and autoclaved silage were greater than that of the initial forage ($P < 0.001$). These significant changes are unexplainable and perplexing. However, these changes may reflect error in the determination of DM.

The observation of no change in DM of ammonia treated corn forage during ensilage is similar to that obtained by Johnson et al. (1982). Huber et al. (1978) using gaseous ammonia treated corn silage found no effect on DM content. A later study (Huber et al., 1980) showed a decrease in DM content when corn forage was treated with aqueous ammonia.

Table 5

Initial and final dry matter content of corn silage ensiled in laboratory silos (trial 1 and 2) with two levels of inocula and/or ammonia as additives and with different environmental manipulations to the forage with or without inoculation.

Experiment 1	Initial (Day 0)	Final (Day 50)	
Treatment	DM%		SD
Control	39.64	39.71	0.63
Inoculum--low[a]	38.04	38.72	0.45
Inoculum--high[a]	39.88	39.03	0.45
Inoculum (low) + NH_3	39.69	38.27	0.77
Ammonia (NH_3)	41.24	38.66	1.33
Experiment 2	**Initial (Day 0)**	**Final (Day 50)**	
Treatment	DM%		SD
Control	38.0	37.1	1.60
Control--inoculated	35.7	36.6	0.64
Watered[b]	32.0	33.4	0.84
Watered--inoculated	36.8	33.6	2.28
Frozen[c]	35.5	34.4	0.84
Frozen--inoculated	35.5	33.3	1.10
Autoclaved[c]	35.8	36.7	0.84
Autoclaved--inoculated	37.2	38.0	0.61
Averages:			
Non-inoculated*	35.33	35.40	
Inoculated*	36.30	35.38	
Non-manipulated**	36.85	36.85	
Manipulated**	35.47	34.90	

*Non-significant ($P < 0.93$). SE = ± 0.264

**$P \leq 0.001$. SE = ± 0.46

SD = Standard deviation of samples throughout each trial.

[a]High and low levels of inoculation, see text.

[b]Environmental manipulation on forage prior to cutting.

[c]Environmental manipulation on forage after chopping.

The result of no difference in DM content of inoculated or control corn silage was similar to that obtained by others (Burghardi et al., 1980; Waldo et al., 1976; and Buchanan-Smith et al., 1981).

In the large cement upright silo experiment, the average DM was 34.13 and 35.59% (Table 6) for the treated and control silage, respectively. There was no significant difference between the two values. Comparing the silages with the initial forage DM, the silage as removed contained less DM than the forage ensiled by 1.6 percent units. These values are within the experimental error.

Both field and laboratory trials indicate that inoculation of the forage did not change the DM content of the resulting silage when compared to non-inoculated silage. In all literature reviewed, however, the inocula treatment had no consistent effects on DM content. There is no known reason why inoculation of a forage as ensiled should alter the DM content of the resulting silage.

b. <u>pH of Ensiled Forage</u>

Addition of ammonium hydroxide raised initial pH to 8.70 (Table 7) and it remained "high" until after day 5. By day 14, the pH was reduced and plateaued at 4.5 to 4.6. This actually was expected because previous investigators (Johnson et al., 1982; Huber, J.T., 1980; Huber et al., 1973; and Huber et al., 1979) found a similar increase followed by decrease in pH values after ammoniation.

Inocula treatment of corn silage did not alter pH during fermentation (day 3 to 14) or final pH (day 50) from that of control silage. These results support those obtained by Burghardi et al. (1980) and Buchanan-Smith et al. (1981).

Table 6

Dry matter of corn silage ensiled in upright silos as removed during five successive periods and the corresponding forage as ensiled.

	Silage as Removed[a]		Forage as Ensiled[a]	
	Control	Treatment	Control	Treatment
Period		DM%		
1	35.15	33.05	37.67	34.54
2	35.60	34.75		
3	35.75	33.30	36.40	34.32
4	36.10	34.05		
5	35.30	35.50	37.63	38.33
Average*	35.6	34.1	37.2	35.7

*$P > 0.05$ (treatment effect).

[a]Values are means of three samples of the forage or silage.

SE = ± 0.174 (of treatment).

Table 7

pH values of corn silage ensiled in laboratory silos in trials 1 and 2, two levels of inocula and/or ammonia as additives and exposed to different environmental factors with or without inocula.

Experiment 4

Treatment	Days after Ensiling 0	3	5	14	50	SD
Control	5.67	4.42	4.25	3.93	4.20	0.68
Inocula (low)	5.74	4.44	4.31	3.97	4.13	0.71
Inocula (high)	5.61	4.54	4.23	3.96	3.98	0.68
Inocula (low) + NH_3	8.77	8.84	6.90	4.56	4.59	2.12
Ammonia	8.70	9.09	7.54	4.64	4.45	2.21

Experiment 2

Treatment	Days after Ensiling 0	2	4	8	30	SD
Control	5.40	4.39	3.78	3.73	3.76	0.72
Control--inoculated	5.20	4.66	3.84	3.66	3.83	0.66
Watered	5.45	4.21	4.06	3.84	3.73	0.69
Watered--inoculated	5.15	4.53	4.05	3.76	3.77	0.59
Frozen	5.39	4.78	4.24	3.93	3.88	0.64
Frozen--inoculated	5.33	4.86	4.16	4.04	3.87	0.62
Autoclaved	5.50	5.41	4.90	4.05	4.60	0.60
Autoclaved--inoculated	5.45	5.13	4.17	3.89	3.70	0.78
Average:						
Non-inoculated*	5.44	4.70	4.25	3.89	3.99	
Inoculated*	5.28	4.80	4.06	3.84	3.79	
Non-manipulated**	5.30	4.53	3.81	3.70	3.80	
Environmentally manipulated**	5.38	4.82	4.26	3.92	3.93	

*SE = ± 0.071 ($P > 0.05$).

**SE = ± 0.147 ($P > 0.05$).

In experiment 2, different environmental manipulations had no significant effect ($P > 0.05$) on initial or final pH values except autoclaved forage and resulting silage had an increased pH compared to control and other treatments. One reason for this may be related to the greater soluble carbohydrate and lower lactic acid contents to be discussed later. Inoculation increased silage pH of autoclaved forage more than for any other treatment. There was no noticeable effect of washing or freezing on the pH from day 3 to 50 when compared to control. Addition of inocula to the control or to the three environmentally altering treatments did not change the pH (Table 7).

In field-scale experiment (Table 8), there was also no significant difference in pH between treated and control silages. The pH of laboratory and cement silo silages were similar even though they were not harvested simultaneously.

