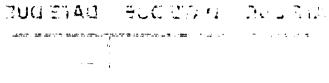
THE RELATION OF SUGAR-ACID RATIOS TO THE RIPENING AND DETERIORATION OF BLUEBERRY FRUIT

> Thests for the Degree of M. S. MICHIGAN STATE UNIVERSITY R. Charles Bowers

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# THE RELATION OF SUGAR-ACID RATIOS TO THE RIPENING AND DETERIORATION OF BLUEBERRY FRUIT

By

R. CHARLES BOWERS

AN ABSTRACT

Submitted to the College of Agriculture, Michigan State University of Agriculture and Applied Science in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

Department of Horticulture

1960 Joeold A. Z. Deeren Approved

ABSTRACT

Overripe blueberry fruit, being susceptable to rapid breakdown and decay, often is responsible for considerable losses during the marketing of this important Michigan crop. Other researchers have proposed a harvest chart for the Jersey variety, based on sugar and acid content of the fruit, for determining the proper time of harvest as an aid to overcoming this difficulty. Since the proposed chart needed further testing, and as Rubel is also an important variety in Michigan, studies were made to (1) further relate the sugar-acid ratio changes of the Rubel and Jersey varieties to ripening, (2) test the proposed index on commercially packed samples of Jersey fruit, and (3) explore the possibilities of a harvest index for fruit of the Rubel variety.

Fruit of known physiological age was obtained by removing all berries that were blue in color from 18 uniform Jersey and 18 uniform Rubel bushes. The next day all berries showing red or blue coloration of the skin were tagged. Random samples of these berries were then harvested at four-day intervals for a period of twenty days. Soluble solid readings of the fruit were taken in the field at the time of harvest. Other berries were frozen the same day of harvest for later analysis of sugar, titratable acid and pH. Sugars increased and acids decreased markedly during the first 8 days after the development of red coloration, and then remained relatively constant for the last 12 days. The

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sugar-acid ratios, however, showed marked and steady increases throughout the 20-day period.

Commercially harvested and packed berries were obtained throughout the 1959 harvest season for testing the Jersey harvest chart. Triplicate samples of each lot were taken immediately for determination of soluble solids and pH of the fruit juices. The rest were stored for evaluation of shelflife; four pints for 6 days at  $75^{\circ}$  F. and 8 pints for 18 days at  $40^{\circ}$  F.

Estimated sugars and estimated acids were determined from soluble solids and pH respectively utilizing conversion charts presented by other workers. The soluble solids, estimated sugar, estimated acid, and the ratios of soluble solids to H-ion and estimated sugar to estimated acid gave low correlation values when compared to deterioration of the fruit.

Samples containing blueberries of variable degrees of ripeness were obtained by a sequence of initial harvests from five commercial plantations in western Michigan to explore the possibilities of devising a harvest chart for the Rubel variety. Twenty uniform and vigorous bushes on each plantation were selected and divided into five plots of four bushes each. A set of four bushes was harvested every four days for a period of sixteen days. No fruit was harvested before the designated time of picking. Eleven

ABSTRACT

pints of fruit were obtained at each harvest. From these, three samples of twenty berries were selected at random for soluble solids measurement at the time of harvest; one pint was frozen for later analysis of sugar, acid and pH; and four pints were held six days at  $75^{\circ}$  F. and six pints were held eighteen days at  $40^{\circ}$  F. for evaluation of shelf-life. The ratios of sugar to acid, soluble solids to H-ion, and estimated sugar to estimated acid were positively correlated in a linear manner to the deterioration of the berries. Of these ratios, the one based on soluble solids and H-ion would serve best as a basis for a harvest chart for the Rubel variety, since both constituents can be readily obtained in the field. Further refinements of the Jersey harvest chart and additional tests with Rubel, however, are desirable prior to devising such a chart.

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#### INTRODUCTION

The fruit of the high bush cultivated blueberry (Vaccinium corymbosum) is harvested periodically according to the personal judgment of the grower based on blue coloration, general flavor, size, and flesh firmness of the fruit, and the quantity of berries ready for harvest. It is believed that overripe berries, which are susceptible to rapid deterioration and decay, should be avoided or eliminated from fresh market packs. Woodruff and Dewey (1959) suggested a harvest index based on the sugar and acid content of the fruit as a possible guide to harvesting the fruit before a large quantity of the berries are overripe. The practicality of their proposal has not been tested. Furthermore, it was determined only for the Jersey variety.

Since further testing of the index proposed by Woodruff and Dewey is essential to ascertain its suitability for evaluating commercial harvests of fruit, and as Rubel is also an important market variety in Michigan, studies were programed to (1) relate the sugar-acid ratio changes of the Rubel and Jersey varieties to ripening, (2) employ the proposed index on commercially packed samples of Jersey fruit to predict their keeping quality, and (3) explore the possibilities of a harvest index for the Rubel variety.

## LITERATURE REVIEW

Researches concerning the chemical and physiological changes associated with ripening of the blueberry fruit were recently reviewed by Woodruff (1959). Further search of the literature has revealed no additional information.

Woodruff's studies, which served as the basis for the present experiment, revealed that marked changes in some fruit constituents occur during the short period of time in which the berries change color from the initial tinges of red to blue, and during the somewhat longer period that the berries may remain on the bushes after becoming blue in color. He found that during these stages of ripening (1) total sugar increased, (2) titratable acid content decreased, (3) sugar-acid ratio increased, (4) pH increased, (5) pectin methylesterase activity increased, coupled with a decrease in soluble pectin content, and (6) intensity of pigmentation increased. There were no significant changes in: (1) starch, (2) acid hydrolyzable polysaccharides, (3) ether soluble material, (4) cellulose, and (5) lignin. There were decreases in calcium, magnesium, manganese, and phosphorous. He concluded that .... "the change of the sugar-acid ratios of the fruit with advancement of physiological age was outstanding and most likely of practical significance for measurement of ripening."

Woodruff and Dewey (1959) reported highly significant correlations between the total sugar content, total

titratable acid content, and the sugar-acid ratio of Jersey blueberry fruit when correlated with percent breakdown of fruit held for 18 days at 40° F. and 6 days at 75° F. The sugar-acid ratio had higher correlation coefficients and lower standard errors with fruit deterioration than either sugar or acid alone. Also it was found that there was a direct correlation between soluble solids and percent sugar, as well as between pH and percent titratable acid. From this information they proposed a tentative harvest chart for Jersey blueberry fruit based on sugar-acid ratios and using soluble solids-pH ratio as a field test for determining whether berries that were blue in color were unripe, ripe or overripe.

