A PRELIMINARY REPORT ON THE EFFECT OF TEMPERATURE AND BEET CONDITIONS ON RESPIRATION AND LOSS OF SUGAR FROM BEETS IN STORAGE*

C. Guinn Barr, Colorado Agricultural Experiment Station E. M. Mervine, Bureau of Agricultural Chemistry and Engineering, U. S. Department of Agriculture

and

R. A. Bice, California Agricultural Experiment Station

*Material and financial support incident to this investigation were provided by the Mechanical Engineering Section of the Colorado Agricultural Experiment Station, the U.S. Beet Sugar Association and the Bureau of Agricultural Chemistry and Engineering, U.S. Department of Agriculture.

Facilities and personnel for the laboratory studies were contributed by the Botany and Plant Pathology Section of the Colorado Experiment Station.

Analyses for sugar content were made by the laboratory staff at the Fort Collins field station of the Division of Sugar Plant Investigations, Bureau of Plant Industry, U.S.D.A.

Introduction

Mechanical methods of thinning and harvesting sugar beets, as contrasted with the regular hand methods may result in more beets of smaller size, in dirtier beets (that is, beets with more leaves and petioles clinging to them), and in a larger number of mechanically cut and bruised beets. In the experiments discussed in this paper, an attempt was made to determine the differences in sugar loss and keeping qualities of beets during storage which may be attributed to these conditions.

Although no significant differences were found in the respiration rates of different sizes and conditions of beets in these experiments, it cannot be safely assumed that no differences actually exist. It is possible that under more severe conditions of test or if more samples were used, significant differences would have been obtained.

Respiration is a biological-chemical process accompanied by the evolution of carbon dioxide and the liberation of energy. The most frequent type of respiration, involving the absorption of atmospheric oxygen, is termed aerobic respiration. Anaerobic respiration occurs in the absence of a supply of atmospheric oxygen. Two of the end products in either case are carbon dioxide and energy in the form of heat.

It is commonly assumed that hexose sugars $(C_6H_{12}O_6)$ are the substances consumed during respiration. Sucrose $(C_{12}H_{22}O_{11})$ is the principal sugar in sugar beets but sucrose is hydrolyzed by the enzyme invertase or sucrase yielding a molecule each of glucose and fructose, both of which are hexose sugars. With theserobic breakdown of hexose, therefore, the carbon dioxide produced may be collected, measured and calculated in terms of sucrose, which is the potential source of respirable material. Thus the rate at which carbon dioxide is liberated becomes an index of the rate of sugar loss. If carbon dioxide liberation is at a uniform rate, it is probably due chiefly to aerobic respiration. If, however, the rate of liberation suddenly and rapidly increases; it is doubtless due to fermentation or rotting caused by the invasion of bacteria or fungi as well as to the respiration of the organisms. Certain complications arise in this connection which will be discussed later.

Several factors operate to control the rate of respiration, one of which is temperature. Respiration rates are uniformly accelerated by rising temperatures over a limited range. Relatively high temperatures favor bacterial and fungal activity which induce rotting and fermentation. Owing to these influences, marked increase in the respiration rate was observed at temperatures of 75°F. and over.

The first practical step in the reduction of storage losses, therefore, appears to be that of keeping the temperature low. By doing so, natural respiration is kept low and there is less rotting. Two conditions affect pile temperature--weather and the heat given off by respiration. Both of these are affected by the circulation of air through the pile. When a high wind occurs, the pile temperature responds quickly to outside temperature due to increased On the other hand, the effect of outside temperature on pile circulation. temperature may be slight if there is little circulation. In this case, respiration probably liberates sufficient heat, even at low temperatures, to raise the pile temperature a measurable amount during a twenty-four hour period. Then, as the temperature rises, the respiration rate increases. When a sufficiently high temperature is attained, rotting starts, respiration increases, and a "hot spot" is the result. In order to reduce sugar loss, therefore, it appears that the pile should be well ventilated and be kept as cool as possible without freezing.

The experiments discussed herein are divided into two main groups: (I) Storage Pile Experiments; and (II) Laboratory Experiments.