Both control and inoculated silages had the greatest pH for silage from the top area of the silo (period 1) and minimum value for silage in the lower part of the silo (period 4 or 5).

c. Lactic Acid Content of Silage

Values for lactic acid concentration (Table 9) in both inocula and ammonia treatment did not differ from that of the control in experiment 1. In experiment 2, the concentrations of lactic acid in the autoclaved silage was the least. Inoculation of autoclaved silage consistently increased lactic acid concentrations. Inoculation of different environmental manipulations increased the overall lactic acid concentrations ($P \leq 0.01$). Although the treatment effects were

Table 8

pH values of corn silage ensiled in cement upright silos removed during five periods and corresponding forage values.

	Silage as Removed Out[a]		Forage as Ensiled[a]	
Period	Control	Treatment	Control	Treatment
1	4.13	4.17	5.52	5.53
2	3.85	3.90		
3	3.82	3.87	5.55	5.55
4	3.74	3.89		
5	3.84	3.87	5.55	5.60
Average	3.88	3.94	5.44	5.56

[a]TRT effect non-significant ($P > 0.5$).

Time effect $P < 0.1$.

not different, the treatment inocula intereaction was highly significant ($P \leq 0.003$). This may have affected the degree of significance of inocula effect since the treatment did not affect lactic acid concentrations. However, values of both experiments throughout the ensiling process were lower than expected. These low values are similar to those obtained by Boman (1980), Britt et al. (1975), and Huber et al. (1979). This may suggest that corn forage might be better preserved when exposed to different environmental factors.

Environmental manipulations may also reduce the endigenous micro-organisms present on the plant (e.g., autoclaving and freezing may kill the endigenous bacteria; drenching may wash bacteria away). When inocula was applied this possible lack of micro-organisms on the forage would be overcome. But inoculation of washed and frozen forage decreased lactic

Table 9

Lactic acid concentrations throughout the experiment of corn forage ensiled in laboratory silos in experiments 1 and 2 with two levels of inocula and/or ammonia as additives and exposed to different environmental factors with or without inocula.

Experiment 1	Days after Ensiling					
Treatment	0	3	5	14	50	SD[a]
	% of DM					
Control	0.30	1.01	2.10	3.09	3.62	1.39
Inocula (low)	0.30	0.94	1.72	2.93	3.46	1.32
Inocula (high)	0.29	1.25	1.87	3.05	3.47	1.30
Inocula (low) + ammonia	0.00	0.00	0.82	3.05	3.48	1.68
Ammonia	0.00	0.08	0.49	2.72	3.12	1.51
Experiment 2	**Days after Ensiling**					
Treatment	0	2	4	8	30	SD[a]
	% of DM					
Control	0.17	1.16	1.43	4.18	2.79	1.56
Control--inoculated	0.09	0.92	1.61	4.45	3.60	1.83
Watered	0.17	1.02	1.25	4.35	3.46	1.77
Watered--inoculated	0.22	1.16	1.39	4.30	2.86	1.60
Frozen	0.10	0.63	1.18	2.93	3.11	1.36
Frozen--inoculated	0.13	1.05	2.19	1.93	3.09	1.13
Autoclaved	0.15	0.17	0.16	1.02	1.10	0.49
Autoclaved--inoculated	0.10	0.44	1.14	2.99	3.64	1.57
Averages[b]						
Non-inoculated*	0.15	0.75	1.01	3.12	2.62	
Inoculated*	0.14	0.89	1.58	3.42	3.30	
Non-manipulated**	0.13	1.04	1.52	4.32	3.20	
Manipulated**	0.15	0.75	1.22	2.92	2.88	

*$P \leq 0.01$. SE = ± 0.043.

**$P > 0.05$. SE = ± 0.076.

[a]SD = Standard deviation

[b]Inocula, treatment interaction ($P \leq 0.003$).

acid concentration, thus a lack of endigenous bacteria was not substantiated.

In the larger cement silos (Table 10), the concentrations of lactic acid were greater than those obtained from laboratory-scale silos. Although the differences between inocula and control were not significant in the large cement silos, the greater lactic acid concentrations than those of laboratory silos may suggest better fermentation and good preservation. The lowest lactic acid concentration was in the top area (period 1) of each silo. There was probably comparatively less anaerobic fermentation in the top position of the cement silos.

Table 10

Lactic acid content of corn silage ensiled in farm upright silos as removed during five periods and corresponding composites as ensiled.

	Silage as Removed Out		Forage as Ensiled	
Period	Control	Treatment	Control	Treatment
	% of DM			
1	2.841	3.941	0.17	0.17
2	5.626	5.978		
3	5.156	6.535	0.30	0.35
4	4.859	6.229		
5	5.932	5.697	0.25	0.28
Average*	4.88	5.68	0.24	0.27

*P = non-significant. SE = ± 1.871.

d. Water Soluble Carbohydrate Contents of Silage

In experiment 1 (Table 11) inocula at two levels did not alter the final water soluble carbohydrate content (WSC). Addition of ammonia alone or in combination with low level inocula did not affect the resultant WSC concentrations. But when ammonia alone was added, the final WSC concentrations were above that of all other treatments. Adding both microbes and ammonia produced WSC concentrations equal to control.

In experiment 2 (Table 11), inoculation increased the final WSC concentrations ($P > .05$). The different environmental manipulations (treatments) increased final WSC concentrations ($P \leq 0.0001$). Inoculated-autoclaved silage had greater WSC content (12.79%) and subsequently greater final WSC concentration. Initial WSC contents in experiment 2, however, were lower than expected and were much lower than in experiment 1. This may have been due to the long time interval between cutting the forage and putting it into laboratory silos.

Carr et al. (1984), Woolfard et al. (1984), and Burghardi et al. (1980) found that the lactic acid production or the rate of acidification which is a function of WSC content of the silage did not differ between control and inoculated silages. The observation of no effect of inocula on final WSC contents of the silages is in agreement with the finding of no effect of inocula on lactic acid concentration (Table 9). It is also consistent with the observation of no effect on inocula on final pH (Table 7). This effect of inocula along with the highly significant ($P \leq 0.0001$) effect of treatments (environmentally manipulating) may explain the highly significant ($P \leq 0.003$)

Table 11

Water soluble carbohydrate content of corn silage ensiled in laboratory-scale silos with two levels of inocula and/or ammonia as additives and exposed to different environmental factors with or without inocula.

Experiment 1 Forage Treatment	Initial (Day 0)	Final (Day 50)	SD[a]
	DM%		
Control	15.48	2.51	5.06
Inocula (low)	11.83	3.45	3.38
Inocula (high)	11.83	2.93	3.49
Inocula (low) + ammonia	12.13	2.97	5.78
Ammonia	11.42	3.91	5.00
Experiment 2 Forage Treatment	Initial (Day 0)	Final (Day 30)	SD[a]
	DM%		
Control	7.31	0.47	2.77
Control--inoculated	7.67	0.68	2.91
Watered	8.13	0.38	3.17
Watered--inoculated	6.57	0.30	2.58
Frozen	8.24	0.37	3.35
Frozen--inoculated	8.10	0.55	2.11
Autoclaved	8.71	3.28	2.31
Autoclaved--inoculated	12.79	4.18	3.40
Averages:			
Non-inoculated*	8.10	1.13	
Inoculated*	8.78	1.43	
Non-manipulated**	7.49	0.58	
Manipulated**	8.76	1.51	

*Non-significant. SE = ± 0.096.