This chart was based on a sugar-acid ratio of 12 as the dividing line between unripe and ripe fruit and a sugar-acid ratio of 17 as the division between ripe and overripe berries.

Woodruff (1959) found that Jersey berries having a ratio of 17 developed about 10 percent deterioration after 18 days at  $40^{\circ}$  F. and about 27 percent deterioration after 6 days at  $75^{\circ}$  F. He also reported that blueberries with sugar-acid ratios below 12 were unpalatable because of excessive tartness.

The sugar and acid changes of many crops are rather inconsistent from year to year so that these constituents are not particularly useful in deriving a maturity index.

This was characteristic for apples (Martin, 1954), grapes (Kasza, 1955; Shoemaker, 1935) and peaches (Haller, 1952). Hilgeman (1941) found that although the ratio of total soluble solids (Brix reading) to acid increased with maturity, it was unsatisfactory as a measure of palatability for grapefruit. He concluded that a combination of percent color, the Brix-acid ratio, and percent juice of grapefruit was a more suitable measure of maturity. Rygg and Getty (1955) tested sugars, acids, soluble solids, pH and sugar-acid ratios as a possible means for a maturity index of grapefruit and also found that the sugar-acid ratios were reliable; however, there was a frequent discrepancy between the ratio and palatability. Allen, et al. (1927) concluded that the soluble solids, as determined by Balling scale hydrometers, varied too greatly to be used as an index for maturity of plums. Fisher and Britton (1940), on the other hand, found a close correlation between soluble solids and the eating quality of Bing and Lambert cherries.

Factors other than maturation and ripening may affect the soluble solids and acids of fruit. For example, Hilgeman (1941) found that large grapefruit usually had a higher Brix to acid ratio than small fruit. An increase in heat units (calculated from mean daily temperatures above  $50^{\circ}$  F.) also caused an increase in the Brix-acid ratio. The application of nitrogen had no effect on total

soluble solids, but it tended to lower the acids of the fruit resulting in a higher Brix-acid ratio. Irrigation appeared to have no effect on the ratio. Lee and Sayre (1946) found that the total solids and acid content of tomatoes varied inversely with the soil moisture content. Also potassium increased the acid and total solids. Neither nitrogen, boron, manganese, and magnesium; nor the method of applying these fertilizers had any affect on either total solids or the acid contents of the tomato fruit. The studies of Caldwell (1928) show that a high sugar content of apples was associated with many hours of sunshine and high temperatures during the growing season. Kenworthy and Mitchell (1952) reported that soil management practices that increase the leaf nitrogen of Montmorency cherry trees cause a decrease in soluble solids content of the fruit. They also found that a deficiency in moisture supply at or prior to harvest increased the soluble solids of the cherry fruit. Ballinger, et al. (1958) found that as the total yield and percent nitrogen in the fruitingshoot leaves of blueberry bushes increased, the soluble solids of the fruit decreased.

Amerine and Roessler (1958) studied sampling methods for maturity testing of grapes in the vineyard. They used three methods of smapling: (1) single berries taken from individual clusters on a large number of vines, (2) single clusters taken from a smaller number of vines, and (3) all

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the fruit collected from a few vines. All samples were tested for degree Balling (soluble solids), Abbe refractometer readings (soluble solids), percent reducing sugar, percent total acidity, and pH. No significant differences were found in these constituents according to methods of sampling.

#### METHODS

The sugar and acid changes of blueberries associated with ripening were ascertained for berries of known physiological age. About the time of first commercial harvest when the berries were ripening in abundance, all of the berries that were blue in color were removed from the same 18 uniform Jersey and 18 uniform Rubel bushes that Woodruff (1959) used in his studies. The next day, on July 21, all berries that had become red or blue in color were individually labelled with 1" x 3/4" tags.

Two pints of tagged fruit of each variety were picked at random from the 18 bushes on the day of tagging, and subsequently at 4-day intervals for a period of 20 days.

Soluble solids readings of the fruit juice were taken in the field at the time of harvest with a Ziess Opton hand refractometer. The berries were selected at random from the harvested fruit, placed into a milk filter and mascerated by squeezing with the fingers. The juice for the reading was obtained by further squeezing the sample after several drops of juice had been discarded. Triplicate soluble solids readings were taken using 20 berries for each reading. The remaining berries were placed in plastic bags and packed in a cooler containing dry ice for transport to East Lansing where they were held at  $-10^{\circ}$  F. for later chemical analysis. Within six months after harvest the pH and percent total acid was determined. Total acids were determined for the frozen sample by the standard A.O.A.C. (1955) method. A 50 gm sample of frozen berries was blended with 150 ml of water in a Waring blender and then filtered through a single thickness of cheese cloth. A 100 ml aliquot was used to determine the pH and total acidity. An initial pH reading was made on the aliquot before titrating to an end point of pH 8.1 with 0.1 N NaOH. All samples were analyzed in duplicate. Total acid was expressed as percent citric acid.

Total sugars were determined by the Lane-Eynon (1923) method. A 50 gm sample of frozen berries was blended with 100 ml of H<sub>2</sub>O and then boiled for 30 minutes with 2 gms of calcium carbonate. The sample was then allowed to cool, transferred to a 500 ml volumetric flask, and made to volume. A 100 ml aliquot was transferred to a 200 ml flask. Three ml of saturated neutral lead acetate was added and the sample allowed to stand for 15 minutes after thorough shaking, made to volume, then filtered into a beaker containing 1 gm dry sodium oxalate and refiltered to remove excessive lead. A 50 ml aliquot was transferred to a 100 ml volumetric flask and hydrolyzed for 12 hours at room temperature with 5 ml concentrated HCl. The sample was then neutralized to a methyl orange end point using 3 N sodium carbonate. The excess carbon dioxide gas given off

from the sample often interferred with the measurement of samples for titration. This problem was eliminated for the last one-third of the samples by using 5 N NaOH instead of  $Na_2CO_3$ . After neutralizing to a methyl orange end point, the sample was made to 100 ml. A 20 ml aliquot was then added to 20 ml of standard Fehlings solution and titrated to a methylene blue end point with 0.5 percent dextrose solution. All samples were analyzed in duplicate.

Samples of commercially packed Jersey berries for testing the harvest chart proposed by Woodruff and Dewey (1959) were obtained from grower packing sheds and from the Michigan Blueberry Growers Association warehouse at Grand Junction. Each sample consisted of twelve pints of berries packed in pulp pint tills and covered with transparent film for fresh market outlets.

