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EFFECT OF VARIETY, NITROGEN FERTILIZATION
AND STORAGE TEMPERATURE ON THE CHANGES
IN MARC, SUCROSE AND DRY MATTER CONTENT
OF SUGARBEETS DURING STORAGE

Thesis for the Degree of M. S.
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ROGER E. WYSE
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ABSTRACT

EFFECT OF VARIETY, NITROGEN FERTILIZATION AND STORAGE TEMPERATURE ON THE CHANGES IN MARC, SUCROSE AND DRY MATTER CONTENT OF SUGARBEETS DURING STORAGE

by Roger E. Wyse

Three sugarbeet varieties grown with 150 and 75 lbs. of nitrogen per acre were stored for 136 days at 35 and 45 F. A supplementary experiment involved seven varieties. The differences between varieties in degree of marc decomposition, dry matter and sucrose losses and the changes in clear juice purity were determined.

At the time of harvest there was a significant difference between varieties in percent marc. Nitrogen fertilization had no effect on the marc and sucrose contents or on the clear juice purity. Beets grown under high nitrogen (150#/A) and stored cold (35 F.) were generally higher in percent of original marc over the entire storage period. The variety x treatment interaction was highly significant for both storage temperature and nitrogen fertilization.

There appeared to be no correlation between the increased unaccounted for impurities in the clear juice and increased impurities in the diffusion juice due to marc solubilization.

Beets grown on high levels of nitrogen and stored cold retained the highest percent of original extractable sugar per ton.

The decrease in dry matter was greater than the decrease in sucrose over the entire 136 day storage period but particularly in the first 93 days indicating that a significant amount of non-sucrose substances were being respired. This may account in part for the increase in clear juice purity which is common in the early stages of storage.

A substantial difference was found between the seven experimental varieties in the solubilization of marc during storage. There was also a significant difference in resistance to rot, mold and sprouting between the seven varieties.

Solubilization of marc during storage is of interest because it increases the pectin content of the diffusion juice and makes filtration of first carbonation juice difficult.

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CONTENT OF SUGARBEETS
DURING STORAGE

By

Roger E. Wyse

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TABLE OF CONTENTS

| | Page |
|---|------|
| ACKNOWLEDGMENTS | ii |
| LIST OF TABLES | v |
| LIST OF ILLUSTRATIONS | vi |
| INTRODUCTION | 1 |
| LITERATURE REVIEW | 3 |
| Sources of Impurities | 3 |
| Breakdown of Marc | 3 |
| Pectic Substances as Sources of Impurities | 3 |
| Changes in the Pectic Substances | |
| During Storage. | 4 |
| Effect of Pectic Substances on Processing | 5 |
| Inversion of Sucrose as a Source of Impurities. | 6 |
| Sucrose Losses | 6 |
| Sucrose Losses Due to Respiration | 6 |
| Sucrose Losses Due to a Decrease in Purity | 8 |
| MATERIALS AND METHODS | 10 |
| Analysis | 11 |
| Marc Determination | 11 |
| RESULTS AND DISCUSSION. | 13 |
| Variables in Marc Determination. | 13 |
| Extraction Temperatures | 13 |
| Length of Extraction Time. | 14 |
| Effect of Nitrogen Fertilization and Variety | |
| on Percent Marc at Harvest | 17 |
| Variety X Nitrogen Experiment | 17 |
| Experimental Varieties. | 17 |
| Results of Beet Storage | 19 |
| Changes in Percent Marc During Storage | 20 |
| Effect of Nitrogen Fertilization | 20 |
| Effect of Storage Temperature | 23 |
| Effect of Variety | 23 |
| Increase in Impurities in the Diffusion | |
| Juice Due to Marc Decomposition. | 29 |

| | Page |
|--|------|
| Relation of Increased Impurities in Diffusion Juice to Unaccounted for Impurities in Clear Juice | 32 |
| Relationship Between the Changes in Dry Matter, Sucrose and Marc During Storage. | 34 |
| Changes in Extractable Sugar During Storage. | 34 |
| Evaluation of the Method of Analysis | 39 |
| SUMMARY AND CONCLUSIONS | 40 |
| BIBLIOGRAPHY | 43 |
| APPENDIX | 46 |

LIST OF TABLES

| Table | | Page |
|-------|---|------|
| 1 | Effect of Extraction Temperature on Percent Marc | 13 |
| 2 | Effect of Varying the Length of Bath Treatment and Extraction Time on Percent Marc | 15 |
| 3 | Percent Marc of Three Varieties at Harvest Adjusted to 25% Dry Matter. | 18 |
| 4 | Percent Marc of Eight Experimental Varieties at Harvest Adjusted to 25% Dry Matter | 19 |
| 5 | Changes in Marc and Dry Matter Content During 140 Days of Storage in Seven Experimental Varieties | 26 |
| 6 | Impurities Added to the Diffusion Juice by Marc Decomposition Over the 136 Day Storage Period. | 31 |
| 7 | The Relationship Between the Impurities Added to the Diffusion Juice by Marc Decomposition and the Increase in Impurities in the Clear Juice After 136 Days of Storage | 33 |
| 8 | Evaluation of Varieties for Storing Ability Using the Concept of Extractable Sugar Per Ton of Beets | 36 |
| 9 | Evaluation of Nitrogen Fertilization and Storage Temperature Using the Concept of Extractable Sugar Per Ton | 38 |

LIST OF ILLUSTRATIONS

| Figure | Page |
|--|------|
| 1 Effect of Treatment Time on Percent Marc. | 16 |
| 2 Effect of Nitrogen Fertilization on Marc Breakdown of Three Varieties During Storage | 21 |
| 3 Effect of Nitrogen Fertilization on Marc Breakdown of Three Individual Varieties | 22 |
| 4 Effect of Storage Temperature on Marc Decomposition of Three Varieties During Storage | 24 |
| 5 Effect of Storage Temperature on Marc Decomposition of Three Individual Varieties | 25 |
| 6 Breakdown of Marc in Three Varieties Over a 136 Day Storage Period. | 26 |
| 7 Effect of Storage on the Breakdown of Marc in all Storage Treatments | 28 |
| 8 Relationship Between Percent Marc at Harvest and Loss of Marc During Storage in Seven Experimental Varieties | 30 |
| 9 Changes in Percent Dry Matter, Percent Sucrose and Percent Marc of Three Varieties During 136 Days of Storage | 35 |

INTRODUCTION

In the past twenty years improved technology has substantially increased the yield of sugarbeets. This coupled with larger acreages and faster mechanical harvest has made it necessary for sugar factories in the United States to lengthen the processing campaign. A longer campaign requires the storage for extended periods of four to five months of a large percentage of the beets delivered.

At the time of harvest the sugarbeet is at its peak in processability, but under present storage conditions the quality decreases rapidly after two or three months storage. The primary effects of storage are (1) a substantial decrease in sucrose, (2) an increase in soluble impurities in the thin juice, (3) an increase in color and lime salts in the thin juice, and (4) an increase in soluble pectic substances in the diffusion juice.

The loss in sucrose can be reduced by lowering pile temperatures thus reducing respirational losses. The increase in impurities increases the production of molasses: an undesirable but tolerable effect. The effect of increased lime salts can be counterbalanced by adding soda ash. The increase in pectins however reduces factory efficiency by substantially reducing filtration and diffuser flow rates.

Previous research on the effects of storage on the soluble pectin content and subsequent decrease in marc on various root and fruit crops has been extensive. In general the results have shown a substantial increase in soluble pectins particularly when stored at warm temperatures. The effect of storage on the breakdown of the insoluble cell wall materials and the subsequent increase in impurities in the diffusion juice in the sugarbeet has yet to be ascertained.

With the removal of the leaves, petioles, and crown at harvest all accumulation of sucrose and minerals within the sugarbeet root is halted. The composition at this time, therefore, serves as a basis for all subsequent metabolic changes in storage.

The important changes which occur in the sugarbeet during storage are those which affect the processability, decrease the recoverable sugar, and cause a solubilization of the marc. These changes are primarily due to respiration and other catabolic processes which increase the amount of soluble impurities and particularly pectic substances, and reduce the sugar content of the beet.

This study was aimed at determining the breakdown of the insoluble cell wall materials and the changes occurring in sucrose and dry matter as affected by variety, cultural practices and storage condition.

REVIEW OF LITERATURE

Sources of Impurities

Breakdown of Marc.--The sugarbeet marc is the insoluble residue which remains after extraction of the beet with water (McGinnis, 1951). The marc is composed primarily of cellulose (~25%), hemicellulose (~25%), and pectic substances (~50%). The pectic substances are stable only in cold water and swell and become soluble in hot water (Silin, 1964).

The marc averages approximately 5% of the fresh weight of the beet (Silin, 1964). In a test of fifteen varieties, however, Owens et al. (1954) found the marc content to vary from 3-6%.

Pectic Substances as Sources of Impurities.--The pectic substances are high molecular weight polymers, derivatives of pectic acid, and made up of galacturonic acid residues. In pectic acid, the carboxyls are free and can readily react with calcium to form insoluble calcium pectate. In pectin and protopectin the carboxyls are esterified with methanol (Bonner and Galston, 1952). In the sugarbeet the free carboxyls may be as high as 50% methylated (Goodban and McCready, 1965). From 6% (Kertesz, 1951)

to 30% (Goodban and McCready, 1965) of the hydroxyls in the two and three positions may be acetylated.

The pectic substances are in two forms: water soluble and water insoluble. The water insoluble form is protopectin, the structure of which is unknown, but which is thought to exist in a matrix complex of pectin, cellulose and hemicellulose. Protopectin upon hydrolysis yields the water soluble pectic acid (Kertesz, 1951 and Joslyn, 1962).

The breakdown of protopectin by water is highly temperature dependent. Protopectin when treated with hot water swells and gradually dissolves in the form of pectin (Silin, 1964 and Owen, 1955). Silin (1931) extracted dried beet pulp and found only a slight increase in solubility up to 80 C., however, the amount of pectin extracted at 90 C. was 30 times greater than that at 80 C.

The soluble forms of the pectic substances are pectin, pectic and pectinic acids (Kertesz, 1951). Pectic and pectinic acids have insoluble calcium salts, but are highly esterified and yield methanol and acetic acid salts when heated in alkali solution.

Changes in the Pectic Substances During Storage.--The soluble pectins after an initial increase in the early stages of growth, decrease steadily until harvest. The amount of total pectic substances is decreased by a moisture deficiency. Fertilizer has no significant effect (Gaponenkov, 1943).