**$P \leq 0.0001$. SE = ± 0.169.

[a]SD = Standard deviation.

effect of treatment-inocula interaction since the treatments ipso facto did not affect final concentrations of lactic acid and pH of the resultant silages.

In the large cement silos (Table 12), the WSC contents were similar for control and inoculated silages ($P > 0.50$). These results are in agreement with those from our model laboratory silos.

Table 12

Water soluble carbohydrate contents of corn silage ensiled in farm upright silos as removed during five consecutive periods and corresponding values of forage as ensiled.

	Silage as Removed Out		Forage as Ensiled	
Period	Control	Treatment	Control	Treatment
	-----------------------	% of DM	-----------------------	
1	2.228	2.739	8.974	8.136
2	1.894	1.972		
3	1.663	2.114	8.285	10.663
4	1.775	1.758		
5	1.965	1.658	9.073	8.229
Average*	1.905	2.046	8.780	9.010

*Non-significant ($P > 0.50$).

e. Nitrogen Content of Silages

Proteolysis occurs in the silage and is due to plant enzymes and action of micro-organisms, although lactic acid bacteria are virtually non-proteolytic. Both WSN and NH_3-N increased during ensiling in all experiments.

In experiment 1, the final concentrations of nitrogen and two nitrogenous fractions are in Table 13 and show little difference except for increase in all categories due to ammoniation. Inoculation did not influence proteolysis or deamination ($P > 0.05$).

In experiment 2, different environmental manipulations ($P > 0.4$), inocula ($P > 0.5$), or interaction of treatment and inocula ($P > 0.8$) did not affect total nitrogen (TN) concentration of the silages. Inoculation did not affect water soluble nitrogen (WSN), but the treatments (environmentally manipulating) decreased WSN significantly ($P \leq 0.001$). Deamination was increased by the inocula addition ($P \leq 9{,}992$) whereas the treatments did not affect the concentrations of ammonia-nitrogen (NH_3-N). There was some decrease in the final NH_3-N concentrations due to treatment effects, but it was non-significant ($P > 0.05$).

In the cement-stone silos (Table 14), TN was not affected by addition of inocula ($P \leq 0.09$). This result is similar to that obtained from our model laboratory silos. Proteolysis was increased by inoculation ($P \leq 0.015$) whereas deamination was not affected by the treatment. The observation of no effect of inocula on the NH_3-N and WSN concentration in the large cement silos is in contrast to that found in the laboratory silos.

Table 13

Nitrogen and nitrogen fractions in corn silage ensiled in laboratory silos with inocula and other variables

Experiment 1	Treatment				
Nitrogen Compounds	Control	Low Inocula	High Inocula	Ammonia + Low Inocula	Ammonia
	% of DM				
Total nitrogen	1.15	1.09	1.15	1.81	2.00
Water soluble nitrogen	0.44	0.42	0.43	0.95	0.91
Ammonia-nitrogen	0.07	0.07	0.07	1.12	1.04

Experiment 2 Nitrogen Component	Treatment	Initial (Day 0)	Final (Day 30)	SD
		% of DM		
Total nitrogen	Control	0.97	1.08	0.08
	Control--inoculated	1.24	1.16	0.06
	Watered	1.17	1.06	0.06
	Watered--inoculated	0.99	1.08	0.03
	Frozen	1.01	1.09	0.04
	Frozen--inoculated	0.99	1.07	0.04
	Autoclaved	0.99	1.12	0.06
	Autoclaved--inoculated	1.09	1.09	0.06
Average		1.06	1.09	
Water soluble nitrogen	Control	0.195	0.469	0.10
	Control--inoculated	0.250	0.430	0.06
	Watered	0.266	0.462	0.07
	Watered--inoculated	0.260	0.288	0.02
	Frozen	0.296	0.447	0.06
	Frozen--inoculated	0.182	0.463	0.10
	Autoclaved	0.190	0.206	0.05
	Autoclaved--inoculated	0.191	0.205	0.04
Average		0.229	0.371	
Ammonia-nitrogen	Control	0.013	0.034	0.01
	Control--inoculated	0.016	0.044	0.01
	Watered	0.013	0.033	0.01
	Watered--inoculated	0.010	0.036	0.01
	Frozen	0.020	0.024	0.01
	Frozen--inoculated	0.018	0.031	0.01
	Autoclaved	0.013	0.004	0.00
	Autoclaved--inoculated	0.015	0.010	0.01
Average		0.015	0.027	± SE
Total nitrogen	Non-inoculated[+]	1.05	1.09	0.02
	Inoculated[+]	1.08	1.10	
	Non-manipulated[++]	1.11	1.12	0.03
	Manipulated[++]	1.04	1.09	
Water soluble nitrogen	Non-inoculated[*]	0.237	0.396	0.06
	Inoculated[*]	0.221	0.347	
	Non-manipulated[**]	0.223	0.450	0.11
	Manipulated[**]	0.231	0.345	
Ammonia-nitrogen	Non-inoculated[†]	0.015	0.024	0.01
	Inoculated[†]	0.015	0.030	
	Non-manipulated[††]	0.015	0.039	0.02
	Manipulated[††]	0.015	0.021	

[+]Non-significant ($P > 0.5$). [*]Non-significant ($P > 0.05$). [†]$P \leq 0.002$.

[++]Non-significant ($P > 0.4$). [**]$P \leq 0.001$. [††]Non-significant ($P > 0.09$).

Table 14

Nitrogen components of corn silage ensiled in farm upright silos as removed at five consecutive periods of two weeks and corresponding values of forage as ensiled.

Nitrogen Component	Period	Silage as Removed Control	Silage as Removed Treatment	Forage as Ensiled Control	Forage as Ensiled Treatment
		% of DM			
Total nitrogen[a]	1	1.290	1.348	1.194	1.152
	2	1.261	1.201		
	3	1.195	1.285	1.030	1.080
	4	1.230	1.320		
	5	1.193	1.357	1.183	1.149
Average†		1.234	1.302	1.136	1.127
Water soluble nitrogen[a]	1	0.718	0.758	0.196	0.211
	2	0.712	0.785		
	3	0.658	0.759	0.209	0.249
	4	0.662	0.738		
	5	0.653	0.649	0.240	0.214
Average*		0.681	0.738	0.215	0.225
Ammonia-nitrogen[a]	1	0.147	0.172	0.016	0.017
	2	0.146	0.140		
	3	0.144	0.152	0.018	0.020
	4	0.145	0.153		
	5	0.141	0.141	0.019	0.015
Average**		0.145	0.152	0.018	0.017

†$P < 0.09$.

