# Method of Test for Sunscreen Agents in Cosmetics

#### 1. Scope

This method is applicable to the determination of benzophenone-3, diethylamino hydroxybenzoyl hexyl benzoate, octocrylene, ethylhexyl dimethyl PABA, ethylhexyl methoxycinnamate, butyl methoxydibenzoylmethane, ethylhexyl salicylate and homosalate in cosmetics.

# 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: Inertsil<sup>®</sup> ODS-2, 5 μm, 4.6 mm i.d.× 25 cm, or an equivalent product.
- 2.1.2. Ultrasonicator.
- 2.2. Chemicals

Methanol, HPLC grade;

Acetic acid, reagent grade;

Deionized water, resistivity  $\geq$  18 M $\Omega$  · cm (at 25°C);

Benzophenone-3, diethylamino hydroxybenzoyl hexyl benzoate, octocrylene, ethylhexyl dimethyl PABA, ethylhexyl methoxycinnamate, butyl methoxydibenzoylmethane, ethylhexyl salicylate and homosalate, reference standards.

## 2.3. Apparatus

- 2.3.1. Volumetric flask: 10 mL.
- 2.3.2. Membrane filter: 0.45 µm, PVDF.

## 2.4. Mobile phase:

- **2.4.1.** Solvent A: Dilute 1 mL of acetic acid with deionized water to 1000 mL, and filter with a membrane filter.
- **2.4.2.** Solvent B: Methanol.

## 2.5. Standard solution preparation

Transfer about 10 mg of benzophenone-3, diethylamino hydroxybenzoyl hexyl benzoate, octocrylene, ethylhexyl dimethyl PABA, butyl methoxydibenzoylmethane (need freshly prepared), ethylhexyl methoxycinnamate, ethylhexyl salicylate and homosalate reference standards accurately weighed into each 10-mL volumetric flask, dissolve and dilute with methanol to volume as the standard stock solutions. Store in the refrigerator and protect from light. When to use, mix appropriate amount of each standard stock solution, and dilute with methanol to 1-50  $\mu$ g/mL for benzophenone-3, diethylamino hydroxybenzoyl hexyl benzoate, ethylhexyl dimethyl PABA, ethylhexyl methoxycinnamate, octocrylene, ethylhexyl salicylate and homosalate, and 2-50  $\mu$ g/mL for butyl methoxydibenzoylmethane as the standard solutions.

2.6. Sample solution preparation

Transfer about 1 g of the well-mixed sample accurately weighed into a 10-mL volumetric flask, add 5 mL of methanol, and ultrasonicate for 30 mins. 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 20  $\mu$ L of the sample solution and the standard solutions into HPLC separately, and operate according to the following conditions. Identify each sunscreen agent based on the retention time and the UV absorption spectrum. Calculate the amount of each sunscreen agent in the sample by the following formula:

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

where,

- C: the concentration of each sunscreen agent in the sample solution calculated by the standard curve (µg/mL)
- V: the final make-up volume of sample (mL)
- M: the weight of the sample (g)

HPLC operating conditions<sup>(note)</sup>:

Photodiode array detector:

Analyte	Quantitative wavelength (nm)
Benzophenone-3, octocrylene, ethylhexyl dimethyl PABA, ethylhexyl methoxycinnamate, ethylhexyl salicylate, homosalate	310

Diethylamino hydroxybenzoyl hexyl benzoate,<br/>butyl methoxydibenzoylmethane350Column: Inertsil® ODS-2, 5 µm, 4.6 mm i.d.× 25 cm.

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

Time (min)	Solvent A (%)	Solvent B (%)
0.0 → 30.0	10	90

Flow rate: 0.8 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

- Limits of quantification (LOQs) are 0.001% for benzophenone-3, diethylamino hydroxybenzoyl hexyl benzoate, octocrylene, ethylhexyl dimethyl PABA, ethylhexyl methoxycinnamate, ethylhexyl salicylate and homosalate, and 0.002% for butyl methoxydibenzoylmethane.
- 2. Further validation should be performed when interference compounds appear in samples.

## Reference

- Wharton, M., Geary, M., O'Connor, N., Curtin, L. and Ketcher, K. 2015. Simultaneous liquid chromatographic determination of 10 ultra-violet filters in sunscreens. J. Chromatogr. Sci. 53: 1289-1295.
- Kim, D., Kim, S., Kim, S. A., Choi, M., Kwon, K. J., Kim, M., Kim, D. S., Kim, S. H. and Choi, B. K. 2012. Simultaneous analysis and monitoring of 16 UV filters in cosmetics by high-performance liquid chromatography. J. Cosmet. Sci. 63: 103-117.

#### **Reference chromatogram**



Figure. HPLC chromatograms of 8 sunscreen agents reference standards.

benzophenone-3; 2. octocrylene; 3. ethylhexyl dimethyl PABA; 4.
ethylhexyl methoxycinnamate; 5. ethylhexyl salicylate; 6. homosalate;
7. diethylamino hydroxybenzoyl hexyl benzoate; 8. butyl methoxydibenzoylmethane.