Studies of Lactic Acid in the Silver Battery

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Introduction

Since the introduction of the continuous diffuser, the beet sugar industry has been confronted with a serious problem of bacterial fermentation in the diffuser process. Not only is sucrose lost directly by fermentation, but there is also undoubtedly some effect on juice purification and molasses production due to impurities introduced by the bacteria.

In recent work at the Western Regional Research Laboratory, United States Department of Agriculture (4)[2], a method has been developed for determination of lactic acid in beet processing liquors. This method has been employed in our work: 1. to determine the point of highest lactic concentration in the Silver chain-type batteries at the Carlton plant, Braw-ley, California, and at Sidney, Montana, and in the Robert battery at Santa Ana, California; 2. to provide a means of measuring actual sugar loss caused by bacterial action in the diffusion process; and 3. to measure the possible effectiveness of treatment of beets by a germicidal agent prior to slicing.

Lactic Concentration in the Battery

Samples of juice for analysis were taken from the juice ducts of the continuous diffusers and from the sample cocks on the Robert cells. A small amount of chloroform was added immediately, and the juices were cooled and quickly analyzed. All analyses for lactic were made by the method of Stark, Goodban, and Owens (4). This method calls for purification and separation of the lactic acid by ion exchange treatment and quantitative determination by the colorimetric procedure of Barker and Summerson (1).

Our results show that the lactic acid concentration in raw juice varies as widely as shown in Table 1. Earlier work (5) has reported concentrations from 82 to 1,610 ppm. (mg./L) .

Table 1.—Lactic Acid Concentration (ppm.) in Raw Juice.

	Carlton	Sidney
Average concentration	149	98
Low concentration	9	45
High concentration	244	136

Within our company we have two continuous diffusers, one located at the Carlton plant, Brawley, California, the other at Sidney, Montana. Both are chain-type, Silver machines. We have obtained data on the lactic acid acid concentration prevailing in the various cells of these diffusers at certain times during the 1953 campaign.

In Figure 1 is shown a comparison of individual runs at the two locations. Of course, the conditions represent only that time at which the samples were collected. However, the curves obtained are typical of the results found in our investigations.

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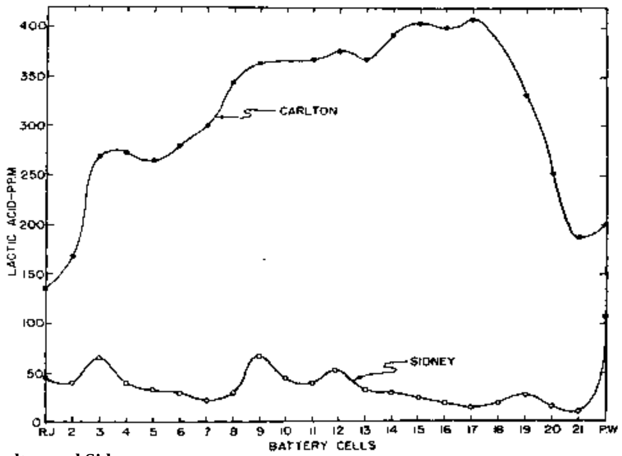
[2] Numbers in parentheses refer to literature cited.

Figure 1.—Lactic Acid Concentration in Various Parts of the Silver battery at



Carlton and Sidney.

The lactic concentration in the Carlton diffuser exhibited a marked peak in the centrally located cells, while no such peak was present in the Sidney battery. The average concentrations at Carlton were much higher than those at Sidney.

From Figure 1 it would be concluded that control measures at Sidney are more effective at the times investigated than those at Carlton. The battery juice temperatures at Carlton ran from about 65° C. on raw juice to a maximum of about 80° C. in cell 3 and back to about 65° C. in cell 21. At Sidney the corresponding temperatures were about 50° C, 60° C, and 40° C, respectively. On the other hand, addition of formaldehyde to the Sidney battery was approximately five times the amount used at Carlton. Thus it would seem that formaldehyde can effectively control fermentation even under temperature conditions quite favorable for bacterial growth. Needless to say, the cost of formaldehyde addition is a factor to be considered.

The geographic locations of Carlton and Sidney are such that extreme differences in climate are involved. Whereas at Carlton day-time temperatures above 100° F. are not uncommon during campaign, at Sidney much of the latter part of campaign can be in subzero wheather. Storage of beets at Carlton is seldom more than 24 hours, while at Sidney beets are often stored for 100 days or more. Neither the temperature in one case nor the long storage in the other is conducive to the best quality of beets. The amount of lactic found in Carlton beets averaged 50 ppm. That found in Sidney beets (late in campaign) was 64 ppm.