*$P < 0.015$.

**Non-significant ($P > 0.09$).

[a]Values are average of three samples of the corresponding treatment.

However, Buchanan-Smith and Yao (1981) found no significant differences for any nitrogen variable measured with respect to inocula treatment. They concluded that the microbial additive they used did not affect fermentation. Similar results and conclusions were obtained by Carr et al. (1984) and Woolfard et al. (1984).

Performance of Lactating Dairy Cows

a. Milk Production

In this experiment, milk production, milk composition (fat and protein), body weight, and feed consumption were measured. Persistency, fat corrected milk (FCM) and milk total solids were calculated. During this experiment, one of the cows became sick and then she and her pairmate were eliminated from all measurements.

Milk production and FCM for the group fed microbial treated corn silage was greater (Tables 15 and 16) than that for those fed untreated corn silage. This increase was not highly significant ($P < 0.25$).

Analysis of data indicated that the interaction of the treatment and block was highly significant ($P \leq 0.001$) (Appendix Table 1a and 2a) for both milk production and FCM. Since blocking the cows was on basis of milk production (during previous 2 weeks), this interaction indicates that the treatment had a differential effect depending on the amount of milk produced. Cows were divided into high and low producers according to their preliminary milk production. The differences between treatment and control in milk production during the experiment were 2.60 lb ($P < 0.2$) and 6.61 lb ($P \leq 0.001$) for high and low producing cows,

Table 15

Milk production in lb/day/cow of 20 cows fed corn silage in five successive periods of two weeks each.[a]

Period Treatment*	1	2	3	4	5	Average	± SE
Treatment	71.53	71.26	69.37	66.08	65.61	68.77	3.39
Control	65.79	66.51	64.56	62.48	61.47	64.16	3.15
Average	68.66	68.89	66.97	64.28	63.54	66.47	0.70

*Non-significant ($P < 0.25$)

[a]Values are averages of the corresponding treatment or period.

Table 16

Fat corrected milk (FCM) in lb/day/cow of 20 cows fed corn silage in five successive periods of two weeks each.[a]

Period Treatment*	1	2	3	4	5	Average	± SE
Treatment	60.24	61.75	64.73	57.68	57.39	60.36	3.24
Control	56.83	57.48	58.03	55.62	63.20	56.23	2.47
Average	58.54	59.62	61.38	56.65	55.30	58.30	1.17

*Non-significant ($P > 0.25$).

[a]Values are averages of the corresponding treatment or period.

respectively (Appendix Table 1b), and in FCM were 2.68 lb ($P \leq 0.5$) and 5.65 lb ($P \leq 0.2$) for high and low producing cows, respectively (Appendix Table 2b). Since cows (i.e., blocks) were originally equal in their milk production ability, we concluded that the treatment had an actual effect on low producers but not on high producers. Milk production during experimental periods is shown in Figure 1. Also, the decrease between preliminary period and period 1 was much greater (7.5 pounds) for control cows than for cows fed inoculated silage (2.2 pounds).

b. Milk Composition

The content of fat, protein, and total solids in milk (Tables 17, 18, and 19) did not differ between treatments for milk protein, milk fat, and total solids. Treatment by block interactions (Appendix Tables 3a, 4a, and 5a) were significant ($P \leq 0.05$, 0.05, and 0.005 for milk protein, fat, and total solids, respectively). Yet, further analysis of these significant interactions (Appendix Tables 3b, 4b, 5b) indicated that the treatment had no effect on high or low producers in terms of milk composition. Only milk protein was primarily different ($P \leq 0.10$).

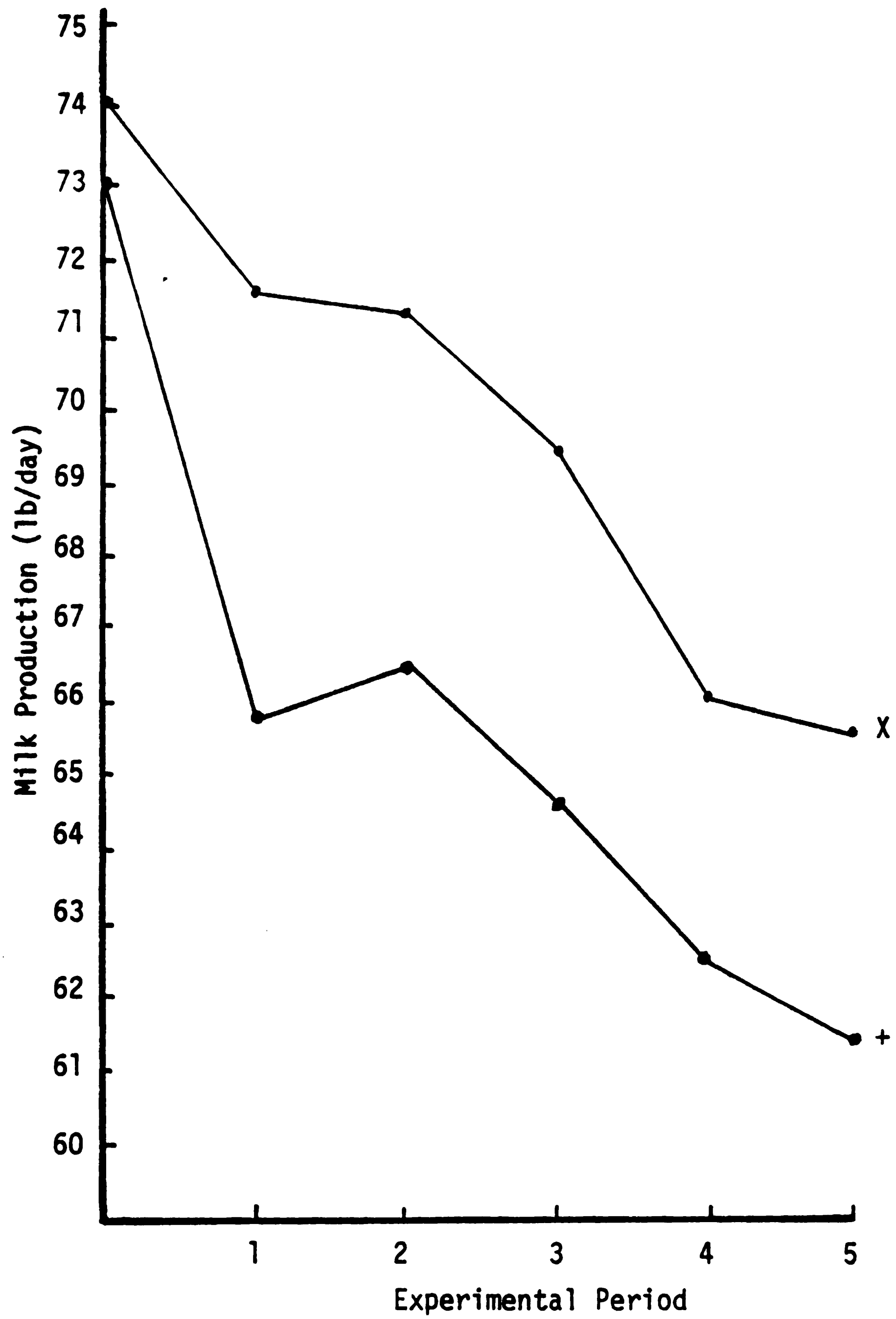

Figure 1. Average milk production (lb/day) of two weeks prior and during the experimental period (five periods of two weeks each) of 20 cows fed either inoculated or non-inoculated corn silage. (X = Treatment; + = Control)

