THE FERMENTATION LOSSES, CHANGES IN BACTERIAL FLORA, AND ACIDITY OF GRASS SILAGE PRESERVED WITH DRIED MOLASSES BEET PULP AND SODIUM METABISULFITE

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INTRODUCTION

The uncertainty of good hay making weather, in areas of high rainfall, has provided a definite place for grass silage in the economy of dairy and livestock farms of the Pacific Northwest. Unlike corn, success is not always certain when ensiling grass and legumes. Some type of preservative is often needed, particularly with young high protein forages.

The ever present need of more economical and more efficient methods has led to the use of new and different preservatives for grass silage. The Oregon Agricultural Experiment Station has pioneered the use of dried molasses beet pulp as a preservative.

Dried molasses beet pulp has proven to be an adequate preservative. In addition to its function of providing fermentable carbohydrates, it has the added advantage of being able to retain a considerable portion of the juice normally lost from the silo through drainage (8, p.1).

In efforts to retain a maximum of the valuable juice, large amounts of beet pulp have been added. Two hundred pounds per ton is common, and at times as high as three hundred pounds per ton have been added. When amounts as

large as two hundred pounds of beet pulp per ton are added to forage, the amount invested in preservative approaches the value of the forage. Since this investment is in the form of a cash outlay, questions arise as to the losses or gains during ensiling. Some workers question the economic advisability of adding any preservatives (23, p.11).

No direct studies have been reported of the fermentation losses occuring in silage preserved with dried molasses beet pulp. Studies have been made of other preservatives, and the reported high losses of some grains, when used as preservatives, indicate the desirability of finding out just what losses are incurred with beet pulp (25, p.8).

This work is an effort to determine the losses that may be expected when dried molasses beet pulp is used as a preservative. Studies were made of the changes in the nutrient content of forage preserved with various amounts of beet pulp, sodium metabisulfite, and without any preservative. To facilitate a fuller understanding of the changes occuring during fermentation, the effects of these various treatments upon the three important bacterial populations and upon the changes in acidity were also studied.

REVIEW OF LITERATURE

1. The use of experimental silos.

The use of experimental silos of various sizes has been generally accepted as useful in the study of silages. Silage made in small silos shows little difference from that made in the usual farm-size silo (1, p.150; 7, p.13; 5, p.784; 11, p.32; 28, p.315).

(1) Types of experimental silos used.

Experimental silos used have varied in size from test tubes (49, p.52), quart jars (28, p.315), two-quart jars (5, p.776), and steel drums (7, p.10; 23, p.1), to miniature silos holding up to one or two tons (11, p.32; 58, p.517).

Autry and associates (5, p.784), using two-quart jars as miniature silos, state, "Desirable fermentation was obtained in all cases except in unwilted, untreated alfalfa silage." They concluded, as the result of experiments, that the use of two-quart glass jars as miniature silos offers a convenient technique for studies in silage preservation, having the important advantage that samples may be taken at any interval desired following the ensiling date.

Eckles and associates (11, p.32) state, "A comparison of silage from a large silo and of silage from the same corn put into a small experimental silo showed the quality to be the same, as judged by appearance and by chemical

analysis. For all purposes, except studying temperature changes, the small silo is believed sufficiently accurate for experimental purposes."

Others place more limitations on the use of experimental silos, believing that for checking nutrient losses at least eight to ten tons should be the minimum size (55, p.73).

Martin, Stoddard and Porter (26, p.1082) state, "Although the rate of spoiling may not be the same in miniature silos as in the large silos, the relative differences between the treatment probably would be similar."

(2) The quality of silage from experimental silos.

The majority of workers' reports indicate that there is little difference in the quality of silage from miniature silos judging by appearances, odor, and chemical analysis, than that from full-sized silos (1, p.152; 5, p.784; 11, p.32; 28, p.315; 49, p.151).

(3) The temperature in experimental silos.

Because of the small volume, experimental silos are subject to more influence by the outside air temperature than the large farm silo with its great mass of material. Obviously, the experimental silo is not usable for temperature experiments (11, p.32). However, the ability to influence the temperature within small experimental silos is an advantage that has been used in silage studies.

Stirling (49, p.151) chose glass test tubes as experimental silos because she could control conditions. Blish (7, p.13) thought jars were superior to large cans as experimental silos because they could be kept where temperature control was possible.

Eckles, Oshel and Magruder (11, p.32) stated that good silage was produced at temperatures ranging from 50°F. to 100°F.

2. Acidity in the silo.

(1) Types of acid.

Silage acid may be classified as volatile and non-volatile acids, of which the non-volatile are the most important. Lactic acid is the most important of the non-volatile acids. Malic and succinic acids are also found in the non-volatile portion (54, p.143).

Volatile acids consist chiefly of acetic. Butyric acid may be present under certain conditions, but when present the silage is not considered to be a first class product (54, p.144).

Volatile acids may be found free or combined with bases, usually ammonia. In measurement it is common to distinguish between the two types of volatile acids. The total content is a better indicator of changes that have taken place in the silo than free acids alone (54, p.144).

Dox and Neidig (10, p.338) established that lactic acid is normally present in silage, and gave the average ratio as being 1.0 part non-volatile lactic acid to 0.75 parts volatile acids.

(2) Source of acid.

Allen et al. (3, p.280) state that lactic acid formation is largely, if not all, brought about by lactobacilli and coliform, particularly the former.

Etchells' and Jones' (12, p.31) bacterial findings indicate that the thermophilic facultative anaerobes were responsible for the development of acidity in steamed potato silage. The usual coliforms and lactics were destroyed in steaming.

Hunter (18, p.787) concluded that the production of acids is chiefly due to microorganisms; plant enzymes were chiefly responsible for hydrolysis of proteins with the formation of amino nitrogen; and that the formation of ammonia was due both to plant enzymes and microorganisms.

Lamb (28, p.330) stated that bacteria are mainly responsible for acid production. Rosenberg (45, p.161) found that dominant anaerobes isolated from grass silage were proteolytic and lactate attacking species. According to Salsbury, Mather and Bender (46, p.901), the desired effect in silage preservation is acid production, by lactic acid bacteria, from carbohydrates.

Malzahen, Bechdel and Stone (31, p.60) found that after six months high levels in the small silos used had a pH above 4.2 and small amounts of reducible sugar and lactic acid. Lower levels of the same silos had pH values below 4.2, more reducing sugar, and averaged 31 per cent more lactic acid.

Stirling (49, p.154), in studying controlled laboratory silages, found that the pH of macerated material dropped to 4 in one to two days, and remained at that level, while in the uncut material a pH of 4.6 was reached only after seven days. She thought that perhaps the practice of bruising the fodder by the chopper, while making farm silage, might have value in accelerating the development of acidity, in addition to facilitating consolidation.

(3) Amount of acid needed.

All acids in the silo combine to give a total acidity in silages. This total acidity is an important value. The greater the total acidity, the better the control over undesirable fermentation (54, p.144). Watson (55, p.87) says that the pH value of the silage is the best index of the degree of preservation.

Huffman, in a review of literature (17, p.922), states, "Most investigations emphasize the importance of a pH of 4.2 or below for good silage making, but excellent alfalfa silages have been produced without any treatment when the

pH ranged from 4.6 to 4.78."

The acceptance of a 4.2 pH as a goal has its basis in the findings of various workers, that butyric acid producers do not multiply in silage with a pH below 4.2 (45, p.161; 3, p.293).

Others have placed the lower limit of hydrogen ion concentration somewhat higher. Blish (7, p.12) places it at about a pH of 4.9.

According to Skaggs and Knodt (48, p.332), who used sulfur dioxide as a preservative, all silages reaching a pH of 4.2 within two weeks suffered little fermentation loss after developing this acidity, even though the pH rose to 5 or higher by the end of 120 days. Very few of their experimental silages maintained the lowest pH for any appreciable length of time. pH values gradually rose until they were, for some silages, almost as high as the pH of the grass when it was put into the silo.

(4) Effect of moisture on pH.

When they lowered the moisture by adding dry hay, Stone, Murdock and Bechdel (50, pp.40-41) found there was no material affect on fermentation of silage. They concluded that, "It is evident that fermentable carbohydrates is a more important factor in the preservation of silage than the adjustment of moisture."

3. Loss of nutrients in the silo.

Because the preservation of forage as silage is dependent upon the use of some of its constituents by microorganisms to produce the necessary acid, some reduction of the original nutrient content is expected. Other losses are the result of juice drainage and spoilage. In efforts to cut losses to a minimum, numerous studies have been made of the different losses.

(1) Drainage losses.

Drainage losses vary with the moisture content of the silage, and constitute a tangible loss readily seen by any operator making silage. Often such losses are lumped with the fermentation losses. Some workers have measured drainage loss, and efforts to reduce it have been made (8, p.2; 22, p.11).

The amount of dry matter lost in the juice varies from about 3 per cent to 10 per cent of the original dry matter (23, p.11; 54, p.371).

