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THE QUALITY OF ALFALFA AND
ALFALFA-BROME GRASS SILAGE WHEN
MADE BY VARIOUS METHODS USING
GLASS JARS AS MINIATURE SILOS

Thesis for the Degree of M. S.

MICHIGAN STATE COLLEGE

Gérard Philippe-Auguste

1952

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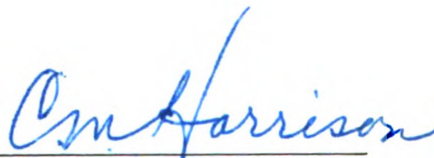
The Quality of Alfalfa and
Alfalfa-Brome Grass Silage When
Made by Various Methods Using
Glass Jars as Miniature Silos

presented by

Gérard Philippe-Auguste

has been accepted towards fulfillment
of the requirements for

M. S. degree in Farm Crops

A handwritten signature in blue ink, reading "C. M. Harrison". The signature is written in a cursive style with a large, stylized "C" and "H".

Major professor

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THE QUALITY OF ALFALFA AND ALFALFA-BROME GRASS
SILAGE WHEN MADE BY VARIOUS METHODS USING
GLASS JARS AS MINIATURE SILOS

By

Gérard Philippe-Auguste

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INTRODUCTION

For many years, farmers and research workers have tried to control completely the important fermentation that occurs following the ensiling of green plants, in order to preserve a feed high in total digestible nutrients, carbohydrates, vitamins, and with a good flavor. At the present time, putrefaction, formation of amines, butyric acid fermentations, growth of molds, and severe loss of dry matter and digestible protein frequently occur. Certain methods and principles have been worked out which aid in successful silage-making, but the effects of certain practices and techniques are still somewhat obscure, which makes the ensiling process occasionally unsuccessful in the hands of farmers. Conditions of soil and climate affect the composition of the plants and, no doubt, their microflora. It seems likely that a complete list of the limiting factors will in time allow a reliable system of silage-making to be described. The present experiment was set up in an attempt to study some of the effects of different methods and chemicals on the quality of silage, using glass jars as miniature silos.

REVIEW OF LITERATURE

Many researches have been conducted in recent years on the different methods of making forage-crop silage. Watson (27, 28, 29) proved that the optimum pH for silage preservation was between 3.5 and 4.0. At that pH level, the undesirable products were not formed, and the decomposition of proteins was prevented. To reach that pH level, research workers in various countries have suggested procedures along the following general lines (5, 6, 16, 17, 14, 18, 19, 22):

1. Addition of various acids to the green material as it is being ensiled;
2. Addition of fermentable carbohydrates, either sugars (as in molasses) or starchy grains;
3. Inoculation of the fodder with a culture of lactic acid-forming bacteria;
4. Partial wilting of the forage to favor greater inclusion of air, higher temperature, and reduction of the activity of undesirable bacteria;
5. The use of carbon dioxide; and
6. Sterilization of the green forages.

The idea of preserving silage by addition of acids was first advanced by Giglioli, in Italy (26). He added hydrochloric acid to beet leaves at the time of ensiling. In the resulting fermentation, there was developed an acidity calculated at 2.2 per cent of lactic acid. There was a loss of 11.8 per cent of the digestible nutrients. The A. I. V. method outlined by Virtanen (26), used mineral acids in ensiling green forage. Here a mixture of equal parts of sulfuric and hydrochloric acids in a 2-normal solution is added to the fodder as it is ensiled. The amounts added to various forages are intended to raise the acidity of the forage to a pH of 3.6 to 4.0. Bender, Bosshardt, and others (2, 5, 6, 13, 30) recommended the use of a mixture of hydrochloric and phosphoric acids, or of phosphorus pentachloride that yields these acids on hydrolysis.

In 1917, Reed and Fitch, at the Kansas Experiment Station (20), were successful in making palatable alfalfa silage by means of adding fermentable carbohydrates in the form of molasses. Their experiments showed that when cowpeas were ensiled alone, a poor quality of silage was obtained, but that a first-class silage resulted when a mixture of cowpeas and corn were ensiled. This finding suggested the role of additional fermentable carbohydrates in the formation of lactic acid. Since that time, many experiments

have been performed in which the beneficial effects of molasses or starchy grains have been demonstrated. Wilson, Webb, Shaw, Wright, Swanson, Tague, and A. King (37, 31, 25, 32, 33, 34, 35, 36, 8) reported that molasses is the best source of carbohydrate, being superior to starchy materials. Alfalfa silage prepared with molasses was found to have a higher acid content, together with a reduced breakdown of protein when compared with silage prepared with no added preservative. Bender (7) proposed a combination of phosphoric acid and molasses as a means of lowering the pH of silage.