Table 17

Milk protein (in percentage) of 20 cows fed corn silage in five successive periods of two weeks each.[a]

Period Treatment*	1	2	3	4	5	Average	± SE
Treatment	3.05	2.93	3.25	2.89	3.25	3.07	0.058
Control	3.15	2.94	3.44	2.98	3.40	3.18	0.095
Average	**3.10**	**2.94**	**3.35**	**2.94**	**3.33**	**3.13**	**0.042**

*$P \leq 0.10$.

[a]Values are average percentages of the corresponding treatment.

Table 18

Milk fat (in percentage) of 20 cows fed corn silage in five successive periods of two weeks each.[a]

Period Treatment*	1	2	3	4	5	Average	± SE
Treatment	2.96	3.12	3.52	3.15	3.15	3.18	0.126
Control	3.13	3.12	3.30	3.34	3.14	3.21	0.213
Average	3.05	3.12	3.41	3.25	3.15	3.20	0.096

*Non-significant.

[a]Values are average percentages of the corresponding treatment.

Table 19

Milk total solids (in percentage) of 20 cows fed corn silage in five successive periods of two weeks each.[a]

Period Treatment*	1	2	3	4	5	Average	± SE
Treatment	11.51	11.71	12.17	11.61	12.17	11.83	0.181
Control	11.70	11.66	12.21	11.86	12.18	11.92	0.262
Average	**11.61**	**11.69**	**12.19**	**11.74**	**12.18**	**11.88**	**0.106**

*Non-significant.

[a]Values are average percentages of the corresponding treatment.

c. Persistency

Persistency of milk production (treatment divided by pre-treatment) was calculated and statistically analyzed by student "t" test (Appendix Table 6). Persistency of the group fed treated silage (93.65) was greater than that of the group fed untreated silage (91.79). The difference between the two groups was not significant. The variation was great within each group. Examination of production over time in Figure 1 indicates that the decrease in milk production from period 1 to 5 was much less for the control group (4.1 lb) than for the treatment group (6.0 lb). The greater persistency calculated for the treatment group is due to the lesser decrease from preliminary period to period 1 for this group.

The satisfactory milk production over 70 days when a mixture of corn silage (50%), chopped hay (10%), and high moisture shelled corn (25%) was fed, is in agreement with other published data (Grieve et al.

1980; Belyea et al., 1975; Thomas et al., 1970; Rumsey et al., 1963; Brown et al., 1965; and Converse et al., 1952).

d. Dry Matter Intake

Dry matter intake (DMI) (Table 20) for the cows fed inoculated corn silage was (57.77 lb/day and for cows fed control silage it was (55.79 lb/day) ($P \leq 0.5$). The DMI, however, increased throughout the experimental period ($P \leq 0.001$, Appendix Table 7a). Also treatment block interaction was highly significant ($P < 0.001$) which indicates that cows in the blocks reacted differently to treatment. In an effort to determine the reason for this interaction, the blocks of high producing cows were compared to the blocks of low producing cows. The DMI throughout the experimental period (Figure 2) indicates that the increase was greater for treatment cows than for controls. Data in Appendix Table 7b indicates that DMI increased significantly for low producers ($P \leq 0.025$). Figure 2 shows an increase in DMI over time ($P \leq 0.001$). The decrease in intake during the third period may have been due to increase in temperature during this period.

The DMI as percentage of body weight (Table 21) ranged from 3.8% to 5.43% and 3.49% to 5.00% of body weight for treated and control silages, respectively.

Our results are in agreement with those obtained by Belyea et al. (1975) and Greive et al. (1980). Carr et al. (1984) treated corn forage with microbial additives (0.05% of the direct-cut forage) and found no difference in DMI between treatment and control. However, the period effects were significant ($P \leq 0.001$), the same significance

Table 20

Dry matter intake of 20 cows fed corn silage during five periods for treatment and control

Period Treatment*	1	2	3	4	5	Average	± SE
	lb/day/cow						
Treatment	55.55	57.13	56.58	59.01	60.60	57.77	2.111
Control	54.36	55.16	54.95	57.33	57.13	55.79	4.854
Average	54.96	56.15	55.77	58.17	58.87	56.78	0.632

*Non-significant ($P \leq 0.5$).

Table 21

DMI as percentage of body weight for 20 cows fed corn silage over a period of 70 days

Time Treatment	1st Five Weeks	2nd Five Weeks
Treatment	3.60 to 4.73%	3.80 to 5.43%
Control	3.84 to 4.94%	3.49 to 5.00%