Silin reports that microbial activity and sprouting increase the quantity of soluble pectins during storage. Walker (1960) found no significant change in the pectin content over a 48 day storage period when beets remained in good condition, but when stored for 161 days at 34 F., the pectin content decreased one-third.

If freezing occurs during storage the pectin, which in a normal healthy beet is a large macromolecule, will be decomposed by enzyme action to a colloidal size (Claassen, 1943).

Effect of Pectic Substances on Processing.--The first noticeable effect of pectic substances in the factory is in the diffuser. Beets which have been exposed to conditions which weaken the protopectin stability must be diffused at lower temperatures to prevent the cosettes from losing their resilience. Loss of resilience will prevent the free flow of water through the diffuser. For frozen or spoiled beets the temperature should not be allowed above 70-75 C. Silin (1964) found a 110% increase in pectic substances in diffusion juices by increasing the temperature from 75.5 C. to 85.1 C.

During defecation the pectinic acid present in the diffusion juice is de-esterified, splitting off methanol and acetic acid. The acetic acid forms soluble calcium acetate and the polygalacturonic acid residue forms an insoluble calcium pectate (Silin, 1964 and Goodban and McCready, 1965).

Inversion of Sucrose as a Source of Impurities.--

Sucrose can be respired to CO_2 or hydrolyzed by invertase to glucose and fructose. Inversion not only increases the amount of invert sugar but glucose and fructose may be fermented to lactic acid in the diffuser or converted to acids in the defecation process.

During the defecation process up to 98% of the invert will decompose if held at 80 C. for 20 minutes in 2% lime (Silin, 1964). The decomposition products are acids most of which give soluble calcium salts on addition of lime.

Sucrose Losses

Sugar losses in storage can occur in three primary ways: directly through respiration and inversion, and indirectly through an increase in C. J. P.* thus reducing the recoverable sugar.

Sugar Losses Due to Respiration and Inversion.--Loss of sucrose due to respiration can be substantial particularly at high storage temperatures (Silin, 1964 and McGinnis, 1951).

Barr et al. (1940) determined the loss of sugar due to respiration at various temperatures over a 47 day storage period and found that the amount of CO_2 evolved accounted for approximately 60% of the total apparent sucrose losses.

*Clear Juice Purity

The percentage accounted for by CO_2 evolution, however, was not constant but varied with temperature.

Stout (1950) stored beets in large drums at 68-70 F. for 45 days and found approximately 80% of the sucrose loss was due to respiration and 20% was due to inversion.

Poor circulation of air through stored beets can cause the build-up of high concentrations of CO_2 thus causing anaerobic respiration to take place. Anaerobic respiration not only reduces the sugar content but greatly increases the amount of impurities produced (Silin, 1964). Dexter (1965) stored beets under water (anaerobically) and in air (aerobically) at 32 F. and found a decrease of 45 lbs. of extractable sugar per ton due to an increase in impurities of 3,955 mg/100S after seven weeks of storage under anaerobic conditions.

A healthy beet root has a natural immunity against invasion by saprophytic organisms (Silin, 1964). Freezing, wilting and bruising, however, will allow invasion of rot and mold organisms. Drought and wilting conditions prior to harvest may also affect the incidence of rotting in storage (Gaskill, 1950a, 1950c, and Larmer, 1937). At high storage temperatures rot and mold will greatly increase respiration losses (Barr, 1940, and Stout, 1950).

Little research has been published where an evaluation of new varieties on the basis of their storing ability has been made (Nelson, 1950 and Gaskill, 1950b). However, Smith (1962) noted a substantial difference between varieties in

resistance to rot when mother beets were stored in polyethylene bags at 38-40 F.

Sugar Losses Due to a Decrease in Purity.--The decrease in recoverable sugar during storage is not solely a result of sucrose lost as CO_2 , but is also due to an increase in impurities in the thin juice (McGinnis, 1951; Silin, 1964; Carruthers, 1962; and Dexter, 1965). Even if the impurities did not increase, a decrease in sucrose in the clear juice would give a lower purity. In the table prepared by Dexter (1965) from the formula proposed by Great Western Research Lab (Sugar Beet Res., 1964) it can be seen that a 1% decrease in clear juice purity will cause approximately a 6 lb. or 2% decrease in extractable sugar per ton of beets, percentage sucrose remaining the same.

In an intensive examination of compositional changes in diffusion juice during storage, Walker (1960) found a decrease in purity from 92.2% to 87.2% in 90 days at 50 F. The apparent impurities calculated from TJP increased from 8,500 to 14,700 mg/100S. The only significant compositional change found, however, was an increase in invert at the expense of sucrose with no loss in total sugars. The 6042 mg/100S increase in invert was approximately equal to the 6200 mg/100S increase in impurities. These results seem very different from those of Barr (1940) and Stout (1950) who accounted for 60 to 80% of their extensive sugar losses by CO_2 evolved and only 20% by inversion.

Carruthers (1962) derived the relationship of $(3.5 \times \text{Na}) + (2.5 \times \text{K}) + (10 \times \text{NH}_2)$ which accounted for a high percent of the total impurities in fresh beets. Dexter et al. (1966) using Carruther's factors for Na, K and αNH_2 in the clear juice of stored beets found that as the apparent impurities increased particularly at warm temperatures the percent of impurities unaccounted for by Na, K and αNH_2 decreased sharply. If this relationship is applied to the data presented by Walker the same trend is noted. Dexter proposed that these unaccounted for impurities may be weak acids with soluble calcium salts formed in the decomposition of sugars and/or cell wall constituents.

MATERIALS AND METHODS

Three varieties of sugarbeets from a variety nitrogen yield experiment conducted by the Monitor Sugar Co., were stored for 136 days at 32 and 45 F. The three varieties (Commercial, 63-194, and 401) had been planted in 28" rows with nitrogen fertilization rates of high (150#/A) and low (75#/A). The average yields for the three varieties were: Commercial, 23 T/A; 63-194, 18.5 T/A; and 401, 19 T/A.

All beets were hand dug and topped by lightly scraping off the petioles and then cutting off the main bud down to a diameter of approximately two inches. Immediately after harvest the beets were bulked by treatment and divided into storage samples of ten beets each. All samples were hand picked to contain only beets of uniform size and to remove broken and diseased beets. The samples were then weighed and placed in mesh bags inside of canvas bags and placed in two walk-in refrigerators at 32-35 F. and 45 F. All samples placed in storage were replicated three times.

In order to further study the variations between varieties in storage based on marc decomposition, a supplementary experiment was conducted. Eight experimental varieties from a variety test conducted by Dr. George Hogaboam were stored for 140 days at 45 F. All samples were machine

harvested and badly bruised when placed in storage. In an attempt to prevent rotting the samples were dipped in a 10% Clorox solution prior to storage. Each sample was placed in a plastic bag. Approximately ten samples were then placed in a large canvas bag for storage.

Analysis

All samples were analyzed for sucrose*, clear juice purity, Na, K, α amino nitrogen, marc and dry matter immediately after harvest and at three removal dates approximately 40 days apart.

Marc Determination.--Brei samples were taken at the time of sugar analysis and immediately frozen for marc analysis later. For analysis samples were thawed at room temperature and 50 gms of brei weighed into a tared 250 ml beaker and dried at 75 C. for 36 hours in a forced air oven. After 36 hours the samples were reweighed to determine the dry matter content. At the same time that the samples were being weighed for drying, two piece buchner funnels were prepared with a 1/4" cellulose filter pad. The filter pads were prepared by pouring a thick slurry of powdered cellulose into the funnels which were then pulled down under vacuum and washed with approximately 250 ml of water. The filters were then dried with the samples for 36 hours and weighed.

*Apparent sucrose, polarimetric

One hundred seventy-five ml of distilled water at 80 C. was added to the dried brei in each beaker. The beakers were then placed in an 80 C. \pm 0.2 C. water bath for thirty minutes. Samples were stirred several times during the thirty minute hot water treatment. After the bath treatment the samples were poured into the tared two piece buckner funnels. The samples were then extracted with 1000 ml of 80 C. \pm 3 C. distilled water added in 200 ml aliquots. The filter was pulled to dryness after the addition of each aliquot. Total extraction time including bath treatment was 45 minutes. The funnels were then dried for 24 hours at 75 C., reweighed and the percent marc calculated.

All values reported for percent sucrose, dry matter, and marc were corrected for shrinkage during storage by multiplying by the ratio of the weight of the sample after storage to the weight of the sample before storage.

RESULTS AND DISCUSSION

Variables in Marc Determination

Extraction Temperatures.--Since the solubility of the pectins which make up approximately 50% of the marc is highly temperature dependent, the percent marc as determined by hot water extraction should also be highly temperature dependent. To determine the effects of temperature in the marc determination a well mixed brei sample was divided into 50 g lots and treated in the normal marc extraction procedure except for the following variations in extraction temperatures (Table 1). All treatments were replicated three times.

TABLE 1.--Effect of Extraction Temperature on Percent Marc.

| Treatment | Bath Temperature (C.) | Extraction Temperature (C.) | Percent Marc |
|-----------|-----------------------------|-----------------------------------|-----------------|
| 1 | 70 | 70 | 4.590 |
| 2 | 80 | 12 | 4.394 |
| 3 | 80 | 80 | 3.844 |
| 4 | 80 | 100 | 3.225 |

The results exemplify the sensitivity of the marc to high extraction temperatures. Increasing both the bath and

extraction temperatures from 70 C. to 80 C. decreased the percent marc by 0.745. Increasing the extraction temperature from 12-80 C. reduced the percent marc 0.550. The increase from 80 to 100 C. reduced the percent marc by 0.619. These results correlate with the rates of protopectin hydrolysis found by Silin (1964). At temperatures below 80 C. the hydrolysis of protopectin was slow, but above 80 C. the breakdown was extremely rapid. In sugar-beet processing, diffusion temperatures do not usually exceed 80 C.

The results indicate the necessity of accurate control of bath and extraction temperatures in determining the percent marc particularly at temperatures near 80 C.

A similar experiment was conducted to determine the effect of the volume of extraction water used. Increasing the volume from 1-2 liters reduced the percent marc 0.158 (3.998 → 3.840). The one liter volume was used in all storage marc determinations to enable a more precise control of extraction temperatures and to reduce the time required for extraction, even though it did not give the lowest percent marc.

Length of Extraction Time.--Solubilization of pectin is not only temperature dependent, but also depends on the length of treatment (Silin, 1964). The extraction time should therefore have an effect on the determination of percent marc.