Considerable attention has been given to the role of bacteria in the fermentation of silage. Particular emphasis is laid on the action of the lactic acid bacteria. Cultures of lactic acid bacteria soon appeared on the market, for addition at the time of silage-making. Watson, and Watson et al. (29, 28, 15, 16, 3, 27, 2) rejected the method for the following reasons:

1. Addition of cultures of lactic acid organisms has never been accompanied by tangible improvement, since the crop usually has a sufficiently large natural flora of these organisms.

2. Fermentation does not vary according to the type of bacteria present, but according to the forage and the conditions

under which it is stored. Therefore, an attempt to control the fermentation by mere addition of bacterial cultures can hardly be effective.

The field-wilting process is the most widely used method on farms in the United States (6, 8, 26). Woodward and Shepherd (38) stated that proper wilting prevents rotting of silage. Archibald and Parsons (4) reported in 1945 that many farm operators had obtained satisfactory silage when the crop was slightly wilted, with no preservative.

The use of carbon dioxide as a preservative involves the replacing of air in the filled silo with carbon dioxide. The method is tedious and uncertain, since it is difficult to know when all air has been replaced. The value of creating an anaerobic condition lies in the fact that the respiration of the plant cells and other aerobic activities, such as those of the molds, will be eliminated (2). Such a condition, however, is favorable for the growth of undesirable, as well as desirable, bacteria.

The use of antiseptics to stop respiration and undesirable fermentation was suggested as early as 1886 (2). Since then, attempts have been made to sterilize the fodder by heat produced by steaming the silage in the silo, and also through the use of electric

currents. Chemical sterilizing agents such as formaldehyde (12, 37, 23, 24) have been tried, but the results did not warrant their further use. According to many workers, it is not practicable to use such a method in silage-making (3, 28, 29).

Other Factors in Silage-Making

The weather may play an important part in silage-making. Dexter (11, 10, 12) found considerable difference in the sugar content of alfalfa plants at various times of day, and pointed out that the highest percentage of sugar was found in alfalfa plants during sunny days. Ahlgren (1) called attention to the fact that alfalfa usually contains about 4.3 per cent sugar, whereas field corn contains about 27 per cent. Santleman (21) investigated the effects of fertilizer, lime, and other soil treatments on the quality of silage produced. Gneist (9) noticed that crushing, or macerating, the forage resulted in smaller losses of nitrogen-free-extract and digestible protein during ensiling. Grazein and Heinzl (9) found that lactic acid fermentation was speeded up by crushing. According to deMan, a pH of 3.9 was obtained with crushed forage, compared with a pH of 5.4 with uncrushed. He suggested a plausible explanation: "It is generally known that the stems of grass have a higher

carbohydrate content than the leaves and a lower protein content; so it might well be that the distribution of the contents of the stems through the silage explains to a certain extent the effect of crushing."

MATERIALS AND METHODS

Experiment 1

A mixture of alfalfa and brome grass cut at early bloom (June 13) provided the material for silage. The green fodder used was divided into two parts. One part, 12 tons, was crushed with an ordinary hay crusher-mower and then chopped at approximately 1 inch with dull field chopper knives. The other half was chopped with sharp knives and not crushed. Each lot was put in an ordinary upright silo. The two silos were filled the same day; no preservative was added, and wilting was avoided. The material was approximately 20 per cent dry matter.

For the purpose of the laboratory experiment, the same material was used on the same day. From each lot, chopped and crushed-chopped, about 200 pounds were taken during filling for special treatments in the fruit jars.

The methods of preparation of the material were:

1. Chopped;
2. Crushed and chopped;
3. Ground.

The grinding was a thorough maceration of the tissues with a meat grinder. Six different treatments in triplicate were applied to each of these lots. These treatments were:

1. Wet untreated (20 per cent dry matter);
2. Partly wilted (24 per cent dry matter);
3. Partly wilted plus sugar (2 per cent sucrose);
4. Partly wilted plus lactic acid culture;
5. Partly wilted plus manganese sulfate (2 lbs. per ton);
6. Fully wilted (30 per cent dry matter).

