Simultaneous Measurement of Dextrose and Sucrose Utilizing External Hydrolysis

I. Introduction

Dextrose (D-glucose) and sucrose concentrations in complex matrices can be measured directly and quickly using the YSI 2700 SELECT Biochemistry Analyzer. YSI's unique enzyme technology provides for rapid dextrose and sucrose measurement. Measurements are virtually unaffected by color, turbidity, density, pH, or the presence of reducing substances.

When a YSI 2700 SELECT Biochemistry Analyzer is equipped with a dextrose membrane, both sucrose and dextrose concentrations can be measured. This is accomplished by first determining the dextrose concentration. The sucrose is then converted to dextrose, and the total dextrose concentration is measured. The difference in the responses corresponds to the sucrose concentration.

After a sample is injected into the sample chamber, the dextrose diffuses to the dextrose membrane, which contains glucose oxidase, and is oxidized to hydrogen peroxide and D-glucono-δ-lactone. The hydrogen peroxide is detected amperometrically at the platinum electrode surface. The current produced is directly proportional to the hydrogen peroxide and dextrose concentrations.

For more information on this system, refer to Section 4 in the Operations Manual.

II. Materials and Setup

A. YSI 2700 SELECT Biochemistry Analyzer - equipped with a 2365 Dextrose Membrane and 2357 Buffer.

B. Dextrose (1.80 g/L) standard solution.

C. Buffer Diluent (40 g/L NaH_2PO_4, 10 g/L Na_2HPO_4 in reagent water).

D. Invertase - Sigma Chemical Company I-4504 recommended.

E. Connect the 2700 SELECT to a suitable power source.

F. Perform the instrument and membrane check described in the Operations Manual (Section 3).

G. Volumetric glassware (Class A recommended).

H. The following instrument setup is recommended:

Sample Size	25 µL
Sample Station #	2
CalMethod	One Station

Black Probe Parameters

Chemistry	Dextrose
Unit	g/L
Calibrator	1.80 g/L
End Point	30 Sec
CalStation#	1

White Probe Parameters

Single channel 2700	N/A
Dual channel 2700	None

Autocal Parameters

Sample Error	ON
Temperature	1°C
Time	15 Min
Sample	5 Sam
Cal Shift	2%

III. Method

In this section two different case studies will be described. These examples can be followed when doing analysis on products that may contain similar concentrations of dextrose and sucrose.

Case #1

Example: The product is a powdered seasoning mix that is believed to contain 14% dextrose and 2% sucrose. The following sample preparation was used:

A. Weigh out about 10 grams of the powder (record exact weight).

B. Transfer the powder to a 100 mL volumetric flask using buffer diluent to rinse and dilute. Fill the flask to the mark with buffer and mix. Allow the solution to equilibrate for about 20 minutes

C. Remove about 3 mL of the solution in B and add ~2 mg of invertase enzyme. Stir gently until dissolved. Cover the sample and allow to incubate at room temperature for 20 minutes.

D. Calibrate the 2700 SELECT with 1.80 g/L dextrose standard solution (at Station #1).

E. Check the linearity of the membrane at least once a day by injection of an appropriate linearity standard. Refer to the Operations Manual (Section 3) for specifications.

F. Assay the sample prepared in B by aspiration into the 2700 SELECT. This is the free dextrose concentration (Dfree).

G. Assay the sample prepared in C (with invertase). The value reported is the sum of the free dextrose and that produced from sucrose hydrolysis (Dtotal).

H. Calibrate frequently as described in the Operations Manual (Section 6).

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Case #2

The product is a hard candy that is believed to contain 13% sucrose and 12% dextrose. With this sample two separate dilutions are necessary. This is due to the dextrose membrane reading both free dextrose and the dextrose produced from the hydrolysis of sucrose. The sum of these two concentrations exceeds the linear range. When analyzing the sample treated with invertase a more dilute sample will be needed. The following sample preparation was used:

A. Grind the sample into a fine powder.

B. Transfer 10 grams (record exact weight) of sample, from step A into a 100 mL volumetric flask using buffer diluent to rinse and dilute. Fill the flask to the mark with buffer and mix. Allow the solution to equilibrate for about 20 minutes.*

C. Transfer 5 grams (record exact weight) of sample, from step A into a 100 mL volumetric flask using buffer diluent to rinse and dilute. Fill the flask to the mark with buffer and mix.

D. Remove about 3 mL of the solution from C and add ~2 mg of invertase enzyme. Stir gently until dissolved. Cover the sample and allow to incubate at room temperature for about 20 minutes.

E. Calibrate the 2700 SELECT with a 1.80 g/L dextrose standard solution (at Station #1).

F. Check the linearity of the membrane at least once a day by injection of an appropriate linearity standard. Refer to the Operations Manual (Section 3) for specifications.

G. Assay the sample prepared in B by aspiration into the 2700 SELECT. This is the free dextrose concentration (Dfree).

H. Assay the sample prepared in C (with invertase). The value reported is the sum of the free dextrose and that produced from sucrose hydrolysis (D total).

I. Calibrate frequently as described in the Operations Manual (Section 6).

IV. Calculations

Case #1

To calculate % dextrose, multiply the reported value (Dfree) by the appropriate dilution factor.

Example: A 10.10 g powdered seasoning mix sample was prepared as described in III. B and C. When assayed, the value reported (Dfree) was 14.6 g/L dextrose.

% Dextrose:

14.6 g/L x 0.100 L/10.10 g = 0.1446 g dextrose/g powdered mix

=14.5% (w/w)

To calculate % sucrose, subtract Dfree from Dtotal and multiply by the appropriate dilution and mass ratio factors.

When the sample containing invertase was assayed, the value reported was 15.8 g/L (Dtotal) dextrose.

% Sucrose:

$$(15.8 \text{ g/L} - 14.6 \text{ g/L}) \times \frac{0.100 \text{ L}}{10.10 \text{g}} \times \frac{342.30 \text{ g/L sucrose}}{180.16 \text{ g/L dextrose}}$$

= 0.0226 g sucrose/g powdered mix

= 2.26% (w/w)

Case #2

To calculate % dextrose, multiply the reported value (Dfree) by the appropriate dilution factor.

Example: A 10.10 g ground hard candy sample was prepared as described in III B. When assayed the value reported (Dfree) was 12.3 g/L dextrose.

% Dextrose:

12.3 g/L x 0.100 L/10.10 g = 0.1218 g dextrose/g candy

= 12.2% (w/w)

To calculate % sucrose, multiply the reported value (Dtotal) by the appropriate dilution factor. Then subtract Dfree from Dtotal and multiply by the mass ratio factor.

Example: A 5.05 g ground hard candy sample was prepared as described in III D. When assayed the value reported (Dtotal) was 13.5 g/L.

% Sucrose:

13.5 g/L x 0.100 L/5.05 g= 0.2673 g dextrose/g candy

= 26.7%

26.7% - 12.2% = 14.5%

$$14.5\% \times \frac{342.30 \text{ g/mole sucrose}}{180.16 \text{ g/mole dextrose}}$$

= 27.5% (w/w) sucrose

V. Ordering Information

YSI No.	
2700	Biochemistry Analyzer
2365	Dextrose Membrane Kit
2747	Dextrose Standard Solution (1.80 g/L)
2357	Buffer Kit
2363	Potassium Ferrocyanide Test Solution
2392	NaCl Solution (for membrane installation)

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