A well mixed brei sample was divided into 50 gm lots and treated with 80 C. distilled water for various bath and extraction times. Table 2 indicates the treatments applied. Lots of six samples were treated in the bath for periods of 10, 30, and 60 minutes after which three samples from each lot were extracted for 10 minutes and three for 20 minutes. The same volume of water was used in both cases.

TABLE 2.--Effect of Varying the Length of Bath Treatment and Extraction Time on Percent Marc.

| Extraction Time | Bath Time | | |
|-----------------|-----------|-------|-------|
| | 10 | 30 | 60 |
| 10 | 4.809 | 4.485 | 4.359 |
| 20 | 4.527 | 4.320 | 4.205 |

A rapid decrease in the percent marc was found between the 10 and 30 minute bath times after which the rate decreased (Figure 1). The twenty minute wash removed a greater proportion of the marc (0.28 compared to 0.16) at the 10 minute bath time than at the 30. However, the greater amount extracted in 20 minutes was a constant between 30 and 60 minutes.

The results indicate that just over 10 minutes is required for the easily solubilized pectins to become completely soluble and to diffuse out of the cell. With the

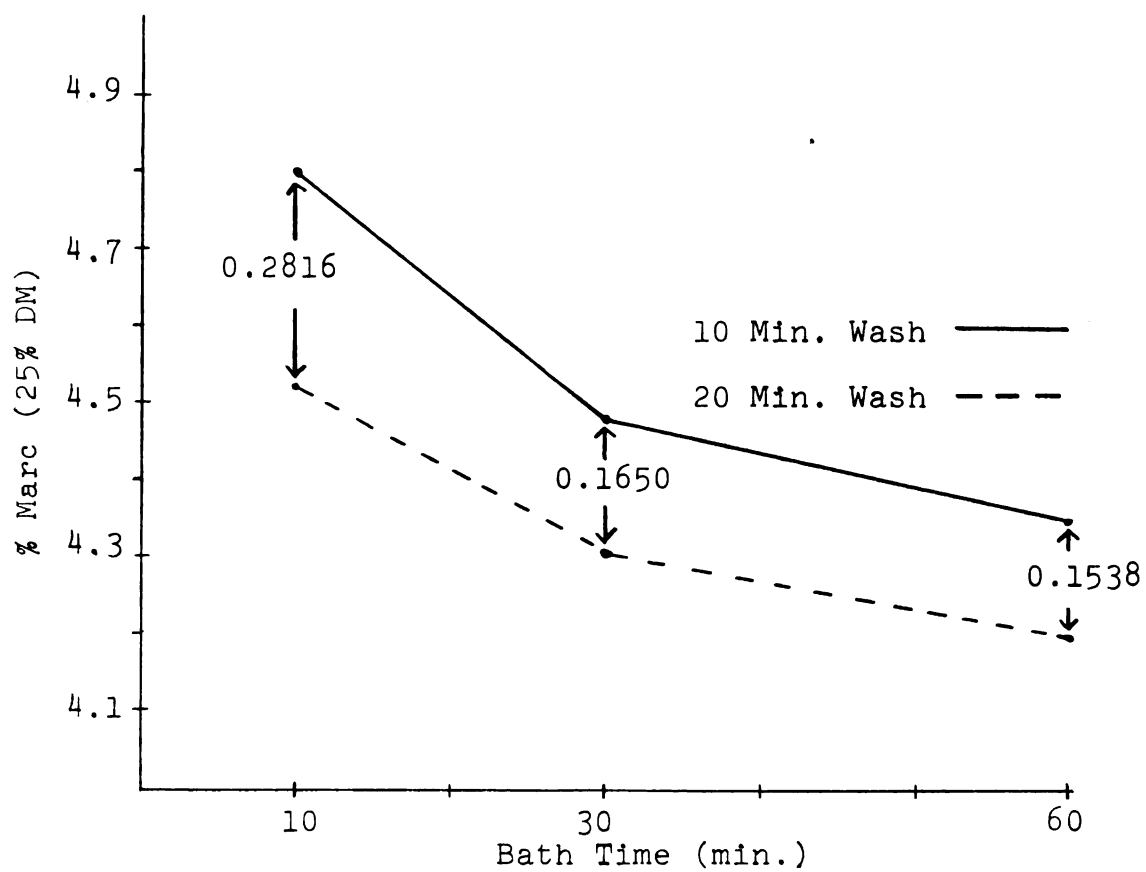


Figure 1.--Effect of treatment time on percent marc.

removal of these pectins the protopectin begins to solubilize at a rate proportional to the extraction time.

Effect of Nitrogen Fertilization and Variety
on Percent Marc at Harvest

Variety X Nitrogen.--Under the conditions of this experiment there was no effect of the rate of nitrogen fertilization on the marc and sucrose contents nor on the C. J. P. at harvest. Previous experiments to determine the effects of nitrogen have shown significant differences between high and low levels of nitrogen. In general lower rates of nitrogen increase the percent sucrose and clear juice purity.

In order to compare marc values directly between varieties, the percent marc at harvest was adjusted to a constant 25% D.M. to eliminate variations due to differences in dry matter content (Table 3). Although a significant difference was found between varieties in percent marc, the rate of nitrogen fertilization had no effect.

Experimental Varieties.--The results of the supplementary experiment to determine the variation in percent marc between eight varieties at harvest are given in Table 4.

At the time of harvest the marc values ranged from a high of 4.604% in O2 clone to 3.853 in variety D. Varieties D and E are identical genetically except for one inbred line and had essentially identical marc values at harvest.

TABLE 3.--Percent Marc of Three Varieties at Harvest
Adjusted to 25% Dry Matter.

| | Nitrogen Treatment | Unadjusted Percent Marc | Percent Dry Matter | Adjusted Percent Marc | Significance |
|-----------------------|-----------------------|----------------------------|-----------------------|--------------------------|--------------|
| Commercial | 150# | 4.501 | 24.656 | 4.563 | |
| | 75# | 4.633 | 24.310 | 4.765 | |
| Average | | 4.567 | 24.483 | 4.664 | ab |
| 63-194 | 150# | 4.826 | 23.651 | 5.101 | |
| | 75# | 4.759 | 23.872 | 4.985 | |
| Average | | 4.793 | 23.762 | 5.043 | a |
| 401 | 150# | 4.336 | 21.824 | 4.967 | |
| | 75# | 4.492 | 23.012 | 4.880 | |
| Average | | 4.414 | 22.418 | 4.923 | b |
| Average high nitrogen | | 4.877 | | | |
| Average low nitrogen | | 4.876 | | | |

TABLE 4.--Percent Marc of Eight Experimental Varieties at Harvest Adjusted to 25% Dry Matter.

| Variety | Unadjusted Percent Marc | Percent Dry Matter | Adjusted Percent Marc |
|---------|-------------------------------|-----------------------|-----------------------------|
| A | 4.240 | 26.101 | 4.061 |
| B | 4.172 | 26.850 | 3.885 |
| C | 3.846 | 24.489 | 3.926 |
| D | 3.944 | 25.551 | 3.858 |
| E | 4.009 | 25.341 | 3.955 |
| F | 4.432 | 26.769 | 4.139 |
| G | 3.837 | 23.126 | 4.147 |
| H | 4.420 | 23.997 | 4.604 |

A - SL(129 x 133) MS x SP(5822-0)
 B - (SP 6121 x FL 31) MS x (SP 5822-0)
 C - SL(129 x 133) MS x (SP 6322-0)
 D - (SL 129 x SP 6121) MS x (SP 6428-0)
 E - (SL 126 x SP 6121) MS x (SP 6428-0)
 F - (SP 5822-0)
 G - SP 6322-0
 H - 02 Clone

Results of Beet Storage

After 136 days of storage all samples of the three varieties were in excellent physical condition. The beets stored warm wilted slightly more than the beets stored cold but no visible rot or mold was found.

Excessive desiccation was noted when samples were removed for the first analysis. This problem was alleviated

by placing the remaining samples inside plastic bags.
(For a complete table of results, see Appendix.)

Changes in Percent Marc During Storage

Effect of Nitrogen Fertilization.--Figure 2 is an average of all varieties and storage temperatures at the high and low nitrogen levels. The results indicate a steady decrease in marc over the entire storage period at both nitrogen levels. Although the difference was not significant the beets grown at the high level of nitrogen were consistently higher in percent of original marc over the entire storage period.

Nitrogen fertilization (Figure 3) had a different effect on each variety. The variety x nitrogen interaction was significant at the 5% level with an LSD of 0.253. In 63-194 the differences between high and low nitrogen were not significant. However, the commercial variety grown on high nitrogen stored significantly better than those on low nitrogen. In variety 401 just the opposite situation existed with the low nitrogen beets losing a smaller percentage of their original marc.

The results indicate very little effect of nitrogen fertilization on the decomposition of insoluble cell wall material in beets during storage. The differences which did occur were consistent for the whole experiment but varied with individual varieties. However, beets grown on high nitrogen were consistently higher in percent of

