COLOR, SOLUTIONS (Spectrophotometric)

PRINCIPLE

When white light passes through a colored solution, certain bands of the spectrum are absorbed allowing the transmitted portion to impart the visual effect of color. The intensity of this transmitted light can be measured by means of a photoelectric cell and a sensitive galvanometer. If the incident light is monochromatic, such as produced by a spectrophotometer, the galvanometer response at various wavelengths when compared to pure water is interpreted as a quantitative estimate of the color, which by definition is calculated as optical density (Note 1).

SCOPE

This method is applicable to corn syrup and starch hydrolyzates, crude and refined sugar solutions (Note 2) which are substantially optically clear, and contain primarily yellow and red colors. When properly standardized, the instrument error should not exceed 0.5% transmittance, absolute. Values should range within ± 10% relative of colors determined visually by an experienced analyst.

SPECIAL APPARATUS

1. Spectrophotometer: An instrument capable of continuously variable wavelength in the visible spectrum, designed to accommodate a pair of 2 x 4 cm cells, is recommended. Spectrophotometers such as the Bausch & Lomb Spectronic 100, the Coleman Universal or Model 14, the Perkin-Elmer Lamda 3, and the Pye-Unicam Model SP-8-150 can be used. Calibrate and operate the instrument according to manufacturer's instructions. A calibration check may be performed with a potassium dichromate solution as outlined under procedure.

2. Sample Cells: Cuvets of optically clear glass, size 2 x 4 cm, having open tops for easy filling and cleaning, are recommended. All cuvets should have matching transmittance values (± 0.5%) under conditions of the test when containing purified water or potassium dichromate solution.

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REAGENTS

Potassium Dichromate Solution, 0.01%: Dissolve 0.100 g of reagent grade potassium dichromate ($K_2Cr_2O_7$) in purified water in a 1000 mL volumetric flask, dilute to volume and mix thoroughly.

PROCEDURE

Calibration: Fill a clean and dry cuvet (Note 3) with purified water, fill another with 0.01% potassium dichromate solution, and place in spectrophotometer so that the light beam passes through the 2 cm cell depth. Ascertain the wavelength which gives exactly 54.5% transmittance (%T) on potassium dichromate solution after the circuit is balanced at 100% transmittance (%T) on purified water. This setting is to be used for all subsequent readings as the corrected 450 nm wavelength. Discard the dichromate solution, rinse and dry the cuvet.

Sample Preparation (Note 4): Dissolve dextrose (anhydrous and hydrate) in hot purified water to obtain a concentration above 17.5° Baumé. Cool solution to room temperature and adjust concentration to 17.5° Baumé at 60 °F (Dilution Factor, DF = 3).

Corn syrup samples, carefully fill cuvet with as is syrup (Note 6).

Dilute concentrated hydrolyzate solutions and greens liquors to 17.5° Baumé at 60 °F. Dilute 10 mL of these solutions to 100 mL with purified water (DF = 30) (Note 5).

Dilute hydrol samples with purified water to 17.5° Baumé at 60 °F. Then dilute 10 mL to 1 L with purified water (DF = 300).

Sample Analysis: Fill a clean and dry cuvet with dilute sample solution, and fill a matching cuvet with purified water (Note 6). Place both cuvets in cell carrier so that light beam passes through the 4 cm cell depth, and set wavelength at the corrected 450 nm position. Determine transmittance (%T) of the sample to the nearest 0.5% immediately after the circuit is balanced at 100% transmittance (T) on purified water. Alternatively, measure absorbance (A).

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Set wavelength scale at 600 nm and again determine transmittance (%T) of the sample immediately after the circuit is balanced at 100% T on purified water (Note 7). Prior to calculation, convert transmittance values to absorbance (A) values as follows: $A = 2 - \log\%T$

CALCULATION (Note 8)

$$\text{Color (Absorbance Difference} \times 100) = \frac{(A_{450\,nm} - A_{600\,nm}) \times 100 \times DF}{\text{Cell Length, cm}}$$

NOTES AND PRECAUTIONS

1. This method was adopted after examination of the C.I.E. (Commission Internationale de l'Eclairage) color specification procedure which describes the primary visual stimuli, dominant wavelength, purity and luminance (luminous transmittance or reflectance). "Dominant wavelength" is that property by which the eye differentiates hue or color type. "Purity" is a measure of color concentration or saturation with respect to a pure spectral standard. "Luminance" expresses the transmittance or reflectance (percentage) of a standard light energy characteristic of normal perception in terms of wavelength and energy. "Luminance," sometimes referred as brightness or brilliance, is indicated on a scale between 0 and 100 representing absolute black and perfect white, respectively.

   Although C.I.E. values are objective, accurate and internationally reproducible, the method is laborious or requires costly equipment. Therefore, the simplified "optical density difference" procedure was developed. Values obtained for corn sugar solutions and other starch hydrolyzates are precisely related to C.I.E. purity.

   In addition, percent transmittance at 550 nm (referred to as brilliance) is substantially numerically equal to C.I.E. luminance (luminous transmittance).

2. Crude and refined sugars include dextrose (anhydrous and hydrate), dextrose solutions, concentrated and refined hydrolyzates, greens liquors (Corn Sugar Molasses) and hydrol (Feeding Corn Sugar Molasses).

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3. Clean cuvets with detergent and warm water. Remove iron stains with dilute hydrochloric acid and grease films with cleaning solution. Cuvets should be dry and free of fingerprints on the outside surface through which the light beam passes, but not necessarily dry inside.

4. Solution concentrations are selected at convenient values to obtain transmittances in the most accurate range of the instrument. Application of the dilution factors provide solution color values corresponding to those for approximately 40 Baumé (80% solids) concentrations of the original samples.

5. The 17.5° Baumé solution diluted 10-fold provides a solution of 2.0° Baumé at 60 °F. If expedient, the solution may be diluted directly to 2.0° Baumé.

6. Avoid introduction of air bubbles when filling cuvets because they reduce transmission of light. However, if not in motion, air bubbles do not seriously affect the optical density difference.

7. Readings at 600 nm are made also so that any reduction in transmittance at 450 nm caused by interfering substances in the sample is compensated for and not included as color.

8. If the observed transmittance (%T) at 600 nm exceeds 100%, or if the absorbance (A) at 600 nm is negative, use a value of zero (0) in the equation for calculating color.

   The absorbance difference per cm is multiplied by 100 to give a value numerically similar to that obtained by the Lovibond system used previously by the corn wet milling industry.

METHOD HISTORY

Combined the Color, Solutions (Spectrophotometric) methods for Corn Syrup (E-16), Corn Sugar (F-14) and Corn Oil (H-12) on 11-9-2010.

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Corn Syrup, Color, Solutions (Spectrophotometric) (E-16), Date of Acceptance 1-25-1957, Revised 4-18-1989.

Corn Sugar, Color, Solutions (Spectrophotometric) (F-14), Date of Acceptance 11-17-1959, Revised 6-20-1991.

Corn Oil, Color, Solutions (Spectrophotometric) (H-12), Date of Acceptance 11-16-64, Revised 10-23-2001.