Archibald and Gunness (4, p.324), using 100 ton silos for seven years, had an average juice dry matter loss of 0.54 per cent with the maximum of 1.08 per cent and a minimum of .12 per cent. They assumed a loss, allowing for that not caught, of 3.25 per cent of the original dry matter, and concluded that juice losses were not as serious as those from other causes.

Watson (54, p.371) in his literature review, concluded that "Drainage is not usually a source of heavy loss of dry matter, seldom exceeding 3 per cent of the original dry matter. Extremely succulent forage or silage subject to leaching by rain may result in an important loss rising to as high as 10 per cent of the dry matter. Crude protein lost in drainage is not excessive, usually consisting almost entirely of the non-protein nitrogenous substances. The ash also contributes a considerable amount to the dry matter of the effluent, particularly when acid solutions have been added."

(2) Spoilage losses.

An important step in silage preservation is the exclusion of air. If this is not properly done, loss results through molding. Such a condition is common at the top, and around openings or cracks in the silo.

The amount of spoilage losses is largely attributable to the construction of the silo and management during filling. Silage that is not tamped and leveled properly, particularly near the top of the silo, will spoil much more readily.

Some investigators ignore top spoilage because it is related to heighth of silo (34, p.669). It is a common practice to differentiate between it and other losses.

(3) Fermentation losses.

The respiration process and microorganisms are responsible for the changes in chemical constituents of the green crop after ensiling. The exact responsibility of each for a given change is not agreed upon by all workers.

Wellson (56, p.541) says that plants cut from roots will continue to respire until every cell is dead. Starch may largely disappear from certain tissues and various sugars increase. In others, sugar may change to starch and respire to CO_2 and H_2O.

Peterson, Hastings and Fred (42, p.1) state, "When green plant material is placed in a closed container, the plant cells continue to respire and produce carbon dioxide and other products. Coincident with the diminishing functions of the plant cells comes an increase in the activities of bacteria, yeasts, and molds."

Plant enzymes have been considered as a source of some losses in the silo. Hunter (18, p.787) states that plant enzymes were chiefly responsible for the hydrolysis of protein with the formation of amino nitrogen. The formation of ammonia was due to both enzymes and microorganisms.

(a) Dry matter.

When measuring dry matter loss two general methods are used, total weight and bag method.

In the total weight method losses are calculated on the total content, using composite samples from all locations in the silo. With the bag method, bags that allow free access to the juices are filled and distributed throughout the rest of the material in the silo. The losses are figured on the changes found in the silage in the bags. The bag method may not take into consideration any difference in quality that may occur due to location in the silo.

Watson and Ferguson (55, p.87), comparing the total weight method with the bag method, found a dry matter loss of 22.2 per cent for the total weight, and 12.9 per cent for the bag method.

Newlander et al. (37, pp.29-30) found that their losses of dry matter figured to total weight, were 15.4 per cent and by the bag method were 9.4 per cent.

Watson (54, p.371), after an extensive review of the literature, concluded that in the silage process approximately one-seventh of the dry matter is lost (14.3 per cent). Slightly less is lost when sugar or similar materials are added to stimulate lactic acid fermentation. Direct acidification with mineral acids shows losses equal to those where stimulation of fermentation is practiced, about one-eighth of the dry matter being lost (12.5 per cent). The loss of dry matter is greatest in silage made from grassland herbage.

Kennedy and Allred (23, p.11) found that the fermentation losses in small silos without preservatives varied from 10 per cent to 20 per cent of the dry matter ensiled. The total loss, fermentation and juices, was about 21 per cent of the dry matter. With molasses as a preservative the loss amounted to about 20 per cent of the dry matter.

Monrow et al. (33, p.247) using one ton silos, observed losses, exclusive of top spoilage, of 5 per cent to 10 per cent of the dry matter.

Neal and Becher (34, p.672), studying soybean silage in large silos, found a dry matter loss of 9 per cent, principally from N-free extract and protein. The average changes were 8.95 per cent dry matter loss, 43.93 per cent loss of crude protein, 20.26 per cent gain of ether extract, 3.09 per cent gain in crude fiber, 17.15 per cent loss nitrogen-free extract, and a gain of 4.54 per cent ash.

(b) Crude protein.

Watson (54, p.371), in his review, states that, on the average, less than one-quarter of crude protein originally contained is lost. In ordinary silage without preservative well over one-third of the protein is lost. In directly acidified and fermentation stimulated silages the loss is only about one-tenth. "In ordinary grass silage about seven-tenths of the 'true' protein of the fresh crop is lost, and in silage made with added sugar or molasses the

loss is still over half, and nearer three-fifths. The use of acid reduces the loss to below half that in the fresh crop, two-fifths only being lost."

(c) Crude fiber.

Neal and Becher (34, p.247) found an increase in crude fiber of 3.09 per cent. Others have noticed this same increase in some silages (6, p.642).

(d) Ether extract.

While the percentage of ether extract in silage is small, an increase during ensiling has been found by many workers. It seems to be normal for most silages to show such an increase. Bender and Bosshardt (6, p.642) report on work that shows increases of as high as 53 per cent in the amount of ether extract. Neal and Becher (34, p.672) reported an increase of 20.26 per cent ether extract in soybean silage. Shepherd et al. (47, p.57) report a 51.8 per cent gain.

4. Additives.

As forage is placed in the silo, various materials may be added to aid in preservation. Roughly, these fall into two classes, direct acidification, and stimulation of lactic acid fermentation.

(1) Direct acidification.

Dilute solutions of mineral acids added in such

proportions as to bring the pH of the mass to a value below 4.0 are used in the A.I.V. method. A limiting pH of 3.0 is placed on such additions, because of possible undesirable effects of very acid materials on animals (54, p.267).

The addition of solutions of acids, either mineral or organic, aids in the control of undesirable fermentation, but does not check all fermentation. Lactic acid and lactic organisms are found in all types of acid silage (54, p.267).

The latest materials used to create conditions unfavorable for undesirable fermentation have been sulfur dioxide and sodium metabisulfite. Both fall in the same catagory, that of increasing the acid by means of an added chemical (9, p.2).

Cowan, Bratzler and Swift (9, p.2) state, "In addition to producing acid, SO_2 gas has other properties which also exert preservative action. The one condition that is absolutely necessary for making good silage is the exclusion of air. Being a powerful reducing agent, SO_2 is able to use up rather large volumes of oxygen and thus is able to contribute to the production of anaerobic conditions. This results in stopping the respiration of the grass very soon after it is ensiled, practically eliminating the chief source of heat in the silo, and thereby retains more green color and carotene and the fresh grass odor and flavor."

The source of heat attributed by Cowan to respiration

does not agree with Hunter (20, p.82) who says that heat in silage is due to bacterial action, and not intramolecular respiration of the plant tissue.

Sulfur dioxide has been used for many years in the preservation of human food. For this purpose it generally has been applied in the form of the acid sulfites of sodium and calcium, which are powders or crystals, and much more convenient to work with than liquid or gaseous SO_2 (9, p.2).

Kennedy and Allred (23, p.11) found that sulfur dioxide cut fermentation losses to half those of forage without any preservative. They found that sodium metabisulfite, although not as effective as sulfur dioxide, was less expensive.

Previous work done at Oregon State College indicates that acceptable silage may be made using sulfur dioxide as a preservative (17, p.2),

Pennsylvania has recommended both sulfur dioxide and sodium metabisulfite as silage preservatives (9, p.2).

(2) Fermentation stimulants.

The practice of mixing forages high in carbohydrates, or materials containing readily available carbohydrates, with forage relatively high in protein and low in carbohydrates has been practiced for many years.

Reed and Fitch (43, p.19), in 1917, concluded that the addition of materials such as molasses, corn chop, and other supplements containing a high percentage of carbohydrate material was effective in preserving alfalfa as silage.

The need for added fermentable carbohydrates is best described by Hunter (19, p.590), who advanced the theory that proteolytic action, which is the cause of the offensive odors characteristic of alfalfa silage, was due in part to the inability of the acid producers to utilize proteins as a source of energy in the absence of carbohydrates.

Molasses is the most universally used preservative (6, p.639) and has been used by many workers as a comparison when checking other preservatives (5, p.775; 8, p.2; 24, p.5; 26, p.15; 25, p.2; 31, p.59).

Grains have also been used to furnish the needed carbohydrates. Some of these have been corn (51, p.242; 32, p.1082; 26, p.19), hominy feed (5, p.775), and barley (25, p.14).

Beet pulp in various forms has been used as a preservative. The first trials were with wet pulp as it came from the sugar mills (15, p.752; 57, p.406). The use of wet beet pulp was not an outstanding success. The wetness of the crop coupled with wet beet pulp caused heavy loss of nutrients.

Watson (54, p.226), discussing the use of wet beet pulp, suggested that perhaps dried molasses beet pulp, which is commercially available, would prove successful.

