CORN-STOVER SILAGE

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INTRODUCTION

The ensiling of corn stover and cured corn fodder is not a new idea. In a few localities farmers have followed this practice to a limited extent for some years, and there have appeared several Experiment Station publications which deal briefly with the process. Experimental data on the subject are, however, very meager; little has been established with reference to the practicability of stover silage, while nothing is known concerning the nature of the fermentation which takes place and the factors operative other than what may be deduced from knowledge of ordinary silage. The present necessity for more economic production, with the conservation of concentrates and the utilization of more roughage in the live-stock and dairy industries, makes a reconsideration of corn-stover silage of especial pertinence.

The object of the present study was partly to test the practicability of ensiling stover, and partly to determine the nature of the fermentation which takes place therein.

PRACTICABILITY OF ENSILING CORN STOVER

The stover used in this experiment was ensiled early in April, 1916; the material had been kept in a shed since fall and was quite dry. The condition of the material was not good; it was moldy in spots and on the whole represented an inferior grade of stover. This stover was run through a silage cutter and packed in the silo by means of tramping. Water was added in a continuous stream through a hose which was carried and the water distributed by the man who did the packing. A water meter was attached to the hose so as to enable the regulation of the amount used. A wooden-stave silo 16 feet in diameter was filled with 32,000 pounds of stover to which were added 66,000 pounds of water.

Samples of the silage, which were taken at frequent intervals, were examined for general appearance, texture, and aroma. The stover was soon observed to undergo a fermentation with the formation of a product quite similar to normal silage made with green corn. The material softened, regained a slightly greenish color, and developed an aroma simulating that of normal silage, though inferior in all these respects to silage made in the usual way from green corn.

Feeding tests made at the end of the experiment showed that cattle ate this silage with little waste and apparently with a relish. While

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it is not believed that the stover silage is as palatable as is that made from fresh corn, it did prove to be a very acceptable feed which was preferred by cattle to any of the dry roughages furnished. Some experiments in which the actual feeding value of stover silage is determined in comparison with ordinary silage and other roughages are desired, and it is hoped that such tests may be carried out at this Station in the near future.

The keeping quality of stover silage appears to be excellent, provided sufficient water is added. Although, as is the case with other types of silage, the surface material undergoes a moldy spoilage accompanied with heat formation, this condition does not extend to more than the ordinary depth. The silage made in this experiment was not all used during the following winter, and the remaining material, at the time of this writing nearly 1½ years old, is still in excellent condition. In view of the very satisfactory results obtained with such an inferior raw product, we do not hesitate to predict success in the ensiling of any stover which is in reasonably good condition.

Probably the most important consideration for the successful production of stover silage is the amount of water to add. This obviously will vary according to the quantity of water contained in the stover, and this factor should be taken into consideration. While it would be more scientific and exact to determine the most desirable amount of water to add by means of moisture tests on the stover, such a recommendation would find no place in farm practice. In our experimental silo the proportion was about 2 parts of water to 1 of stover, but the stover was probably somewhat drier than would usually be the case. As may be seen from the moisture determinations, which are reported in another section (Table I), the quantity of water added was none too much; in general appearance and to the touch some of the samples seemed to be considerably below the most desirable point.

A laboratory test was carried out upon this point by making stover silage in small jars with varying quantities of water and examining after about one month for general appearance and condition of moisture. The stover used in these tests was very dry. Samples which were made with equal parts by weight of water and stover, as well as those made with 1½ parts of water to 1 of stover were too dry to undergo a typical fermentation and form good silage. Those which had water in the proportion of 2 parts to 1 of stover made good silage, but did not appear to have as much moisture as would be best. Samples put up with 2½ and with 2¾ parts by weight of water to 1 part of stover were in good condition when opened and apparently did not contain an excess of water.

It seems that in general, when reasonably fresh stover is used, about 2 parts of water by weight to 1 of stover would be advisable, while for older and drier stover a slightly larger proportion of water may be more

desirable. It should be kept in mind that these tests were all made with rather dry stover. In the case of ensiling soon after the corn is husked, 2 parts of water might be too much. However, it appears from our observations that there is less danger of adding too much water than of getting too small an amount, and that considerable water may be added above the required amount without injury to the product.