I. Storage Pile Experiments

A. Purpose

There were three things to be observed in this division of the experiments: The first observation was the relative sugar loss and keeping qualities of four types of beets placed in the storage pile. The second was the temperature fluctuations in the storage pile at various points, leading to information regarding the rate of air movement in the pile, the correlation between external conditions and pile temperature, and the effect on pile temperature of size and condition of beets. The third was to determine the carbon dioxide content of the atmosphere in the pile, to find out if carbon dioxide accumulated in the atmosphere surrounding the four different types of beets.

B. Apparatus.

The apparatus consisted of twenty wood-and-wire-mesh cages, each having a capacity for approximately one ton of beets; thirty-six thermocouples and leads connected with a potentiometer; and ten one-eighth-inch metal pipes, thirty feet long, equipped with guards at one end, to sample the atmosphere for carbon dioxide in ten of the cages.

C. Procedure

Four classes of beets were arbitrarily established:

(1) Standard, comprising 90 percent (by number) over 3" in diameter and 10 percent between 3" and 14". (2) Small, consisting of 50 percent over $3^{"}$ and 50 percent between $3^{"}$ and $1\frac{1}{4}^{"}$.

(3) Dirty, composed of standard sized beets 25 percent of which were topped high. Each high-topped beet was accompanied by one leaf.

(4) Bruised - Standard sized beets which were dropped 30" onto a corrugated iron surface.

Twenty samples - five each of the four different classes - were counted and weighed in and out of storage. They were placed in cages and set at randon in five replicates in the pile. Representative sugar and dry weight samples were taken of each type before and after storage in order that the quantity of sugar lost during the storage period might be estimated on the dry weight basis. This is necessary to prevent confusion of moisture loss with sugar loss.

One thermocouple was placed in each of the twenty samples and sixteen were placed in a pile cross section. (See Fig. 1, p. 55.) The temperatures of all thermocouples were taken daily. In addition the cross section temperatures were taken hourly for one twenty-four hour period and one forty-eight hour period.

A record of the outside temperature over the period of storage was obtained from the recording thermometer of the Great Western Sugar Company factory near the pile.

A wind record over the storage period was obtained from the Meterrological Laboratory of the Colorado Agricultural Experiment Station located on the Colorado State College campus about two miles from the pile.

The pipes for carbon dioxide collection led from the first ten cages to the outside of the pile. From these, carbon dioxide analyses were made twice during the storage period.

D. Results.

Among the four classes of beets studied, no significant differences were found in sugar losses or in shrinkage. The shrinkage averaged 7.9 percent for the storage period of 47 days.

The sugar lbss of the samples in the storage pile was determined by analyzing representative samples of beets at the beginning and at the end of the storage period. The errors involved in determining the sugar loss are such that an accurate measure of sugar loss cannot be made, but within the limits of the statistically standard error the sugar loss was not greater than the .06 pound per ton of beets per day of storage as determined from the respiration curves.

The temperature results are plotted on the graphs of Figures 2 and 3.

Circulation of air through the pile apparently did not cause the temperature to fluctuate with the change in outside temperature, unless the temperature remained at the changed level for an extended time or unless the wind was blowing.





THERMOCOUPLE TEMPERATURES IN CROSS SECTION OF STORAGE PILE, 1939 (without switch)

4)) (9 L. 9

8 10 KT 15

EFFECT OF WIND VELOCITY AND VERAGE OUTSIDE TEMPERATURE ON PILE TEMPERATURE

3 3

0 11

5

E 16



Fig. 4

Referring to Fig. 3, it appears that outside temperature fluctuations when accompanied by high wind velocities had rapid effect on pile temperatures. On the 28th and 29th of October an outside temperature drop accompanied by wind resulted in a drop in the pile temperature of about 8° F.

There was no precipitation over the storage period, so that the effect of rain or snow on the pile temperature was not a factor to be considered. No significant temperature difference among the four classes of beet samples was observed. There was no carbon dioxide accumulation in the pile that could be detected with the instruments used.