The miniature silos used were quart glass jars with metallic covers. Six hundred thirty grams of crushed and ground silage and 550 grams of chopped silage were preserved in each jar. In the lots crushed and ground, the 630 grams were exposed to the air at room temperature until that weight was brought down to 525 grams for the "partially wilted," and down to 427 grams for "fully wilted." The 550 grams used for the lot "chopped" was reduced to 458 grams for partly wilted, and down to 367 grams for fully wilted. In all the cases, only the "partly wilted" material was treated chemically. Lactic acid culture was added at the rate of one 4-ounce bottle of Ericsson's lactic acid starter per ton of wet silage. One cubic centimeter of suspension was put on per quart jar and thoroughly mixed.

The manganese sulfate was applied in solution; 5 cubic centimeters per jar, equivalent to 2 pounds of dry manganese sulfate per ton.

Experiment 2

A second experiment was started 26 days later, on July 9. Alfalfa of prebloom stage was harvested with a tractor-mounted mower between ten and eleven o'clock in the morning. The material was preserved in one-quart glass jars, and the leaves and stems were chopped in the laboratory with a paper cutter. The alfalfa contained 28.9 per cent dry matter, and each sample was made of 500 grams of material. The experiment consisted of seven treatments, each replicated three times. The treatments were:

1. Ensiled at once;
2. Unwilted, warm, in the dark;
3. Unwilted, cold, in the dark;
4. 0.5 per cent of H_3PO_4 (commercial sirupy phosphoric acid);
5. 2.0 per cent sugar;
6. Wilted in the dark;
7. Wilted in sunlight.

The alfalfa which was unwilted and kept warm in the dark was placed unchopped in a closed box at room temperature for 9 hours. When reweighed at the end of the period, it showed a loss in weight of 20 grams that was corrected by addition of water. The material was then chopped into small pieces at once and the jars filled. The unwilted alfalfa which was kept cold in the dark differed from the first only by being kept in a refrigerator for the same length of time.

The alfalfa which was wilted in the dark was placed in a closed box, while the wilting in sunlight was accomplished by exposing the alfalfa to the sun long enough to bring the weight down to 420 grams.

Experiment 3

Alfalfa at the 1/4 bloom stage, harvested on July 29 at three different times of day, provided material for this experiment. The times of cutting were: 1:30 p.m., 5:00 p.m., and 5:00 a.m. the following morning. Two mechanical treatments--"chopped" and "ground"--for each cutting were made in triplicate. The green fodder was divided into two parts; one part was ground and the

other chopped and put up at once, and 500 grams of each were preserved in glass jars.

Measuring the Quality of the Silage

In all three experiments, the same measurements were made and the same code-system was used. Odor desirability was scored. (1) excellent, (2) good, (3) fair, (4) offensive, (5) poor, et cetera. The quality of the preserved silage was judged additionally by the pH level. For the determination of pH, 5 grams of silage were put in a 50 cubic centimeter beaker, where it was mixed with 25 cubic centimeters of distilled water. That mixture was stirred to facilitate the diffusion of the silage juice, and the pH was taken with a Beckman pH meter with extension glass electrodes. In addition, the buffer capacity between pH 3 to 11 was determined. A 5-gram sample was put in a Waring blender containing about 10 cubic centimeters of distilled water for 2 minutes. The sample was completely removed from the blender by washing the latter with the remaining 15 cubic centimeters of distilled water. The buffer capacity was studied by using a solution of sodium hydroxide and another of hydrochloric acid, both being of equal normality--0.2563. The natural pH of the sample

was taken first and recorded. Then, the pH was brought up to 11 by addition of the solution of sodium hydroxide, and brought down to pH 3 by adding the hydrochloric acid solution. The amount of hydrochloric acid required to reach pH 3 was the buffer capacity of that particular sample.

RESULTS AND DISCUSSION

Experiment 1

The results of Experiment 1 are presented in Table I. Analysis of variance of the data, presented in Table II, was made in order to determine whether or not the treatments and methods were significantly different. This analysis showed significant differences between the methods and between the treatments. In either case, these differences were highly significant at the 1 per cent level.

The "ground" alfalfa resulted in the best silage, followed by "crushed" and then "chopped."