The water should be added uniformly as the silo is being filled so that all the dry cut stover becomes thoroughly wet down. If this precaution is not taken, the water may follow channels down through the silage and waste away at the bottom of the silo. In such an instance spoiled silage might result in some parts of the silo because of an insufficient amount of water.

FERMENTATION OF STOVER SILAGE

To obtain information on the nature of the fermentation which takes place in silage made from stover, determinations were made of the volatile and nonvolatile acids, temperatures, and numbers and types of bacteria at various stages of the ripening process.

Samples for examination were obtained by means of a 2-inch auger provided with an extension shaft of 8 feet, thus making it possible to penetrate to the center of the silo. By repeatedly boring in a short distance and withdrawing the auger until the center of the silo was reached, no difficulty was experienced in securing sufficient material for the different tests. The sample obtained in this way represented the silage mass from the wall to the center of the silo. Not more than one boring was made in one place, the different samples being removed at points all the way around the silo and from 3 to 8 feet from the ground. The material so obtained was subjected to pressure in an ordinary lard press and sufficient juice collected for the various examinations made.

ACID FORMATION.—There can be no doubt that the amount and character of acids in silage influence its quality profoundly. Since it has been the general experience that extremely green corn produces a very sour silage, while corn more nearly mature produces a silage with less acid and of a much better quality, the acid fermentation in silage made from dry material is of interest.

The volatile acidity was determined by subjecting a 100-gm. sample of juice to steam distillation under reduced pressure until 4 liters of distillate were secured. These were titrated directly after collection with *N*/10 barium hydrate, with phenolphthalein as the indicator. The nonvolatile acidity was obtained by the difference between the volatile acidity and a total acid determination made by the titration of 20 gm. of juice, diluted to 500 c. c. with carbon-dioxid-free water, against *N*/10 barium hydroxid. In Table I the nonvolatile and volatile acids (calculated as lactic and acetic, respectively) are reported in terms of percentage of air-dry material.

TABLE I.—*Acid formation in stover silage*

Age.	Total solids.[a]	Acid in total solids.		Ratio of nonvolatile to volatile.
		Nonvolatile.[b]	Volatile.[c]	
Weeks.	*Per cent.*	*Per cent.*	*Per cent.*	
3/7	28.9	Trace	0.51	.....
1	35.5	0.16	.87	1 : 5.40
2	26.7	.95	1.36	1 : 1.43
3	26.8	1.36	1.41	1 : 1.04
4	27.1	1.54	1.49	1 : .97
5	29.7	1.51	1.55	1 : 1.02
6	31.2	2.00	1.69	1 : .85
8	27.5	2.53	1.92	1 : .75
10	32.1	2.64	1.82	1 : .75
12	25.8	3.15	2.24	1 : .71

[a] Air-dried. [b] Calculated as lactic acid. [c] Calculated as acetic acid.

It is seen from the figures in Table I that for the first week the volatile acidity was greatly in excess if the nonvolatile portion; that from the second to the fifth week the two were apparently present in about equal amounts; and that from the sixth week on, the nonvolatile acids were in excess, the proportion of nonvolatile acidity increasing to the end of the experiment. The total acidity obtained was somewhat lower than is usually found in ordinary silage. This is probably to be expected, in view of the chemical differences in the raw materials. In this connection, however, it should be noted that in this experiment samples were not taken after the twelfth week. It is not unlikely that the acidity increased some after the last sample was secured. In regard to the proportion of nonvolatile to volatile acids, if we accept the ratio 1 to 0.75 reported by Dox and Neidig (*5*)[1] as representing a general average for ordinary silage, it will be seen that our results on corn stover silage indicate a remarkable agreement in this respect between these two types of ensilage.

FERMENTATION TEMPERATURES.—Temperature records were obtained by means of four resistance bulbs with about 60 feet of insulated cable attached to each which were buried in the silage as the silo was filled. The bulbs were located as follows:

No. 1. About 2 feet from the bottom and in the center of the silo.

No. 2. About 6 feet from the bottom and about 3 feet from the center of the silo.

No. 3. At the same height as No. 2 but about 3 feet from the center in the opposite direction.

No. 4. About 12 feet from the bottom and in the center of the silo.