II. Laboratory Experiments

A. Purpose.

This set of experiments had a two-fold purpose: First, to obtain a temperature-respiration curve and a temperature - sugar-loss curve, and second, to determine the sugar loss and relative respiration rates as influenced by different types of beets in storage.

B. Apparatus.

Thirty-four respiration chambers were built in the following manner:

A fifty-five gallon metal drum was equipped with a demountable top. It was covered with one or two thicknesses of $1/2^{\mu}$ balsam wool insulation, the amount depending on the temperature gradient it was to maintain.

Two lengths of one-eighth-inch metal pipe were welded into the top, one reaching to within two inches of the bottom of the drum, and one extending about three inches below the top. Each pipe extended about six inches above the top for attaching the unit to the aspiration train.

A one hundred-watt heating element was placed in the bottom of the drum. The temperature was controlled by means of a bimetal thermostat acting as a switch in the heating circuit. A small seven-watt signal bulb was placed on the outside of the drum to indicate whether or not the heating element was functioning. Inside the drum and over the heating element was placed a tripod stand fitted with a heat deflector which supported a wire basket of about thirty pounds capacity. The top was sealed by means of a glycerin soaked sponge rubber gasket and a compression clamp. A thermometer was placed in a hole in the center top of the drum.

To free the intake air of carbon dioxide, two bubbler bottles ordinarily were used. One contained two-normal sodium hydroxide solution. Connected in series between this and the respiration unit was a bubbler bottle containing barium hydroxide solution which served as a check and indicator to insure complete removal of all carbon dioxide from the air entering the respiration chamber. The drums were connected in parallel to a line of reduced pressure and aerated with carbon dioxide-free air throughout the period of the experiment. The carbon dioxide produced by respiration of the beets for a twenty-four-hour period was collected at regular intervals. Absorption of carbon dioxide was accomplished by bubbling the exhaust air through standard one-normal sodium-hydroxide solution. Either three or four sodium hydroxide bubblers were used, the number depending on the temperature of the sample. The quantity of carbon dioxide evolved by the respiring sample was determined by differential titration. Test solutions of standard barium hydroxide were used in all cases to insure complete absorption. Screw clamps were used to adjust the aeration to an approximate uniform rate so that the air volume of the units was changed once in about every six hours.

C. Procedure.

For the laboratory studies, the beets were washed free of soil and allowed to dry until the surface moisture was completely evaporated. The samples were weighed to check within = 0.20 pound and, after counting the number of beets, were placed in the baskets in the drums.

A total of thirty-four respiration units were set up and experiments were performed on the following beet samples:

Standard beets (as defined previously) held at 35°, 45°, 55°, 65°, 75°, 85°, 95° F.; small bruised and dirty beets at 45° F.; small bruised and dirty beets at 65°F.; low topped beets, standard beets treated and bruised beets treated at 65°F. The treatment consisted of dipping in "Dowwax." Temperature control was facilitated by holding the lower temperature units in a cold storage room, while the higher ones were set up in the basement of one of the college buildings.

These samples were all run in duplicate except the standard beets at 65° for which four samples were used. Carbon dioxide samples were collected from the lots held at 75°, 85° and 95° twice weekly and from all the others once a week.

At the beginning of the experiment representative samples of unstored beets were analyzed for sugar content and calculations were made on the dry weight basis. At the end of the storage period sugar analyses were again made on the stored beets and the loss in pounds of sugar per pound of dry matter was calculated. For obvious reasons this was necessary since the loss in fresh weight during storage is due largely to loss in moisture content.

Relative respiration rates by the measurement of carbon dioxide produced were determined, and the calculations were made to grams of carbon dioxide per pound of fresh material per day.