It has been commonly observed that good silage should have a pH of 4.2 after a few days of fermentation. In Table I, it can be seen that the method "ground" untreated approaches this figure, while the "crushed" and the "chopped" untreated are at about pH 4.7. A clear-cut difference between these silages becomes more evident as time proceeds, since bacterial fermentation is still active. After seventeen, twenty-four, thirty-three, and fifty-two days, the value of the grinding method had become definitely established, since

TABLE I

pH OF SILAGES MADE UP OF YOUNG ALFALFA-BROME
GRASS AND TREATED DIFFERENT WAYS,
AFTER 4, 17, 24, 33, AND 52 DAYS

Days	Treatment					
	Wet Un- treated	Partly Wilted Un- treated	Partly Wilted + Sugar	Partly Wilted + Lac- tic Acid	Partly Wilted + MnSO ₄	Fully Wilted Un- treated
<u>Chopping Method--Replicate I</u>						
4	4.71	5.88	4.68	6.22	5.86	6.28
17	4.38	5.70	4.38	5.74	5.84	5.74
24	4.50	5.80	4.37	5.94	6.00	5.59
33	4.62	5.92	4.47	5.82	5.91	5.72
52	5.50	5.78	4.31	5.70	5.60	5.72
<u>Chopping Method--Replicate II</u>						
4						
17	5.21	5.68	4.24	5.74	6.20	6.42
24	5.45	5.78	4.31	5.75	6.15	6.36
33	5.70	5.89	4.34	5.56	5.89	5.87
52	5.60	5.51	4.29	5.51	5.64	5.89
<u>Chopping Method--Replicate III</u>						
4						
17	4.78	5.69	4.31	5.74	6.70	6.00
24	5.22	5.78	4.42	6.00	6.09	5.83
33	5.78	6.03	4.38	5.91	5.94	5.80
52	4.50	5.59	4.25	5.65	5.60	5.60

TABLE I (Continued)

Days	Treatment					
	Wet Un- treated	Partly Wilted Un- treated	Partly Wilted + Sugar	Partly Wilted + Lac- tic Acid	Partly Wilted + MnSO ₄	Fully Wilted Un- treated
<u>Chopping Method--Average Quality</u> <u>After Given Number of Days</u>						
4	1	4	2	3	2	2
17	1	3	1	4	3	3
24	2	3	1	4	5	5
33	2	3	1	4	5	3
52	3	3	1	5	4	3
<u>Crushing Method--Replicate 1</u>						
4	4.83	5.96	4.77	5.90	6.02	6.06
17	4.67	5.94	4.36	5.83	5.78	5.77
24	4.51	5.92	4.40	5.71	5.70	5.98
33	4.73	5.83	4.55	5.74	6.82	6.07
52	5.49	5.50	4.24	5.49	5.60	5.80
<u>Crushing Method--Replicate 2</u>						
4						
17	4.33	6.02	4.24	5.92	6.03	5.97
24	4.39	5.53	4.36	5.89	5.92	6.59
33	4.43	5.75	4.28	5.78	5.82	5.96
52	5.59	5.51	4.51	5.50	5.52	5.72

TABLE I (Continued)

Days	Treatment					
	Wet Un- treated	Partly Wilted Un- treated	Partly Wilted + Sugar	Partly Wilted + Lac- tic Acid	Partly Wilted + MnSO ₄	Fully Wilted Un- treated
<u>Crushing Method--Replicate III</u>						
4						
17	4.50	5.08	4.30	5.87	5.00	5.87
24	5.02	5.52	4.38	5.84	5.84	6.00
33	5.28	5.76	4.34	5.66	5.00	6.00
52	4.59	5.50	4.15	5.51	5.59	5.00
<u>Crushing Method--Average Quality</u> <u>After Given Number of Days</u>						
4	2	4	2	4	2	2
17	1	5	1	5	5	5
24	1	5	1	5	4	4
33	1	5	1	5	5	5
52	3	4	1	4	5	4
<u>Grinding Method--Replicate I</u>						
4	4.48	5.28	4.50	5.41	5.29	5.28
17	4.22	6.16	4.10	5.07	6.19	5.38
24	4.28	5.85	4.22	5.32	6.30	5.50
33	4.18	5.35	4.09	5.79	6.02	5.80
52	4.01	4.13	4.19	5.79	5.80	5.79

TABLE I (Continued)