Soluble solid and pH readings were taken for each flat at the time the berries were obtained. Approximately 20 berries were taken at random from the 12 pints and squeezed in a milk filter pad to obtain several drops of juice for the refractometer. Fifty gms of berries, selected at random from the flat, were blended with 100 ml of  $H_20$  and tested for pH with a portable meter. All samples were run in duplicate.

The remaining berries were placed the same day in holding rooms at Michigan State University. Four pints were held at 75° F. for 6 days and 8 pints held at 40° F. for 18

days prior to examination for deterioration and decay.

Woodruff (1959) found that breakdown consisted of stem scar decay and "runny soft" berries. Stem scar decay was characterized by a slight indentation at the stem end with some browning of the skin and flesh occurring. The flesh of "runny soft" berries was completely disintegrated, and upon slight pressure from the fingers the berries would completely fall apart. There was some mycelium associated with the stem scar decay but no indication of secondary infection could be found with "runny soft" berries. Shelflife was expressed as percent breakdown on a fresh weight basis. Shriveled fruit was not considered as breakdown.

Five plots of Rubel bushes, one at each of five locations, were selected for study of the effect of time of initial harvest upon the sugar-acid ratio of ripe berries. Each plot consisted of 20 full grown, vigorous bushes from which no fruit had yet been harvested during the current season.

Five pickings at four day intervals were made at each plot. Berries were harvested from four bushes at each of the harvest dates, thus each harvest represented an initial picking. The harvest dates were initiated as soon as 11 pints could be obtained from the four bushes. The first initial picking of the Allegan plot was four days later than the plot at Bangor, the plot at Lacota four days later than the plot at Allegan, the Douglas plot eight days later

than Lacota, and the Holland plot four days later than that at Douglas.

Clusters of berries were selected at random for harvest of berries blue in color. The sample included berries that had ripened throughout the season up to the time at which the harvest was made. Berries were picked into standard blueberry picking pails and randomly poured into 11 pint boxes. The fruit of one pint was sealed in a plastic bag for later chemical analysis. These samples were refrigerated with dry ice until placed in the freezer room at East Lansing late the same day. On the day of harvest six pints were placed at  $40^{\circ}$  F. for 18 days and 4 pints were placed at  $75^{\circ}$  F. for 6 days for evaluation of susceptibility to breakdown and decay.

Three soluble solid readings were taken at the time of harvest using 20 berries for each reading. Total sugar, total acid and pH was determined from the frozen samples, as described previously.

#### RESULTS

The sugar changes of Jersey and Rubel fruit, which had been tagged at the time of development of red coloration and harvested periodically for 20 days, are shown in Figure 1. The berries of both varieties had marked increases in sugar between 0 and 4 days after red coloration of the skin of the fruit. There were further increases until 12 days, at which time they leveled off and remained fairly stable for the next eight days.

The acids showed a pattern similar to the sugar except that they decreased with ripening (Figure 2). There was a sharp decrease in acidity the first eight days in both varieties and then a gradual leveling off until the twentieth day.

Both sugars and acids were higher in fruit of the Rubel variety than in the Jersey fruits.

The sugar-acid ratios at four-day intervals calculated from the above sugar and acid contents are given in Table 1. The ratios in Jersey increased from 0 to 16 days then leveled off, but showed a further increase between 16 and 20 days after coloration.

Usually the ratios were higher for Jersey fruit than for Rubel fruit, especially at eight days and longer after red coloration. This is clearly demonstrated in Figure 3 by a steeper line of the sugar-acid ratio increase with time of harvest. FIGURE 1. Changes in the Total Sugar Content of Rubel and Jersey Blueberry Fruit Harvested 0, 4, 8, 12, 16, and 20 Days After the Development of Red Coloration of the Skin.

FIGURE 2. Changes in the Titratable Acid Content of Rubel and Jersey Blueberry Fruit Harvested O, 4, 8, 12, 16, and 20 Days After the Development of Red Coloration of the Skin.

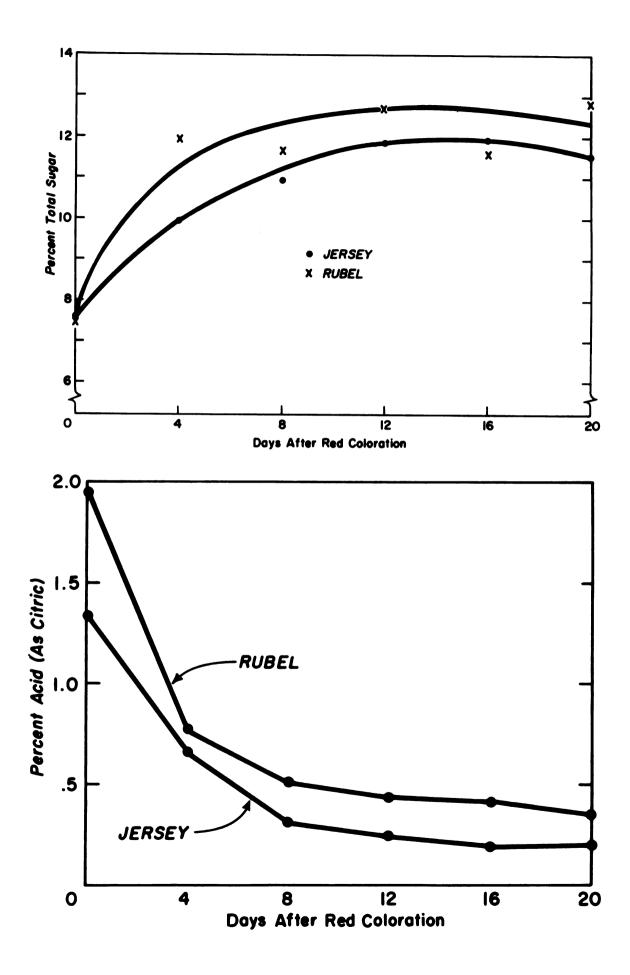
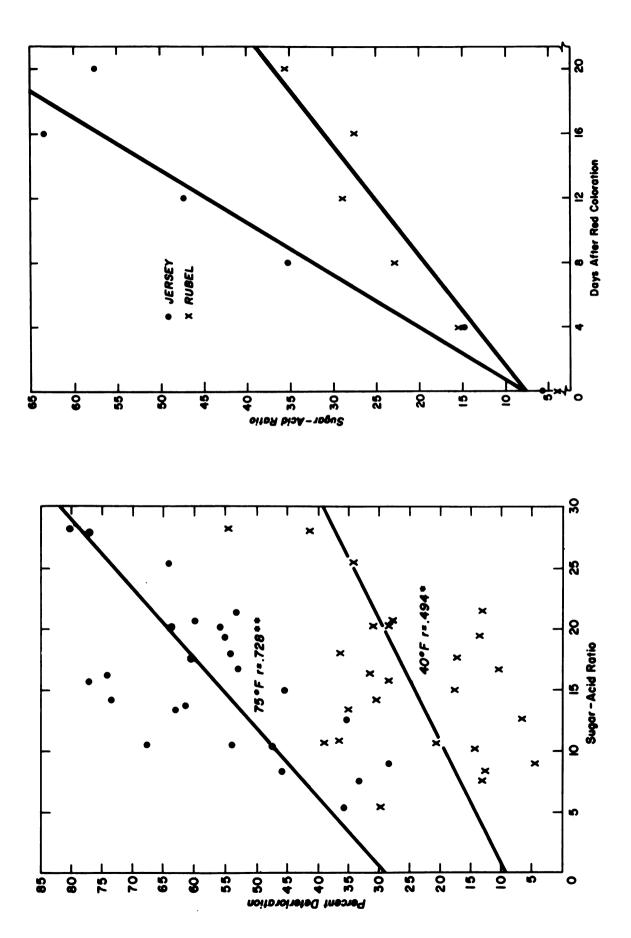