D. Results.

Theoretical sugar loss, as sucrose, was calculated from the equation $C_{12H_{22}O_{11}} + 12O_2 \longrightarrow 12 CO_2 + 11H_2$. Hence from the value of carbon dioxide obtained the quantity of sugar used could be determined by a simple calculation, if we assume that all the carbon dioxide was liberated in aerobic respiration. It is probable, however, that considerable anaerobic respiration or fermentation took place, in which case the breakdown of sucrose might yield moderate quantities of carbon dioxide together with alcohol, organic acids, etc., instead of carbon dioxide and water. The probable reaction is as follows:

 $\begin{array}{r} \begin{array}{r} \underline{Hydrolysis} \\ C_{12}\underline{H}_{22}O_{11} \\ C_{6}\underline{H}_{12}O_{6} \\ (glucose) \\ \underline{C}_{6}\underline{H}_{12}O_{6} \\ \underline{C}_{6}\underline{$

-59 -

In the destruction of one mol of sucrose (342 gms) by aerobic respiration, 12 mols (528 gms.) of carbon dioxide would be produced, while by anaerobic respiration one mol of sucrose would yield only four mols of carbon dioxide or 176 grams. Consequently, by assuming that all the carbon dioxide produced resulted from aerobic respiration only, the apparent sugar loss would be about 33 per cent of the value obtained if all the carbon dioxide resulted from anaerobic respiration alone. It should be kept in mind that the alcohol, etc., produced in anaerobic respiration probably would not be oxidized to carbon dioxide and water.

Since the amount of oxygen consumed in these experiments was not measured, no respiratory quotients could be calculated to determine the type of respiration in progress. The carbon dioxide which was measured represents that produced by both aerobic and anaerobic respiration and no attempt was made to evaluate each one separately. It is clear, therefore, that the sugar loss, if calculated on the basis of the carbon dioxide produced by aerobic respiration, would necessarily be something less than the actual loss as measured by sugar analysis. This was especially true at the higher temperature where anaerobic respiration, fermentation and rotting were doubtless more rapid.

The results of the respiration studies are shown in Tables 1 and 2 and Figs. 4 and 5. The data showed no significant difference in the respiration rate of the various types of beets used. There was, however, a close correlation between temperature and respiration as shown by the standard beets.

The data in Table 1 show the effect of temperature on the respiration rate of standard beets (1) in terms of carbon dioxide (grams) per pound fresh weight per 24 hours, (2) the loss of sugar (pounds per ton fresh weight) per 24 hours as calculated from the carbon dioxide produced, and (3) the loss of sugar as determined by analyses. The data given in this table are presented graphically in Figure 4.

There was a gradual increase in the respiration rate with increasing temperature up to 18.5°C. (65°F.), as shown by the carbon dioxide values, (A, Fig. 4). As the temperature increased above 18.5°C., the rate of carbon dioxide production was rapidly accelerated. This rapid increase at the higher temperatures suggests invasion by fungi, resulting in decomposition.

The values of the sugar lost in respiration and rotting at the higher temperatures, if calculated from the carbon dioxide produced, are misleading since the loss of sugar is actually greater than seems apparent from the carbon dioxide values. This is clearly shown by the curves in Fig. 4. At the lower temperatures the sugar losses, as calculated from carbon dioxide produced and as determined by analyses, have a common difference but that difference increases as the temperature rises. Hence the differences between the curves probably represent anaerobic respiration.

Rot caused by fungi started at the highest temperature (35°C.) within twelve days after the start of the experiment. At 29.5°C. rotting and fungal action was delayed 14 days and at 24°C. this was delayed still another 14 days.

Tests for carbon dioxide from the samples in the large pile failed to show accumulation of the gas, which suggests unexpectedly rapid natural aeration.





Effect of Temperature (Degrees C.) on Respiration Rate of Sugar Beets in Storage. (Ave. for 47 days).

- A. Respiration expressed in grams of carbon dioxide liberated per pound of fresh material per 24 hours. Values shown at right of figure.
- B. Respiration calculated from carbon dioxide liberated in terms of pounds of sugar lost per ton of beets per 24 hours. Values shown at left of figure.
- C. Respiration in pounds of sugar lost per ton of beets per 24 hours. Values shown at left of figure.

Temperature	Carbon Dioxide(1)	Sugar loss calculated Carbon dioxide (2)	from Sugar loss by analysis (3)
2-3°C. (35°F.)	0.039	0.113	0.230
7°C. (45°F.)	.065	.166	.269
12.8°C. (55°F.)	.078	.224	.384
18.5°C.(65°F.)	.142	.405	.514
24°C.(75°F.)	.235	.669	.882
29.5°C.(85°F.)	.361	1.032	1.610
35°C. (95°F.)	.625	1.783	2,610

Table 1.--Effect of Temperature on Respiration Rate of Beets in Storage*. (Average for 47 days.)