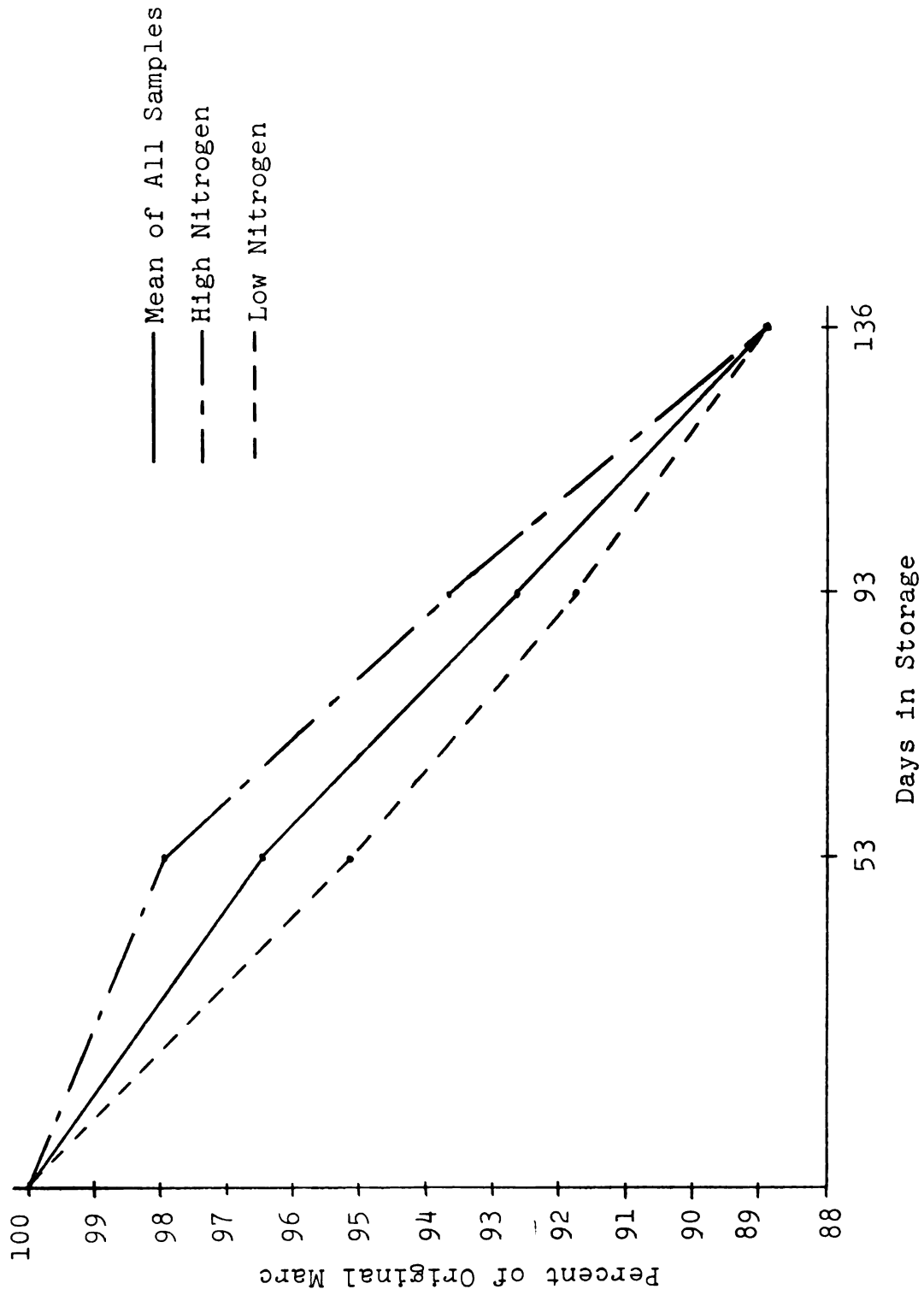


Figure 2.--Effect of nitrogen fertilization on marc breakdown of three varieties during storage.

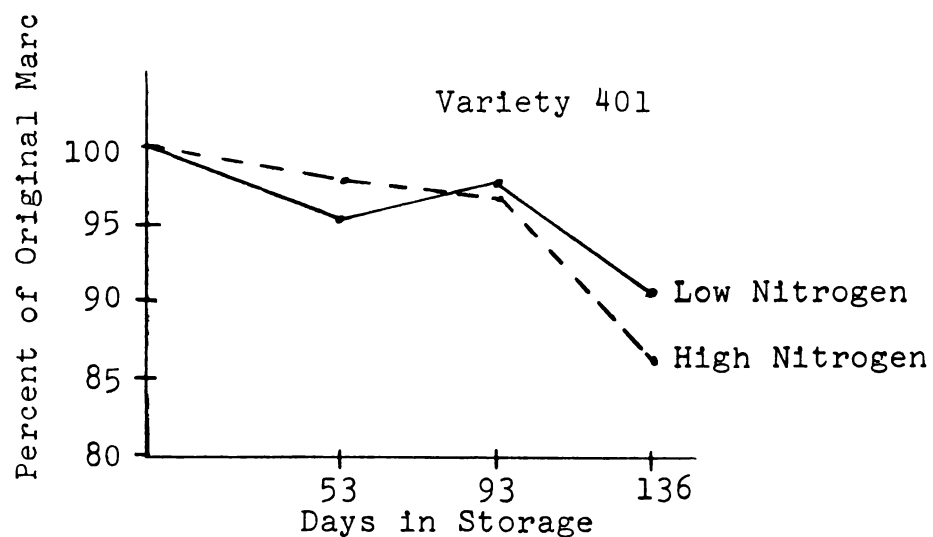
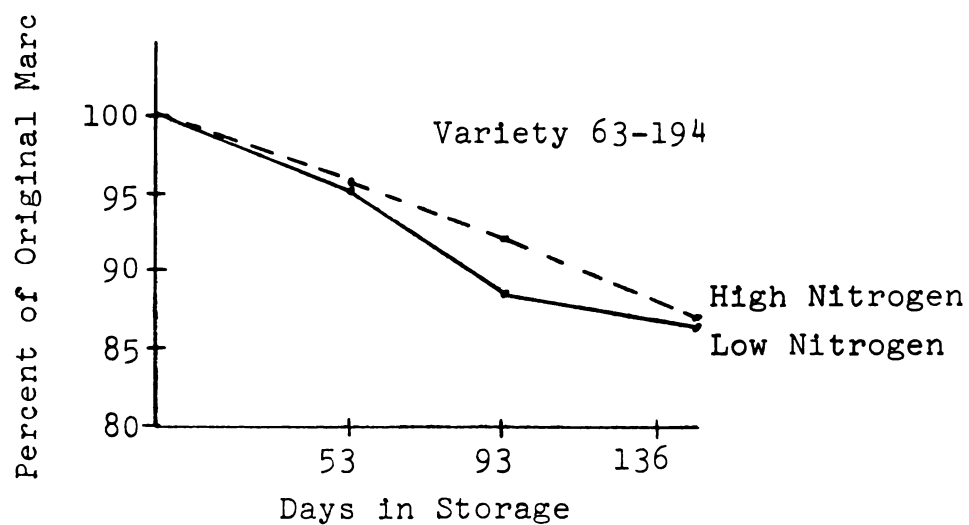
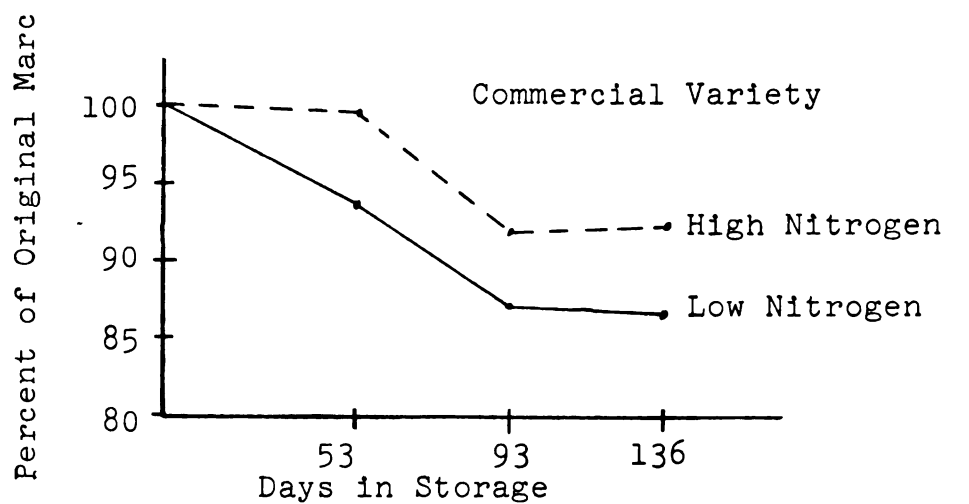


Figure 3.--Effect of nitrogen fertilization on marc breakdown of three individual varieties.

original marc remaining, indicating that a high level of nitrogen fertilization may contribute to marc stability during storage.

Effect of Storage Temperature.--Figure 4 is an average of all varieties and nitrogen treatments at each storage temperature. The final difference between the two storage temperatures was significantly different at the 10% level. The temperature x variety interaction was also significant at the 10% level. A breakdown into individual varieties indicates little effect due to storage temperature in the commercial and 63-194 varieties. Variety 401, however, stored better at 35 F. than at 45 F.

Although the final difference between the two temperatures was small, the beets stored at 32 F. stored with consistently less decomposition of marc than the beets stored at 45 F., particularly in the first 93 days of storage.

Individual varieties (Figure 5), showed a significant difference in storing ability. Long term storage may require the proper combination of fertilization, variety and temperature.

Effect of Variety.--After 136 days of storage an analysis of variance indicated a significant difference in percent marc between the three varieties (Figure 6). Variety 401, however, did not lose a significant amount of marc until after 93 days of storage. The other two varieties decreased in a linear fashion over the entire storage period.

