

Method of Test for Preservatives in Cosmetics (2)

1. Scope

This method is applicable to the determination of 2,4-dichlorobenzyl alcohol, benzyl alcohol, bromochlorophene, chlorocresol, chlorphenesin, *p*-chlorophenol, phenoxyethanol and benzoyl peroxide in cosmetics.

2. Method

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

2.1. Equipments

2.1.1. High performance liquid chromatograph.

2.1.1.1. Detector: photodiode array detector.

2.1.1.2. Column: ACQUITY UPLC[®] BEH C18, 1.7 μ m, 2.1 mm i.d. \times 100 mm; or an equivalent product.

2.1.2. Ultrasonicator.

2.2. Chemicals

Methanol, HPLC grade;

Formic acid, GR grade;

Deionized water, resistivity $\geq 18 \text{ M}\Omega\cdot\text{cm}$ (at 25°C);

2,4-Dichlorobenzyl alcohol, benzyl alcohol, bromochlorophene, chlorocresol, chlorphenesin, *p*-chlorophenol, phenoxyethanol and benzoyl peroxide, reference standards.

2.3. Apparatus

2.3.1. Volumetric flask: 10 mL, 20 mL, 50 mL and 100 mL.

2.3.2. Membrane filter: 0.22 μ m, Nylon.

2.4. Mobile phase

2.4.1. Solvent A: Dilute 1 mL formic acid with deionized water to 1000 mL, and filter with a membrane filter.

2.4.2. Solvent B: Methanol.

2.5. Standard solution preparation

Accurately weigh 50 mg of 2,4-dichlorobenzyl alcohol, benzyl alcohol, bromochlorophene, chlorocresol, chlorphenesin, *p*-chlorophenol, phenoxyethanol, and benzoyl peroxide to

each 10-mL volumetric flask, dissolve and dilute with methanol to volume as standard stock solutions. When to use, mix appropriate volume of each standard stock solution and dilute with methanol to 10-500 µg/mL for benzyl alcohol, 5-100 µg/mL for 2,4-dichlorobenzyl alcohol, phenoxyethanol, and benzoyl peroxide, and 1-50 µg/mL for bromochlorophene, chlorocresol, chlorphenesin, and *p*-chlorophenol respectively as the standard solutions.

2.6. Sample solution preparation

Transfer about 1 g of the well-mixed sample accurately weighed into a 20-mL volumetric flask and add 10 mL of methanol, ultrasonicate for 30 min. Dilute to volume with methanol and filter with a membrane filter. Take the filtrate as the sample solution.

2.7. Identification and Quantitation

Accurately inject 2.5 µL of the sample solution and the standard solutions into the HPLC separately, and operate according to the following conditions. Identify each preservative based on the retention time and the UV absorption spectrum. Calculate the amount of each preservative in the sample by the following formula.

$$\text{The amount of each preservative in the sample (\%)} = \frac{C \times V}{M} \times 10^{-4}$$

Where,

C: the concentration of each preservative in the sample solution calculated by the standard curve (µg/mL).

V: the make up volume of sample (mL).

M: the weight of sample (g).

HPLC operating conditions:

Photodiode array detector: the quantitative wavelength for each preservatives is as follows.

Analyte	Wavelength (nm)
Benzyl alcohol	260

Phenoxyethanol	
Benzoyl peroxide	
2,4-Dichlorobenzyl alcohol	
Bromochlorophene	
Chlorocresol	280
Chlorphenesin	
<i>p</i> -Chlorophenol	

Column: ACQUITY UPLC[®] BEH C18, 1.7 μ m, 2.1 mm \times 100 mm.

Mobile phase: a gradient program of solvent A and solvent B is as follows.

Time (min)	Solvent A (%)	Solvent B (%)
0.0 \rightarrow 2.0	65 \rightarrow 65	35 \rightarrow 35
2.0 \rightarrow 4.0	65 \rightarrow 50	35 \rightarrow 50
4.0 \rightarrow 7.0	50 \rightarrow 30	50 \rightarrow 70
7.0 \rightarrow 11.0	30 \rightarrow 25	70 \rightarrow 75
11.0 \rightarrow 13.0	25 \rightarrow 0	75 \rightarrow 100
13.0 \rightarrow 17.5	0 \rightarrow 0	100 \rightarrow 100
17.5 \rightarrow 18.5	0 \rightarrow 65	100 \rightarrow 35
18.5 \rightarrow 20.0	65 \rightarrow 65	35 \rightarrow 35

Flow rate: 0.2 mL/min

Injection volume: 2.5 μ L

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

Remark

1. The limits of quantitation (LOQs) are as follows: 0.02% for benzyl alcohol, 0.01% for phenoxyethanol, 2,4-dichlorobenzyl alcohol and benzoyl peroxide, and 0.002% for bromochlorophene, chlorocresol, chlorphenesin and *p*-chlorophenol.
2. Further validation shall be done when interference compounds appear in samples.

Reference

1. Wu, T., Wang, C. and Ma, Q. 2008. Simultaneous determination of 21 preservatives in cosmetics by ultra performance liquid chromatography. *Int. J. Cosmet. Sci.* 30: 367-372.
2. Baranowska, I. and Wojciechowska, I. 2012. Development of SPE/HPLC-DAD to determine residues of selected disinfectant agents in surface water. *Pol. J. Environ. Stud.* 21: 269-277.
3. European Commission. 1999. Reports of the Scientific Committee on Cosmetology. ISBN 92-828-8951-3.