The ends of the cables were located at a convenient place on the outside of the silo so as to allow easy attachment for temperature readings. Table II gives the temperatures obtained from April 4, the day the silo was filled, until June 16.

[1] Reference is made by number (italic) to "Literature cited," p. 600.

TABLE II.—*Fermentation temperatures of stover silage*

Date.	Temperature (°F.).				
	Bulb 1.	Bulb 2.	Bulb 3.	Bulb 4.	Atmosphere.
Apr. 4	42.0	42.0	42.0	.....	33.5
5	42.5	49.5	47.0	.....	42.0
6	46.5	50.5	48.0	46.5	43.0
7	46.5	51.0	48.5	47.5	46.5
8	47.0	52.5	49.5	48.0	33.0
9	47.5	54.0	51.0	49.5	33.0
10	49.0	54.0	51.0	50.5	37.0
11	49.0	54.5	51.5	51.0	42.0
12	49.5	54 5	52.0	52.5	48.0
13	50.0	56.0	52.5	52.5	58.0
14	50.5	56.0	53.5	55.0	51.0
15	50.0	56.0	53.5	55.0	49.0
16	50.0	56.0	53.5	55.0	50.0
17	50.0	56.0	53.5	55.5	52.0
18	50.0	56.5	54.0	56.0	49.5
19	50.0	57.0	54.5	56.0	46.0
20	50.0	57.0	54.5	56.0	55.0
21	50.0	57.0	54.5	55.0	49.5
22	49.5	57.0	54.5	56.0	49.5
23	49.5	57.5	55.0	57.0	41.5
24	50.0	57.5	55.0	57.0	47.0
25	50.0	57.5	55.0	57.0	47.0
26	49.5	57.5	55.0	57.5	48.5
27	49.5	57.5	55.0	57.5	48.5
28	50.0	57.5	55.5	57.5	47.0
May 1	49.5	57.0	55.5	58.5	66.5
2	49.0	57.5	55.5	59.5	63.5
4	50.0	58.5	56.5	59.5	53.5
6	50.0	58.5	56.5	59.5	60.5
8	50.0	58.0	56.0	59.0	65.5
16	50.5	58.0	56.0	59.0	65.0
22	50.5	58.0	56.5	59.0	55.0
31	51.0	58.0	57.0	61.0	64.0
June 16	52.0	58.5	57.5	63.0	63.5

Recent investigations have furnished abundant proof that high temperatures are not essential in silage preservation, and, in fact, do not occur except at the surface, which undergoes an aerobic decomposition. Bechdel (*3*) has recorded an instance in which the maximum temperature attained in the center of a concrete silo during the curing period was only 60° F. As has been shown by Eckles, Oshel, and Magruder (*6*), the atmospheric temperature at time of filling influences greatly the temperature attained during the fermentation of the silage.

Table II shows that the temperature at the start was 42° F. and gradually increased until the readings were discontinued. The maximum temperature attained was 63° F. in the case of bulb 4; but bulb 1, which was buried to a depth slightly below the surface of the surrounding soil, showed a maximum temperature of only 52° F. An examination of the column giving the atmospheric temperature during this period suggests that the continued increase in the silage temperature during the latter part of the time may be accounted for by a similar increase in

the outside air. However, the more rapid increase during the early part of the period was entirely independent of this factor. A comparison of the temperature records secured in this experiment with the data which have been obtained on ordinary silage at this and other Experiment Stations indicates that there is no wide difference, if any, between the rate and amount of increase in temperature in silos in which ordinary silage and corn-stover silage have been stored.

BACTERIOLOGICAL OBSERVATIONS.—A quantitative bacterial examination was made on each of the samples taken. The juice obtained was plated in proper dilutions and the counts obtained were reported as numbers per cubic centimeter of juice. Lactose agar was used, and the plates were counted after six days' incubation at 33° C. As may be seen from Table III, it appears that the bacterial count increases during the first week and is followed by a continued decrease thereafter.

