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Method of Test for Thioglycolic Acid and Cysteine in Permanent Wave

1. Scope

This method is applicable to the determination of thioglycolic acid and cysteine in permanent wave.

2. Method

After extraction, analytes are determined by high performance liquid chromatography (HPLC).

2.1. Equipment

2.1.1. High performance liquid chromatograph.

2.1.1.1. Detector: photodiode array detector.

2.1.1.2. Column: LiChrospher® 100 RP-18 endcapped, 5 μm, 4.6 mm i.d. × 25 cm, or an equivalent product.

2.1.2. Ultrasonicator.

2.2. Chemicals

1-Octanesulfonic acid sodium salt, GR grade;

Phosphoric acid (85%), GR grade;

Deionized water, resistivity ≥ 18 MΩ • cm (at 25°C);

Thioglycolic acid and cysteine, reference standards

2.3. Apparatus

2.3.1. Volumetric flask: 10 mL and 20 mL.

2.3.2. Membrane filter: 0.45 μm, Nylon.

2.4. Mobile phase

2.4.1. Solvent A:

Dissolve and dilute 0.8 g of 1-octanesulfonic acid sodium salt and 1 mL phosphoric acid with deionized water to 1000 mL, then filter with a membrane filter.

2.4.2. Solvent B: Acetonitrile.

2.5. Standard solution preparation

Transfer about 20 mg of thioglycolic acid and 10 mg cysteine reference standards (cysteine need freshly prepared) accurately weighed into 20-mL and 10-mL volumetric flask, respectively. Dissolve and dilute with deionized water to volume as the standard stock solutions. Store in the refrigerator. When to use, mix appropriate volume of each standard stock solution, and dilute with deionized water to 100 - 800 μg/mL for thioglycolic acid and to 50 - 250 μg/mL

for cysteine as standard solutions, respectively.

2.6. Sample solution preparation

Transfer about 0.5 g of the well-mixed sample accurately weighed into a 10-mL volumetric flask, add 5 mL of deionized water, and sonicate for 20 mins. Add deionized water to volume. Filter with a membrane filter, and take the filtrate as the sample solution.

2.7. Identification and quantification

Accurately inject 20 μL of the sample solution and the standard solutions into HPLC separately, and operate according to the following conditions. Identify each analyte based on the retention time and the UV absorption spectrum. Calculate the amount of thioglycolic acid or cysteine in the sample by the following formula (%):

The amount of thioglycolic acid or cysteine in the sample (%) =

$$\frac{C \times V}{M} \times 10^{-4}$$

where,

C: the concentration of thioglycolic acid or cysteine in the sample solution calculated by the standard curve (μg/mL)

V: the final make-up volume of sample (mL)

M: the weight of sample (g)

HPLC operating conditions(note):

Photodiode array detector: quantitative wavelength 214 nm.

Column: LiChrospher® 100 RP-18 endcapped, 5 μm, 4.6 mm i.d. × 25 cm.

Column oven temperature: 40°C.

Mobile phase: an isocratic elution program of solvent A and solvent B is as follows.

Time (min)	Solvent A (%)	Solvent B (%)
0.0 → 20.0	90	10

Flow rate: 1.0 mL/min.

Injection volume: 20 μL

Note: All the parameters can be adjusted depending on the instruments used if the above conditions are not applicable.

Remark

1. Limits of quantitation (LOQs) is 0.2% for thioglycolic acid and 0.1% for cysteine.
2. Further validation should be performed when interference compounds appear in samples.

Reference

1. Kim, Y and Na, D. H. 2019. Simultaneous determination of cysteamine and cystamine in cosmetics by ion-pairing reversed-phase high-performance liquid chromatography. Toxicol. Res. 35: 161-165.
2. Kuśmierek, K. and Bald, E. 2008. Determination of N-acetylcysteine and thioglycolic acid in human urine. Chromatographia 67: 23-29.

Reference chromatogram

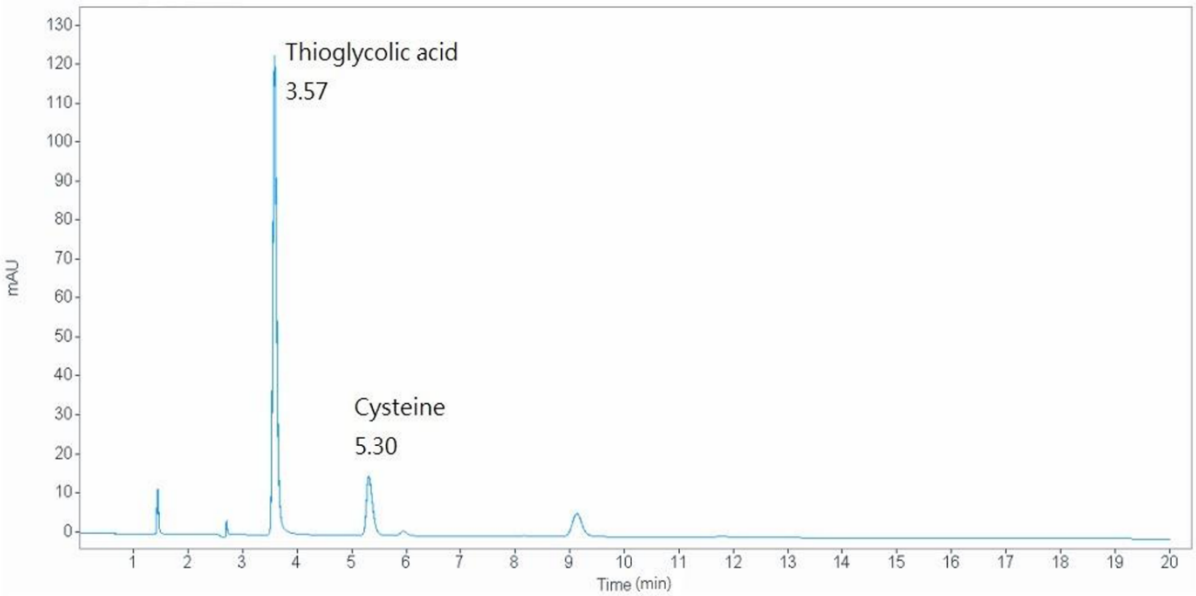

Figure. HPLC chromatogram of thioglycolic acid and cysteine reference standards.