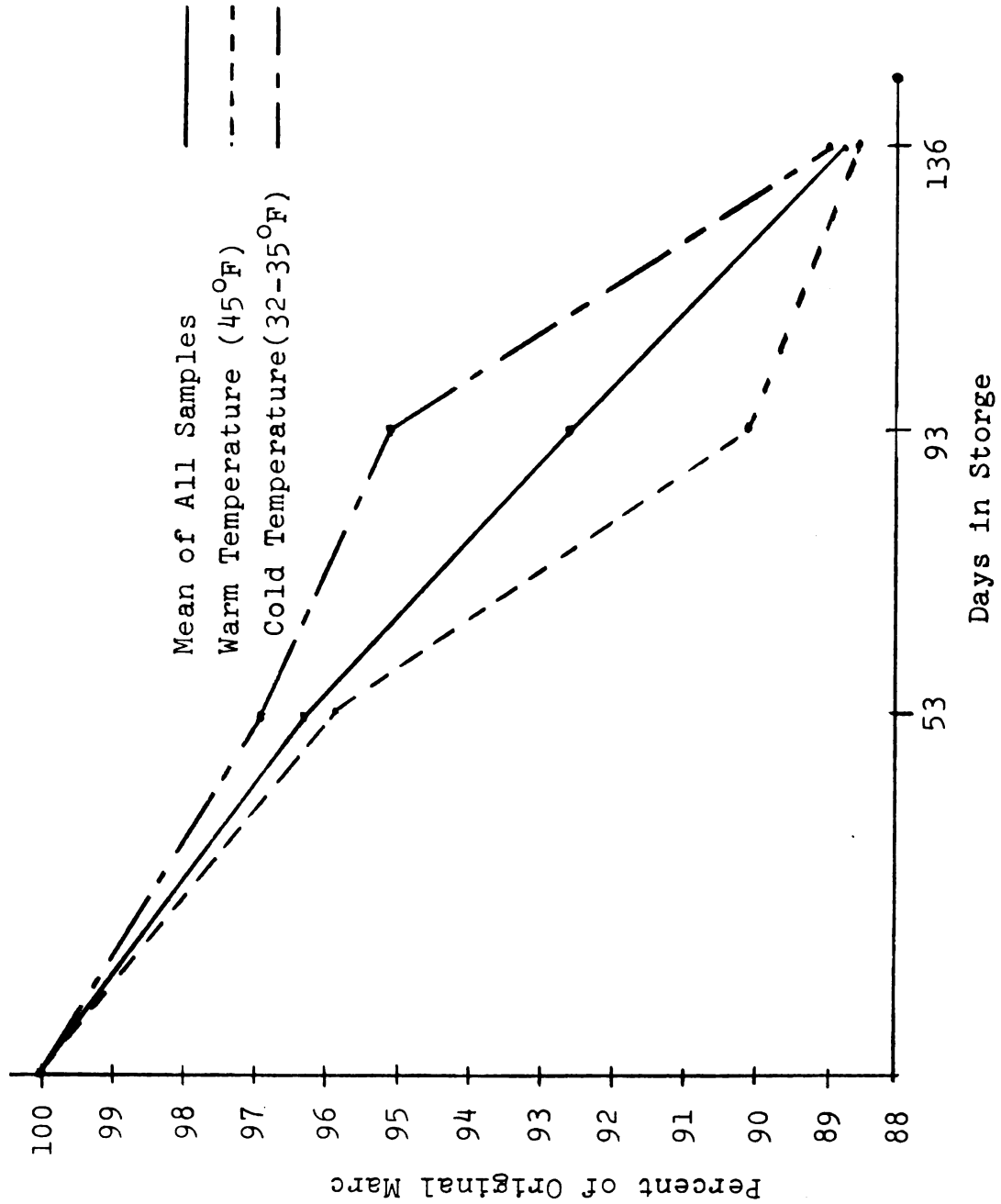


Figure 4.--Effect of storage temperature on marc decomposition of three varieties during storage.

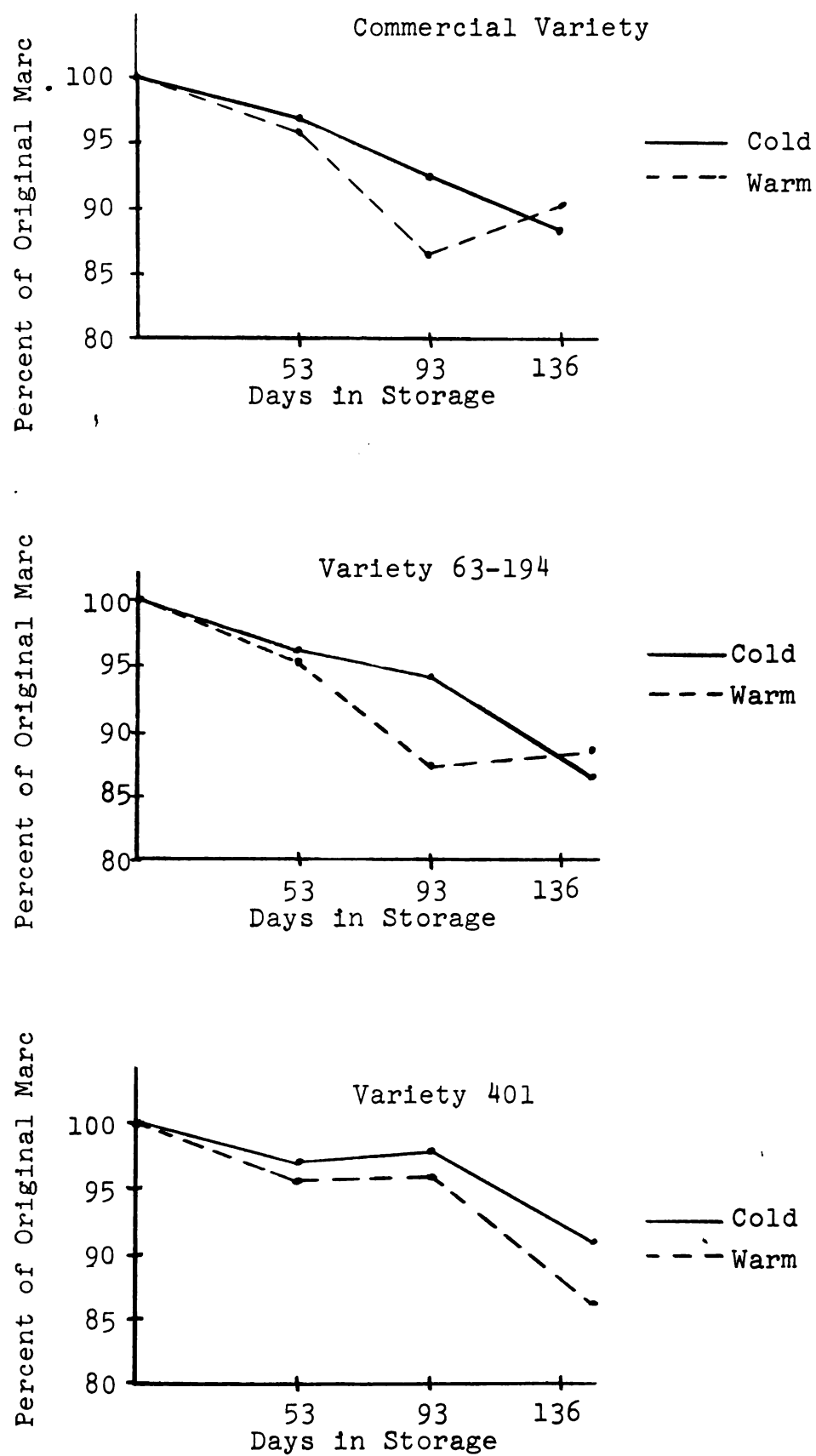


Figure 5. Effect of storage temperature on marc decomposition of three individual varieties.

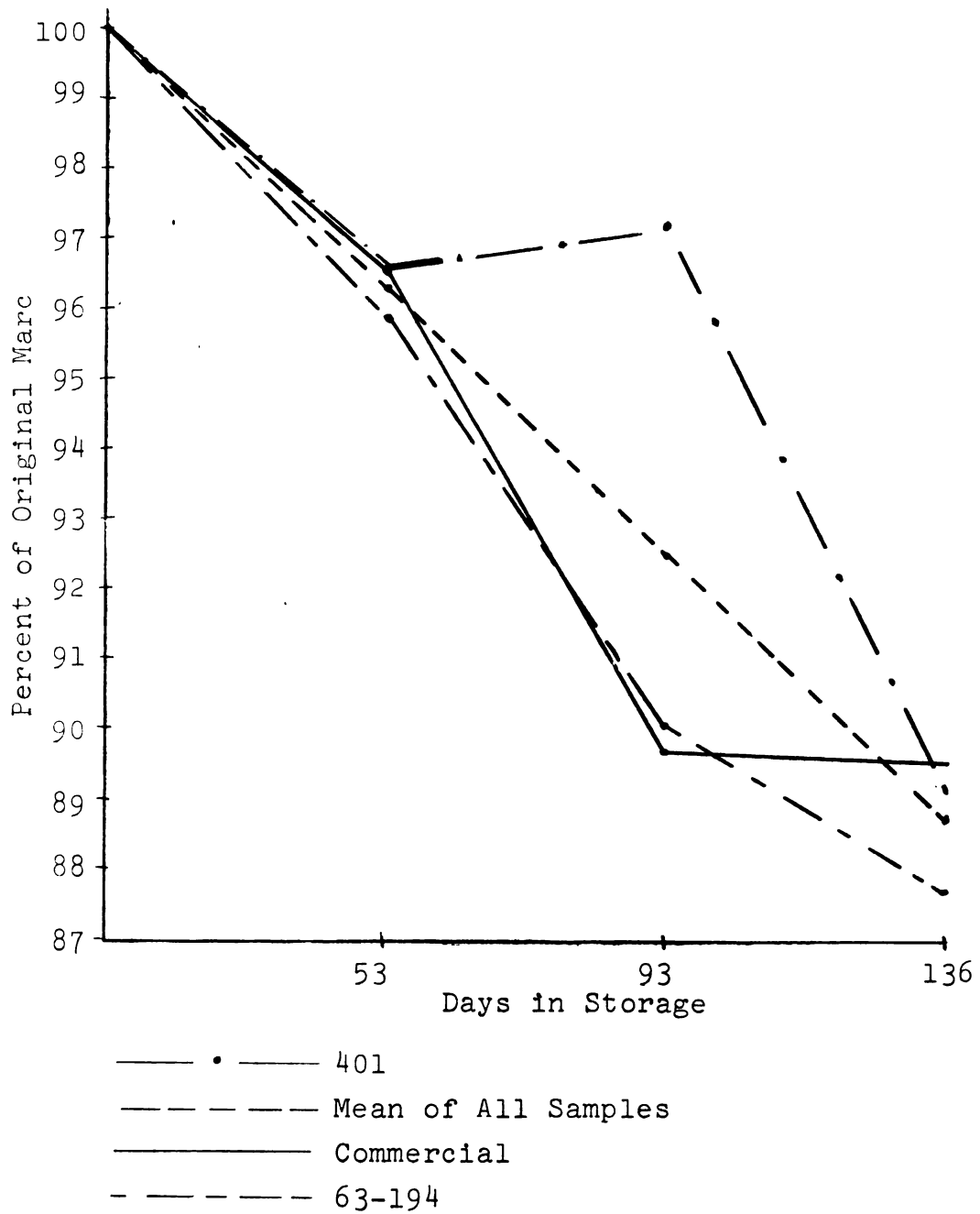


Figure 6.--Breakdown of marc in three varieties over a 136 day storage period.

Figure 7 is a composite of all marc values at each removal date. The decrease was essentially linear over the entire storage period. All varieties retained approximately 89% of their original marc.

Table 5 shows the changes in marc which occurred over the 140 day storage period for the seven experimental varieties stored at 45 F.