(1) Grams carbon dioxide liberated per pound of beets (fresh weight) per 24 hours. (See curve A of Fig. 4).

(2) Sugar loss calculated from carbon dioxide in pounds of sugar per ton of beets (fresh weight) per 24 hours. (See curve B of Fig. 4)

(3) Sugar loss (by analysis) in pounds of sugar per ton of beets (fresh weight) per 24 hours. (See curve C of Fig. 4).

*Data based on average of duplicate 30-beet samples in all cases.

In Table 2 and Figure 5 are presented data on the effect of time of storage on the respiration rates of standard beets. In the figure the rate of sugar loss is plotted against time of storage.



Fig. 5

Effect of Time of Storage from October 16 to November 27 at Various Temperatures on Respiration Rate of Beets.

(Respiration expressed in grams of carbon dioxide liberated per pound of fresh beets per 24 hours).

Table 2.---The Effect of Time of Storage on Respiration Rates of Beets. (Results expressed in grams of carbon dioxide liberated per pound of beets (fresh weight) per 24 hours. Average of Standard 30-beet samples made in duplicate in all cases.)

Temperature	Date Samples									,	
	10/16	10/23	11/6	11/13	11/20	11/27					
2-3°C.(35°F.) 7°C.(45°F.) 12.8°C.(55°F.)	•054 •056 •070	.027 .062 .058	•042 •057 •098	.024 .063 .078	•049 •051 •086	.041 .098 .079		- 6.1	and a second		
18.5°C.	10/16	10/19	10/23	10/26	10/30	11/2	11/9	11/13	11/16	11/20	11/23
(65°F.) 24°C.(75°F.) 29.5°C.(85°F.) 35°C.(95°F.)	.155 .182 .300 .262	.112 .160 .262 .263	.157 .180 .475	•152 •244 •246 •569	.134 .255	.089 .278 .275	•258 •672	.230	.222	,202	•528

Temperature readings showed no fluctuations within the pile. The temperatures remained uniformly low, around 3° to 7°C.

Isolations from the interior of the rotten tap roots yielded colonies of Rhizopus nigricans. This organism is not pathogenic to beets at ordinary temperatures.

Calculations indicate that the air, by natural circulation under the conditions of this experiment, moved through the pile at the rate of 16 feet a minute.

Since the results obtained in these experiments indicate a sugar loss lower than was expected, and since natural circulation is apparently faster than was suspected, the feasibility of blowing low temperature night air into the pile to cool it merits further investigation.

In order to determine the keeping qualities of beets under more extreme conditions of size, dirtiness, injury and weather, more investigation under controlled conditions will be necessary.

Summary

At 18.5°C. (45°F.) the standard, bruised, dirty and small beets showed no significant difference in respiration or loss in sugar content. At temperatures below 24°C. the standard beets showed a close relationship between temperature and respiration rate or sugar loss. At temperatures of 24°°C. and higher, the rate of respiration increased more rapidly in proportion to the temperature rise than at the lower temperatures. This behavior was in close agreement with the condition of the beets at the various temperatures, since observations showed that the lower temperatures retarded or inhibited mold and rot, while rapid decomposition took place at the higher temperatures.

Under the conditions which obtained in this experiment, it may be concluded that wounding, or the presence of reasonable quantities of foreign material mixed with sugar beets, caused no important change in the rate of respiration or sugar loss.

RESULTS OF FIELD TRIALS OF BORON AND TREATED SEED IN THE GREAT LAKES COMPANY TERRITORY

M. W. Sergeant1/

(a) Field Trials of Boron

General Notes

Beets are among the high boron containing plants, according to the French investigator, G. Bertrand.

When boron is known to be needed, about 20 pounds of borax per acre is a good general recommendation. Since all boron is not used by the first crop subsequent applications of 5 to 8 pounds per acre will suffice when beets are

1/ Great Lakes Sugar Company