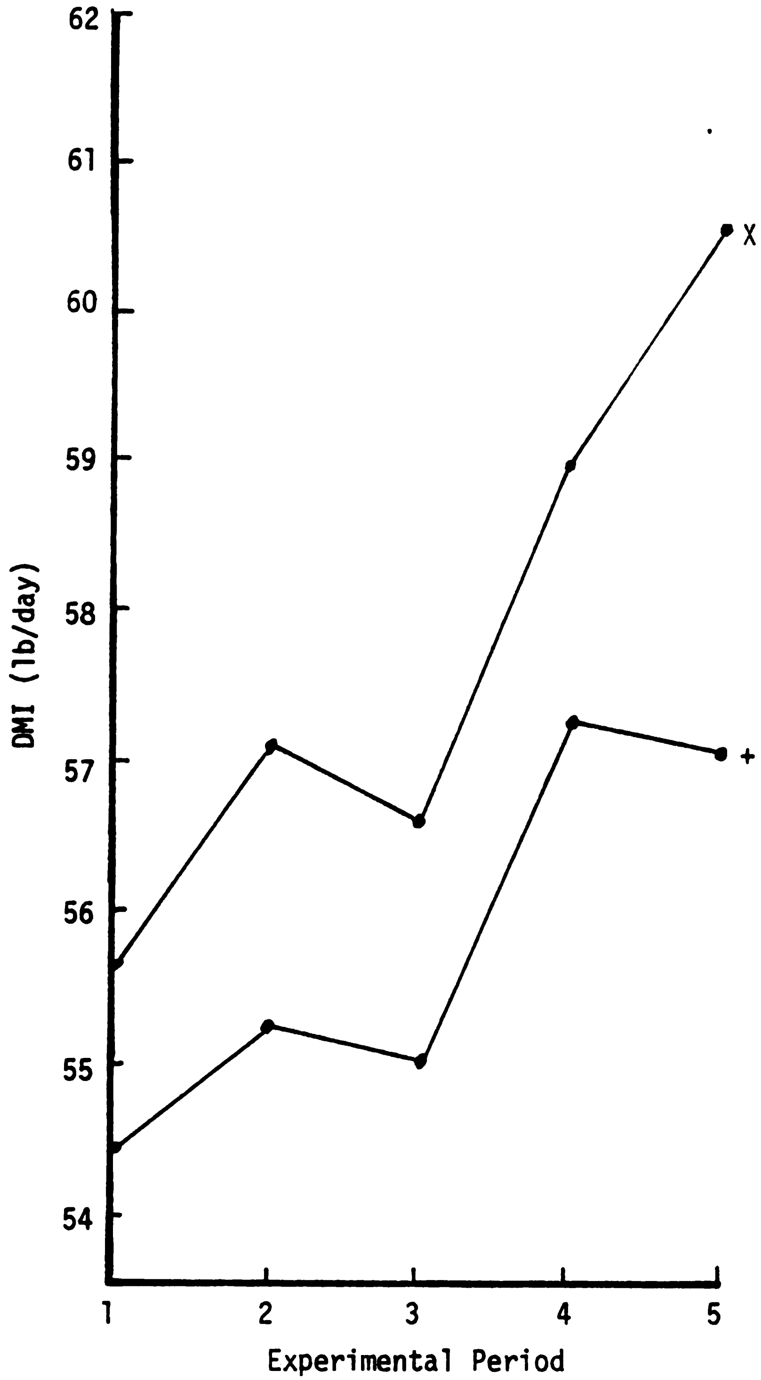

Figure 2. Dry matter intake (DMI) in lb/day during experimental period (5 periods of two weeks each) of 20 cows fed either inoculated or non-inoculated corn silage. (X = Treatment; + = Control)

level that we found in our experiment. They attributed this trend to the association of both acclimation to silage and the onset of cold weather. We also found a similar interaction of treatment and period.

A relationship between animal performances and chemical ingredients of the silage can be made. Silage in the upper area of the silo (period 1) had less anaerobic fermentation and less lactic acid and greater pH indicating a lower quality silage than that in lower areas of the silo. Silage quality measurements and probable nutritive value increased as the silage depth increased indicative of more anaerobic fermentation and better preservation. There was little difference in DM at different silage depths (Table 5). Performance of the animals increased with silo depths. The large decrease in milk production during period 1 may have been related to low quality of silage from the top area of the silo. The increase in dry matter intake in both cows and steers as the experiment progressed supports the idea of increased nutritive value of the silage as silage depth increased.

e. Body Weight Changes

Cows fed inoculated corn silage had a greater DMI (Table 20) and although this difference was not significant, the body weight gains were different ($P \leq 0.0001$). Average daily gains (ADG) were 0.8 and 0.58 lb for treatment and control, respectively (Appendix Table 8). The average age for the two groups was similar. That excludes age from influencing ADG's. Belyea et al. (1975) noted a large increase in body weights during lactation, especially from lactation one to lactation two, but this was highly associated with the fact that cows were young growing. Thomas et al. (1970), however, reported less weight loss during lactation by cows fed corn silage compared with alfalfa hay as the only forage source. The difference in body weight gains in favor of the inoculated silage is difficult to interpret. This amount of gain is not desirable during lactation.

Steers Experiment

a. Dry Matter Intake

Thirty-two growing steers were grouped randomly into groups of eight and randomly assigned to inoculated or control silages which were provided ad libitum. The DMI of the steers (Table 22) averaged 17.62 and 19.02 lb/day/steer for treatment and control, respectively $P \leq 0.10$). Inoculated silage decreased the intake of steers. Steers on control silage had a greater DMI but the difference was not significant ($P \leq 0.10$). Steers on control silage consistently consumed more DMI than those on treated silage. The difference between the two groups was greater in period 1 and 2 (1.91 and 2.08 lb/day/steer). This finding is slightly similar to that found by Burgardi et al. (1980). They found that DMI was identical (5.83 and 5.82 Kg/day/steer) for treated and control groups, respectively. In our experiment, steers consumed more DM than those of Burghardi et al. (1980).

However, we found that the time effect (Appendix Table 9) was significant ($P \leq 0.05$). This increase in DMI over time (Table 22) may be related to growth and adaptation of steers to the environment or to changes in silage quality.

Table 22

Dry matter intakes (lb/day/steer) for 32 steers, grouped in four pens and fed microbial treated or control corn silage during four periods of two weeks each

Treatment* Period**	Treatment	Control	Average
1	16.73	18.64	17.69
2	18.01	18.62	18.32
3	18.12	19.12	18.62
4	17.63	19.71	18.67
Average	17.62	19.02	

*$P \leq 0.10$. SE of treatment = ± 0.449

**$P \leq 0.05$. SE of period = ± 0.370

b. Average Daily Gains

Average daily gains (ADG) of the growing steers are in Table 23. The ADG for the treatment group was 2.06 lb/steer/day and for control group was 2.60 lb/steer/day. In an effort to statistically test this difference, "preliminary F test" was used by pooling the confounded effects of pen/treatment and steer/pen effects (Appendix Table 10). We found that it was significant at $P \leq 0.025$ and this allowed us to give the test more power by testing the effects of treatment against error 1 (pen/treatment). This statistical test showed that the treatment was not effective in increasing ADG. This was expected since the treatment effects on DMI was not significant and had the same significance level ($P \leq 0.10$) as ADG.

Table 23

Average daily gains (ADG) in lb/steer/day for 32 steers grouped in four pens, fed microbial treated or control corn silage during four periods of two weeks each

Period* Treatment**	1	2	3	4	Average	± SE
Treatment	1.44	2.32	2.34	2.14	2.06	0.419
Control	2.05	2.21	2.84	3.30	2.60	0.103
Average	1.75	2.27	2.59	2.72	2.33	0.938

*Preliminary F test accepted at ($P \leq 0.025$) and significance level was $P \leq 0.10$.

**Preliminary F test was not accepted at ($P \leq 0.50$).

Treatment group gained more than control during period 2 (2.32 vs. 2.21 lb/steer/day). The time effects also were analyzed using the same approach. The "Preliminary F test" was not accepted at the first place ($P \leq 0.5$), so we did not proceed further than that since the basic F test was not significant. Time effects might be or expected to be different. This is not always true and time effects were not significant in this trial.

