THE DETERMINATION OF SUCROSE IN MIXED JUICE AND CLEAR JUICE BY VARIOUS METHODS

By P. J. LAUBACHER.

Introduction.
Any modification of the Clerget method of determining sucrose in juices can only be correct if there are no substances present in the juice that have different optical activities under the conditions at which the direct and invert polarizations are read, and if no optically active substances are formed from substances other than sucrose during the inversion process.

The question has often been asked whether the Jackson and Gillis No. 4 method, which is the official Natal method, gives correct results under all circumstances. It has been suggested that certain non-sugars characteristic for Natal juices might influence the result, and that abnormal fluctuations sometimes observed in the Boiling House Recovery data of Natal mills should be ascribed to this influence. In the last season, an attempt was made to answer this question.

Method of checking the Sucrose determined by Jackson and Gillis Method No. 4.

To determine whether the Jackson and Gillis No. 4 method of sucrose determination, as described in the "Recommended Methods of Chemical Control" of the South African Sugar Technologists' Association, was sometimes in error, this method was compared with the rapid invertase method, as described by Browne and Zerban in "Sugar Analysis," pp. 433-434, and also with the chemical method based on the determination of reducing sugars in the juice before and after inversion, as described by Douwes Dekker in the South African Sugar Journal, March 1950, p. 165. In addition, the apparent sucrose (direct polarization) and sucrose by the baryta method were also determined.

Sucrose was determined by the baryta method at a few mills for comparison with the other methods mentioned, at the request of the management of Umfolozi. The baryta method of sucrose determination is used by Umfolozi's test laboratory and is as follows:

100 ml. of de-leaded clarified juice is boiled for exactly 5 minutes with 2 gms. of baryta under reflux. After cooling, the pH is brought to 4.9 ± 0.1 with acetic acid, 5 drops of alumina cream are added and the volume is made up to 200 ml. The solution is then filtered and polarized.

The sucrose determinations were done at all factories at different periods of the season in both mixed juice and clear juice.

To convert the readings for the direct and invert polarizations in the Jackson and Gillis method to the polarization of the sucrose present in the solution, the tables in the "Recommended Methods of Chemical Control" were used. In the invertase method the polarization of the sucrose in the solution was determined by using the formula:

\[ \text{Polarization} = \frac{P - I}{142.1 + 0.073(m-13) - \frac{t}{2}} \]

where \( P - I \) = Arithmetical sum of the direct and invert polarizations corrected for the polarization of the invertase used.

\( m \) = Number of grams of solids present per 100 ml. of the invert solution.

\( t \) = Temperature in degrees centigrade.

All determinations were done in duplicate and all glassware used was standardized at the Sugar Milling Research Institute. The solutions used were all standardized periodically.

Results.

The results of the sucrose determination are given in Tables I and II.

Discussion of Results.

In Tables I and II the columns \((b-d)\) give the differences in percentage sucrose by the invertase method and the percentage sucrose by the Jackson and Gillis method No. 4. The mean, maximum and minimum differences \((b-d)\) were +0.043 per cent., +0.145 per cent. and -0.05 per cent. respectively for mixed juice, and +0.056 per cent., +0.125 per cent. and -0.065 per cent. for clear juice.

The apparent sucrose percentage was on the whole also lower than the sucrose percentage determined by the invertase method. The mean, maximum and minimum differences for the percentage sucrose by the invertase method and the percentage sucrose by the direct polarization method were +0.10 per cent., +0.275 per cent. and -0.015 per cent. for mixed juice, and +0.056 per cent., +0.125 per cent. and -0.065 per cent. for clear juice respectively. These differences are found in the columns \((b-f)\).

The percentage sucrose determined chemically was always considerably higher than that determined by the invertase method and the sucrose percentage determined by the baryta method was somewhat
lower than that determined by the invertase method. The differences between the percentage sucrose by the invertase method and the chemical method are found in columns (b-h).

The differences in percentage sucrose by the invertase method and by the baryta method are in the columns (b-j). There are, however, insufficient figures for sucrose determinations by the baryta method to draw any conclusions about this method.

Column (b-d), Table I, shows that on the average the sucrose percentage of Natal mixed juice determined by the invertase method was 0.043 higher than the Jackson and Gillis result, i.e. 0.30 per cent. on this last result. Taking into consideration the complicated nature of the analytical procedure, this agreement is quite satisfactory and demonstrates the general reliability and usefulness of the Jackson and Gillis method No. 4.