Days	Treatment					
	Wet Un- treated	Partly Wilted Un- treated	Partly Wilted + Sugar	Partly Wilted + Lac- tic Acid	Partly Wilted + MnSO ₄	Fully Wilted Un- treated
<u>Grinding Method--Replicate II</u>						
4						
17	4.26	6.18	4.09	6.18	6.33	5.69
24	4.29	6.02	4.20	6.10	6.54	6.05
33	4.22	5.68	4.05	5.88	6.10	5.83
52	4.10	5.50	4.21	5.59	5.83	5.82
<u>Grinding Method--Replicate III</u>						
4						
17	4.24	6.17	4.11	6.07	6.29	5.52
24	4.27	6.21	4.20	6.11	6.33	5.56
33	4.18	5.79	4.31	5.80	6.10	5.98
52	4.12	5.51	4.22	5.68	5.80	5.80
<u>Grinding Method--Average Quality After Given Number of Days</u>						
4	2	2	1	2	2	3
17	1	5	1	4	4	3
24	1	5	1	3	5	3
33	1	5	1	5	6	3
52	1	2	1	5	4	2

TABLE II

ANALYSIS OF VARIANCE OF THE DATA FOR EXPERIMENT I

	D.F.	S.S.	M.Sq.	F_1
Total	269	130.08		
Replicates	2	0.44	0.22	
Methods	2	2.95	1.48	74.0**
Replicates x Methods	4	0.23	0.06	
Treatments	5	104.60	20.92	1046.0**
Methods x Treatments	10	4.81	0.48	
Error (b) Replicate x Method x Treatment	30	0.66	0.021	
Error (c)	216	16.39	0.08	

** Significant at the 1% level.

the silage prepared by this method was obviously of far higher quality than the silage prepared by chopping or crushing.

Mechanical preparation of the forage before ensiling apparently affected the fermentation process that is responsible for the production of good silage. By grinding, cell walls were broken, and a solution of nutrients was available to bacterial action without the delay of diffusion from the interior of the plant tissues. Thus, a very rapid action of lactic acid bacteria was possible in the case of the ground silage and high acidities were produced promptly, which could inhibit the action of bacteria which produce the weaker butyric acid and other undesirable materials.

The difference in the behavior of the silages might be attributed additionally to a difference in the cycle of carbohydrate utilization and formation. In the "ground" silage, the readily fermentable carbohydrates would be quickly converted into lactic acid, with the production of high acidity promptly. This high acidity would inhibit the action of bacteria and enzymes that hydrolyze proteins into amino-acids, and would prevent their breakdown into basic substances (amids, amines, ammonia) which would neutralize the acids previously formed. In the meantime, enzyme action would gradually transform the higher carbohydrates, such as starch, into

simpler fermentable forms, which would be converted into acids, with gradual increase of acidity.

In the "crushed" and "chopped" samples, fermentation to give high acidity was delayed, permitting degradation of the proteins and continual neutralization of acids. The formation of butyric rather than lactic, acid was favored. At the higher pH, later fermentation of available sugars into acids was slower than the formation of basic material by proteolysis, and the pH rose. Thus, grinding the silage appears to have been beneficial due to rapid initial lactic acid fermentation, which prevented the development of an adverse proteolytic and butyric fermentation.

The t test of the statistical analysis revealed that the difference to be significant between treatments was 0.06. The treatments with sugar staying at a pH of 4.4, 4.43, and 4.24 for the three methods "chopped," "crushed," and "ground," respectively, were by far the best.

The sugar treatment (Figure 1) for the three methods may be considered as proof of the action of the microorganisms upon the readily fermentable carbohydrates and the value of the "ground" technique. As seen in Table I, the sugar treatment was most helpful in the methods "chopped" and "crushed," but was not different