CONCLUSIONS AND RECOMMENDATIONS

Results obtained in this study, although demonstrated a possibility of improved fermentation under field conditions, need to be established more clearly through conducting more ensiling trials. Woolford et al. (1984b) mentioned that although a logical approach to the control of fermentation, the possibility of success is hindered because any such cultrue has to compete for substrates with the indigenous microflora, in addition to the inefficient acid-producing bacteria within the silage. The elimination of unwanted micro-organisms by sterilization prior to inoculation is impractical. In spite of the application of sufficient lactic acid producing micro-organisms, some of the resultant silages had less dry matter content. This may imply, in part, dry matter loss is due to environmental manipulations.

The success of preserving the silage, however, depends on the potential of fodder for ensilage (Woolfard et al., 1984c). Legumes are invariably ensiled less easily than grasses since they generally possess lower concentrations of DM and reserves of fermentable substrates and have greater buffering capacity. To preserve low DM grass, 0.08% of DM as WSC is required (Wieringa, 1961) whereas with legumes at least 0.12% DM as WSC is required (Zelter, 1960). It seems a logical development to conduct fermentation trials on corn forage with greater DM than that we used in this study. This might provide sifficient substrates for lactic acid bacterial cultures to ensure the dominance of these organisms over the indigenous microflora. This is true since Thomas (1978) mentioned that one limiting constituent in many forages for proper ensiling be a sufficient amount of readily fermentable carbohydrates. In addition, the greater the dry matter (i.e., over 30% DM), the better the preservation of the plant material. This will prevent the clostoridial fermentation, encourage decrease in proteolysis, increase in ash and fiber concentrations (Thomas, 1978). Hence, this will result in well preserved nutrients. Weise (1969) has shown an inverse relationship between original microbial numbers and their rate of growth. Thus, when there are few lactic acid bacteria present in the forage, they grow faster than when there are greater numbers present on the forage (Thomas, 1978). So, the rate of acidification and the rapid drop in pH will be achieved more rapidly when fewer numbers of lactic acid bacteria is present. The recommendation here is that high levels of microbial inocula is not useful. The indigenous bacteria may be enough to preserve the silage adequately.

APPENDIX

APPENDIX

Table 1a. Analysis of milk production in (lb/day) of 20 cows fed control or inoculated corn silages over five periods of two weeks each. Design of experiment was split-plot.

Source of Variation	Degrees of Freedom	Sums of Squares	Means of Squares	F_{ratio}	Critical Values	Significance Levels
Treatment	1	530.9799	530.9799	2.14	1.57	(0.25)
Block	9	10013.4553	1112.6061	113.04	4.315	(0.001)
Period	4	485.6578	121.4144	7.93		
Treatment, Block	9	2232.0581	248.0065	25.20	4.315	(0.001)
Treatment, Period	4	12.8922	3.2230	0.33		
Block, Period	36	550.9219	15.3034	1.55		
Treat., Block, Period	36	354.3419	9.8428	No pure Error		
Total	99					

Table 1b. Further analysis of the highly significant treatment, block interaction using Student "t" test.

	Control	Treatment	Difference*
High producers (block 1-5)	74.26	76.86	+2.60**
Low producers (block 6-10)	54.06	60.67	+6.61+

*Difference = treatment - control
**P $\leq$ 0.2
+P $\leq$ 0.01

Table 2a. Analysis of fat corrected milk (FCM) of 20 cows fed control or inoculated corn silages over five periods of two weeks each. Design of experiment was split-plot.

Source of Variation	Degrees of Freedom	Sums of Squares	Means of Squares	F_{ratio}	Critical Value	Significance Level
Treatment	1	425.2256	425.2256	2.29	1.51	0.25
Block	9	7998.9866	888.7763			
Period	4	460.6381	115.1595			
Treatment, Block	9	1377.3566	153.0396	5.58	4.205	0.001
Treatment, Period	4	57.1402	14.2851			
Block, Period	36	1513.1986	42.0333			
Treat., Block, Period	36	986.6772	27.4077			
Total	99					

Table 2b. Further analysis of the highly significant treatment, block interaction using Student "t" test.

	Control	Treatment	Difference*
High producers (blocks 1-5)	64.41	67.09	+2.68**
Low producers (blocks 6-10)	48.05	53.70	+5.65⁺

*Difference = treatment - control
**$P \leq 0.5$
⁺$P \leq 0.2$

Table 3a. Analysis of milk protein of 20 cows fed control or inoculated corn silages over five periods of two weeks each. Design of experiment is split-plot.

Source of Variation	Degrees of Freedom	Sums of Squares	Means of Squares	F_{ratio}	Critical Value	Significance Level
Treatment	1	3.04×10^{-6}	3.04×10^{-6}	3.652	3.36	0.10
Block	9	5.80×10^{-5}	6.40×10^{-6}			
Period	4	3.22×10^{-4}	8.05×10^{-5}			
Treatment, Block	9	7.50×10^{-4}	8.30×10^{-6}	2.358	2.15	0.05
Treatment, Period	4	9.60×10^{-6}	2.40×10^{-6}			
Block, Period	36	7.90×10^{-5}	2.20×10^{-6}			
Treat., Block, Period	36	1.30×10^{-4}	3.50×10^{-6}			
Total	99					

Table 3b. Further analysis of the significant treatment, block interaction using Student "t" test.

	Control	Treatment	Difference*
High producers (blocks 1-5)	0.0298	0.0292	-0.0006**
Low producers (blocks 6-10)	0.0310	0.0304	-0.0006**

*Difference = treatment - control
**Non-significant

Table 4a. Analysis of milk fat of 20 cows fed control or treated corn silages over five periods of two weeks each. Design of experiment is split-plot.

Source of Variation	Degrees of Freedom	Sums of Squares	Means of Squares	F_{ratio}	Critical Value	Significance Level
Treatment	1	1.6×10^{-6}	1.6×10^{-6}	0.040	---	N.S.
Block	9	8.1×10^{-4}	9.0×10^{-5}			
Period	4	1.6×10^{-4}	3.9×10^{-5}			
Treatment, Block	9	3.6×10^{-4}	4.0×10^{-5}	2.171	2.15	0.05
Treatment, Period	4	5.6×10^{-5}	1.4×10^{-5}			
Block, Period	36	6.1×10^{-4}	1.7×10^{-5}			
Treat., Block, Period	36	6.6×10^{-4}	1.8×10^{-5}			
Total	99					

Table 4b. Further analysis of the significant treatment, block interaction using Student "t" test.

	Control	Treatment	Difference*
High producers (blocks 1-5)	0.031468	0.031348	-0.00012**
Low producers (blocks 6-10)	0.03262	0.032232	-0.00039**

*Difference = treatment - control
N.S., **Non-significant

Table 5a. Analysis of milk total solids of 20 cows fed control or inoculated corn silages over five periods of two weeks each. Design of experiment is split-plot.