Byers and Jones (8, p.1) have shown that dried molasses beet pulp is an excellent preservative. In addition to

providing needed carbohydrates, dried molasses beet pulp conserves, by absorption, plant juices that often drain from the silo. Each pound of beet pulp will take up about 2 pounds of juice (8, p.2) which contains about 10 per cent dry matter high in protein, sugars, and mineral matter (8, p.1).

(3) Losses of nutrients from additives.

The addition of carbohydrates increases the cost of each ton of silage fed, and the natural question is how much preservative will be still intact at feeding time. Many workers have apparently ignored such losses, or assumed that they would be in the neighborhood of the normal losses encountered in silage preservation.

Kennedy and Allred (23, p.11) are extremely critical of the use of preservatives. They concluded from experiments that:

1. Untreated silage lost 21 per cent of dry matter by fermentation or juice drainage.
2. Molasses did not significantly decrease the losses, and 20 per cent of the molasses disappeared.
3. Ground grains decreased juice losses slightly, but 15 per cent of the grain disappeared, exceeding the forage saved. A net loss was sustained in its use.

4. Sulfur dioxide and sodium metabisulfite may be justified if good odor is important. Silages preserved with either of them were always of good odor.

King (26, p.10) found that the loss of nutrients in corn meal silage was approximately 10 per cent in excess of normal silage losses.

King (25, p.17) states, "The total digestible nutrients and metabolizable energy of ground barley grass silage were higher than those of molasses grass silage, but not so high as might be expected. The lower nutrient content of ground barley grass silage was evident in the feeding trial, in chemical composition of the silage, in the coefficient of digestibility of the dry matter and nitrogen-free extract of the barley silage, in the total digestible nutrients, and in the metabolizable energy values. The loss amounted to about 30 per cent of the nutrient of the ground barley or 10 per cent of the nutrient of the entire silage."

5. Bacteriology of silage.

The main function of microorganisms in silage making takes place after the cells are dead. However, microbiological activity begins very soon after filling the silo, with the organisms using the exuded sap (54, p.136).

(1) Source of silage bacteria.

Workers using all types of forage material have observed that silage undergoes a typical lactic acid fermentation in all cases, regardless of treatment, although some forages may change the type of fermentation after the lactic acid producing population is no longer dominant (50, p.44; 3, p.280; 5, p.784; 19, p.588).

Thomas (53, p.4), upon washing uninjured leaves of alfalfa, clover, or timothy, found that the bacteria on the fresh forage were distinctly different from those found in silage. The same media, Trypsin digest or tomato juice agar, when inoculated with washings from uninjured plants, seldom showed a typical silage organism on the surface of the plates. He attributed this change in population to constituents in the plant juice which change the existing organisms to secondary forms that bear little or no resemblance to the original species. Instead of bacteria adapting and changing slowly through a long series of transfers to growth in a different medium, the action is immediate (53, pp.17-18).

Stirling (49, p.154) found that the number of silage bacteria increased more rapidly when the grass put into the test tubes was finely minced instead of uncut. It is interesting to note that the curves of colony counts show an immediate increase without a decline in numbers as was

shown for uncut grass, at the twenty-four hour sampling period. The increased colony count was also reflected in the rapid build-up of acidity in the minced grass samples.

Fresh plant juice is not a prerequisite of silage fermentation, as dry forage will undergo a normal fermentation when water is added (20, p.82).

(2) Types of silage microorganisms.

The bacterial population of silage is heterogenous and may vary in composition with each silage made, or even from sample to sample (46, p.901). Any successful silage will show a restricted flora. The diverse flora found in green forage has almost entirely disappeared as the acidity builds up, and the true anaerobic flora remains practically unchanged or disappears entirely (54, p.136; 42, p.8; 3, p.293).

The important populations of microorganisms in the preservation of silages are the yeasts and molds, which are anaerobic; the lactic acid producers and proteolytics which are facultative anaerobes; and the butyric acid formers, also proteolytic, which are obligate anaerobes.

The yeasts and molds cease to be of concern in silage making as soon as the air is excluded, and as long as sufficient silage is removed each day during feeding (42, p.8).

The lactic acid producers form the most important population in silage. In successful silage they produce the acid and are the dominant type of bacteria. They increase rapidly within the first twenty-four hours, and remain at a high level throughout fermentation (42, p.30; 3, p.280).

Watson (45, p.139) says, "All investigators are agreed on the fact that the lactic acid organisms are the most important in making good silage by the ordinary process, and indeed that their rapid development is an essential feature of successful silage practice."

Allen et al. (3, p.281) found that the lactobacilli in silage were types that produced lactic acid as their chief product, producing only small amounts of volatile acid, almost entirely acetic.

The undesirable fermentation within the silo is due to the obligate anaerobes and is usually noted by the formation of butyric acid, which, when excessive, is accompanied by putrefactive changes (54, p.140).

Allen et al. (3, p.281) found that anaerobic spore formers occurred after the eighth day, and continued increasing as long as the degree of anaerobiosis was favorable and the pH was not inhibiting. The predominant species was Clostridium sporangenes. In other studiesthey found that this was the only type of obligate anaerobe found at all

stages of fermentation.

For practical purposes the primary interest in silage bacteria is in the effect of populations and the results of that effect. Even though each silage may have a population varying in composition, the results are the same in a large number of cases (46, p.901).

(3) Action of bacterial populations in silages.

All silage undergoes a typical initial fermentation, regardless of the type of forage.

Hunter (19, p.558) says that alfalfa siloed alone underwent a typical fermentation caused by microorganisms practically identical with those in normal corn silage.

Stone, Murdock and Bechdel (50, p.41), who also used alfalfa, say that silages underwent a typical acid fermentation in all cases regardless of treatment, and lactobacilli of the heterofermentative type were isolated at all stages of the fermentation.

These same authors (50, pp.41-42) draw a clear picture of the interaction of bacterial populations in silage, and the need for adding readily fermentable carbohydrates. "A control silage with no special treatment developed about 1.5 per cent lactic acid in a few days. But as the reducing sugar was depleted, increasing amounts of acetic acid formed at the expense of lactic acid with an accompanying rise in pH and degredation of quality."

The degredation of quality at a high pH is usually caused by the obligate anaerobes, producing butyric acid and attacking the proteins. These are not as tolerant of acid as are the lactics and fail to multiply, or disappear as the acid is built up by an increasing lactic population.

The literature shows that anaerobic bacteria tend to multiply in later stages of silage fermentation, and that some of the butyric acid producing types attack lactate, but they cannot multiply at pH levels below 4.0 to 4.2 (45, p.161).

METHODS AND MATERIALS

1. Silos.

Six experimental silos and two wood stave silos of conventional size were used. The six experimental silos were made from 55-gallon steel drums. Each drum was inverted, the bottom cut out and replaced with a wooden follower which served as a cover. The original bungs served as drainage points for excess juice which was to be conducted to five-gallon tins by means of plastic tubing. Sampling was provided for by a one and one quarter inch nipple welded in the side of each drum. After being filled, the drums were placed under cover and in a rack that held pressure on their contents. This pressure was applied by a hydraulic jack, and held, between applications, by blocks. These drum silos were designated D1, D2, D3, D4, D5, and D6.

The wood stave silos were of conventional construction, thirty-two feet high and twelve and one-half feet in diameter. Each was built on a concrete base which had a center drain to separate concrete tanks located below and beside the silos. Concrete gutters on each silo designed to catch any drainage through the walls, were also connected to these tanks. The wooden silos were designated W1 and W2, respectively.

2. Forage.

The forage used in all silos came from the same field, and consisted of a mixture of first cutting Ladino clover and grasses from irrigated pastures.

3. Additives.

The additives used were dried molasses beet pulp and sodium metabisulfite.

4. Bacteriological and chemical methods.

(1) Sampling.

Samples were taken by means of a soil auger from W1 and W2, and by means of a long forceps from the drum silos. CO_2 was introduced into the sampling port after each sample was taken, to offset the effect of any introduced air. Each sample was placed into a sterile flask for transportation to the laboratory.

(2) Masceration.

The Waring Blender was used to prepare the samples for dilution.

(3) Acidity determinations.

The Beckman pH meter with glass electrodes was used for pH determinations. One-tenth normal sodium hydroxide was used in titrations against the Beckman pH meter in determining total acidity.

(4) Media used.

Difco Trypsin Digest Agar, altered by the addition of 0.10 grams of yeast extract, was used for determining the total count.

The proteolytic count was carried out using Frazier's Nutrient Gelatin Agar (13, p.42). This medium was prepared as follows:

A.	Distilled H_2O	100	cc.
	NaCl	5	g.
	KH_2PO_4	0.5	g.
	K_2HPO_4	1.5	g.
B.	Distilled H_2O	400	cc.
	Gelatin	4.0	g.
	Dextrose	0.05	g.
	Peptone	0.1	g.
	Veal Infusion	5	cc.

Pour A and B together, heat in flowing steam, and mix with 500 cc. of 3 per cent Agar (washed) solution. Adjust to a pH of 7.0 (13, p.42).