The individual deviations from this average difference indicate that the result of one or of both methods is, to a slight extent, influenced by the peculiarity of the analyzed juice. In one extreme case, the difference (b-d) was 0.145, i.e. 1.01 per cent. on the Jackson and Gillis result. In the other extreme case the difference was -0.05, i.e. -0.40 per cent. on the Jackson and Gillis result.

Assuming the invertase method to yield accurate results, a systematical difference of 0.30 per cent. for the Jackson and Gillis method has to be accepted. Individual deviations from this average difference up to 0.7 per cent. in both directions appear to be possible.

It is, however, hardly likely that such extreme deviations will characterise all mixed juice samples, the analyses of which are used for the computation of a mill's weekly Boiling House Recovery data. The influence of these deviations contributing to a fluctuation of the Boiling House Recovery should therefore be not more than a few tenths of a unit.

It is obvious that such a fluctuation is considerably smaller than the fluctuations due to unavoidable errors in the stock taking and can be ignored. In clarified juice the results are similar.

The average difference in the sucrose percentage is 0.056, i.e. 0.40 per cent. on the Jackson and Gillis result. The extreme deviations are +0.41 per cent. in one direction and -0.87 per cent. in the other direction.

Conclusions.

No proof has been found that the sucrose determined by the Jackson and Gillis method No. 4 is subject to serious errors due to substances in the juice other than sucrose having different optical activities under the conditions at which the direct and invert polarizations are read. The Jackson and Gillis method has been found to be generally reliable and useful for the determination of sucrose in mixed juice.

Acknowledgments.

The author wishes to thank the managements and staffs of the sugar mills for their kind assistance and the opportunity given to carry out these tests.

Sugar Milling Research Institute,
Durban.
February, 1951.
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Average ... 14.13 14.07 0.056 0.036 —0.50 0.11

(a) Invertase.  (c) Jackson and Gillis.  (e) Apparent Sucrose.  (g) Chemical Sucrose.  (i) Baryta Method.  
(b) Invertase Average.  (d) Jackson and Gillis Average.  (f) Apparent Sucrose Average.  (h) Chemical Sucrose Average.  (j) Baryta Method Average.
Mr. Hendry said that in the first tests at Esperanza there was no difference between D and F. In the second test the difference was 0.01. In view of these results he would like to have the opinion of those present on the necessity for applying the Jackson and Gillis method to mixed juice samples.

Mr. Dymond said that for many years the accuracy of the methods of sucrose determination had been queried in an attempt to explain the occasional serious fluctuations in undetermined losses. At Darnall the Jackson and Gillis method was carried out throughout the season on both mixed juice and clarified juice. Fluctuations occurred, but on the whole there were no outstanding irregularities as now demonstrated by Mr. Laubscher.

Mr. Dymond illustrated by means of a graph the daily differences in purity between the mixed and clarified juices. Analyses by simple polarization and by Jackson and Gillis No. 4 showed a few cases when the former was higher or equal. The highest difference was when the Jackson and Gillis method was higher by 0.06. This comparative series, while revealing nothing seriously abnormal, did show the effect of stale cane on clarification, which usually occurred at the weekend when cane had been lying over. Little if any rise in purity was observed during such periods.

Mr. Phipson asked whether Mr. Dymond found any difference between the determination by Jackson and Gillis method and the invertase method during the different periods of the season.

Mr. Dymond replied that the invertase method had been accepted as the most accurate method of sucrose determination. He believed that when large differences between the methods occurred, they would be found in the higher ranges.

Dr. Douwes Dekker said that the difference between the results of the direct polarization and the Clerget method was mainly to be ascribed to the presence of reducing sugars. The rotation of the mixture of reducing sugars depended on the ratio between dextrose and levulose present in the juice, and by making juice alkaline, this ratio, and therefore the polarization, was affected. (Reaction of Lobry du Bruyn and Alberda van Ekenstein.) For this reason it was useless to attach too much value to the difference in purity of mixed and clarified juice, since the former was true and the latter apparent purity. He thought Mr. Dymond’s statement about the difference in results of the Jackson and Gillis and the invertase method in high purity juice had not yet been confirmed by experience in other countries. The point, however, was of some importance and it was hoped that Mr. Dymond would publish his data.