TABLE 5.--Changes in Marc and Dry Matter Content During 140 Days of Storage in Seven Experimental Varieties.

| Variety | Harvest | | Last Removal | | Percent of Original Marc Remaining at Storage End | |
|--------------|--------------|--------|--------------|--------|---|--------|
| | % Dry Matter | % Marc | % Dry Matter | % Marc | % Dry Matter | % Marc |
| A | 26.101 | 4.240 | 22.982 | 3.690 | 88.05 | 87.02 |
| B | 26.850 | 4.172 | 21.747 | 3.323 | 80.99 | 79.65 |
| D | 25.551 | 3.944 | 22.055 | 3.358 | 86.32 | 85.14 |
| E | 25.341 | 4.009 | 22.738 | 3.609 | 89.73 | 90.03 |
| F | 26.769 | 4.432 | 23.948 | 3.979 | 89.46 | 89.77 |
| G | 23.127 | 3.837 | 21.295 | 3.363 | 92.08 | 87.65 |
| H | 23.997 | 4.420 | 20.662 | 3.816 | 86.10 | 86.33 |
| Average Loss | | | | | | 13.49 |

When removed from storage varieties G and H had only small amounts of rot but were excessively sprouted. Variety E had no rot and very few sprouts. Variety C was almost completely rotted and was discarded. The other varieties

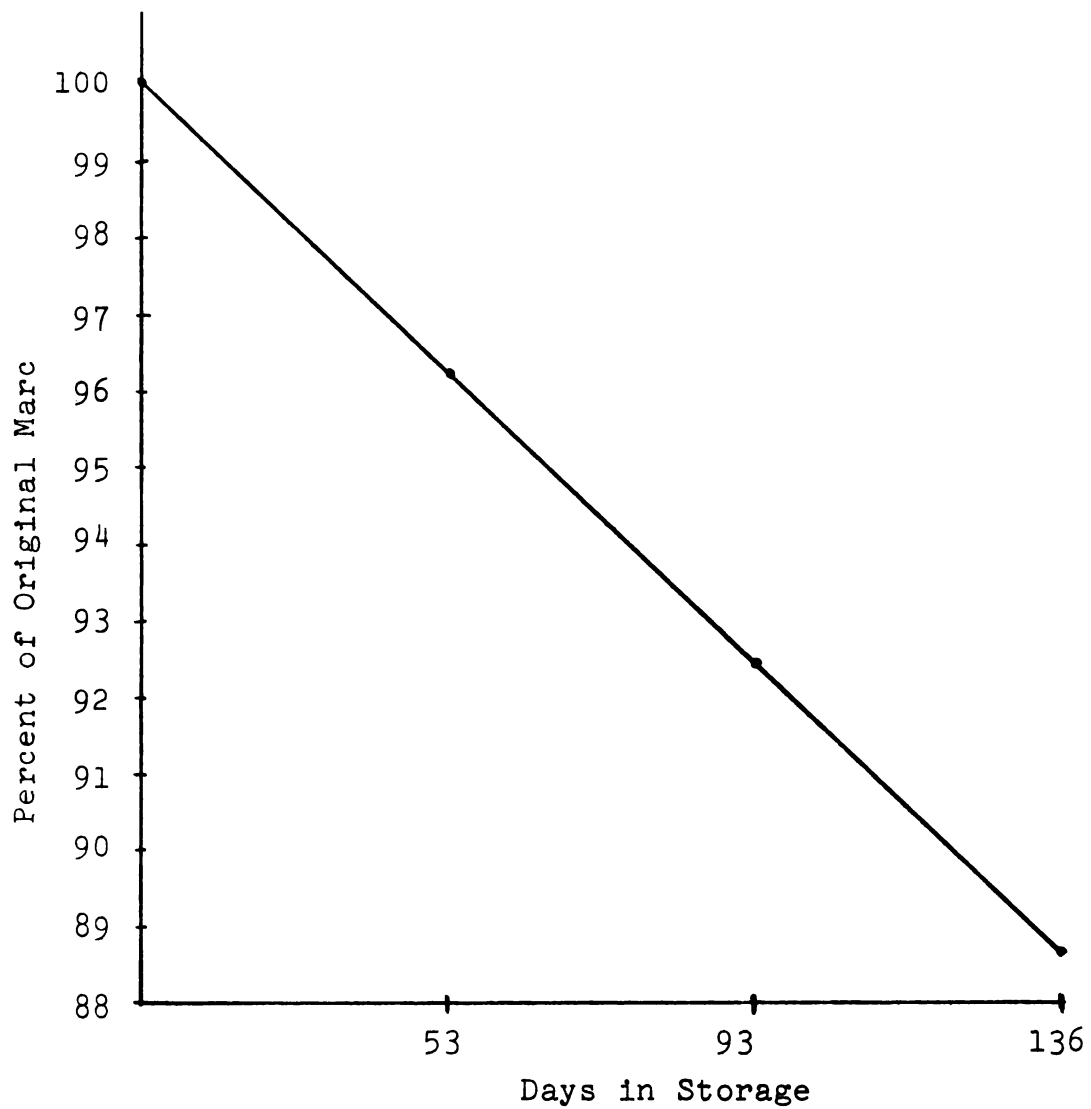


Figure 7.--Effect of storage on the breakdown of marc in all storage treatments.

were intermediate between these two extremes but all contained excessive rot in the crown area. Before obtaining brei for marc determinations all rotting tissue was removed. A wide range was found within the varieties in the percent of marc solubilized. Variety B losing 20.35% of its original marc was definitely the inferior variety and variety E the superior losing only 9.98%.

The other varieties were grouped at about 13.49% loss which corresponds rather closely to the 11% average loss in the variety-nitrogen experiment.

There appeared to be little connection between original percent marc and the loss of marc during storage. Figure 8 is a plot of the original percent marc for the seven varieties adjusted to 25% dry matter plotted against the decrease in storage. The two varieties B and D which were low in percent marc had the greater loss in original marc. However, there appeared to be little connection between original percent marc and loss during storage.

Proper selection of variety for long storage may become a factor to consider in future breeding programs.

Increase in Impurities in the Diffusion Juice Due to Marc Decomposition

The dry matter in the beet, before or after storage, might be considered to be (1) insoluble marc, (2) sucrose, and (3) solubles other than sucrose in the diffusion juice.

A logical means of expressing the effect of marc decomposition on sugarbeet processing is on the increased

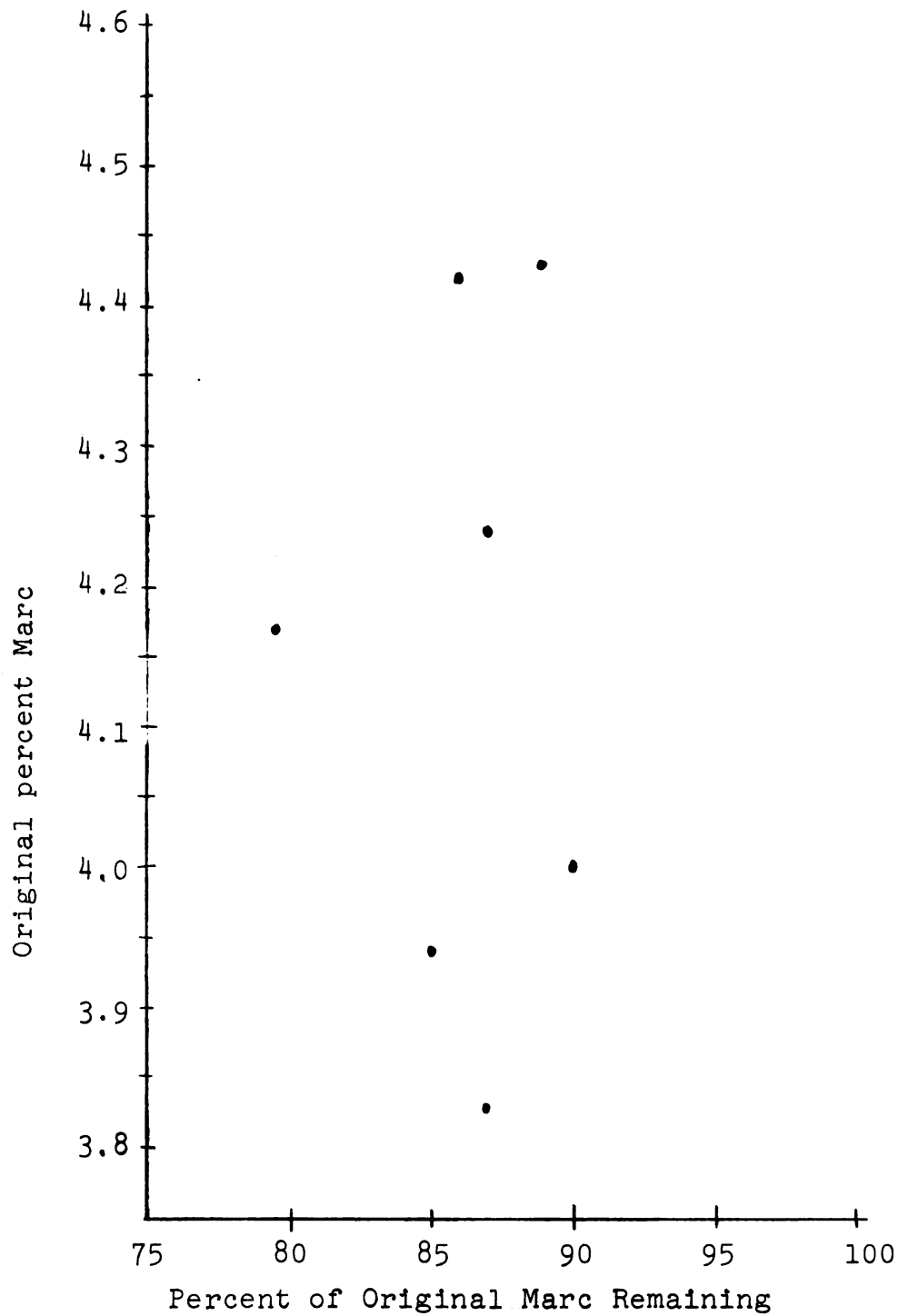


Figure 8.--Relationship between percent marc at harvest and loss of marc during storage in seven experimental varieties.

non-sucrose impurities in the diffusion juice which must be removed in the purification process. In Table 6 the increased impurities due to the breakdown of marc are expressed in mg/100S.