FIGURE 3. The Relationship of Sugar-Acid Ratio Changes to Days After Red Coloration for Rubel and Jersey Blueberry Fruit.

FIGURE 4. The Linear Relationship of Deterioration and the Sugar-Acid Ratios of Rubel Blueberry Fruit. (D.F. = 23; at 75° F., Y = 29.32 + 1.73x; at 40° F., Y = 9.15 + 1.01x).



| TA | BI | E | 1 |
|----|----|---|---|
|    |    |   |   |

Changes in pH, Soluble Solids, Total Sugar, Titratable Acid, and Sugar-Acid Ratio During Ripening of Rubel and Jersey Blueberry Fruit.

| Days after | Percent | Percent | pH        | Percent    | Sugar- |
|------------|---------|---------|-----------|------------|--------|
| red        | soluble | total   |           | titratable | acid   |
| coloration | solids  | sugar   |           | acid       | ratio  |
|            |         | Jerse   | <u>y</u>  |            |        |
| 0          | 12.4    | 7.6     | 2.98      | 1.34       | 5.7    |
| 4          | 13.9    | 9.9     | 3.32      | .67        | 14.8   |
| 8          | 13.6    | 10.9    | 3.73      | .31        | 35.2   |
| 12         | 14.6    | 11.8    | 4.16      | .25        | 47.2   |
| 16         | 14.3    | 11.9    | 4.29      | .19        | 62.6   |
| 20         | 13.6    | 11.5    | 4.18      | .20        | 57.5   |
|            |         | Rube    | <u>el</u> |            |        |
| 0          | 12.8    | 7.5     | 2.82      | 1.95       | 3.9    |
| 4          | 13.3    | 11.9    | 3.30      | .77        | 15.5   |
| 8          | 14.1    | 11.7    | 3.59      | .51        | 22.9   |
| 12         | 15.3    | 12.7    | 3.74      | .44        | 28.9   |
| 16         | 14.5    | 11.6    | 3.85      | .42        | 27.6   |
| 20         | 13.8    | 12.8    | 3.89      | .36        | 35.6   |

Berries harvested 0 and 4 days after coloration were tart and unpalatable to the taste. Some of the berries started to shrivel on the bush at 12 days after red coloration.

The pH and soluble solids of the extracted juice of Jersey fruit obtained from commercial packs were determined for possible prediction of shelf-life using the harvest chart proposed by Woodruff and Dewey (1959). These data, tabulated by degree of ripeness as estimated from the chart, together with fruit breakdown upon holding these lots at  $40^{\circ}$  F. and 75° F. are given in Table 2.

No consistent relationship of ripeness to percentage breakdown seemed apparent at either temperature. Estimated sugars and estimated acids were then determined with the conversion charts presented by Woodruff and Dewey (1959) for calculation of the estimated sugar-estimated acid ratios for each sample (Table 2). Correlation analysis showed no relation of estimated sugar-estimated acid to deterioration for berries held at  $40^{\circ}$  F. for 18 days, but there was a significant correlation for berries held at  $75^{\circ}$  F. for 6 days (Table 3).

Estimated sugar, estimated acid, soluble solids and soluble solids-pH ratio were also compared with percent breakdown (Table 3). The only significant relationships were between estimated percent sugar and percent breakdown (r = -.375) for berries held at 40° F. for 18 days, and soluble solids and percent breakdown (r = -.383) for berries

## TABLE 2

The pH, Soluble Solids and Estimated Sugar-Estimated Acid Ratio at Harvest and Percent Breakdown After Holding for 6 Days at 40° F. and 18 Days at 75° F. of Commercially Packed Jersey Blueberry Fruit in Relation to Ripeness