TABLE III.—*Number of bacteria in stover silage at different stages of curing*

Age.	Number of bacteria per cubic centimeter.	Age.	Number of bacteria per cubic centimeter.
Weeks.		*Weeks.*	
3/7	528, 000, 000	5	510, 000, 000
1	3, 630, 000, 000	6	235, 000, 000
2	1, 850, 000, 000	8	186, 000, 000
3	975, 000, 000	10	98, 000, 000
4	400, 000, 000	12	71, 000, 000

Direct microscopic examinations of the silage juice were made in order to follow in a general way any marked changes which take place in the bacterial flora during the curing process. This at best could only give suggestive data, but such examinations are sometimes important in connection with cultural studies. At first a great variety of cells were observed. During the first two weeks rods and cocci were apparently present in about equal numbers, after which the rods became increasingly predominant. Toward the end of the experiment practically nothing but rods were found in the microscopic preparations. Because of the high acidity it is not likely that the cocci were active nearly as long as they appeared under the microscope. But it is probable that the acid medium would tend to peserve the cells so that they would appear for some time after they were inactive or even dead.

A qualitative bacterial study was also carried out. All of the colonies from a representative lactose-agar plate from each sample were isolated and subjected to a cultural study. For the present purpose they may be divided roughly, according to their action on litmus milk, into acid-forming, casein-digesting, alkali-forming, and inert groups. The acid formers may be further divided according to whether they produced sufficient acid to cause coagulation of the milk. The distribution of these groups in silage at various stages of its fermentation is shown in Table IV.

TABLE IV.—*Groups of bacteria present at different stages of curing*

Age.	Percentage of total number.				
	Acid-coagulating group.	Acid-noncoagulating group.	Casein-digesting group.	Alkali-forming group.	Inert group.
Weeks.					
3/7	10	53	10	4	23
1	14	57	9	6	14
2	20	71	0	0	9
3	11	63	5	0	21
5	21	67	0	0	12
6	60	30	0	0	10
8	68	32	0	0	0
10	60	40	0	0	0
12	56	39	5	0	0

Table IV shows that the rather complex bacterial flora which is present at the beginning of the process gives way to one which is almost entirely acid-producing as the fermentation progresses. The proportion of acid-forming and coagulating organisms to the noncoagulating ones also increases as the curing period advances. A comparison of these figures with those given in Table III indicates that the change in flora is not to be accounted for by an actual increase in the high acid-forming organisms during the latter part of the fermentation period, but rather to the fact that they do not decrease as rapidly because of their greater resistance to the unfavorable hydrogen-ion concentration.

The division of the acid-forming organisms into coagulating and noncoagulating types, though convenient and significant for the present purpose, probably does not separate them into natural groups. From early in the fermentation the predominating organisms were acid formers, most of which probably belonged to the same general group. We have found cultures which were apparently identical, as indicated by the cultural and fermentative reactions studied, but which varied in the amount of lactic acid produced in milk from only 0.3 to more than 2.0 per cent. All of these probably belonged to the same general group as the aciduric bacteria which have previously been noted as occurring abundantly in silage. In the first two samples examined organisms of the colon-aerogenes group were found and also a few cultures which were probably *Streptococcus lacticus*, but tests were not applied which would definitely identify the latter.

NATURE OF SILAGE FERMENTATION

Until recently the cell-respiration theory of silage fermentation established by Babcock and Russell (*1*, *2*) has not been seriously challenged. During the past year, however, several publications have appeared in support of the bacterial explanation of this phenomenon. In view of the recent contributions to the subject, it is not out of place to examine

critically the present status of the question. As the older literature has been reviewed so many times that further elaboration is not necessary, we shall pass in review only those papers of very recent date.

Hunter and Bushnell (*8*) have demonstrated the presence in silage of large numbers of high acid-producing bacteria, and have furnished strong evidence that these organisms are mainly responsible for the acid fermentation. Although their work is a most valuable one, it should be borne in mind that the evidence is circumstantial and perhaps not conclusive. It is rather doubtful if the data submitted justify the positive conclusion that—

> The present investigation warrants the statement that acid production, common to all normal silage, is largely the result of fermentation by the Bulgarian group of bacteria.

The fact that these bacteria formed considerable acetic acid when grown in alfalfa extract to which was added 1 per cent of glucose hardly warrants the assumption that—

> Although these organisms evidently do not produce all of the acetic acid found in normal silage, they must be responsible for a large per cent of it.

Sherman (*13*) also noted the presence of large numbers of the aciduric bacilli in silage and reported some observations which indicated that they are of significance in the fermentation process. The evidence, however, was not direct and by no means conclusive.