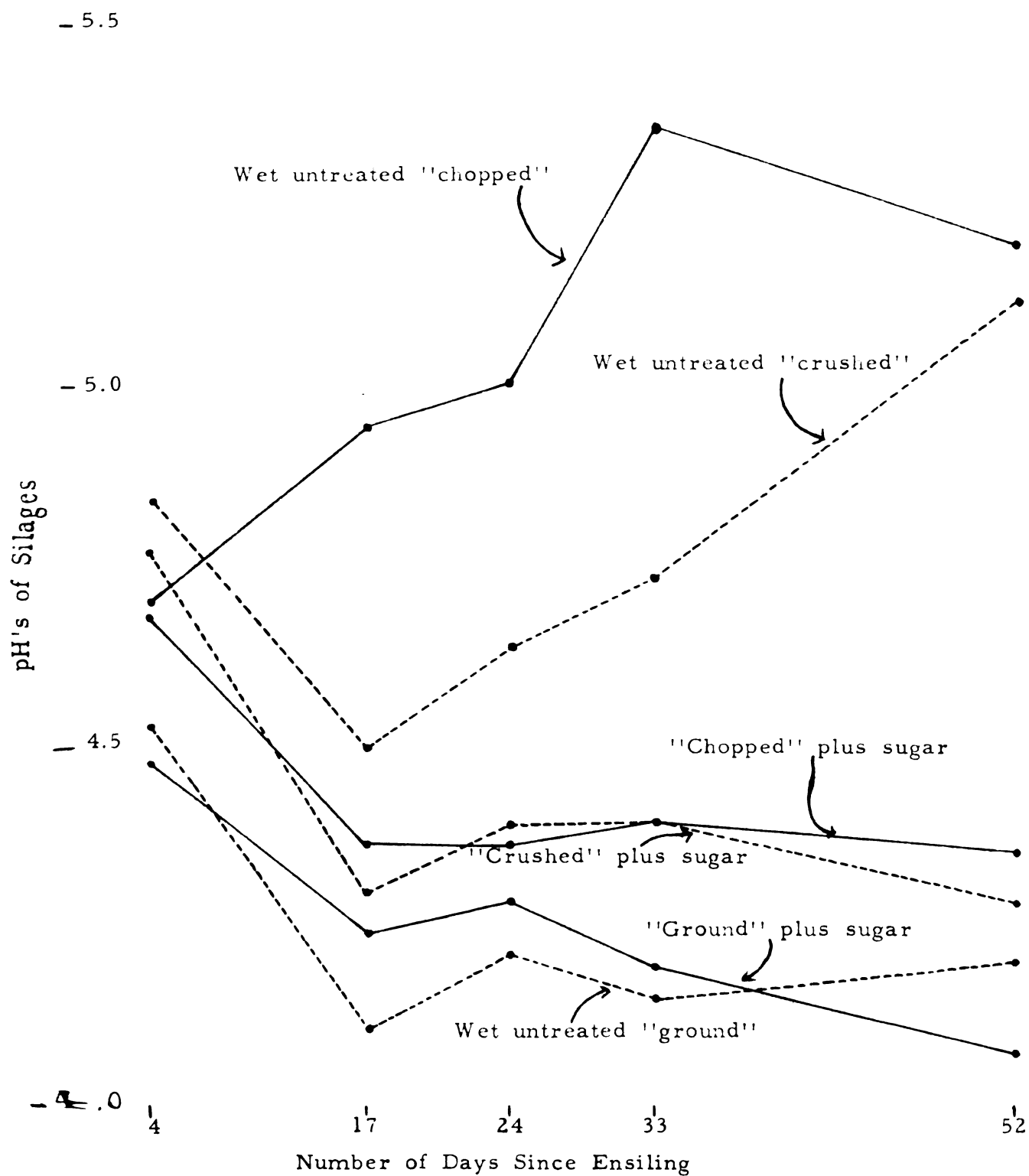


Figure 1. The fluctuations of the pH of the silage during the storage period.

from "wet untreated" in the method "ground." The statistical analysis did not show any significant difference between wet, untreated ground and partly wilted sugar ground. In this experiment sugar was always helpful in making good silage when the partly wilted green fodders were "chopped" or "crushed" or "ground." It also looks probable, as shown in Figure 1, that a "ground" wet silage treated with sugar could stay unspoiled longer than one treated mechanically the same way, but put up without sugar. As shown in Table III, the treatments with sugar had the lowest buffer capacity after thirty-three days, but no difference was observed between this treatment and the "wet untreated," followed by the "lactic acid," "partly wilted untreated," "manganese sulfate," and "fully wilted untreated." The "wet untreated" material in all three methods was better than either the chemically treated ones or those wilted except where sugar was used.

The material which was partly wilted but untreated in the methods "chopped" and "crushed" was found to be better than the fully wilted material. The "ground" silage which was partly wilted was almost equal to the fully wilted.

The lactic acid culture treatments, in all three methods, were better than both manganese sulfate and fully wilted treatments.