| Ripeness             |      |         | Ratio of es-  |                       |           |
|----------------------|------|---------|---------------|-----------------------|-----------|
| according            |      | Percent | timated sugar | Percent               | breakdown |
| to matur-            |      | soluble | to estimated  | 6 days                | 18 days   |
| ity index <u>a</u> / | pH   | solids  | acid          | at 75 <sup>0</sup> F. | at 40°F.  |
| UNRIPE               | 3.06 | 12.6    | 7.3           | <b>45.7</b>           | 30.9      |
|                      | 3.18 | 12.9    | 9.5           | 40.6                  | 18.3      |
|                      | 3.11 | 12.9    | 8.7           | 59.3                  | 35.0      |
|                      | 3.23 | 12.8    | 10.1          | 74.9                  | 64.0      |
|                      | 3.25 | 13.0    | 10.5          | 43.8                  | 37.1      |
|                      | 3.08 | 14.1    | 8.9           | 39.2                  | 24.1      |
|                      | 3.20 | 13.6    | 10.2          | 18.2                  | 9.9       |
|                      |      | Averag  | e 9.3         | 46.0                  | 31.3      |
| RIPE                 | 3.37 | 14.0    | 13.3          | 49.2                  | 10.1      |
|                      | 3.24 | 14.0    | 11.2          | 34.2                  | 15.1      |
|                      | 3.41 | 14.6    | 14.5          | 37.5                  | 18.6      |
|                      | 3.41 | 13.6    | 13.6          | 39.9                  | 11.2      |
|                      | 3.47 | 12.5    | 13.2          | 44.1                  | 17.5      |
|                      | 3.29 | 14.5    | 12.2          | 37.5                  | 20.2      |
|                      | 3.45 | 12.4    | 13.4          | 58.5                  | 20.1      |
|                      | 3.47 | 13.5    | 14.8          | 57.0                  | 23.5      |
|                      | 3.42 | 12.7    | 13.2          | 10.6                  | 4.1       |
|                      | 3.54 | 12.6    | 15.5          | 38.2                  | 27.1      |
|                      | 3.21 | 14.0    | 10.5          | 19.0                  | 8.4       |
|                      |      | Averag  | e 13.2        | 38 <b>.7</b>          | 16.0      |
| OVERRIPE             | 3.78 | 14.6    | 24.7          | 66.4                  | 20.1      |
|                      | 3.61 | 14.3    | 22.7          | 36.6                  | 10.9      |
|                      | 3.66 | 14.3    | 20.3          | 29.9                  | 5.0       |
|                      | 3.65 | 14.4    | 20.1          | 57.1                  | 18.0      |
|                      | 3.75 | 13.9    | 22.7          | 58.6                  | 28.1      |
|                      | 3.84 | 13.4    | 25.1          | 70.0                  | 28.3      |
|                      | 3.72 | 14.1    | 22.0          | 37.3                  | 8.8       |
|                      | 3.60 | 13.9    | 18.0          | 51.6                  | 18.1      |
|                      | 3.77 | 13.3    | 22.2          | 54.2                  | 14.4      |
|                      | 3.94 | 12.9    | 27.5          | 29.3                  | 14.4      |
|                      | 3.46 | 14.6    | 15.9          | 32.3                  | 13.6      |
|                      | 3.62 | 12.9    | 17.6          | 62.1                  | 25.2      |
|                      | 3.94 | 13.1    | 27.8          | 46.4                  | 21.6      |
|                      | 3.75 | 12.6    | 20.5          | 81.5                  | 33.9      |
|                      | 4.32 | 13.4    | 14.7          | 78.1                  | 38.3      |
|                      |      | Averag  | e 21.5        | 52.8                  | 19.9      |

a/ Index proposed by Woodruff and Dewey (1959).

## TABLE 3

The Relation of Deterioration to Fruit Constituents Ascertained by Soluble Solids and pH Measurements for Commercially Harvested Jersey Blueberries Held 18 Days at  $40^{\circ}$  F. and 6 Days at  $75^{\circ}$  F.

|   | FRUIT<br>18 days at        | -                   | ION UPON HOLI<br>6 <b>days</b> at |                     |
|---|----------------------------|---------------------|-----------------------------------|---------------------|
| Fruit<br>constituents   | Correlation<br>coefficient | S.E. of<br>estimate | Correlation<br>coefficient        | S.E. of<br>estimate |
| Soluble solids (percent)  | -0.383*                    | 11.20               | -0.111                            |                     |
| Soluble solids-   | .180                       |                     | .196                              |                     |
| H-ion (ratio)<br>Estimated sugar<br>(percent) a/                      | 375*                       | 11.24               | .210                              |                     |
| Estimated acid  | .125                       |                     | •342                              |                     |
| (percent) <u>a</u> /<br>Estimated sugar-<br>estimated acid<br>(ratio) | .023                       |                     | <b>.</b> 394 <b>*</b>             | 15.88               |

\*Significant at the 5 percent level.

a/ Estimated according to method of Woodruff and Dewey (1959).

held at the same temperature.

The Rubel fruit of the maturity plots at five locations was harvested in a commercial manner in that all blue colored fruit was taken for the experimental sample. Overall ripeness was varied by a sequence of initial harvest dates. The sugars and acids of these samples at harvest and their subsequent deterioration upon holding at  $40^{\circ}$  F. and  $75^{\circ}$  F. are presented in Table 4.

The sugar content of the fruit varied considerably according to harvest date and there was no consistent pattern of increase with time, as was characteristic of the tagged fruit. The acid content of the fruit of all plots tended to decrease as the time of initial harvest was delayed.

The sugar-acid ratios increased throughout the harvest season for fruit from three of the five plots tested. The fifth picking at Bangor and the second picking at Holland were exceptions. For the latter, a marked increase in the ratio followed the decrease at the second harvest date. All sugar-acid ratios fell within the range of sugar-acid that occurred between 0 and 12 days after red coloration for the tagged Rubel berries.

Correlation analysis of percent breakdown at both temperatures to sugars, acids, and their ratios are presented in Table 5. The sugar-acid ratio and percent breakdown at 40° F. and 75° F., as illustrated in Figure 4 (page 16), were significantly related. Otherwise, only percent acid

| 4     |  |
|-------|--|
| TABLE |  |
|       |  |

Sugar and Acid Contents, Sugar-Acid Ratios, and Breakdown The Soluble Solids. pH.