Hunter (*7*) in his work on heat production in silage has added further weight to the bacterial theory of silage fermentation. Unfortunately it is not possible to evaluate properly some of the interesting points contained in his paper, as they are obscured by insufficient description. For example, some data are given which show the difference in heat formation between green kafir heated and green kafir inoculated with *Bacillus bulgaricus*. The exact treatment in this case is not clear: If the inoculation was made into heated kafir, the results are of utmost significance; if, on the other hand, as the caption of the graph would indicate (*see 7, fig. 9*), the inoculation was made into unheated kafir and that compared with heated kafir, the test contributes nothing to the solution of the moot question. In his assumption that cell respiration can play no part in the fermentation of silage made from dry forage, Hunter has arrived, we think, at conclusions which are, in part at least, erroneous.

In a very interesting paper Lamb (*9*) concludes that both factors are of importance, but that microorganisms play the larger part, especially in the production of acid. Following the suggestion of Rahn (*10*), he has attempted to determine the cause of the process by the rates of change in the fermentations studied. The course of the curve obtained when such data were plotted was interpreted as indicating whether the action was of bacterial or enzymic origin.

Although it is true that purified enzym preparations acting under favorable conditions give time curves which follow, with certain modifications, the law of mass action, while curves representing bacterial action take an entirely different course because of an increase in the active mass with the multiplication of the organisms, we are inclined to believe that Lamb has placed undue confidence in this method, especially when we consider the complexity of the material studied and the factors concerned. A few considerations will suffice to illustrate some of the possibilities of error in such a method. In the first place conditions in silage are not constant, but are undergoing continual change. For example, the temperature and acidity, which are of utmost significance in enzym action, increase as the fermentation progresses. In view of the great increase in activity of some enzyms as the temperature is increased, and the stimulating effect on some enzyms of an increased hydrogen-ion concentration (within certain limits), it is not at all impossible that these factors might so modify the course of action of an enzym as to produce a curve resembling that typical of bacterial action.

Again, the phenomenon of adsorption and the action of the so-called antienzyms in many cases may so suppress the activities of an enzym during the early stages of the reaction as to cause it to follow a course not at all characteristic of enzym action. This has been beautifully illustrated by Rosenthal (*11*) in his work on the antitryptic action of egg albumen. It was shown that the trypsin was at first suppressed, but gradually regained its power and increased in activity so as to give the appearance of an increase in the "active mass" as indicated by a curve convex to the axis of abscissæ (the typical bacterial curve). On the other hand, the same trypsin preparation when acting on egg albumen which had been previously heated to destroy the antitrypsin gave a time curve characteristic of enzym action.

These illustrations will suffice to show the fallacy of such a method, but the number of possibilities of error in its application to such a complex mixture as silage might be increased almost indefinitely. That the limitations of this method of studying biochemical phenomena were appreciated by Rahn (*10*) is shown in the following paragraph from his valuable paper:

> It is hardly necessary to mention that the curve of a process will be an absolute means of discussion only in case of pure cultures. In natural fermentations, there is always the possibility that different processes taking place at the same time destroy the regular form of the curve. A simple example would be the growth of an acid-producing and an alkali-producing organism in the same liquid. It is also possible that an enzymic curve under certain conditions shows the form of a fermentation curve. We can imagine that an enzym is acting slowly at first, because of an unsatisfactory acidity of the medium. By a chemical or microbial process, independent of the enzymic action, the acidity may be made more suitable for the enzyme, and this will cause an increased rate of action of the enzyme and give the type

of a fermentation curve without the presence of organisms. The value of the curve is, therefore, not an absolute one and no conclusions ought to be drawn without consideration of the possibilities of error.

It would indeed seem that the application of such methods to the study of silage fermentation is entirely without foundation.

To the evidence in support of the bacterial theory of silage fermentation should be added the very suggestive observation of Clark (*4*) that the hydrogen-ion concentration of mature silage is coincident with the limiting hydrogen-ion concentration obtained in cultures of *Bacillus bulgaricus*, which organisms are considered by some workers as of paramount importance in the ripening process. In support of the respiration theory, on the other hand, it is pertinent to call attention to the recent work of Round (*12*), which indicates that cell respiration is of greater importance in the fermentation of sauerkraut than has been generally recognized.