Mr. du Toit said that, as Dr. Douwes Dekker had explained, the Clerget method was really a method of getting sucrose irrespective of the reducing sugar which was the variation the Clerget method took account of. The Jackson and Gillis No. 4 method he felt was excellent, if the correct formula was used, for giving correct results of sucrose if sugars only were present. If other substances, such as reversion products and amino acids were present, then the Jackson and Gillis No. 4 method could be wrong and could give results either too high or too low. The best method available was undoubtedly the invertase method if the differences were small, but it would be wrong to say that the Jackson and Gillis method was 100 per cent. correct for research purposes. Mr. Laubscher was using t/2. That was a temperature correction of 0.5, whereas if he had used the tables for Jackson and Gillis No. 4 he would use a correction of 0.53 t. He could see no justification for using a different temperature correction for the two methods.

Referring to the effects of amino acids and reversion products on the Jackson and Gillis No. 4 method he said if amino acids were present a sucrose result would be obtained that was too low; reversion products would give results that were too high. He had never been able to see how it was possible to get reversion products in mixed juice, but the amino acids would be present. To get an idea of the ordinary amounts of amino acids present it would not do to compare the invertase method with the Jackson and Gillis No. 4 because the latter suffered both from the effects of the amino acids and the reversion products. An estimate of amino acids in terms of optical activity could best be made by comparing the results obtained by the Jackson and Gillis methods Nos. 4 and 2. He would like to see an investigation carried out to determine the amount of amino acids and reversion products present in final molasses by choosing the correct methods and expressing the results in terms of optical activity. An investigation into the quantities and identities of the different amino acids present would be found more valuable.

Mr. Laubscher said that in connection with what Mr. du Toit said about using the temperature correction t/2 instead of 0.53t, he would like to point out that this was not the only difference between the formulae for calculating the Clerget factor. The term: 0.073 (m—13) in the formula given was, he believed, also different from the corresponding one in the formula used for calculating the Jackson and Gillis tables.
Calculating the Clerget factor from the formula 
\[(142.1 + 0.073 (m - 13) - t/2)\] and comparing it with 
the factor found in the Jackson and Gillis tables, 
differences of up to 0.4 per cent. were sometimes 
found. These differences in the Clerget constant 
might be due to the different methods of inversion. 
It was thus difficult to decide on the formulae to 
be used for calculating the Clerget factors for each 
method.

Being unable to decide how to arrive at the proper 
inversion constant for each method for purposes of 
comparison, it was decided to use in each case the 
formula and table recommended in the books where 
the methods were found. Possible differences in the 
sucrose determined by making use of the Clerget con­ 
stants obtained in these ways, was not of the order 
of the differences looked for in this investigation.

Mr. Rault suggested that in view of the length of 
the season and the variations in temperature during 
the seven months, as well as during the day and 
night periods, and the influence of temperature on 
polarization, causes of errors in polariscope work 
could be obviated by erecting constant temperature 
rooms.

Mr. Phipson said he found when testing by the 
Jackson and Gillis method that if the temperature 
dropped a lower result was obtained. Sugar polariza­ 
tion was found to be 0.2° lower than the refinery, but 
when the temperature correction was applied the 
figures agreed with the refinery. He felt that the 
errection of constant temperature rooms at the mills 
should be included in the recommended methods.

He asked whether a correction for temperature 
could be used when polarizing sugar.

Mr. Dymond said that the subject was a complex 
one and would only be finally solved by continuous 
and co-operative research whereby each laboratory 
was apportioned part of the work. The improvement 
of working conditions in mill laboratories, together 
with the installation of constant temperature rooms, 
was well worthy of investigation.

The President said he had been informed that the 
cost of a low temperature room would be about £120 
but the humidity was not controlled. A small 
constant temperature room had been put up at the 
Experiment Station at a cost of £150.

Mr. du Toit expressed the view that in the case of 
factories the cost would be perhaps £200 to £250.

Dr. Douwes Dekker reiterated that, for better 
information on the rise in purity during the clarifi­ 
cation process, the Jackson and Gillis method No. 4 
should generally be applied in clarified juice. This 
would also allow a more accurate non-sucrose balance 
to be calculated, by which a more reliable check on 
the amount of final molasses produced would be 
possible.

Speaking on the desirability of a constant tem­ 
perature laboratory, Dr. Douwes Dekker said that 
this should be considered as part of a survey of the 
available laboratory facilities at the mills. He 
certainly would not recommend building constant 
temperature laboratories without a proper investi­ 
gation into the present status of laboratory efficiency.