TABLE 6.--Impurities Added to the Diffusion Juice by Marc Decomposition Over the 136 Day Storage Period.

| Treatment | Final Percent Sucrose | Decrease in Marc During Storage | Impurities* Added to the Diffusion Juice |
|------------------|-----------------------------|---------------------------------------|--|
| | | gm/100gm beets | mg/100S |
| Variety: | | | |
| Commercial | 16.05 | .472 | 2941 |
| 43-194 | 15.33 | .594 | 3875 |
| 401 | 14.19 | .478 | 3368 |
| High Nitrogen | 15.35 | .506 | 3301 |
| Low Nitrogen | 15.04 | .523 | 3477 |
| 45 F. - Warm | 14.69 | .520 | 3540 |
| 32 F. - Cold | 15.69 | .509 | 3368 |

* (Decrease in Marc/Final % Sucrose)100,000

De-esterification of soluble pectins will yield acids with soluble calcium salts which will pass into the thin juice, be concentrated in the thick juice, and precipitate out, forming calcium deposits on the evaporation pans.

High nitrogen fertilization and cold storage temperatures reduced the amount of impurities in the diffusion juice due to a solubilization of marc although the differences between treatments was small. The commercial variety was superior to both 401 and 63-194.

Since the soluble pectic substances are the portion of the marc which is easily degraded in storage, a part of the increase in impurities in the diffusion juice can safely be assumed to be pectinaceous materials that precipitate with lime.

Relation of Increased Impurities in
Diffusion Juice to Unaccounted for
Impurities in Clear Juice

From the purity of the thin juice the total impurities in mg/100S were calculated. As found previously by Dexter (1966) the percent of these impurities accounted for by Carruthers between Na, K + α NH₂ decreased during storage. However, the increase in unaccounted for impurities showed little relation to the increased impurities in the diffusion juice due to the decomposition of the marc (Table 7).

A rigorous study of the impurity changes which occurred in the diffusion juice, the clear juice and Ca precipitated impurities could not be made pending a determination of raffinose and inverts. Such a study is required to correct the pol. reading for these optically active compounds before valid conclusions can be drawn. When the extract from the marc determination was treated with CaO

the supernatant did not reveal the presence of any pectic substance using the carbazole test of McComb and McCready (1952) for Anhydronic Acid.

TABLE 7.--The Relationship Between the Impurities Added to the Diffusion Juice by Marc Decomposition and the Increase in Impurities in the Clear Juice After 136 Days of Storage.

| Variety | Increase in Unaccounted for Impurities in Clear Juice (mg/100S) | Increase in Apparent Impurities in Clear Juice (mg/100S) | Change in Marc (mg/100S) |
|------------|---|---|--------------------------------|
| Commercial | 3034 | 2336 | 2975 |
| 63-194 | 1066 | 738 | 3870 |
| 401 | 2402 | 2111 | 3429 |

At present the insoluble protopectin is thought to exist in a complex with the hemicellulose arabans and galactans (Joslyn, 1962). The breakdown of protopectin during storage would also cause the arabans to become soluble (Owens and Goodban, 1956). Since the arabans are not precipitated with calcium they would pass through the factory into the molasses (Silin, 1964). Therefore, the absence of pectic substances in the clear juice does not imply that all the impurities added to the diffusion juice due to marc breakdown were precipitated with lime and did not contribute to the impurities in the clear juice.

Relationship Between the Changes in Dry Matter, Sucrose and Marc During Storage

Figure 9 is a composite of all varieties and treatments and gives the actual changes in marc, dry matter and sucrose during the 136 day storage period.

Although the total DM and apparent sucrose losses are much greater than the loss of marc, the relative decrease is much greater in the marc. The total decrease in dry matter (1.45 gm/100gm beets) is greater than the loss in apparent sucrose (1.17 gm/100gm beets). Since it is currently thought that a large part of the sucrose losses during storage are due to inversion (Stout, 1950 and Barr, 1940) which would not result in a D.M. decrease, this phenomena is even more surprising. If we assume that all the sucrose was lost by respiration, the decrease accounts for only 67% of the total dry matter lost after 93 days of storage and 80.6% after 136 days.

The greater decrease in dry matter would indicate that non-sucrose compounds are being respired. This may in part account for the increase in purity during the early stages of storage. However, before valid conclusions can be drawn a correction for raffinose and invert must be made. Analyses for "apparent sucrose" are not sufficient.

Changes in Extractable Sugar During Storage

A comparison of the three varieties in recoverable sugar lost (Table 8) indicates that the commercial variety,

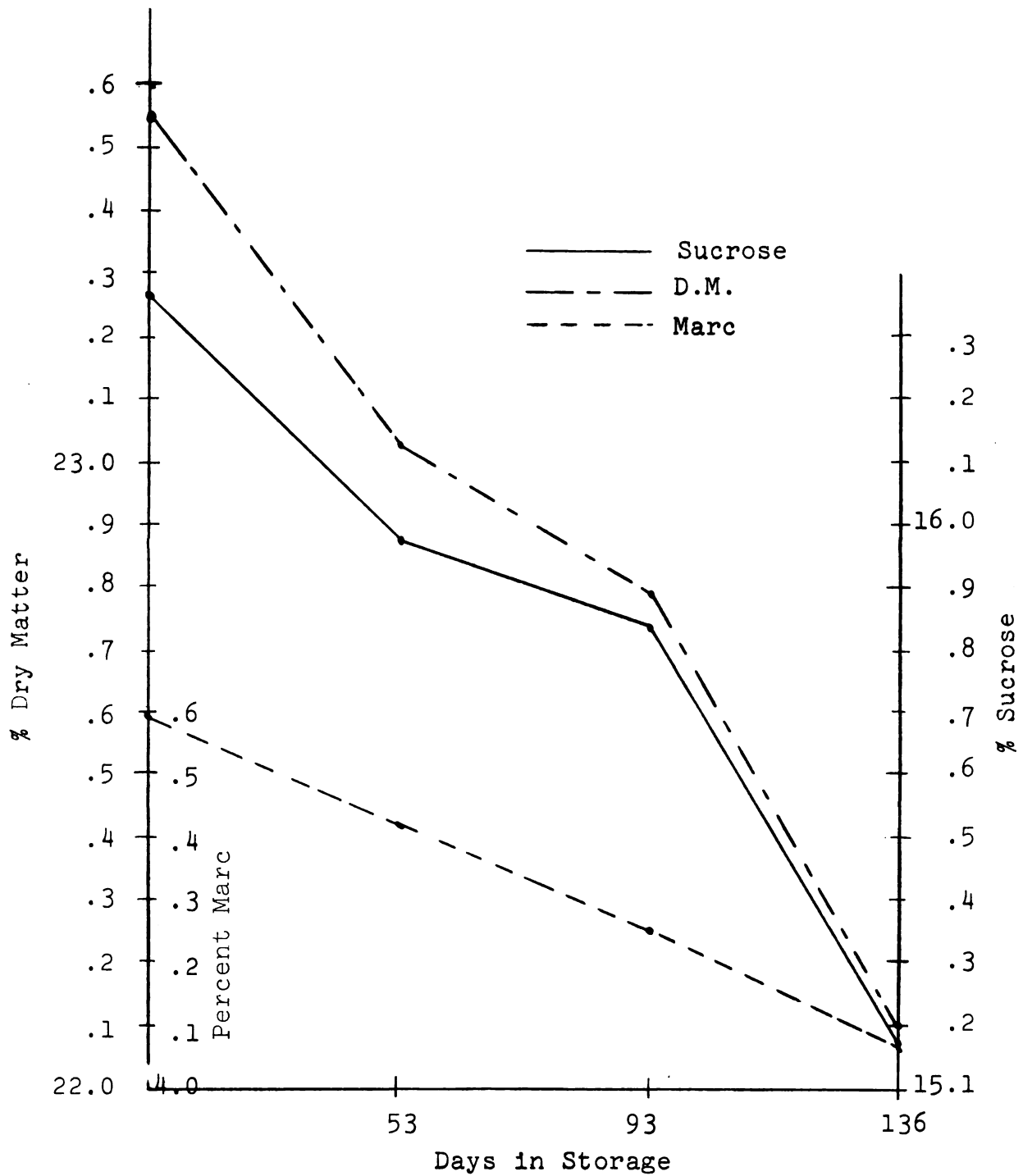


Figure 9.--Changes in percent dry matter, percent sucrose and percent marc of three varieties during 136 days of storage.

TABLE 8.--Evaluation of Varieties for Storing Ability Using the Concept of Extractable Sugar per Ton of Beets (Average for 32 F. and 45 F. Storage, High and Low Nitrogen).

| Variety | Removal | | | | | | | | | | | |
|------------|----------|---------|----------|---------|----------|---------|----------|---------|-----|-------|-------|-----|
| | Harvest | | First | | Second | | Third | | | | | |
| | Sucrose* | C.J.P.* | Sucrose* | C.J.P.* | Sucrose* | C.J.P.* | Sucrose* | C.J.P.* | | | | |
| Commercial | 17.55 | 93.05 | 302 | 16.96 | 93.47 | 294 | 16.52 | 92.38 | 280 | 16.05 | 91.79 | 268 |
| 63-194 | 16.04 | 90.60 | 260 | 15.73 | 91.76 | 262 | 15.74 | 90.86 | 257 | 15.33 | 90.02 | 245 |
| 401 | 15.50 | 90.94 | 253 | 15.25 | 91.66 | 253 | 15.26 | 91.22 | 251 | 14.19 | 89.65 | 224 |

*Apparent Sucrose, C. J. P., and ESPT (Extractable Sugar per Ton)

although containing a higher percent sucrose at harvest, and after storage had a larger percentage of loss. Varieties 63-194 and 401, however, did not decrease significantly in recoverable sugar in the first 93 days of storage. Over the entire 136 day storage period the commercial and 401 both lost approximately 11% of their original recoverable sugar, while variety 63-194 lost only 5.2%. Marc decomposition was greatest in 63-194. Therefore, although a high percent of recoverable sugar was maintained, the overall processability may be lower due to the increased pectinaceous materials caused by marc decomposition.

In comparing the two storage temperatures (Table 9), the decrease in extractable sugar was essentially the same in the first 93 days of storage, however, in the final 43 days the decrease was extremely rapid with the warm temperature resulting in three times more extractable sugar being lost than the cold stored beets.