It was thought at the beginning of this experiment that a study of the fermentation in silage made from dry stover would throw much light on the nature of the process in ordinary silage. The belief was held that the activity of the plant cells (which have been demonstrated to play an important part in the fermentation of silage made from green corn) would be eliminated, to a large extent at least, in the stover silage. But, as may be seen from an examination of the foregoing results, the fermentation of ensiled stover appears to resemble, in its main characteristics, that which takes place in green-corn silage. In an effort to determine the factors responsible for the fermentation, laboratory tests were made by ensiling stover under different conditions. Glass jars containing 175 gm. of cut stover and 400 gm. of water were used. Some were untreated, some put up with antiseptics, while some were sterilized in the autoclave and reinoculated with 1 per cent of raw-silage juice. The results of this study are given in Table V.

TABLE V.—*Fermentation of stover ensiled under different conditions*

Sample No.	Treatment.	Age.	Acidity.	Bacterial count.
		Weeks.	*Per cent.*	
1	Untreated	4	15.5	210,000,000
2	....do	4	10.4	240,000,000
3	Sterilized and inoculated	7	2.4	320,000,000
4	....do	5	3.3	36,000,000
5	2 per cent of toluene	6	4.3	180,000
6	....do	6	4.6	460,000
7	2 per cent of ether	5	10.8	7,500,000
8	....do	8	12.4	Not made.

The main points brought out by this test are that stover silage is capable of undergoing a fermentation when preserved with ether, while bacteria alone are apparently unable to produce the typical fermentation, even though conditions are favorable for their active development. The predominating organisms in the sterilized and inoculated samples

were of the same type as those characteristic of normal silage. The organisms found in the samples preserved with antiseptics, on the other hand, were a more miscellaneous group; and, although many probably belonged to the same group as the aciduric bacilli of normal silage, the cultures isolated were mostly very weak acid producers. The fact that fermentation took place under ether indicates that the activity of the plant cells, whether it be called "respiration" or "autolysis," is present in silage made from dry stover. As silage preserved with ether fermented, whereas in that kept with toluene the process was checked suggests that conclusions drawn from experiments conducted with only one antiseptic are of doubtful value. When opened, the ether-preserved samples, after the evaporation of the ether, appeared to resemble the untreated material, while the sterilized and inoculated silage were "flat" and lacking in the characteristic aroma. The results with ether were checked by another test in which triplicate samples were preserved, and again an active fermentation took place as was indicated by the development of acidity in each case.

Although our results would tend to support the respiration theory of silage-curing rather than the bacterial, we do not feel that the data thus far collected warrant definite conclusions on this point. It is difficult to believe that such active acid-forming organisms should occur in silage in large numbers without taking some part in the acid fermentation; perhaps they supplement in some important way the action of the plant cells. It is not inconceivable that a preliminary cleavage due to cell respiration is an essential prerequisite for the vigorous action of the aciduric bacteria. In fact, the continued increase in the ratio of nonvolatile to volatile acidity as the fermentation progressed (see Table I) might lead one to suspect that such was the case. On the other hand, the great increase in the nonvolatile acidity from the fifth to the twelfth week, during which time the bacterial count was rapidly decreasing, might be interpreted as strong evidence against that view. It is clear that microorganisms are not solely responsible for the fermentation of silage, and the weight of evidence at the present time, in our opinion, indicates that their rôle is not as important as that of the plant cells.

Although not committing ourselves definitely on the nature of silage fermentation in general, in regard to the present problem we do maintain that the fermentation which takes place in stover silage is similar in its essential points to that of ordinary silage and is caused by similar factors.

SUMMARY

Corn stover when ensiled with a suitable quantity of water undergoes fermentation with the production of a palatable silage of good keeping quality, which resembles ordinary corn silage in aroma and appearance.

The fermentation which takes place in corn-stover silage appears to be essentially the same as that of silage made from green corn, as is

indicated by the total acidity developed, the ratio of nonvolatile to volatile acids, temperature observations, and bacterial studies.

From a review of the present status of the question as to whether bacteria or plant cells are mainly responsible for silage fermentation, it is concluded that the data thus far published are inconclusive. Although the results of the present study tend to support the cell-respiration theory, conclusions on this point are withheld.

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