TABLE III
 BUFFER CAPACITY AFTER 33 DAYS
 (cc. of N/4 acid required to bring 5 gms.
 of silage from pH 11.0 to pH 3.0)
 Experiment 1

Treat- ments	Silage						Quality
	Chopped		Crushed		Ground		
	pH	B.C.*	pH	B.C.*	pH	B.C.*	
Wet un- treated	5.70	18.0	4.43	18.2	4.18	17.3	1
Partly dry un- treated	5.89	20.4	5.76	19.5	5.68	19.0	3
Partly dry + sugar	4.40	15.1	4.28	17.1	4.09	17.3	1
Partly dry + lactic acid	5.56	18.6	5.78	20.1	5.88	17.9	3
Partly dry + MnSO ₄	5.89	20.5	5.90	19.4	6.10	17.7	3
Fully dry un- treated	5.72	21.0	6.07	21.4	5.33	20.1	4

* Buffer capacity after 33 days.

** "Partly dry" and "fully dry" mean partly wilting and fully wilting, respectively.

The addition of lactic acid bacteria starters did not show any advantage in silage-making. It appeared that the forage as harvested carried enough of this type of bacteria to perform the desirable fermentation.

The addition of manganese sulfate was not an effective way of improving grass silage quality.

In general (Table I), within the three methods the treatments can be classified from a pH standpoint from the lowest to the highest as follows:

1. Partly wilted plus sugar;
2. Wet untreated;
3. Partly wilted untreated;
4. Partly wilted plus lactic acid;
5. Partly wilted plus manganese sulfate;
6. Fully wilted untreated.

The odor of the silage did not invariably indicate the pH level, since it was found that the "fully wilted silages" had a better odor than did the "lactic acid" or the "manganese sulfate" treatments.

Experiment 2

The results of this experiment are recorded in Table IV. An analysis of variance of the data (Table V) shows differences highly significant between the treatments.

Silage "ensiled at once" was more acid eleven days after ensiling than it was five days after ensiling, but showed a slight decrease in acidity by the twenty-sixth day. However, the quality of the silage was not poor, nor its odor offensive, in spite of a pH around 6.

Silage prepared from forage stored "unwilted, cold, in the dark" (about 16 hours at around 40° F.) was more acid than forage similarly stored at about 75° Fahrenheit. Again, pH did not adequately indicate quality, since none was offensive.

The effects of sugar and phosphoric acid were conspicuous, though neither one had, even after twenty-six days, a pH in the vicinity of the ideal (4.2). The phosphoric acid showed the lowest pH 4.79 after eleven days which stayed almost unchanged as time proceeded. It seems that the amount of acid added to the material was enough to favor the action of the lactic acid bacteria and inhibit that of the proteolytic enzymes and other microbes since even after twenty-six days this silage was still mild. Table VI shows



TABLE IV

pH OF SILAGE MADE UP OF SECOND CUTTING ALFALFA
TREATED IN DIFFERENT WAYS, AFTER
5, 11, AND 26 DAYS
Experiment II

Treatments	Replicate I			
	Days			Quality After 26 Days
	5	11	26	
Ensiled at once	5.86	5.45	6.00	1
Unwilted, warm in the dark	6.08	6.20	6.09	2
Unwilted, cold in the dark	5.86	5.20	5.87	2
0.5% phosphoric acid	5.21	5.49	5.61	1
2% sugar	5.74	5.32	5.39	2
Wilted in the dark	5.58	5.32	6.60	2
Wilted in sunlight	5.57	4.85	5.39	2

TABLE IV (Continued)

Replicate I				Replicate III			
Days			Quality After 26 Days	Days			Quality After 26 Days
5	11	26		5	11	26	
4.87	5.20		1	5.63	5.87		1
5.75	5.82		2	6.08	6.05		2
5.71	5.49		2	5.67	6.22		2
4.79	5.75		1	4.80	4.81		1
5.59	4.82		2	5.95	5.10		2
5.51	4.39		2	5.62	5.69		2
4.55	5.19		2	5.35	5.30		2

TABLE V

ANALYSIS OF VARIANCE OF THE DATA FOR EXPERIMENT II

	D.F.	S.S.	M.Sq.	F ₁	F ₂
Total	62	11.93			
Replicates	2	0.74	0.370	3.135	3.34-5.45
Treatments	6	4.45	0.742	6.286**	2.44-3.53
Replicates x treatments	12	1.16	0.096		
pH	2	0.80	0.400	3.390	3.34-5.45
pH x treatments	12	1.45	0.120		
Error	28	3.33	0.118		

