

Phenols in Nutmeg and Mace

Purpose: To distinguish between nutmegs or maces grown in the West Indies from those grown in the East Indies.

A. Apparatus:

1. Centrifuge.
2. Centrifuge tubes, conical, 12 mL.
3. Spectrophotometer, capable of accurately measuring absorbance at 400 m μ .
4. Test tube, 20 x 150 mm.

B. Reagents:

1. p-Nitroaniline, recrystallized.
2. Sulfuric acid, 24 N. (Dissolve 660 mL. of H₂SO₄, Sp. gr. 1.84, in distilled water and dilute to 1 liter). (Note 1)
3. Sodium hydroxide, solid.
4. Sodium hydroxide, 10% solution.
5. Sodium nitrite, 0.2N. (Dissolve 13.8 g of NaNO₂ in distilled water and dilute to 1 liter.) *Use only freshly prepared solution.*
6. Chloroform.
7. Sodium sulfate, anhydrous.

C. Preparation of Sample:

1. Use steam volatile oil nutmeg or mace from Method 5.0, 5.1 or 5.2.

Method 19.0

Phenols in Nutmeg and Mace

D. Procedure:

1. Pipette 1 mL of the steam-volatile nutmeg or mace oil (Method 5.0 or 5.1) into a 12 mL conical centrifuge tube, add 6 mL of chloroform and 4 mL of 10% sodium hydroxide. Mix thoroughly and centrifuge briefly to separate the two layers.
2. Pipette 2 mL of the upper (basic) layer into a 22 x 150 mm. test tube, cool to 0°C. in an ice water bath and keep cold.
3. Diazonium salt solution--Dissolve 0.276 g of p-nitroaniline in 5 mL of 24 N. sulfuric acid in a 250 mL beaker and add 3 g of solid sodium hydroxide. Cool, add 20 g of ice made with distilled water and place the beaker in an ice-salt bath. Add 10 mL of 0.2 N. sodium nitrite to the solution slowly (over 15 minutes) with continuous stirring. Stir for 15 minutes after the addition of nitrite; then add 20 g of ice and 40 mL of ice-cold water. Keep the solution cold. *Prepare in hood.*
4. Add slowly with shaking 4 mL of the diazonium salt solution from a cold pipette. Keep the test tube cold during the addition of diazonium salt so as to allow coupling of phenols.
5. Pipette 1 mL of the coupled phenol solution to a 10 mL volumetric flask and dilute to the mark with distilled water.
6. Measure the absorbance of the solution at 400 mμ against a blank derived from 2 mL of 10% sodium hydroxide carried through steps 4 and 5. (See Note 2 for interpretation)

E. Calculation:

N/A

F. Statistics:

TBD

Phenols in Nutmeg and Mace

G. Note:

1. Dissolving sulfuric acid in water is an exothermic reaction. Exercise caution when performing this step.
2. Interpretation:
 - a. West Indian nutmeg and mace oils normally have absorbances less than 0.020.
 - b. East Indian nutmeg and mace oils normally have absorbances between 0.060 and 0.150.

H. References:

N/A