The use of this medium was suggested by Dr. C.M.Gilmour of the Oregon State College Bacteriological Department.

Corn liver mash was used to determine the presence of butyric acid-formers. The medium contained 1.5 per cent liver extract and 5 per cent corn meal. The liver chunks and corn meal were steamed in 100 ml. of distilled water for one hour, after which the medium was tubed in 10 ml. amounts, and autoclaved for two hours.

(5) Chemical study.

Samples were placed in plastic bags and transported to the Agricultural Chemistry Department for chemical analyses.

PROCEDURES

1. Filling the silos.

The forage for all silos was cut from the same field by means of a field chopper, and blown into W1 and W2. In W1, 8 pounds of sodium metabisulfite per ton was applied at the blower by means of a ten gallon milk can equipped with an adjustable slide in the bottom. This apparatus was mounted on the blower, and the vibration of the blower kept a steady stream flowing. Two hundred pounds of dried molasses beet pulp per ton was added to W2 by spreading on top of the forage before unloading it into the blower.

The preservatives used in the drum silos were: no preservative in D1; 50 pounds of dried molasses beet pulp per ton in D2; 100 pounds of dried molasses beet pulp per ton in D3; 150 pounds of dried molasses beet pulp per ton in D4; 200 pounds of dried molasses beet pulp per ton in D5; and 8 pounds of sodium metabisulfite per ton in D6.

The drums were filled with forage taken from the wagon, mixed with the appropriate amount of preservative, and then tamped into the drums. The only exception was D6 in which hand mixing would have been highly inaccurate because of the small amount of preservative to be used. Therefore, it was decided to use forage forked down from

the large silo, W1, that already contained sodium meta-bisulfite.

2. Sampling.

Bacteriological counts were made at 0, 12 and 24 hours, 2, 3, 5, 7, 10, 20, and 37 days, and 7 months on samples taken in sterile flasks. Each sample was weighed, to determine the amount removed, and processed as soon as possible. After drawing each sample, CO_2 was introduced into the sample hole and the cap replaced. Pressure was then applied to the drums to compress the forage. This procedure, up until the 10th day, moved fresh material in front of the sampling port. After the seventh day there was very little decrease in the volume of the contents of the drums. By that time the volume had decreased by approximately one-third.

3. Dilution.

A 10 gram silage sample was placed in a Waring Blender along with 90 ml. of sterile water. After two minutes, the liquid portion was placed in a sterile container and used for preparing further dilutions, and for pH and titratable acidity determinations.

4. Plating.

Dilution blanks were prepared and used to inoculate

the three media: Trypsin Digest, Nutrient Gelatin, and corn liver mash.

5. Incubation.

All plates and tubes were incubated at 37°C. The 0 hour and 12 hour plates were incubated for five days. A four day incubation period was used on subsequent platings, because of the presence of spreaders.

6. Counting.

After the number of colonies were counted to obtain the proteolytic count, the plates containing Frazier's Nutrient Gelatin were treated as follows: one plate was flooded with a 1 per cent tannic acid solution and the other with acid mercuric chloride solution ($HgCl_2$ 15g, and HCl (con.) 20 cc, add to 100 cc H_2O). If the gelatin has been changed, a clear zone appears about the colony to which $HgCl_2$ has been added. This required fifteen to thirty minutes. The tannic acid plate will show the amount of decomposition of the gelatin (13, p.42).

7. pH and titratable acidity.

pH determinations were made on the 1:10 dilutions prepared for bacteriological studies, using a Beckman pH meter with glass electrodes.

Titratable acidity determinations were made with 10 ml. of the 1:10 dilution, plus 10 ml. of distilled water.

Titrations were made with .10193 normal sodium hydroxide to a pH of 8.5, using the Beckman meter to determine the end point. The meter was used because the green silage juice masked the colors of indicators.

8. Chemical analyses.

All chemical analyses were performed by the Department of Agricultural Chemistry, using official O.A.O.C. methods.

Dry matter - - - - - - - - (30, p.342, paragraph 22.3)
Total nitrogen - - - - - (30, p.13, paragraph 2.23)
Ash - - - - - - - - - - - (30, p.343, paragraph 22.9)
Crude Fiber - - - - - - (30, p.347, paragraph 22.31)
Crude Fat - - - - - - - (30, p.346, paragraph 22.25)

Losses were determined on the total content of the drum silos, neglecting the top spoilage. It was assumed that the amount and percentage of spoilage is related to the size, construction, and filling procedure at the silo.

RESULTS AND DISCUSSION

1. Fermentation losses.

Analyses were run on the forages, with the experimental silo D1, containing forage without a preservative, serving as a control. At the time of filling, analyses were run on samples of the beet pulp used as a preservative, on the forage, and on the forage mixed with the preservatives. Upon opening, representative samples were taken, after removal of spoilage, from the mixed contents of each individual experimental silo. The results of these experimental silage analyses are shown on an as-fed basis in Table I.

No attempt was made to evaluate the changes in the seventy-five ton silos W1 and W2, because it was impossible to measure the amounts put in and taken out of these silos. A comparison of the analyses of silages from the large silos and their experimental duplicates is shown in Table II. The analyses of silages from W1 and W2 are based on averages of composite samples taken during various digestion trials. Due to an extremely wet season these silages were, at times, rained on before feeding. This is reflected in the low dry matter figures for W1 and W2.

Table I

Approximate Analyses of Experimental Silages
on an As-Fed Basis

Dried Molasses	Water %	Dry Matter %	Ash %	Crude Protein %	Crude Fat %	Crude Fiber %	N.F.E. %
			Forage and Preservatives				
Beet Pulp	13.18	86.82	4.42	9.18	0.40	13.67	59.16
D1	73.05	26.95	1.75	1.90	0.50	8.00	14.80
D2	69.75	30.25	1.73	2.34	0.74	8.45	17.09
D3	67.91	32.09	1.81	2.58	0.64	8.33	17.13
D4	67.91	32.09	1.78	2.55	0.59	8.41	18.76
D5	66.85	33.15	1.90	2.80	0.63	8.67	19.16
D6	71.15	28.85	1.98	2.11	0.66	8.03	16.07
			Silages				
D1	74.67	25.33	1.89	2.02	0.67	7.98	12.77
D2	71.78	28.22	1.98	2.45	0.90	8.39	14.49
D3	70.40	29.60	1.84	2.52	0.83	8.52	15.90
D4	69.79	30.21	1.83	2.69	0.76	8.57	16.36
D5	67.85	32.15	2.07	2.94	0.72	8.88	17.52
D6	75.36	24.64	2.11	2.03	0.72	8.21	11.57

Table II

Comparative Analyses of Silages from Large Silos and Experimental Duplicates on an As-Fed Basis

Silo	Water	Dry Matter	Ash	Crude Protein	Crude Fat	Crude Fiber	N.F.E.
W1	79.8	20.2	1.45	2.38	0.81	6.49	9.17
D6	75.3	24.64	2.11	2.03	0.72	8.21	11.57
W2	77.2	22.8	1.51	2.24	0.61	6.90	11.54
D5	67.8	32.1	2.07	2.94	0.72	8.88	17.52

Table III shows the relative losses, in pounds, of the different constituents, and also the percentage of loss. These losses were taken on the total contents of the silos, including top spoilage.

(1) Total weight.

The loss of total weight ranged from a low of 3.3 per cent to a high of 6.1 per cent of the total weight. On opening, D1, containing no preservative, was noticeably more moist than any of the other silages, particularly near the bottom. D6, preserved with sodium metabisulfite, although not as moist as D1, was damper than the silos preserved with dried molasses beet pulp. As far as could be determined by inspection, there was no noticeable difference in moisture content of the silages containing beet pulp.

During the storage period there was no juice drainage from any of the six experimental silos. Presumably

all of the non-volatile products and by-products of fermentation were still in the silo at the time of opening.

The loss of weight was least in D2, which contained dried molasses beet pulp at the rate of 50 pounds per ton. The losses became greater as the amount of beet pulp was increased. All beet pulp preserved silages lost less than the silage without any preservative. Sodium meta-bisulfite silage lost slightly more total weight. The computation of nutrients in Table II indicates that the loss of total weight had no bearing upon the actual loss of dry matter. Others have also found a lack of correlation between total weight changes and loss of nutrients (54, p.318).

(2) Dry matter.

Loss of dry matter ranged from 7.9 per cent for silage preserved with 200 pounds dried molasses beet pulp per ton, to 19.8 per cent in that preserved with approximately 8 pounds of sodium meta-bisulfite per ton.

The silage showed a steady decline in loss of dry matter, from silage with no preservative to that with 200 pounds beet pulp per ton, decreasing as the amount of preservative was increased. D3 with 100 pounds of beet pulp per ton was the only exception. It has a dry matter loss equal to that of the silage without any preservative.