Since our Santa Ana, California, plant processes Imperial Valley beets in the spring, we had a unique opportunity to compare lactic acid formation in the Robert battery at Santa Ana with that formed in the continuous diffuser at Carlton at a time when both plants were operating on beets from the same area.

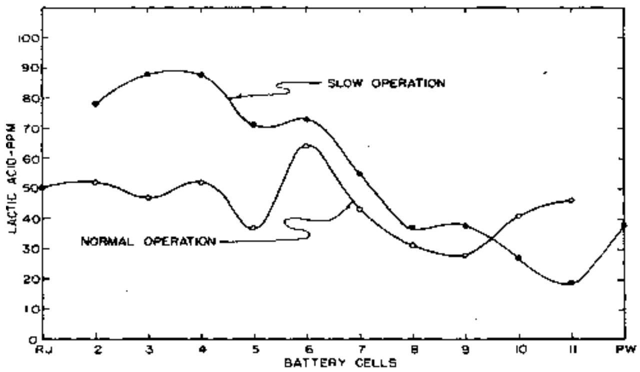


Figure 2.—Lactic Acid Concentration in Various Parts of the Robert Battery at Santa Ana.

Figure 2 shows the results obtained at Santa Ana when the battery was operating normally with about 45 minutes residence time for cossettes in the cell and when operating very slowly with about three hours hold-up of cossettes in each cell. The significant difference between these curves seems to be the peak in the curve for slow operation. During normal operation a relatively constant lactic concentration was obtained with little evidence of any lactic fermentation. Apparently the three hours' time during slow operation, along with somewhat lower temperatures, was sufficient to allow lactic fermentation to begin, as evidenced by the large increase in lactic with a maximum at cells 3 and 4. On the whole, lactic acid concentrations in the Robert battery bear out the contention that little or no fermentation takes place therein.

It was observed in our work that in those cases where a maximum lactic peak was found in the battery this peak fell into a rather narrow range of juice solids concentration of about 2.0-3.5 percent R.D.S. (Refractometer Dry Substance) . This was true of the case where a peak was found in the Robert battery as well as those in the continuous diffuser. It would appear that the bacteria tend to thrive at these levels of solids concentration.

The relationship between lactic acid and pH in the various cells was an inverse one, as would be expected. The patterns of high lactic and low pH followed each other closely during any one run, although the relative amounts of lactic could not be correlated with any given pH. It was felt that pH was a satisfactory control measure for factory operations provided changes in pH and particularly dips in the pH curve in the battery were carefully watched. Occasional checks of lactic acid levels should be helpful in factory control.

At Carlton in several of the runs made lactic was compared with reducing sugars obtained by the triphenyltetrazolium chloride method of Matt-son and Jensen (2) . Generally the reducing sugars decreased as lactic acid increased. This would probably be expected, since many bacteria attack reducing sugars in preference to sucrose.

Ratio of Sugar Lost to Lactic Acid Produced

If we can assume that the lactic acid produced is a measure of total bacterial activity and thus of the sucrose consumed by this activity, a ratio of the amount of sucrose lost to lactic acid produced would be of great value in calculating the sucrose losses in the battery. Such a determination is difficult since duplication of exact battery conditions is impossible and since calculation of loss of sucrose involves subtraction of two large values to obtain a small one. However, at Sidney several determinations of such a factor were made by analyzing fresh diffusion juice for lactic acid and true sugar (by single inversion) , placing some of this juice in a sterile container, allowing it to stand for 24 hours at battery temperature (about 60-65° C.), and again analyzing for lactic acid and true sugars. The ratio was then determined from the differences of these two analyses. The results were ratios (by weight) of sugar to lactic of 0.81-4.44 with an average ratio of 2.14.

The figure is, of course, an arbitrary one, applicable with certainty only to the juices and conditions under which it was obtained. Such a ratio may vary from area to area, year to year, and perhaps even cell to cell within the battery. However, it does provide a figure which is in the right order of magnitude, as one mole of lactic produced from a mole of sucrose would give a weight ratio of 3.8 and a maximum of 4 moles of lactic from one of sucrose gives a ratio of 0.95, the average between the two extremes being 2.38.

As an example of the application of this ratio, let us calculate the average amount of sugar lost at Carlton per day under the conditions encountered. Assume that 3,000 tons of beets are sliced per day and that the draft on the diffuser is 130. The average lactic acid concentration in the entering cossettes is 50 ppm., and that in the raw juice is 149 ppm.