| The Soluble<br>of Rubel Bl | Soluble Solids,<br>Rubel Blueberries  | ph, sugar<br>3 Harveste | ana acı<br>d Period | ically                     | ents, out  | Location | ns in Western  | rn Michigan.         |
|----------------------------|---|-------------------------|---------------------|----------------------------|------------|----------|----------------|----------------------|
|                            | Date of   | Percent<br>soluble      | Percent             |                            | Percent    | Sugar:   | Ferc<br>6 days | Breakdown<br>18 days |
| Plantation                 | harvest   | solids                  | sugar               | μd                         | -1 1       | 5        | <u></u>        | - #C                 |
|                            | Dilv 9  |                         | •                   | 8                          | 9          | ٠        | 2.             | 29.9                 |
|                            | -   |                         | •                   | •                          | 1.30       | 7.6      | 33.2           | 5                    |
| Ren con                    | 170   |                         | •                   | -                          | ਼          | ۲        | <b>2</b>       | NI I                 |
| Daugo.                     | 1   |                         | •                   | 4                          | 5          | ີ່ຄ      | ~              | ω.                   |
|                            | 8<br>8<br>8<br>1<br>9<br>1<br>9<br>1<br>9<br>1<br>9<br>1<br>9<br>1<br>9<br>1<br>9<br>19<br>19<br>19<br>19 | 14.8                    | 0                   | 3.52                       | .67        | •        | 3              | •                    |
|                            | דו יוויד  | 5                       | 6                   | <u></u>                    |            | •        |                | 39.1                 |
|                            |   | 20                      | N                   | 4                          | Ô          | 3        | ÷              | •<br>•               |
| 2000 C C C                 | 10  | - α                     | 2                   | 9                          | 9          | 0        | .0             | Ļ.                   |
| TTABAILA                   | 1 K<br>2 O  | ) r                     | 2 0                 | Ő                          |            | 28.0     | 77.1           | -                    |
|                            | 0 0<br>2 0<br>2   | 15.8                    | 11.9                | 3.97                       | .42        | ω.       | 2              | 4                    |
|                            | 1   | L u                     | -                   | C                          | 2          | •        | ω              | 4.                   |
|                            | LT ATNC   | •                       | •                   | ) –                        | 20         | 0        | 67.9           |                      |
| -                          |   |                         | ວັດ                 | 5                          | 9          | 6        | į.             | -                    |
| Lacota                     |   | •<br># ব                | 2                   | )4                         |            | •        | 4              |                      |
|                            |   |                         | 11.5                | 3.51                       |            | •        | 3.             | C.03                 |
| 1                          | Aug. ~  | •                       |                     |                            |            |          | -              |                      |
|                            | Julv 25   |                         | 0                   | 0.<br>10<br>10<br>10       | 00         | 12.01    | 35.5           | .0                   |
|                            |   | •                       | •                   | •                          |            |          | ີ<br>ເລ        | -                    |
| חחוות אי                   | AU2. 2  | •                       | -                   | ົ່                         |            |          | 4              | ٠                    |
| <b>DO4644</b>              | 9   | 15.1                    | ٠                   | ຸດ                         |            |          | С              | ອ                    |
|                            | 10  | •                       | i.                  | •                          |            | 5        |                | ١.                   |
|                            |   | Ι.                      | -                   | 4                          | 9          | 0        | 53.1           | 10.5                 |
|                            |   |                         | •                   |                            | Ś          | 2        | м.             | •                    |
|                            | Aug. 2  | 6                       | ຄໍ່ດ                | ዛሪ                         | ) (C       | 5        | ີ<br>ເມ        | ເ                    |
| Holland                    |   | -                       |                     | יאר                        | • 0 •      |          | 3.             |                      |
|                            | <b>1</b> 0  | 16.7                    | 7.º 2T              | 0<br>0<br>0<br>0<br>0<br>0 | • 46<br>46 | 25.4     | 4.             | 4                    |
|                            | 14  | <u>.</u>                | -                   | •                          |            | - 1      |                |                      |

and percent breakdown after six days at 75° F. were related.

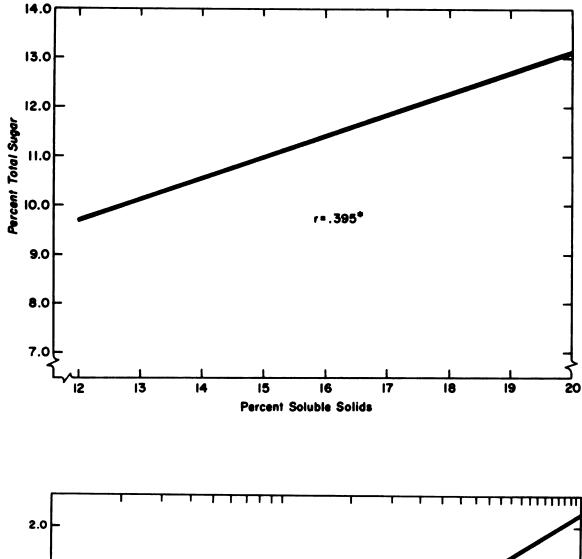
Since sugar and acid contents can be readily determined by measurement of soluble solids and pH, respectively, for Jersey berries (Woodruff and Dewey, 1959), these measurements were tested for Rubel. There was a significant correlation coefficient of 0.395 and a standard error of estimate of  $\pm 1.28$  (Figure 5). There was a high correlation coefficient of pH to percent titratable acid; it was 0.979 with a standard error of estimate of  $\pm 0.07$  (Figure 6).

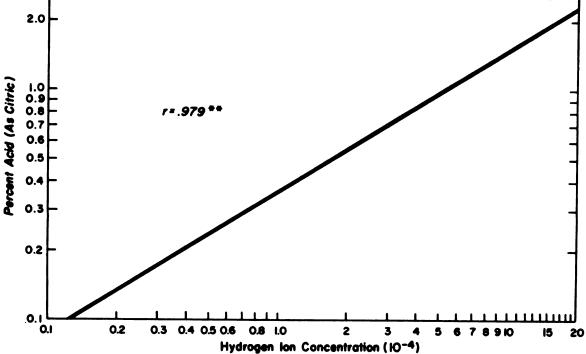
As noted in Table 5, estimated sugar as calculated from Figure 5, was not related to percentage breakdown at either temperature. However, the estimated acid (determined from Figure 6) was significantly related to percentage breakdown of berries held 18 days at 40° F. ( $\mathbf{r} = -.502$ , S.E. = ±11.01) and highly significantly related to percent breakdown of fruit held six days at 75° F. ( $\mathbf{r} = -.754$ , S.E. = ±9.72). The ratios of estimated sugar and estimated acid are correlated to percentage breakdown in Table 5. Highly significant correlation coefficients existed for fruit held at 40° F. (0.593, S.E. ±10.25) and at 75° F. (0.686, S.E. ±10.76).

Since soluble solids and pH might serve as field methods of determining sugars and acids, their ratio was compared to breakdown of the fruit after harvest. Highly significant coefficients were found to exist between the ratios and percent breakdown upon holding the fruit 18 days at  $40^{\circ}$  F. (r = .635, S.E. = ±9.83) and six days at  $75^{\circ}$  F. (r = .636,

FIGURE 5. The Linear Relationship of Total Sugar and Soluble Solids of Rubel Blueberry Fruit. (D. F. = 29, Y = 4.63 + .42x)

FIGURE 6. The Linear Relationship of Acid (Expressed as Citric) and Hydrogen Ion Concentration of Rubel Blueberry Fruit. (D. F. = 29; Y = 0.25 + 0.11x)





## TABLE 5

The Linear Correlation Coefficients of Sugar and Acid Measurements to the Breakdown of Rubel Blueberry Fruit Held 18 Days at 40° F. and 6 Days at 75° F.