Table III

Nutrient Losses in Experimental Silages
(Gains indicated by Positive Sign)

		Total Weight	Dry Matter	Ash	Crude Protein	Crude Fat	Crude Fiber	N.F.E.
D1 No Preservative								
	in	234	63.1	4.1	4.4	1.1	18.7	34.6
Pounds	out	220	55.7	4.2	4.4	1.5	17.6	28.1
	Difference	14	7.4	+0.1	0	+0.4	3.1	6.5
	% Loss	5.98	11.7	+2.4	0	+26.4	16.5	18.8
D2 50# beet pulp/ton								
	in	242	73.2	4.2	5.7	1.8	20.5	41.4
Pounds	out	234	66.0	4.6	5.7	2.1	19.6	33.9
	Difference	8	7.2	+0.4	0	+0.3	0.9	7.5
	% Loss	3.3	9.8	+9.5	0	+14.2	4.4	18.1
D3 100# beet pulp/ton								
	in	224	71.9	4.1	5.8	1.4	18.7	38.4
Pounds	out	214.5	63.5	3.9	5.4	1.8	18.3	34.1
	Difference	9.5	8.4	0.3	0.4	+0.4	0.4	4.3
	% Loss	4.2	11.7	7.3	0	+22.2	2.1	9.95

Table III (Continued)

		Total Weight	Dry Matter	Ash	Crude Protein	Crude Fat	Crude Fiber	N.F.E.
D4	150# beet pulp/ton							
	in	230	73.8	4.1	5.9	1.4	19.3	43.2
Pounds	out	220.5	66.6	4.0	5.9	1.7	18.9	36.1
	Difference	9.5	7.2	0.1	0	+0.3	0.4	7.1
	% Loss	4.1	9.7	2.4	0	+17.6	2.1	16.4
D5	200# beet pulp/ton							
	in	214	70.9	4.1	6.0	1.3	18.6	41.0
Pounds	out	203	65.3	4.2	6.0	1.5	18.0	35.6
	Difference	11	5.6	+0.1	0	+0.2	0.6	5.4
	% Loss	5.1	7.9	+2.4	0	+13.3	3.2	13.2
D6	8# sodium meta-bisulfite							
	in	262	75.6	5.2	5.5	1.7	21.0	42.1
Pounds	out	246	60.6	5.2	5.0	1.7	20.2	28.5
	Difference	16	15.0	0	0.5	0	0.8	13.6
	% Loss	6.1	19.8	0	9.1	0	3.8	32.3

However, the loss in these five silos was not excessively heavy. The high was a loss of 11.7 per cent and the low a loss of 7.9 per cent. All fall within the expected limits of good silage. Thus, it may be assumed that no preservative was necessary for successful preservation of this forage under the conditions encountered during preservation.

To determine the loss of dry matter that may be charged to dried molasses beet pulp, the expected dry matter loss of the grass in each silo was assumed to be equal to the 11.7 per cent loss sustained by the forage without any preservative. Using the data in Table III, the expected loss was subtracted from the actual loss and the difference was taken as the loss of beet pulp dry matter. The average loss above the expected loss for all four silages was about zero, and the highest loss 11.4 per cent for silo D3. Such a loss is no higher than that normally assumed to be unavoidable during a successful silage fermentation.

While the loss chargeable to beet pulp did not show a progressive pattern coinciding with amounts added, it may be assumed that no excessive loss of beet pulp dry matter should be expected when it is used as a silage preservative. The gains shown by some of the silages can probably be attributed to the beet pulp absorbing juices

fairly high in dry matter. Dried molasses beet pulp has been shown to absorb approximately two pounds of juice, containing some 10 per cent dry matter per pound (8, p.2). However, because of the small number of silos, lack of duplicates, and absence of drainage, this can remain only a hypothesis until further studies have been made.

(3) Ash.

Changes in the amount of ash varied from a 9.8 per cent gain for silo D2, to a 7.3 per cent loss for silo D3.

Since there was no drainage from any experimental silo, a loss of ash is difficult to explain, other than through the sampling and computing of such small amounts. This same variation in ash content has been reported by many workers.

(4) Protein.

The only silage to show a loss of protein was that from D6 which had a 9.1 per cent loss. This is well within the loss of protein usually expected. Watson (54, p.371) gives the expected loss of protein for silage with preservatives as about 10 per cent, and those without preservatives as about 30 per cent.

There was no evidence of breakdown to satisfy fermentation demands, unless the 9.1 per cent loss found in the silo D6 can be considered as used in lieu of carbohydrates. It is more probable that the higher loss in D6 is the

result of the delay in acid development in that silo. This work does not have enough evidence to support any definite conclusion.

(5) Crude fat.

All silos except D6 showed an increase in crude fat, ranging from a 13.3 per cent gain for D5 to a 26.4 per cent gain for D1. Silo D6 showed no loss of crude fat.

The production of crude fat as a by-product of fermentation is to be expected. It is not surprising that very little loss or even a gain in this constituent would be apparent. Reports of increases in crude fat are common in the literature.

(6) Crude fiber.

The loss of crude fiber was slight in all silages except for W1, which had a 16.5 per cent loss.

(7) Nitrogen free extract.

Nitrogen free extract was calculated by difference, and varied from a loss of 13.2 per cent in silo D5, preserved with 200 pounds dried molasses beet pulp per ton, to 32.3 per cent in D6, which utilized sodium metabisulfite as the preservative. There was a general decline of nitrogen free extract losses which followed the increased amounts of dried molasses beet pulp. The exception was D3 which showed only a 9.95 per cent loss.

D6 also showed the highest loss of protein as well as the highest loss of nitrogen free extract. This could be interpreted as evidence that the utilization of available carbohydrates in this silo was not as efficient as in the other silos. It is possible that the continued high population of proteolytics used up some of the nutrients that a more rapid developing population of lactics could have converted into acid more efficiently. The relative development of these two bacterial populations is shown in Figure 1.

2. Changes in bacterial flora.

In order to extend the evaluation of fermentation losses, studies on the important microorganisms of silage were planned. The lactic acid, proteolytics and the butyric acid bacteria were considered, and media known to encourage their growth were selected.

(1) Total count.

The total count on Trypsin Digest medium in all silages showed a general build up of numbers for the first five days, after which there was a gradual decline that continued until the silos were opened.

Throughout the study the same general type colonies appeared on the plates. The predominant colonies were pinpoint in size, and about equally divided between a smooth, translucent colony, and an irregular brownish

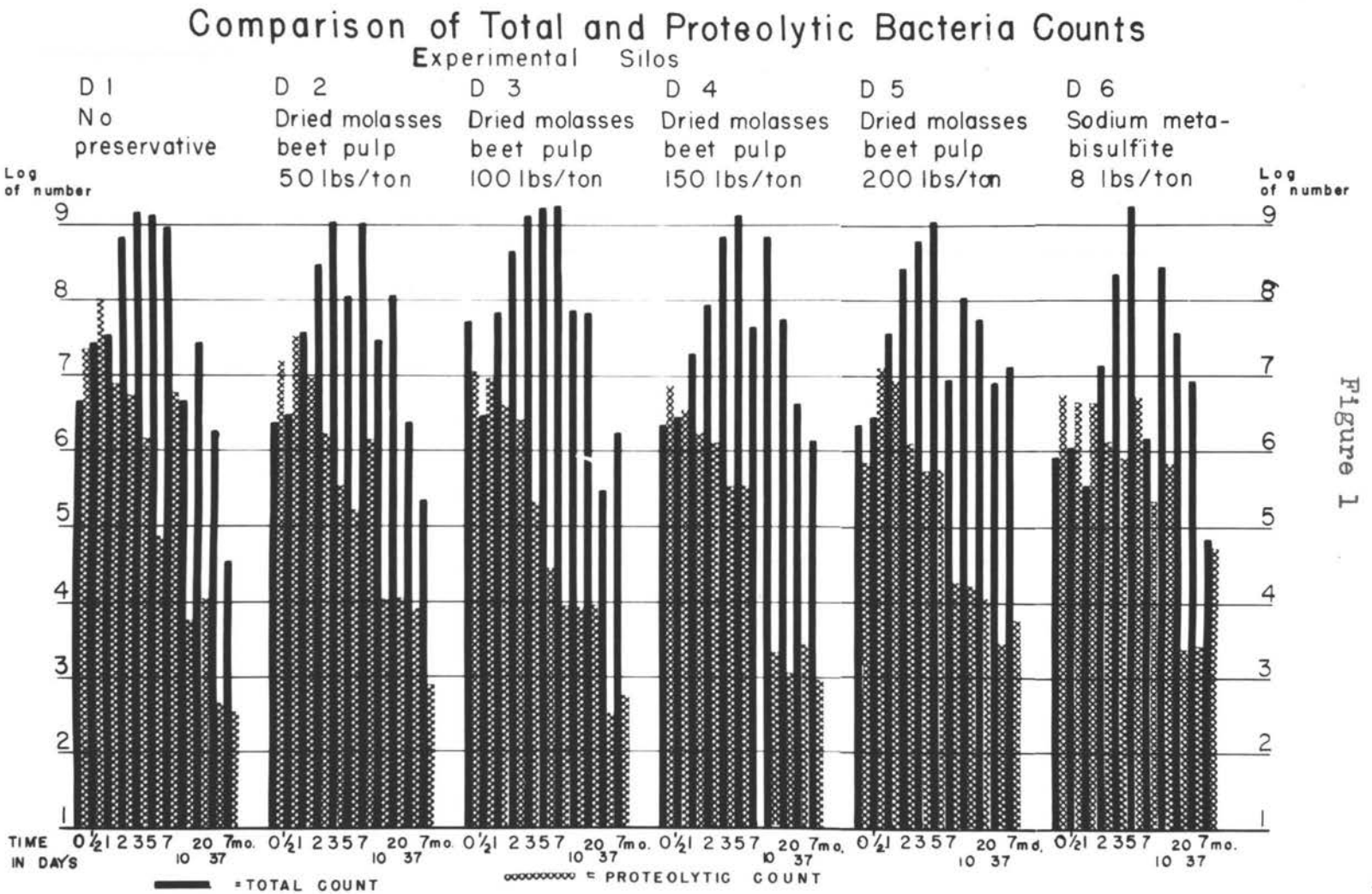
Comparison of Total and Proteolytic Bacteria Counts
Experimental Silos
D 1
No preservative
D 2
Dried molasses beet pulp
50 lbs/ton
D 3
Dried molasses beet pulp
100 lbs/ton
D 4
Dried molasses beet pulp
150 lbs/ton
D 5
Dried molasses beet pulp
200 lbs/ton
D 6
Sodium meta-bisulfite
8 lbs/ton
Log of number
9
8
7
6
5
4
3
2
1
TIME IN DAYS
0 ½ 1 2 3 5 7 10 20 37 7mo.
= TOTAL COUNT
= PROTEOLYTIC COUNT

