



Food & Beverage Series

SIMULTANEOUS MEASUREMENT OF GLUCOSE AND SUCROSE
IN SWEETENED CONDENSED MILK

Application Note 216LS
YSI Life Sciences



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INTRODUCTION

Dextrose (D-glucose) and sucrose concentrations in complex matrices such as sweetened condensed milk can be measured directly and quickly using the YSI 2900 Series Biochemistry Analyzer. YSI's unique enzyme technology provides for rapid glucose and sucrose measurements. Measurements are virtually unaffected by color, turbidity, density, pH, or the presence of reducing substances.

When a 2900 Series Biochemistry Analyzer is equipped with a glucose and a sucrose membrane, simultaneous measurement of both analytes is possible. Because glucose interferes with sucrose analysis, it is necessary to follow this protocol when analyzing for sucrose in the presence of glucose.

When a sample is injected into the sample chamber, the sucrose diffuses to the sucrose membrane, which contains invertase, mutarotase, and glucose oxidase. The sucrose is hydrolyzed to α -D-glucose and fructose. The mutarotase allows for the quick equilibrium of glucose between its α and β forms. In the presence of glucose oxidase, the β -D-dextrose (glucose) is oxidized to hydrogen peroxide and D-glucono- δ -lactone. The hydrogen peroxide is detected amperometrically at the platinum electrode surface. The glucose in the sample diffuses to both the glucose and sucrose membranes, which contain glucose oxidase, and is oxidized as well. Subtracting the response of the glucose membrane from the response of the sucrose membrane yields the response due to sucrose alone. The glucose response is taken directly from the glucose membrane. The algorithm in the instrument software calculates the net concentrations. For more information on this system, refer to the Operations Manual.

I. MATERIALS & SETUP

- YSI 2900 Series Biochemistry Analyzer - equipped with a 2703 Sucrose Membrane, a 2365 Glucose Membrane and 2357 Buffer.
- Glucose (2.50 g/L, 25.00 g/L) and Sucrose (5.00 g/L, 25.0 g/L) standard solutions.

- Buffer Diluent (40 g/L NaH_2PO_4 , 10g/L Na_2HPO_4 in reagent water).
- Connect the 2900 Series instrument to a suitable power source.
- Perform the instrument and membrane daily checks described in the Operations Manual.
- Volumetric glassware (Class A recommended).
- The following instrument setup is recommended:
Sample Size 10 μL

Probe A Parameters

Chemistry	Glucose
Unit	g/L
Calibrator	2.50
End Point	30 Sec

Probe B Parameters

Chemistry	Sucrose
Unit	g/L
Calibrator	5.00 g/L
End Point	30 Sec

Autocal Parameters

Temperature	1°C
Time	30 Min
Sample	5 Sam
Cal Shift	2%

II. METHOD

- As with all applications, make sure the sample is homogeneous. In the case of a canned sample of sweetened condensed milk, warm the can in a water bath that is between 35 - 45°C. Shake the can before opening. Transfer the contents of the can to another container. Scrape the top, bottom and sides of the can, making sure to remove any sugar that may have settled. Stir entire contents.
- Weigh up to 1.000 g of sweetened condensed milk to be analyzed.
- Transfer the sample to a 100 mL volumetric flask
continued

using buffer diluent to rinse and dilute. Fill the flask to the mark with buffer and mix. Allow the solution to equilibrate for about twenty minutes before analysis.

- D. Calibrate the 2900 Series instrument with 2.50 g/L glucose and 5.00 g/L sucrose standard solutions.
- E. Check the linearity of the membranes at least once a day by injection of glucose (25.0 g/L) and sucrose (25.0 g/L) linearity check solutions. Refer to the Operations Manual for specifications.
- F. Assay the sample prepared in B by aspiration into the 2900 Series instrument.
- G. Calibrate frequently as described in the Operations Manual.

Limitations:

The linear range of the system is 0.05 to 25.0 g/L for both glucose and sucrose. The combined concentration of glucose + sucrose cannot exceed 25.0 g/L. If the sum of the values reported exceeds this, further dilution of the sample is required.

If the glucose concentration exceeds the sucrose concentration, accuracy and precision may be compromised due to the software algorithm that subtracts glucose from sucrose. To avoid compromising accuracy, refer to Application Note 204LS.

III. CALCULATIONS

To calculate % glucose and sucrose, multiply the values reported by the appropriate dilution factor.

Example: 1.0135 g of sweetened condensed milk was diluted to 100 mL in a Class A volumetric flask. When assayed, the values reported were 0.096 g/L glucose and 4.47 g/L sucrose.

$\begin{aligned} &\% \text{ Glucose:} \\ &0.096 \text{ g/L} \times 0.100\text{L} \\ &/1.0135 \text{ g} \end{aligned}$	$\begin{aligned} &= 0.0095 \text{ g glucose/g milk} \\ &= 0.95\% \text{ (w/w)} \end{aligned}$
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$\begin{aligned} &\% \text{ Sucrose:} \\ &4.47 \text{ g/L} \times 0.100\text{L} \\ &/1.0135 \text{ g} \end{aligned}$	$\begin{aligned} &= 0.4410 \text{ g sucrose/g milk} \\ &= 44.1\% \text{ (w/w)} \end{aligned}$
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ORDERING INFORMATION

YSI Part Numbers:

- 2900 Biochemistry Analyzer
- 2365 Glucose Membrane Kit
- 2776 Glucose Standard Solution (2.50 g/L)
- 2777 Glucose Standard Solution (25.0 g/L)
- 2703 Sucrose Membrane Kit
- 2780 Sucrose Standard Solution (5.00 g/L)
- 2778 Sucrose Standard Solution (25.0 g/L)
- 2357 Buffer Kit
- 2363 Potassium Ferrocyanide Test Solution
- 2392 NaCl Solution (for membrane installation)

For further information, please contact:

YSI Life Sciences

1725 Brannum Lane | Yellow Springs, Ohio 45387

Website: ysi.com | Email: support@ysi.com

Telephone: (937) 767-7241 | Fax: (937) 767-9320



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YSI Life Sciences develops and manufactures scientific instruments, sensors and systems that serve a variety of scientific and industrial markets worldwide. YSI has a long history in the life sciences and bioanalytical markets, most notably with our introduction of the world's first commercial whole blood glucose analyzer in 1975. Today there are over 10,000 YSI instruments installed around the world, trusted in critical situations to provide the most accurate data in the shortest time.

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