3,000 x 2,000	=	6,000,000 pounds of beets per day
6.0 x 50	=	300 pounds of lactic acid enter in beets
6,000,000 x 1.30	=	7,800,000 pounds of raw juice per day
7.8 x 149	=	1,162 pounds of lactic acid in raw juice
1.162 — 300	=	862 pounds of lactic formed in the battery
862 x 2.14	=	1,845 pounds of sugar lost per day

$$\frac{1,845}{3,000 \times 2,000} \times 100 = 0.031 \text{ percent on beets}$$

Chemical Treatment of Beets

Since most of the bacteria which enter the battery come in on the surface of the beets, it was our thought that treatment of the beets with a germicidal agent before slicing might be of help in holding down fermentation in the battery. Several tests were made at Sidney to determine whether any of a number of chemicals might be promising in this respect.

Samples of 5 or 6 beets were taken from the picking rolls following the beet washer. The beets were cut in half longitudinally, with one-half being sliced by hand immediately and the other half being treated by immersion in the germicidal solution. Treatment was made at about 55°-60° C. for a two-minute period in solutions at the concentration recommended by the manufacturer. The treated beet halves were also sliced by hand and the cossettes produced in each case were placed in a sterilized container, covered with water, and allowed to stand at 60°-65° C. for 24 hours. The lactic acid content of the resulting juice was taken as a measure of the effectiveness of the germicide.

Table 2 shows the results of these tests.

. Table 2.—Treatment of Beets with Germicidal Agents.

Germicide	Lactic Acid in Juice		pH of Juice	
	Untreated	Treated	Untreated	Treated
	ppm.	ppm.		
Chlorine compound No. 1	120	220	5.5	5.2
Quaternary amine	230	140	5.2	5.3
Wetting agent	140	115	5.3	5.3
Commercial detergent	320	345	4.9	4.8
Phenolic germicide A	540	270	4.9	5.3
Phenolic germicide P	545	20	5.1	6.1
Chlorine compound No. 2	350	245	5.3	5.3
Polymixin	240	230	5.2	5.0
Bromine	385	320	4.9	4.9

It will be seen that the quarternary amine compound and the commercial phenolic germicides appear to be quite effective in lowering the fermentation potential of the beets tested. Not only is the lactic acid reduced by these agents, but also the pH of the resulting juice is somewhat higher than the corresponding untreated samples. It must be concluded that treatment of beets by a method similar to that employed may hold promise in helping to reduce sugar losses due to bacterial action provided that other factors within the battery itself can be controlled satisfactorily.

Control of Fermentation in the Battery

At the present time there are two principal methods of control of bacterial action in the continuous diffuser: 1. maintaining a high enough temperature to discourage bacterial growth, and 2. addition of a germicide to the battery cells. Maudru (3) has shown that a good bacteria kill results if battery temperatures can be maintained at 70° C. throughout, but that a temperature of 80° C. is necessary to obtain complete destruction. The addition of formaldehyde to the battery has proven effective, but in some cases large amounts are required, and such practice can be costly.

It is a well known fact that there are a great many locations within the continuous diffuser where cossettes can accumulate and lie stagnant. This held up material undoubtedly becomes an ideal breeding place for bacteria and a source of inoculation of the juice. We have found the concentration of lactic acid in some of these held up materials to be more than 2,000 ppm. as compared with about 50 ppm. in the original cossettes. Elimination of these stagnant areas is a most formidable task, yet it is felt that accomplishment of this will yield the most benefit in the control of fermentation in diffusion.

We list the following steps as necessary to control completely bacterial action in the continuous diffuser:

1. Elimination of stagnant cossettes.
2. Maintenance of battery temperature as high as possible consistent with satisfactory pressability of pulp and quality of raw juice.
3. Treatment of beets with an efficient germicide prior to slicing. Such treatment might well be advantageous, especially early in campaign, even if stagnant areas cannot be eliminated. Cost must necessarily be a factor in the choice of the proper germicide.
4. Addition of germicide to the battery stream when necessary, but only when other measures are not completely effective.

Summary

Data are given which show the lactic acid concentrations found in continuous and cell-type diffusers operating under widely different conditions. A determination is given of the ratio of sugar lost to lactic produced under arbitrary experimental conditions. Treatment of beets prior to slicing was shown to be promising in conjunction with other measures of control of fermentation in diffusion.

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