High nitrogen beets decreased steadily in extractable sugar over the entire storage period. The low nitrogen beets however increased slightly after 53 days of storage but then decreased rapidly. In this instance, although the differences between high and low nitrogen beets were small at harvest, the low nitrogen beets were slightly higher in extractable sugar. However, these higher quality beets had the greatest decrease in extractable sugar which is in conflict with the previous findings of Dexter (1966) where

TABLE 9.--Evaluation of Nitrogen Fertilization and Storage Temperature Using the Concept of Extractable Sugar Per Ton of Beets.

| Treatment | Removal | | | | | | | | | | | |
|---------------|---------|--------|------|---------|--------|------|---------|--------|-------|---------|--------|------|
| | First | | | | Second | | | | Third | | | |
| | Sucrose | C.J.P. | ESPT | Sucrose | C.J.P. | ESPT | Sucrose | C.J.P. | ESPT | Sucrose | C.J.P. | ESPT |
| Cold | 16.36 | 91.53 | 272 | 16.05 | 92.62 | 262 | 15.62 | 91.99 | 262 | 15.69 | 91.44 | 260 |
| Warm | 16.36 | 91.53 | 272 | 15.91 | 92.30 | 269 | 15.73 | 90.98 | 257 | 14.87 | 89.69 | 235 |
| High Nitrogen | 16.33 | 91.27 | 269 | 15.75 | 91.91 | 264 | 15.73 | 90.93 | 257 | 15.33 | 90.53 | 248 |
| Low Nitrogen | 16.40 | 91.80 | 274 | 16.21 | 92.69 | 276 | 15.95 | 92.05 | 268 | 15.05 | 90.44 | 243 |

Change in
ESPT

high quality beets stored with a smaller decrease in ESPT.*

Although the calculation of extractable sugar per ton is an excellent method of combining the factors which enter into the quality of freshly harvested sugarbeets into one number, it does not incorporate the factor of processability.

Evaluation of the Method of Analysis

Although the accuracy of the procedure was demonstrated in duplicate marc determinations on a large, well-mixed brei sample, the replications in the storage experiment occasionally contained marc values which were obviously out of line. However, these variations may have been due to sampling errors and not procedure. At the time of the last removal LSD values of 0.35, 0.25, and 0.439 were required for significance at the 10% level between varieties, interactions and treatments respectively.

The procedure used in marc determination was a rather severe one and may have partially obstructed treatment differences by removing a large amount of substances which were insoluble prior to the extraction thus blanketing differences.

A possible solution would be to reduce the bath temperature to 70 C. and wash with cold water. This would also remove the error due to small temperature differences at 80 C. (see pg. 13).

*Extractable sugar per ton.

SUMMARY AND CONCLUSIONS

The effects of sugar beet variety, nitrogen fertilization and storage temperature on the decomposition of the insoluble cell wall materials and the changes in apparent sucrose and dry matter in a 136 day storage period were studied. Three varieties--commercial, 63-194, and 401--were grown under high (150#/A) and low (75#/A) nitrogen fertilization and stored at 32-35 F. and 45 F. Percentages of marc, dry matter, apparent sucrose and C. J. P. were determined at approximately 40 day intervals.

Marc and dry matter were determined at harvest and after 140 days of storage with seven additional varieties to further study the difference in storing ability between varieties.

The study yielded the following conclusions:

1. In this experiment only slight differences in sucrose and C. J. P. were found between lots of beets fertilized with 75 vs 150 lbs. of nitrogen per acre.
2. At the time of harvest there was a significant difference between varieties in percent marc. Nitrogen fertilization had no effect.
3. A rise in temperature from 35 F. to 45 F. significantly increased the rate of marc

decomposition. There was also a significant variety X temperature interaction.

4. The rate of nitrogen fertilization did not affect the solubilization of marc in the overall experiment. However, the variety X nitrogen interaction was highly significant. In variety 63-194 marc decomposition was the same at both levels of nitrogen. In the commercial variety, however, marc decomposition was significantly lower at the high nitrogen rate than at the low. Variety 401 was just the reverse of the commercial variety.
5. After 136 days of storage there was a significant difference between varieties in the percent of original marc lost. Variety 401 was definitely superior in storing ability for the first 93 days of storage.
6. A comparison of seven experimental varieties stored at 45 F. indicated a significant difference between varieties in resistance to rot, mold and sprouting. The variety most resistant to mold retained the highest percent marc. Sprouting appeared to increase solubilization of marc. There was little correlation, however, between percent marc at harvest and the decrease in marc during storage.

7. The loss in marc was 11.3 and 13.5 percent in the Variety x Nitrogen and Variety Test respectively.
8. There was little correlation between the increase in soluble solids in the diffusion juice due to marc decomposition and the increased impurities in the clear juice. However, no analysis of raffinose and invert were conducted to correct for errors in the apparent sucrose values.
9. The absolute decrease in dry matter was significantly greater than the decrease in apparent sucrose. This was particularly true at warm storage temperatures, indicating the rate of non-sucrose substrate respiration to be substantial.
10. Beets stored cold and beets grown with high nitrogen (150#/A) stored with the least loss of extractable sugar.

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APPENDIX

| | Original | | | | First Removal | | | | Second Removal | | | | Third Removal | | | |
|-----------|----------|-------|-------|-------|---------------|-------|-------|-------|----------------|-------|-------|-------|---------------|-------|-------|-------|
| | DM | Marc | %S | CJP | DM | Marc | %S | CJP | DM | Marc | %S | CJP | DM | Marc | %S | CJP |
| 1 HC | 24.656 | 4.501 | 17.89 | 93.62 | 24.414 | 4.510 | 17.34 | 93.59 | 23.410 | 4.183 | 16.95 | 92.80 | 23.875 | 4.158 | 17.31 | 92.41 |
| 1 LC | 24.339 | 4.634 | 17.22 | 92.49 | 23.456 | 4.377 | 16.59 | 94.15 | 23.963 | 4.264 | 16.96 | 92.41 | 22.740 | 3.948 | 15.99 | 92.09 |
| Ave. | 24.483 | 4.575 | 17.55 | 93.05 | 22.935 | 4.443 | 16.96 | 93.56 | 22.686 | 4.224 | 16.95 | 92.60 | 23.310 | 4.052 | 16.65 | 92.25 |
| 1 HW | 24.656 | 4.501 | 17.89 | 93.62 | 22.442 | 4.455 | 17.00 | 95.17 | 22.942 | 4.192 | 16.95 | 91.67 | 23.422 | 4.162 | 16.31 | 91.28 |
| 1 LW | 24.339 | 4.634 | 17.22 | 92.49 | 22.443 | 4.377 | 16.57 | 92.45 | 22.667 | 4.231 | 16.95 | 92.24 | 21.596 | 4.113 | 16.75 | 92.69 |
| Ave. | 24.483 | 4.575 | 17.55 | 93.05 | 22.443 | 4.377 | 16.78 | 93.81 | 22.804 | 4.211 | 16.95 | 91.96 | 22.509 | 4.137 | 16.53 | 92.19 |
| Var. Ave. | 24.483 | 4.575 | 17.55 | 93.05 | 22.935 | 4.443 | 16.96 | 93.47 | 22.686 | 4.184 | 16.95 | 92.56 | 22.639 | 4.095 | 16.33 | 91.79 |
| 2 HC | 23.651 | 4.826 | 15.99 | 91.24 | 22.942 | 4.525 | 15.34 | 92.84 | 23.402 | 4.341 | 15.42 | 91.37 | 22.869 | 4.163 | 15.33 | 91.26 |
| 2 LC | 23.872 | 4.579 | 16.39 | 90.67 | 22.835 | 4.286 | 15.71 | 92.34 | 22.657 | 4.243 | 15.77 | 91.75 | 22.558 | 4.192 | 15.33 | 91.65 |
| Ave. | 23.761 | 4.703 | 16.34 | 90.63 | 22.888 | 4.406 | 15.53 | 92.59 | 23.029 | 4.292 | 15.59 | 91.56 | 22.713 | 4.178 | 15.33 | 91.44 |
| 2 HW | 23.651 | 4.826 | 15.99 | 91.24 | 22.942 | 4.525 | 15.34 | 91.39 | 22.959 | 4.341 | 15.42 | 90.71 | 22.922 | 4.307 | 15.35 | 88.71 |
| 2 LW | 23.872 | 4.579 | 16.39 | 90.67 | 22.634 | 4.421 | 15.43 | 91.91 | 22.547 | 4.236 | 15.36 | 91.91 | 22.905 | 4.173 | 14.75 | 88.60 |
| Ave. | 23.761 | 4.793 | 16.34 | 90.95 | 22.788 | 4.534 | 15.79 | 91.80 | 22.743 | 4.289 | 15.74 | 91.80 | 22.874 | 4.240 | 14.95 | 88.61 |
| Var. Ave. | 23.761 | 4.493 | 16.34 | 90.65 | 22.942 | 4.406 | 15.53 | 91.37 | 22.888 | 4.286 | 15.59 | 91.66 | 22.639 | 4.192 | 15.33 | 90.82 |
| 3 HC | 21.824 | 4.336 | 15.11 | 89.86 | 21.513 | 4.269 | 14.82 | 90.00 | 21.925 | 4.229 | 14.91 | 90.96 | 22.441 | 3.743 | 14.11 | 90.16 |
| 3 LC | 23.012 | 4.402 | 15.89 | 92.03 | 22.555 | 4.236 | 16.41 | 93.64 | 22.621 | 4.248 | 16.91 | 92.74 | 22.769 | 4.276 | 15.42 | 91.39 |
| Ave. | 22.418 | 4.414 | 15.50 | 90.94 | 22.399 | 4.266 | 15.62 | 91.77 | 22.278 | 4.247 | 15.46 | 91.53 | 21.395 | 4.237 | 14.76 | 90.62 |
| 3 HW | 21.824 | 4.336 | 15.11 | 89.86 | 22.074 | 4.224 | 14.38 | 90.47 | 20.905 | 4.131 | 14.63 | 89.42 | 22.144 | 3.703 | 13.52 | 88.79 |
| 3 LW | 23.012 | 4.402 | 15.89 | 92.03 | 22.475 | 4.238 | 15.10 | 92.66 | 22.335 | 4.350 | 15.10 | 91.63 | 20.630 | 3.969 | 13.70 | 88.58 |
| Ave. | 22.418 | 4.414 | 15.50 | 90.94 | 21.669 | 4.236 | 15.34 | 91.56 | 21.650 | 4.240 | 15.36 | 90.62 | 20.412 | 3.835 | 13.61 | 88.68 |
| Var. Ave. | 22.418 | 4.414 | 15.50 | 90.94 | 21.874 | 4.261 | 15.35 | 91.66 | 21.854 | 4.293 | 15.26 | 91.22 | 20.873 | 3.936 | 14.19 | 89.65 |

1 - Commercial H - 15 # N/A W - 45 F.
 2 - 63-174 L - 7 # N/A C - 52 F.
 3 - 401

Results of Storage of Three Varieties Grown at Two Levels of Nitrogen and Stored Warm and Cold