|                                    | FRUIT DETERIORATION        |                     |                            |                     |
|------------------------------------|----------------------------|---------------------|----------------------------|---------------------|
|                                    | 18 Days a                  | t 40° F.            | 6 Days at                  | 75 <sup>0</sup> F.  |
| Index                              | Correlation<br>Coefficient | S.E. of<br>Estimate | Correlation<br>Coefficient | S.E. of<br>Estimate |
| Percent sugar                      | 0.109                      |                     | 0.277                      |                     |
| Percent acid                       | 389                        |                     | 704**                      | 10.51               |
| Sugar:Acid                         | •49 <b>4</b> *             | 11.07               | •728 <b>**</b>             | 10.15               |
| Soluble solids                     | .283                       |                     | .273                       |                     |
| Soluble solids:<br>H-ion           | •635 <sup>**</sup>         | 9.83                | •636 <sup>**</sup>         | 11.43               |
| Estimated sugar                    | .286                       |                     | .279                       |                     |
| Estimated acid                     | 502*                       | 11.01               | 754**                      | 9.72                |
| Estimated sugar:<br>Estimated acid | .593**                     | 10.25               | •686**                     | 10.76               |

\*Significant at the 5 percent level. \*\*Significant at the 1 percent level. S.E. =  $\pm$ 11.43). Soluble solids alone were not related to breakdown of berries held at either temperature. Since the pH was so highly correlated to percent acid, correlation analysis between pH and breakdown were not run.

#### DISCUSSION

The marked changes in the sugar and acid content of Rubel and Jersey fruit during the four days following the development of red coloration in the skin indicate that the ripening processes are rapid at this time. The maximum level of sugar and the minimum acid at 8-12 days following red coloration suggests that full ripeness was then attained. Berries picked at eight days were of acceptable market quality, as determined by taste; whereas those picked at 0 and 4 days were unsatisfactory. Many berries at 0 days were still red in color and excessively tart to the taste. Even though berries at four days were blue in color, they were still unpalatable because of tartness.

Changes in sugars and acids followed a pattern similar to those recorded by Woodruff (1959). However, sugars and acids in Jersey were lower in 1959 than in 1958, and the overall change in these constituents was less in 1959 than in the previous year. These yearly differences slightly affected the sugar-acid ratios so that in 1959 the sugaracid ratios were higher and increased more rapidly with time and were indicated by a steeper line than in the previous year. The sugar-acid ratios of the berries of the Rubel variety were the same at 0 days, higher at 4 and 8 days, and lower at 12, 16, and 20 days after red coloration in

1959 than in 1958. Therefore, the slope of the line of sugaracid ratios to time had a smaller angle from the horizontal axis in 1959.

There was rainfall on 9 days during the 1959 harvest period and on 6 days in the 1958 season. The average maximum and minimum temperatures were  $79^{\circ}$  F. and  $60^{\circ}$  F. during the testing period in 1958, and 80° F. and 62° F. in 1959. These small differences in climatic conditions during the harvest periods would probably not account for the seasonal variations in sugars and acids. However, other factors which were not measured, such as number of hours of sunshine. berry size, yield, pruning practices, nutritional status. and irrigation may have affected these constituents. Caldwell (1928) found that the total sugar content of an apple crop was highest in the year that received the largest number of hours of sunshine. Hilgeman (1941) reported that large grapefruit had higher soluble solid contents than smaller fruit. It has been shown that the soluble solids content of blueberry fruit is affected by yield and nitrogen content of the fruiting-shoot leaves (Ballinger, et al., 1958). Also blueberries from thick wood are considerably larger than berries from thin or medium wood (Shutak, et al., 1957). Lee and Sayre (1946) found that total solids and acid content of tomatoes varied with soil moisture and the amount of potassium applied. Soluble solids in the fruit of the sour cherry also are affected by soil moisture and nutrient status

of the trees (Kenworthy and Mitchell, 1952).

Varietal differences in sugars and acids found by Woodruff in 1958 were confirmed in 1959. In both years the Rubel fruit had a higher acid content, and usually a slightly higher sugar content than Jersey (Figures 1 and 2). These resulted in lower sugar-acid ratios for the Rubel variety than for Jersey (Figure 3).

The general agreement of the sugar-acid ratios obtained in these tests to those obtained by Woodruff for 1958 fruit of known physiological age is good evidence that the ratio should be valid for measuring the degree of ripeness of the fruit. The chart devised by Woodruff and Dewey (1959) for utilizing the ratio of sugar and acid contents as a measure of ripeness, however, did not prove satisfactory for commercially harvested fruit (Table 2). The low correlation values of the ratio to deterioration of the fruit upon holding at  $40^{\circ}$  F. and 75° F. (Table 3) indicate that the ratio either did not measure the degree of ripeness of these commercially harvested berries, or that deterioration was affected by factors in addition to ripeness.

The percent deterioration of Jersey fruit was considerably higher in 1959 than in 1958 even though the ratios of estimated sugar to estimated acid was lower this year than the actual sugar-acid ratios were the previous year.

It is likely that variations in the methods of harvesting and handling were primarily responsible for the failure to relate sugar:acid to ripening. There was no control over the source of fruit so that the fruit in a commercial flat containing 12 pints could have been harvested from one bush or many bushes. Also there was no control on the lengths of time elapsed between harvesting and testing of the various fruit samples for soluble solids and pH, and between harvesting and placing the samples in holding rooms. These factors, especially on days of high temperatures, could have seriously affected the accuracy of the tests.

The care exercised in selecting berries by pickers during the harvest operation could have greatly affected the subsequent extent of fruit breakdown. For example, fruit harvested by a careful picker who excluded berries having a reddish tinge and soft, overripe fruit could have the same sugar-acid ratio as berries picked by a careless person who included these unripe and overripe fruit in the pack. The latter lot, however, would likely develop a higher percentage of deterioration due to the greater quantity of overripe berries.

Another variable could have been the method in which the pickers handled the berries; for instance, some pickers compress the berries more than others during the harvesting procedure. Berries with skin breaks or juice exudation at the stem scar are more susceptible to decay and breakdown than berries handled with greater care.

It is suspected also that the method of determining percent deterioration could have contributed to the inconsistent results for the two years. It is thought that stem scar decay is a secondary infection rather than a primary breakdown as classified for these experiments. Furthermore, the determination of breakdown was subjective. In the future, when determining deterioration, breakdown and decay should be classified separately.

The correlation coefficient of soluble solids to sugar was not as high for Rubel fruit in these studies as reported by Woodruff and Dewey (1959) for Jersey (r = .395 vs r = .619); however, the correlation coefficient of H-ion to percent acid was about the same for the two years (r = .979; vs r = .916). Apparently soluble solid readings do not yield a consistent measurement of sugar content for the blueberry fruit. It was found that soluble solids was within three percent of the sugar content of tagged berries (Table 1), but ranged up to seven percent greater than the sugar of berries of mixed degrees of ripeness (see Table 4). Woodruff (1959) found that soluble solids of blueberries was 2-3 percentage points higher than sugar, and Allen (1932) reported that the soluble solids content of plums, peaches, and pears was approximately twice that of total sugars. Removal of the non-sugar constituents of soluble solids, perhaps by their precipitation from the fruit juice with cation and anion resins, would make soluble solids a more accurate measure for total sugars.