Source of Variation	Degrees of Freedom	Sums of Squares	Means of Squares	F_{ratio}	Critical Value	Significance Level
Treatment	1	1.9×10^{-5}	1.9×10^{-6}	0.235	---	N.S.
Block	9	1.2×10^{-3}	1.8×10^{-4}			
Period	4	6.4×10^{-4}	1.6×10^{-4}			
Treatment, Block	9	7.4×10^{-4}	8.2×10^{-5}	3.656	3.335	0.005
Treatment, Period	4	3.3×10^{-5}	8.2×10^{-6}			
Block, Period	36	6.0×10^{-4}	1.7×10^{-5}			
Treat., Block, Period	36	8.0×10^{-4}	2.2×10^{-5}			
Total	99					

Table 5b. Further analysis of the highly significant treatment, block interaction using Student "t" test.

	Control	Treatment	Difference*
High producers (blocks 1-5)	0.118552	0.117428	-0.001124**
Low producers (blocks 6-10)	0.119884	0.119248	-0.000636**

*Difference = treatment - control
N.S., **Non-significant

Table 6. Analysis and data of persistency of 20 cows fed control or inoculated corn silages over five periods of two weeks each, using "paired-data" design.

Pair #	Cow #	Treatment	Persistency %	Difference $D=(Y_A-Y_B)$	Remarks
1	1871	B	91.40	- 3.48	
	1669	A	87.92		
2	1796	A	82.10	+ 3.78	$t_{test}=1.865/\sqrt{160.869/10}$
	1857	B	78.32		$=0.465$*
3	1883	B	100.69	- 6.71	
	1801	A	93.98		
4	1744	B	125.34	-14.84	
	1738	A	110.50		
5	1752	B	98.08	-10.8	
	1873	A	87.28		
6	1809	B	60.50	+23.18	
	1829	A	83.68		
7	1687	A	90.64	+ 2.68	
	1805	B	87.96		
8	1963	A	86.68	+ 0.99	
	1874	B	85.69		
9	1877	A	97.67	+ 0.87	
	1889	B	96.80		
10	1944	B	93.09	+ 22.98	
	1953	A	116.07		

Average of difference $(\overline{Y}_D)$=1.85. Variance of difference(S_D^2)=160.869.
Standard deviation(S_D)=12.683.
*Non-significant. A = treatment, B = control

Table 7a. Analysis of dry matter intakes of 20 cows fed control or inoculated corn silages over five periods of two weeks each. Design of experiment is split-plot.

Source of Variation	Degrees of Freedom	Sums of Squares	Means of Squares	F_{ratio}	Critical Value	Significance Level
Treatment	1	98.8633	98.8633	0.887	0.49	0.5
Block	9	1826.9399	202.9933			
Period	4	220.5263	55.1316			
Treatment, Block	9	1002.7011	111.4112	13.967	4.205	0.001
Treatment, Period	4	15.3154	3.8289			
Block, Period	36	313.2755	8.7021			
Treat., Block, Period	36	287.159	7.9767			
Total	99					

Table 7b. Further analysis of the highly significant treatment, block interaction using Student "t" test.

	Control	Treatment	Difference*
High producers (blocks 1-5)	60.85	60.87	+0.02**
Low producers (blocks 6-10)	50.73	54.67	+3.94+

*Difference = treatment - control
**Non-significant
+$P \leq 0.025$

Table 8. Analysis and data of average daily gains in pounds of 20 cows fed control or inoculated corn silages over five periods of two weeks each, using "paired-data" design.

Pair #	Cow #	Treat-ment	Body Weight Before Ave.*	Body Weight At the End*	Body Weight Gain	Difference $Y_D=Y_A-Y_B$	Remarks
1	1871	B	1279.5	1305.5	+26	+ 2.5	Average A** = +561 Lb
	1669	A	1517	1545.5	+28.5		
2	1796	A	1596	1688.5	+92.5	+28.5	Average B** = 408.5 Lb
	1857	B	1363	1427	+64		
3	1883	B	1271.5	1306	+34.5	+28	$\overline{Y}_D$ = 152.5 Lb
	1801	A	1331	1393.5	+62.5		
4	1744	B	1259.5	1285.5	+26	-46	Variance $(\overline{S}_D^2)$ = 848.958
	1738	A	1346.5	1326.5	-20		
5	1752	B	1236	1269.5	+33.5	+43.5	
	1873	A	1223.5	1300.5	+77		Standard Deviation = 29.137
6	1809	B	1273.5	1281	+ 7.5	+ 6.5	of the difference $(\overline{S}_D)$
	1829	A	1479	1493	+14		
7	1687	A	1327	1396	+69	- 8	t_{test} =
	1805	B	1298.5	1375.5	+77		
8	1963	A	1307	1418	+111	+38	$152.5/\sqrt{S_D^2/10}$
	1874	B	1309	1382	+73		= 16.553^x
9	1877	A	1110	1201.5	+91.5	+52.5	
	1889	B	1156	1195	+39		
10	1944	B	1129	1157	+28	+ 7	
	1953	A	1170.5	1205.5	+35		

*Values are averages of 2 consecutive days.
A = treatment, B = control.
^{x} $P < 0.0001$.
**$\overline{ADG}$ = 0.8 Lb (for treatment), ADG = 0.58 Lb (for control).

Table 9. Analysis of DMI of 32 steers in four pens fed inoculated or control corn silage during four periods of two weeks each.

Source of Variation	d.f.	S.S.	M.S.	F_{ratio}	Significance Level
Treatment	1	7.87083	7.87083	9.743	$P \leq 0.10$
Pen/Treatment	2	1.615657	0.80783		
Period	3	2.449177	0.81639	5.966	$P \leq 0.05$
Treatment x Period	3	1.502678	0.50089	3.660	$P \leq 0.10$
(Pen/Treatment) x Period	6	0.821017	0.13684		

Table 10. Analysis of average daily gains (ADG) of 32 steers in four pens fed inoculated or control corn silage during four periods of two weeks each.

Source of Variation	d.f.	S.S.	M.S.	F_{ratio}	Preliminarly F (Significance Level)	F test (Significance Level
Treatment	1	9.3042195	9.3042195	prelim*=5.844	P $\leq$ 0.025	P $\leq$ 0.10
Pen/Treatment	2 } 30	1.3500953	0.67504765	f_{test}=13.783		
Steer/Pen	28	25.6760172	0.9170006143			
Period	3	18.1767398	6.058913267	prelim*=0.934	P $\leq$ 0.5	P $\leq$ 0.5
Treatment/Period	3	6.5611962	2.186698733			
Pen x Period	6 } 90	33.7723141	5.628719017			
Steers x Period	84	72.1764906	0.859243936			

*Preliminary F test.

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