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Application Note
YSI Life Sciences • 222LS-02

Food & Beverage Series

DETERMINATION OF COOK IN CEREAL

Introduction

The degree of cook of extruded cereal products can be determined using the YSI 2900 Series Biochemistry Analyzer. YSI's unique enzyme technology provides for specific glucose measurement. Measurements are virtually unaffected by color, turbidity, density, pH or the presence of reducing substances.

A portion of a sample is solubilized in cold water and a portion is autoclaved. The samples containing starch are treated identically with glucoamylase. Glucose produced from this reaction is measured with the YSI 2900 Series instrument. The ratio of glucose in the cold water sample to glucose in the autoclaved sample yields % cook.

When a sample is injected into the sample chamber, the glucose diffuses into the membrane containing glucose oxidase. The glucose is immediately oxidized to hydrogen peroxide and D-glucono-δ-lactone.

The hydrogen peroxide is detected amperometrically at the platinum electrode surface. The current flow at the electrode is directly proportional to the hydrogen peroxide concentration, and therefore is directly proportional to the glucose concentration.

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I. Materials & Setup

A. YSI 2900 Series Biochemistry Analyzer - equipped with a 2365 Glucose Membrane and 2357 Buffer.

B. Glucose standards (2.50 g/L, 9.00 g/L).

C. 1N Acetate buffer.

D. Glucoamylase solution.

E. 25% Trichloracetic Acid.

F. Phosphate diluent buffer (40 g/L NaH_2PO_4, 10 g/L Na_2HPO_4 in reagent water, pH 5.9).

G. Volumetric glassware (Class A recommended).

H. Connect the 2900 Series instrument to a suitable power source.

I. Perform the instrument and membrane daily checks described in the Operations Manual.

H. The following instrument setup is recommended:
Sample Size 25 µL

Probe A Parameters		**Autocal Parameters**	
Chemistry	Glucose	Temperature	1°C
Unit	g/L	Time	30 Min
Calibrator	2.50	Sample	5 Sam
End Point	30 Sec	Cal Shift	2%

II. Method

A. Cold water sample:
1. Into a 125 mL Erlenmeyer flask disperse ~ 0.50 g sample into ~ 43 mL reagent water. Record the exact weight of the sample.
2. Add 5 mL of 1N Acetate buffer, pH 4.2.
3. Add 2.5 mL of 30% glucoamylase solution
4. Cover with aluminum foil and incubate, in water bath, for one hour at 40°C.
5. Add ~ 3.2 mL of 25% Trichloracetic Acid immediately after the incubation and swirl the contents.
6. Allow the solution to cool to room temperature. Transfer the solution to a 100 mL volumetric flask and dilute with phosphate diluent buffer (pH 5.9). Shake vigorously.

B. Autoclaved sample:
1. Dilute the sample as in A1. Cover with aluminum foil and autoclave at 15-20 psi, ~124°C ± 3°C for one hour. Cool to 40°C.
2. Repeat steps A2 - A6 above.

C. Blank sample:
Since glucoamylase may contain free glucose, perform steps A1 - A6 without using the sample containing starch. Both the cold water sample and the autoclaved sample should be corrected using this value.

D. Calibrate the 2900 Series instrument with a 2.50 g/L glucose standard solution.

E. Check the linearity of the membrane at least once a day by injection of a glucose linearity check solution (9.00 g/L). Refer to the Operators Manual for specifications.

F. Determination of Blank: Assay the blank prepared in C by aspiration into the 2900 Series instrument.*

G. Determination of Cooked Starch: Assay the sample prepared in A by aspiration into the 2900 Series Instrument.*

H. Determination of Total Starch: Assay the sample prepared in B by aspiration into the 2900 Series Instrument.*

I. Calibrate frequently as described in the Operations Manual.

* The linear range of the system is 0 to 9.00 g/L glucose. If the value reported exceeds this, further dilution is required.

Note: If the sample contains free glucose, both the cold water and the autoclaved sample will have to be corrected with this value. Weigh 0.5 grams of sample into 100 mL volumetric flask and dilute to the mark with phosphate diluent buffer. Mix the sample until dissolved and analyze.

III. Calculations

To calculate % cook, multiply the reported value by the appropriate dilution factor. The value of the blank (measured step F) should be subtracted from the cooked starch (measured in step G) and the total starch (measured in step H).

Since 1.1 g of glucose is produced when 1.0 g of starch is hydrolyzed, the glucose concentration of the sample should be multiplied by 0.9.

Example: 0.52 g of pet food was diluted to 100 mL in a Class A volumetric flask. The sample was prepared using the cold water procedure. When assayed, the value reported was 1.45 g/L glucose.

0.52 g of pet food was diluted to 100 mL in a Class A volumetric flask. The sample was prepared using the autoclaved procedure. When assayed, the value reported was 1.82 g/L glucose.

The blank contained 0.01 g/L of glucose.

$$\% \text{ Cook} = \frac{[\text{Cooked Starch}]}{[\text{Total Starch}]} \times 100\%$$

or

$$\% \text{ Cook} = \frac{[(\text{Step G} - \text{Step F}) \times 0.9]}{[(\text{Step H} - \text{Step F}) \times 0.9]} \times 100\%$$

Cold water starch: 1.45-0.01 g/L x 0.9 x 0.100L/0.52 g	= 0.249 g starch/g food = 24.9% (w/w)
Total starch: 1.82-0.01 g/L x 0.9 x 0.100L/0.52 g	= 0.313 g starch/g food = 31.3% (w/w)
% Cook: 24.9% / 31.3% x 100%	= 76.6%

Ordering Information

2900	Biochemistry Analyzer
2365	Glucose Membrane Kit
2776	Glucose Standard Solution (2.50 g/L)
1531	Glucose Standard Solution (9.00 g/L)
2357	Buffer Kit
2363	Potassium Ferrocyanide Test Solution
2392	NaCl Solution (for membrane installation)

YSI 2900 Biochemical Analyzer

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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