** Significant different at the 1% level.

TABLE VI
 BUFFER CAPACITY AFTER 5 DAYS
 Experiment II

Treatments	Per Cent of Dry Matter	pH	Buffer Capacity	Quality
Ensiled at once	28.90	5.86	19.10	1
Unwilted, warm in the dark	-	6.08	21.90	1
Unwilted, cold in the dark	-	5.86	22.50	1
5% phosphoric acid	28.90	5.21	21.20	1
2% sugar	29.85	5.74	20.75	1
Wilted in the dark	-	5.98	25.30	1
Wilted in sunlight	22.15	5.58	19.00	1

their different buffer capacity. The addition of sugar to the second cutting alfalfa did not develop a low pH as it did when the treatment was made with immature alfalfa in Experiment 1. This might have been due to a lack of water in the woody alfalfa (70 per cent water) which became a limiting factor to enzymes and microorganisms that were present in the material.

The treatments "wilted" in the dark and sunlight were not very different in odor, but they were remarkably different to the standpoint of acidity. The material kept in sunlight was definitely more acid. After eleven days, the bacteria built a pH level between 4.85 to 5.35. The same material kept in the dark did indicate a decrease of the pH only after twenty-six days. This difference may be explained by the fact that the material kept in the dark could not continue to build simple carbohydrates through the photosynthetic process, while the one kept in sunlight could. Therefore, the sunlight-treated silage had a better chance to produce a lower pH after a few days, or to be kept longer in storage without spoilage.

Experiment 3

The results of this experiment are recorded in Table VII. In every case, silage from "ground" material was better than silage from "chopped" forage.

When judged on a pH level, the silage prepared from forage cut at 1:30 p.m. appeared better than that from forage cut at 5:00 p.m. the same afternoon. Silage prepared from forage cut at 5:00 a.m. the following morning appeared better than silage prepared from forage cut in the late afternoon of the day before.

In spite of an apparently inadequate acidity, all the silages had a mild, inoffensive odor, and did not deteriorate on extensive storage. This is in striking contrast to the results in Experiment 1. In this experiment, pH does not seem an adequate criterion of quality.

TABLE VII

pH OF SILAGE MADE OF WOODY ALFAIFA AT DIFFERENT
TIMES OF DAY, AFTER 10 DAYS
Experiment III

Time of Day Silage Made	Treatments	pH After 10 Days			Quality
		Repli- cate	Repli- cate	Repli- cate	
		I	II	III	
1:30 p.m. (Aug. 4)	Chopped	-	5.50	5.04	1
	Ground	5.08	5.09	5.05	1
5:00 p.m. (Aug. 6)	Chopped	5.63	5.50	5.59	1
	Ground	5.31	5.10	5.38	1
5:00 a.m. (Aug. 5)	Chopped	5.10	5.53	5.20	1
	Ground	4.35	4.49	4.40	1

SUMMARY AND CONCLUSION

Immature first-cutting alfalfa in mixture with brome grass was "chopped," crushed and chopped (or "crushed"), and chopped and "ground" before ensiling. In all cases, "ground" silage appeared more desirable in pH and odor than the others. Silage made with 2 per cent added sugar was much better than untreated, partly wilted silage, whether "chopped," "crushed," or "ground," but was not better than "ground, wet untreated." Wilting before ensiling in glass jars was always detrimental to quality. Lactic acid bacterial cultures were not helpful, nor was addition of manganese sulfate. Silages at pH values greatly above 4.2 were highly offensive in odor.

Second-cutting alfalfa was wilted in sunlight, in the dark, at warm and at cool temperatures in various combinations. Wilting or storage in sunlight or at low temperature was better than similar exposure in the dark or at higher temperature. Addition of 2 per cent sugar was remarkable ineffective in lowering the pH of these silages. Silages were inoffensive in odor, even at pH values of 5.5.

Silage was made from alfalfa cut at various times of day. In all cases "ground" silage was lower in pH than "chopped." Effects due to time of day appeared inconclusive. All silages were inoffensive in odor even at pH of 5.6. The pH was not an effective criterion of silage quality.

It is suggested that by grinding, all membranes are broken, and plant juices are made free to bacterial action. Prompt fermentation of the free solution results. This produces a high initial acidity which inhibits butyric and proteolytic fermentation. By avoiding protein splitting, neutralization of silage acids is prevented and preservation is assured.

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