It should be pointed out, also, that the chart was devised for determining the proper time of harvest from random field samples of fruit, rather than for classifying fruit already harvested by commercial pickers, as was done in these tests.

A harvest index for the Rubel variety based on the sugar to acid ratio of the fruit seems feasible. Fruit picked at a sequence of initial dates at five locations had increasing ratios and amounts of deterioration upon holding as the time of initial harvest was delayed (Table 4). Although deterioration at both 40° F. and 75° F. was significantly correlated to sugar-acid ratio (Figure 4), a higher correlation value existed for the higher holding temperature. It is believed that the fruit held at 75° F. had a higher amount of "runny soft" breakdown of the berries, whereas, much of the deterioration at 40° F. was decay at the stem scar. As discussed above, this type of deterioration may not be associated necessarily with ripeness of the berries at harvest. The Rubel fruit harvested from the maturity plots showed greater amounts of breakdown than was found for the Jersey variety in the previous year. No records of the types of deterioration were obtained.

Since this study confirmed that the sugar-acid ratio is definitely associated with shelf-life under controlled methods of harvest, it was desirable to explore more simple or rapid methods of determining this ratio. It was found that estimated sugar-estimated acid ratios, as well as soluble solid-

estimated acid ratios, were directly correlated to percent breakdown of Jersey fruit (Woodruff and Dewey, 1959). Since estimated sugar and estimated acid were determined from soluble solids and H-ion readings, respectively, it would seem logical that the ratio of these two measurements are closely related to breakdown. Accordingly, the ratio of soluble solids to H-ion concentration was correlated with the percentage breakdown. Highly significant correlations were found to exist between these ratios and percent deterioration of fruit held 18 days at 40° F. and 6 days at 75° F., with r equal to 0.635 and 0.636 respectively (Table 5).

Since percent sugar could be estimated from soluble solids (Figure 5) and percent acid from pH (Figure 6), the ratio of these two estimates were tested with percent breakdown for Rubel. Highly significant relationships existed between the estimated sugar-estimated acid ratios and percent breakdown after 18 days at 40° F. ( $\mathbf{r} = 0.593$ ) and 6 days at 75° F. ( $\mathbf{r} = 0.686$ ). These findings are in agreement with those of Woodruff and Dewey (1959) for the Jersey variety.

Sugars alone, whether expressed as percent total sugar, soluble solids, or estimated percent sugar, were not related to percent breakdown (Table 5). However, percent of actual acid was highly related to percent deterioration after 6 days at 75° F.; and estimated acid (from pH) was significantly associated with percent breakdown after 18 days at  $40^{\circ}$  F. and significantly associated at the 1 percent level with

breakdown after 6 days at 75° F. (Table 5).

The highest correlation coefficient (and the lowest standard error of estimate) occurred for the relation of estimated acid and deterioration at  $75^{\circ}$  F., however, the r value for estimated acid and deterioration at  $40^{\circ}$  F. was significant only at the 5 percent level. The ratios of sugars to acids, whether determined as soluble solids:H-ion, or as estimated sugar:estimated acid were highly significant at both temperatures. The ratios, therefore, were of higher accuracy than single factors for estimating the degree of fruit ripeness.

Soluble solids and pH instrument readings determined in the field can be used directly without further calculation for determining the ratio. A harvest chart based on these readings, as devised earlier for the Jersey variety (Woodruff and Dewey, 1959) seems suitable for the Rubel variety. Before a chart is recommended, however, further refinements of the Jersey chart and additional tests with Rubel are desirable.

#### CONCLUSIONS

The continued ripening of Rubel and Jersey blueberry fruit subsequent to the appearance of red color in the fruit skin was related in a linear and positive manner to the ratios of sugar and acid contents of the fruit.

The harvest index chart previously developed for the Jersey variety did not prove to be a satisfactory measure of ripeness and subsequent deterioration for blueberries already harvested and packed by commercial methods. Apparently, its application is limited to use in the field for determining the time of harvest.

The ratios of total sugar to total acid, soluble solids to H-ion, and estimated sugar to estimated acid of Rubel blueberry fruit at various dates of initial harvest were statistically related to fruit deterioration upon holding at  $40^{\circ}$  F. and  $75^{\circ}$  F. Since the soluble solids and H-ion of the fruit juice can be easily determined in the field, a time of harvest chart based on a ratio of these measurements seems feasible. Further refinements of the Jersey harvest chart and additional tests with Rubel berries are recommended before a Rubel harvest chart is devised.

#### SUMMARY

Jersey and Rubel blueberry fruit of known physiological age in respect to ripening were analyzed for sugar and acid contents. It was found that the ratios of these constituents were related in a linear and positive manner for 20 days subsequent to the time of red color development in the fruit. Rubel berries had higher contents of total sugars and titratable acids than Jersey berries at comparable stages of ripeness.

The harvest index chart for Jersey berries based on the sugar to acid ratio of the fruit proposed by other workers did not prove suitable for predicting the shelf-life of fruit already harvested and packed by commercial procedures. Correlation analysis showed no relation of the estimated sugar content (by soluble solids reading) to estimated acid content (by pH) ratio of these berries to the amounts of breakdown upon holding at 40° F. for 18 days; there was a significant correlation of this ratio to breakdown for berries held at 75° F. for 6 days. Variations in methods and times of picking and handling were suspected to be responsible for the failure of the chart for this purpose. These difficulties would not be encountered when the chart is used, as recommended, for determining the time to begin harvest operations.

Samples of Rubel blueberry fruit containing berries of variable degrees of ripeness were obtained in a sequence of initial harvests of bushes from five commercial plantations in Western Michigan. The fruit soluble solids, determined at harvest, and total sugars, titratable acids, and pH, determined from berries frozen immediately after harvest, were related to deterioration of the fruit when held for 6 days at 75° F. and 18 days at 40° F. Significant linear and positive correlations occurred for sugar-acid, soluble solid-H-ion, and estimated sugar-estimated acid ratios to fruit breakdown at both temperatures. A harvest chart for the Rubel variety utilizing field tests for soluble solids and pH is conceivable. Further testing, however, is recommended before such a chart is devised.

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