READILY CARBONIZABLE SUBSTANCES(Spectrophotometric)

PRINCIPLE

This method describes a test for various impurities called readily carbonizable substances. The quality of the sample is determined by the color that results from the treatment of the sample being tested with concentrated sulfuric acid in a strictly defined manner. Carbohydrates, such as glucose, are largely responsible for the yellow color developed in citric acid during this test. The carbohydrates dehydrate to form furfural species that can subsequently undergo condensation with citric acid to produce colored condensation products having a λ_{max} of 470 nm. Instead of visually evaluating the color of the sample, this method describes the spectrophotometric determination of visible light absorbance at 470 nm.

SCOPE

This method applies to citric acid final product samples, both liquids and solids.

SPECIAL APPARATUS

1. Water or oil bath set at 90°C ($\pm$1°C)
2. Test tubes (18x150 mm as per FCC)
3. Test tube rack
4. Analytical balance
5. Spectrophotometer capable of measuring the appropriate wavelength - prism or grating, with a 1-cm quartz or glass cuvette (or matching round cuvette, 19 x 150 mm, Fisher Cat. 14-385-900E)
6. Vortexing mixer (VWR Vortexer)
7. Timer
8. Sonicator bath
9. Volumetric pipet, 1.0 mL and 10 mL

REAGENTS

98% minimum Reagent Grade Sulfuric Acid (low Nitrate less than 1 ppm maximum)

READILY CARBONIZABLE SUBSTANCES — continued

SAFETY PRECAUTIONS

1. Sulfuric acid is considered to be a particularly hazardous substance by the Hazard Evaluation for Highly Hazardous Substances. Review the Material Safety Data Sheet (MSDS) to familiarize yourself with the hazards associated with this chemical. Some symptoms of overexposure are as follows:
   - inhalation - severe irritation or burns of respiratory system
   - skin/eyes - physical contact may cause severe second and third degree skin burns

2. Mists containing sulfuric acid are known as human carcinogens by IARC.

3. To adequately control the health hazards associated with this chemical:
   - use only in a fume hood (ensure fume hood is working prior to use)
   - use proper gloves (latex gloves for use up to 5 minutes, neoprene/latex gloves for up to 1 1/2 hours, Stanzoil Neoprene for up to 8 hours) and splash-proof safety glasses
   - properly dispose of any generated waste
   - decontaminate designated area upon completion

4. Stanzoil Neoprene gloves, chemical-resistant apron and splash-proof safety goggles must be worn when changing out the concentrated sulfuric acid bottle.

5. Whenever possible, complete the portion of this procedure on which you are working without disruption. Should an interruption occur, immediately remove your gloves to avoid contamination of other objects.

6. The test tube racks which are used have been known to deteriorate over time under the harsh acidic conditions of this test; check the integrity of the rack prior to each use.

7. Perform this procedure carefully, keeping the safety of yourself and others top priority at all times.

READILY CARBONIZABLE SUBSTANCES — continued

PROCEDURE

**********Familiarize yourself with precautions before proceeding**********

Weigh 1.00 ± 0.01 g of sample (see Note 1) into a clean test tube and record the exact weight used.

Carefully dispense 10 ± 0.1 mL of concentrated sulfuric acid into the sample test tube and an empty test tube (the 'blank'). Vortex to mix for one minute.

Place the test tubes in a water bath maintained at 90 ± 1°C for one hour (see Note 1). Carefully shake the tube a few times during the process to ensure that citric acid is dissolved and gaseous decomposition products are allowed to escape to the atmosphere. A recommendation is to very carefully shake the tube at the 5 and 30 minute marks.

When the reaction is complete, remove the samples, and cool to ambient temperature (Note 1).

Set the spectrophotometer to 470 nm. Fill a 1 cm cuvette with the 'blank' solution, place in the spectrophotometer, and adjust to zero absorbance. Carefully discard the sulfuric acid solution. Refill the same cuvette with subsequent samples and record the absorbance and transmittance of the solution at 470 nm in a 1 cm cuvette.

CALCULATIONS

Record the absorbance and transmittance of the solution at 470 nm.

NOTE

1. Both the weight of the sample used and the reaction time are important since the reaction of readily carbonizable substances with sulfuric acid is non-linear. After 1 hour, promptly remove the samples from the 90°C water bath and cool to quench the reaction. Prolonged reaction times will result in high RCS results.

READILY CARBONIZABLE SUBSTANCES — continued

REFERENCE

Food Chemicals Codex, Fifth Edition, Food and Nutrition Board, Institute of Medicine, National Academy of Sciences, National Academy Press, Washington, D.C. (1996), General Tests and Assays, page 115 and Appendix II, pages 845-846.

METHOD HISTORY

Lactic Acid, Readily Carbonizable Substances, (Spectrophotometric) (L-2), Date of Acceptance 9-08-2006.