Figure 1

colony. During the first platings, a few colonies showed pigmentation, but most pigmented colonies disappeared after the second day.

In all of the silos, excepting the large silo (W2) containing dried molasses beet pulp as a preservative, the total count remained above that of the zero hour plating, for the first twenty days. Usually the number of viable colonies did not drop below the initial number until some time after the thirty-seventh day. These results are presented in Figures 1 and 2, which show the viable count on Trypsin Digest Agar as a solid bar.

(2) Proteolytic count.

A true proteolytic count is very difficult to secure. However, Frazier's Nutrient Gelatin can be expected to give a fairly accurate picture of the proteolytic population, except for the obligate anaerobes. The cross-hatched bars on Figures 1 and 2 show the changes in viable proteolytic numbers.

When making studies using mercuric chloride and tannic acid, it was found that approximately 90 per cent of the colonies developing on Frazier's medium were from strongly to weakly proteolytic. Therefore, a correction factor of 10 per cent of the total count is subtracted to give the total proteolytic count.

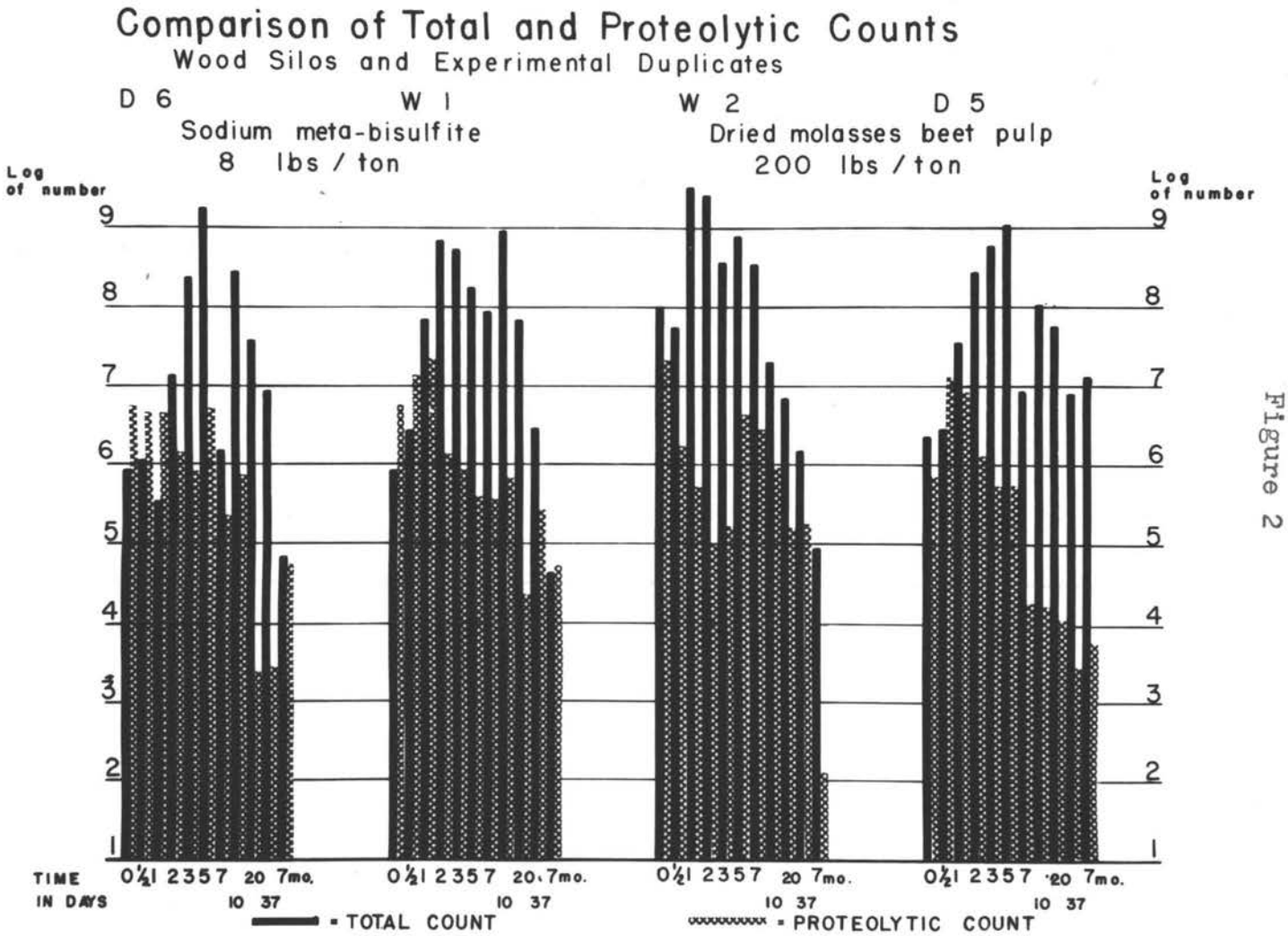
Comparison of Total and Proteolytic Counts
Wood Silos and Experimental Duplicates
D 6
W 1
Sodium meta-bisulfite
8 lbs / ton
W 2
D 5
Dried molasses beet pulp
200 lbs / ton
Log of number
Log of number
9
8
7
6
5
4
3
2
1
TIME IN DAYS
0½1 2357 20 7mo.
10 37
= TOTAL COUNT
= PROTEOLYTIC COUNT

Figure 2

The zero hour platings on Frazier's Nutrient Gelatin showed approximately the same total count as that found on the Trypsin Digest plates. This might be expected, as the lactics in this period are not a dominant population.

As may be seen in Figure 1, subsequent platings after the twenty-four hour sample showed a general trend toward a reduction in numbers until about the tenth day when the rate of decline leveled off.

Throughout the study, Streptomyces occasionally appeared on plates from all silages. The occurrence of these colonies tended to increase in later platings. At the twenty-day period, all silages showed Streptomyces in varying numbers ranging from a few to as high as 90 per cent of the viable colonies in silage from W2. All Streptomyces colonies were highly proteolytic as measured by mercuric chloride and tannic acid.

Small pinpoint colonies were usually in the majority on these plates, and exhibited varying degrees of proteolysis. It was often difficult to determine if a small colony was proteolytic, or if the gelatin had been changed by a neighboring colony. Many isolated pinpoint colonies changed the gelatin in an area many times greater than that occupied by the colony itself.

