METHOD #: 420.3 (Issued 1978)

TITLE: Phenolics (Spectrophotometric, MBTH With Distillation)

ANALYTE: Phenolics

INSTRUMENTATION: Spectrophotometer

STORET No. 32730

1.0 Scope and Application

1.1 This method is applicable to the analysis of drinking, surface and saline waters, domestic and industrial wastes.

1.2 The method is capable of measuring phenolic materials at the 2 μg/L level when the colored end product is extracted and concentrated in a solvent phase using phenol as a standard.

1.3 The method is capable of measuring phenolic materials that contain from 50 to 1000 μg/L in the aqueous phase (without solvent extraction) using phenol as a standard.

1.4 It is not possible to use this method to differentiate between different kinds of phenols.

2.0 Summary of Method

2.1 This method is based on the coupling of phenol with MBTH in an acid medium using ceric ammonium sulfate as an oxidant. The coupling takes place in the p-position; if this position is occupied, the MBTH reagent will react at a free o-position. The colors obtained have maximum absorbance from 460-595 nm. For phenol and most phenolic mixtures the absorbance is 520 and 490 nm.

3.0 Comments

3.1 For most samples a preliminary distillation is required to remove interfering materials.

3.2 Color response of phenolic materials with MBTH is not the same for all compounds. Because phenolic type wastes usually contain a variety of phenols, it is not possible to duplicate a mixture of phenols to be used as a standard. For this reason phenol has been selected as a standard and any color produced by the reaction of other phenolic compounds is reported as phenol. This value will represent the minimum concentration of phenolic compounds present in the sample.

4.0 Sample Handling and Preservation

4.1 Biological degradation is inhibited by the addition of 1 g/L of copper sulfate to the sample and acidification to a pH of less than 4 with sulfuric acid. The sample should be kept at 4°C and analyzed within 24 hours after collection.

5.0 Interference

5.1 Interferences from sulfur compounds are eliminated by acidifying the sample to a pH of less than 4.0 with H_2SO_4 and aerating briefly by stirring and adding $CuSO_4$.

5.2 Oxidizing agents such as chlorine, detected by the liberation of iodine upon acidification in the presence of potassium iodide, are removed immediately after sampling by the addition of an excess of ferrous ammonium sulfate (6.5). If chlorine is not removed, the phenolic compounds may be partially oxidized and the results may be low.

5.3 Phosphate causes a precipitate to form; therefore, phosphoric acid should not be used for preservation. All glassware should be phosphate free.

5.4 High concentrations of aldehydes may be an interference.

6.0 Apparatus

6.1 Distillation apparatus: All glass consisting of a 1 liter pyrex distilling apparatus with Graham condensor

6.2 pH meter.

6.3 Spectrophotometer.

6.4 Funnels.

6.5 Filter paper.

6.6 Membrane filters.

6.7 Separatory funnels.

7.0 Reagents

7.1 Copper sulfate solution: Dissolve 100 g $CuSO_4 \cdot 5H_2O$ in distilled water and dilute to 1 liter.

7.2 Sulfuric acid, 1 N: Add 28 mL of conc. H_2SO_4 to 900 mL of distilled water, mix and dilute to 1 liter.

7.3 MBTH solution, 0.05%: Dissolve 0.1 g of 3-methyl-2-benzothiazolinone hydrazone hydrochloride in 200 mL of distilled water.

7.4 Ceric ammonium sulfate solution: Add 2.0 g of $Ce(SO_4)_2 \cdot 2(NH_4)_2SO_4 \cdot 2H_2O$ and 2.0 mL of conc. H_2SO_4 to 150 mL of distilled water. After the solid has dissolved dilute to 200 mL with distilled water.

7.5 Buffer solution: Dissolve in the following order, 8 g of sodium hydroxide, 2 g EDTA (disodium salt) and 8 g boric acid in 200 mL of distilled water. Dilute to 250 mL with distilled water.

7.6 Working buffer solution: Make a working solution by mixing an appropriate volume of buffer solution (7.5) with an equal volume of ethanol.

7.7 Chloroform

7.8 Stock phenol: Dissolve 1.00 g phenol in 500 mL of distilled water and dilute to 1000 mL. Add 1 g $CuSO_4$ and 0.5 mL conc. H_2SO_4 as preservative. 1.0 mL = 1.0 mg phenol.

7.9 Standard phenol solution A: Dilute 10.0 mL of stock phenol solution (7.8) to 1000 mL. 1.0 mL = 0.01 mg phenol.

7.10 Standard phenol solution B: Dilute 100.0 mL of standard phenol solution A (7.9) to 1000 mL with distilled water. 1.0 mL = 0.001 mg phenol.

8.0 Procedure

8.1 Distillation

8.1.1 To 500 mL of sample add 5 mL of copper sulfate solution (7.1) and adjust the pH to approximately 4 with 1 N sulfuric acid solution (7.2).

8.1.2 Distill over 450 mL of sample, add 50 mL of warm distilled water to flask, and resume distillation until 500 mL has been collected.

8.1.3 If the distillate is turbid, filter through a prewashed membrane filter.

8.2 Concentration above 50 μg/L.

8.2.1 To 100 mL of distillate or an aliquot diluted to 100 ml, add 4 mL of MBTH solution (7.3).

8.2.2 After 5 minutes, add 2.5 mL of ceric ammonium sulfate solution (7.4).

8.2.3 Wait another 5 minutes and add 7 mL of working buffer solution (7.6).

8.2.4 After 15 minutes, read the absorbance at 520 nm against a reagent blank. The color is stable for 4 hours.

8.3 Concentration below 50 μg/L.

8.3.1 To 500 mL of distillate in a separately funnel, add 4 mL of MBTH solution (7.3).

8.3.2 After 5 minutes, add 2.5 mL of ceric ammonia sulfate solution (7.4).

8.3.3 After an additional 5 minutes, add 7 mL of working buffer solution (7.6).

8.3.4 After 15 minutes, add 25 mL of chloroform. Shake the separatory funnel at least 20 times. Allow the layer to separate, and pass the chloroform layer through filter paper.

8.3.5 Read the absorbance at 490 nm against a reagent blank.

9.0 Calculation

9.1 Prepare a standard curve by plotting absorbance against concentration values.

9.2 Obtain concentration value of sample directly from prepared standard curve.

10. Precision and Accuracy

10.1 Precision and accuracy data are not available at this time.

Bibliography

1. Friestad, H.O., Ott, E.E., and Gunther, F.A., "Automated Colorimetric Micro Determination of Phenol by Oxidative Coupling with 3-Methyl-2- benzothiazolinone Hydrazone", Technicon International Congress (1969).
2. Gales, M. E., "An Evaluation of the 3-Methyl-2 benzothiazolinone Hydrazone Method for the Determination of Phenols in Water and Wastewater", Analyst, 100, No. 1197, 841 (1975).