The general pattern of changes in bacteria numbers was the same for all silos. Figure 2 shows the changes in

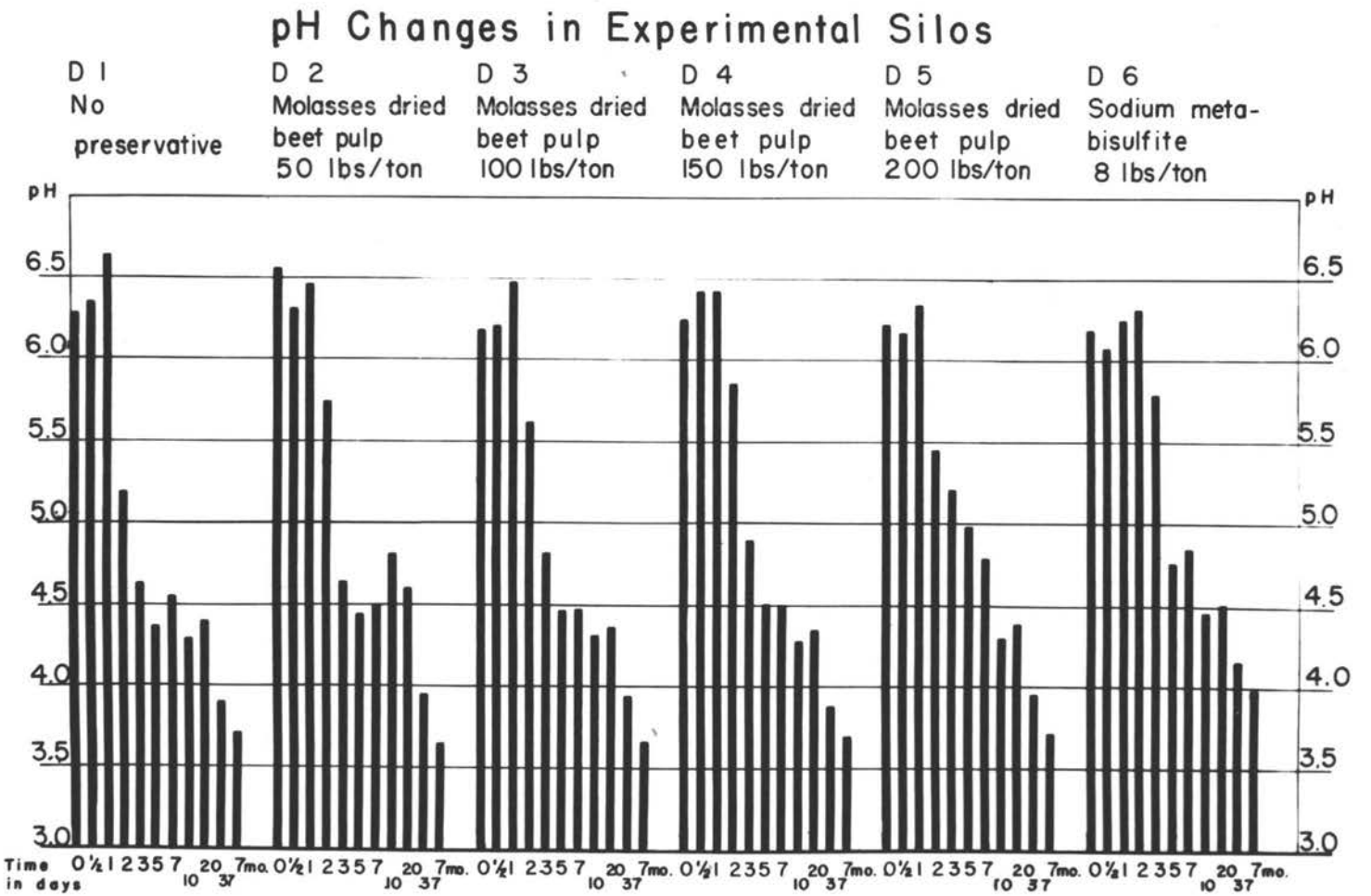
pH Changes in Experimental Silos
D 1
No
preservative
D 2
Molasses dried
beet pulp
50 lbs/ton
D 3
Molasses dried
beet pulp
100 lbs/ton
D 4
Molasses dried
beet pulp
150 lbs/ton
D 5
Molasses dried
beet pulp
200 lbs/ton
D 6
Sodium meta-
bisulfite
8 lbs/ton
pH
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
Time in days
0 ½ 1 2 3 5 7 10 20 37 7mo.

Figure 3

numbers for each large silo and its experimental duplicate. It is apparent that the preservative had more effect than the size of the silo.

(3) Butyric acid formers.

There was no growth in any of the tubes containing corn liver mash. After the first three days, all inoculations were made from the 1:10 dilution. There was no evidence of butyric acid in any silo so far as could be detected by olfactory means, or by gas production in bacteriological media.

3. Acidity determinations.

Titratable acidity and pH determinations were made at each sampling from the 1:10 dilution. The changes in pH of the six experimental silages are shown in Figure 3, and of regular silos W1 and W2 in Figure 5. All samples except silo W2 showed an increase in pH through the twenty-four hour period. This period of slight increase was followed by a rapid drop in pH that continued until the fifth day before leveling off. Figure 4 shows the approximate relationship of the changes in bacterial counts to the changes in pH in silo D1. These relative changes are typical of those in all the silos. The rapid increase in total count on Trypsin Digest Agar is followed by a corresponding decrease in pH, and in the proteolytic count.

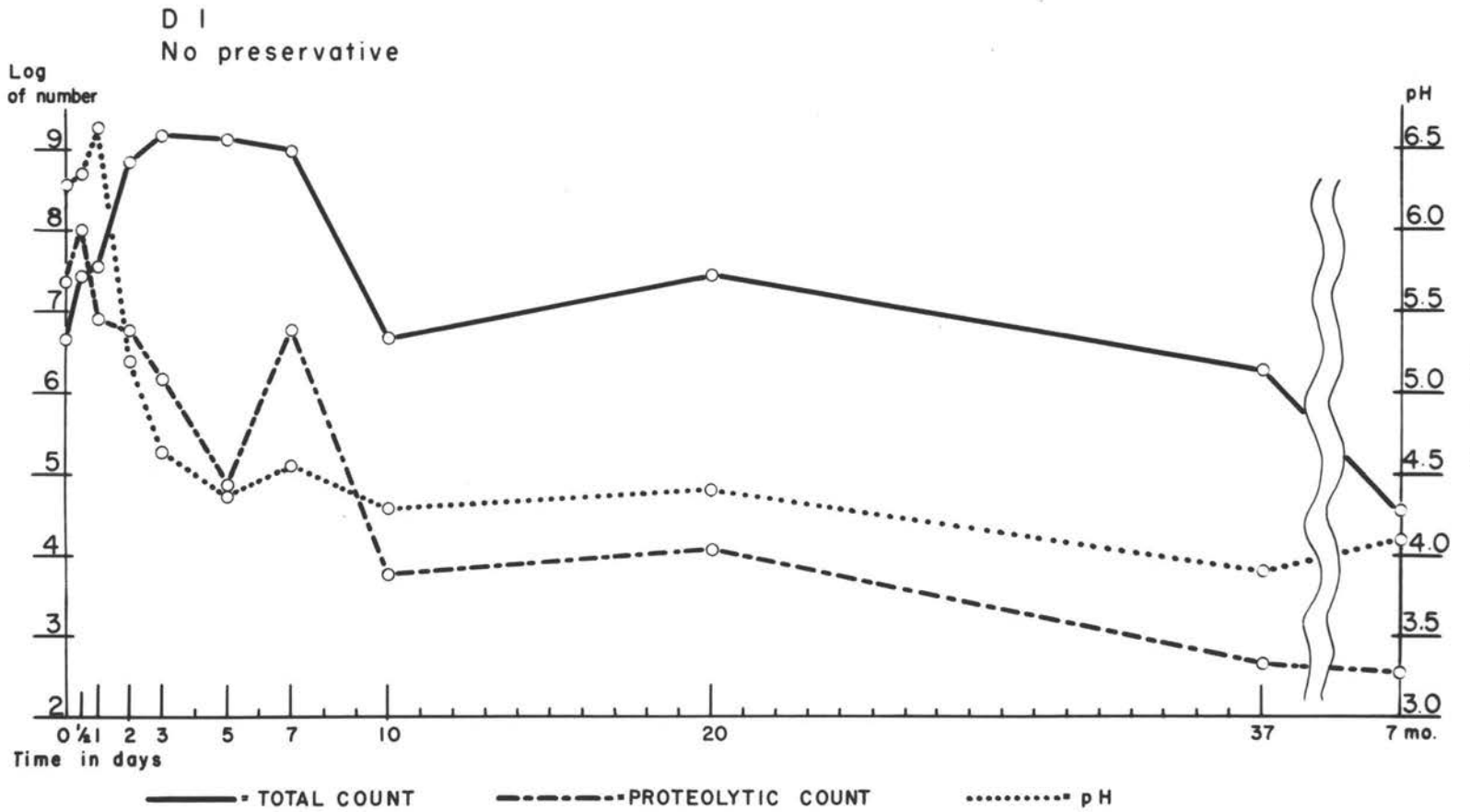
Relationship of Bacteria Population and pH
D I
No preservative
Log of number
9
8
7
6
5
4
3
2
pH
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
0
½
1
2
3
5
7
10
20
37
7 mo.
Time in days
TOTAL COUNT
PROTEOLYTIC COUNT
pH

Figure 4

Generally an increase of titratable acidity followed the drop in pH. However, there was no clear cut picture of a given amount of acid for a given pH. This was particularly true in later periods of storage. Table IV shows the two values at thirty-seven days and again in seven months.

Table IV

pH and Titratable Acidity

	Titratable acid in ml of 0.1 normal NaOH							
37 days	D1	D2	D3	D4	D5	D6	W1	W2
pH	4.10	4.05	3.98	4.00	4.25	4.30	4.40	3.95
Tit. acidity	2.90	2.90	3.30	2.70	2.80	1.70	1.20	2.80
7 months	D1	D2	D3	D4	D5	D6	W1	W2
pH	3.70	3.65	3.67	3.70	3.72	3.99	3.87	3.48
Tit. acidity	3.80	4.50	4.00	3.90	3.90	2.30	2.80	3.90

The relatively large difference in titratable acidity for small changes in pH is explained by a buffering action that appeared below a pH of about 4.40.

There seemed to be little difference in the pattern of pH and titratable acidity of the silages without any preservative and those with various amounts of beet pulp. However, there was a distinct difference in build up of acidity in experimental silages from silos D6 and large

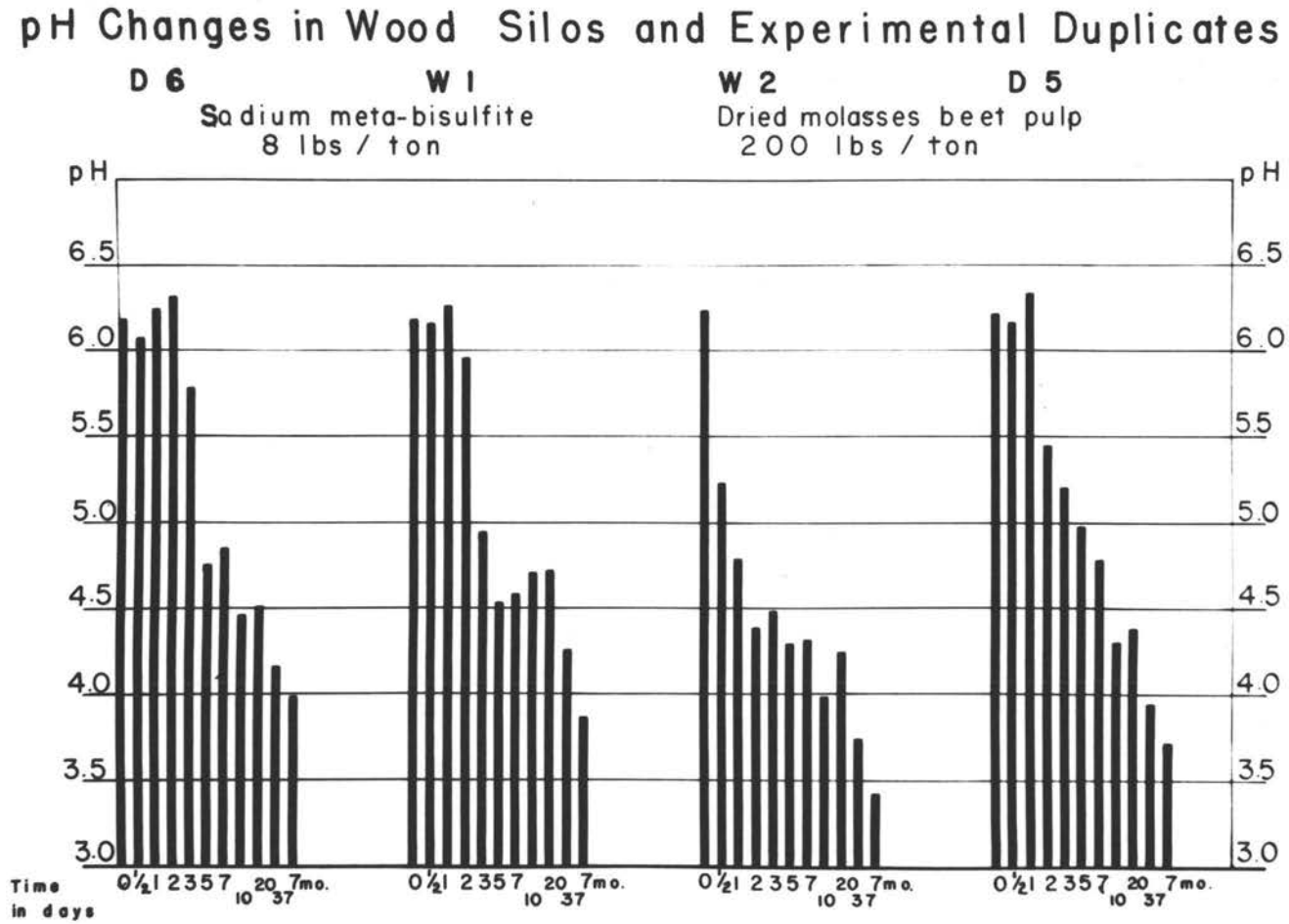
pH Changes in Wood Silos and Experimental Duplicates
D 6
W 1
Sodium meta-bisulfite
8 lbs / ton
W 2
D 5
Dried molasses beet pulp
200 lbs / ton
pH
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
Time
in days
0 ½ 1 2 3 5 7 10 20 37 7mo.

Figure 5

silo W1, preserved with approximately 8 pounds of sodium meta-bisulfite per ton. These silages were noticeably slower in the dropping of pH and proteolytic count, and in the production of acids. The relative values shown in Table IV are typical of the picture throughout the fermentation period.

The changes in pH of the large silos, W1 and W2, are shown in Figure 5, where they are charted with the experimental duplicates. The same delay in drop of pH can be seen for both silages preserved with sodium meta-bisulfite. The development of titratable acidity in these silages was about the same. The relative values for the 37 days and seven months are given in Table IV.

The changes in bacterial numbers and pH are about what would be expected as the result of successful silage fermentation. There was no apparent significant rise in pH during the later periods of storage such as were reported by Skaggs and Knodt (48, p.332) in studies on sulfur dioxide silage.

4. Quality.

(1) Color and odor.

The two silages preserved with sodium meta-bisulfite maintained a greener color throughout storage than did the other silages, and never developed a true silage odor.

Their odors could be described as rather bland, and not unpleasant. The retention of green color was particularly pronounced in silo W1, until the third day when the temperature of the silage increased to about 100°F. This increase in temperature lasted until the tenth day when it dropped back to about 80°F. The rise in the temperature may have been due to a mechanical breakdown that delayed completely filling the silo for about three days. D6 did not begin to lose its bright color until the tenth day.

Upon opening, D1 was wetter and had a darker green color near the bottom than the other experimental silages. The top had approximately the same yellow-brown color as did the large silo W1 and the other experimental silos containing beet pulp. D6 and W1, the silages containing sodium meta-bisulfite, were lighter, brighter, apparently more yellow than the others.

(2) Palatability.

All silages were of good quality, judging from appearance and odor. A set of identical twin cows was offered the silages free choice. All silages were consumed, but were relished in the following order: beet pulp preserved silage, sodium meta-bisulfite preserved silage, and silage with no preservative. The preference for beet pulp in the silage was emphasized by the order in which they

ate these silages. The higher the content of beet pulp, the more quickly the silage disappeared.

SUMMARY

Loss of total dry matter was less in silages preserved with dried molasses beet pulp than it was in silage without any preservative. The loss of total dry matter decreased as the amount of preservative increased.

Loss of total dry matter was greater in silages preserved with approximately 8 pounds of sodium meta-bisulfite than it was in the silages preserved with beet pulp or that made without any preservative.

In silages preserved with beet pulp the total dry matter losses varied from 11 per cent less to approximately 11.4 per cent more than the expected loss of dry matter in the grass portion of the silage.

The pH values were a good indication of the development of the lactic acid producing population and the decreasing proteolytic population.

There was no apparent development of butyric acid producing bacteria in any of the silages.

The development of acidity was slower in the silages preserved with approximately 8 pounds of sodium meta-bisulfite. The titratable acidity in these silages remained lower than that of the other silages.

The shift of the microorganisms from a dominantly proteolytic population to dominantly lactic producing was slower in the sodium meta-bisulfite preserved silage.

The silages preserved with sodium meta-bisulfite retained more of the original grass color than the other silages. Upon opening, they did not have a typical silage odor. However, their odor is more acceptable by human standards than a typical silage odor.

The silages preserved with beet pulp and the silage without any preservative developed a typical silage color and odor.

All silages were of good quality and acceptance to the cows with the silages preserved with dried molasses beet pulp being preferred.

CONCLUSIONS

The data indicate that dried molasses beet pulp may be used in varying amounts as a silage preservative without losing an excessive amount of its feed nutrients.

More studies are needed, under conditions that actually require the use of a preservative, to establish the overall efficiency of dried molasses beet pulp as a silage preservative.

Under the conditions encountered in this experiment, sodium meta-bisulfite appeared to delay the normal development of silage fermentation, but resulted in acceptable silage.

Frazier's Nutrient Gelatin Agar appeared to be an excellent medium for study of the proteolytic micro-organisms found in silage.

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