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**Determination of forbidden bleach in toothpaste - High
performance liquid chromatography method**

牙膏中禁用漂白剂的测定 高效液相色谱法

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Determination of forbidden bleach in toothpaste - High performance liquid chromatography method

1 Scope

This Standard specifies high performance liquid chromatography method to determine the content of forbidden bleach - benzoyl peroxide - in toothpaste.

This Standard is applicable to the determination of the content of forbidden bleach - benzoyl peroxide - in toothpaste.

The method detection limit of this Standard is 0.5mg/kg.

2 Normative references

The following referenced documents are indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

GB/T 6682-2008, *Water for analytical laboratory use - Specification and test methods*

3 Principle

Acetonitrile for samples: After water extraction (80:20, volume ratio), after the supernatant is filtered through a 0.45 μ m filter membrane, use post-column derivatization-high performance liquid chromatography-fluorescence detection method to determine. Use external standard peak area method to quantify.

4 Reagents and standard materials

Unless otherwise specified, all reagents used are analytically-pure AND water is grade one water specified in GB/T 6682-2008.

4.1 Acetonitrile: chromatographically pure.

4.2 Hemin: mass fraction $\geq 98.0\%$.

4.3 P-hydroxyphenylacetic acid: mass fraction $\geq 98.0\%$.

(250mm×4.6mm×5μm).

- b) Fluorescence detector wavelength: excitation wavelength is 315nm, emission wavelength is 400nm.
- c) Mobile phase: methanol with 0.1% acetic acid: water (85:15, volume ratio).
- d) Flow rate: 0.6mL/min.
- e) Column temperature: 20°C.
- f) Injection volume: 10μL.
- g) Derivative conditions: The derivatization reagent are 8μmol/L hemin and 80μmol/L p-hydroxyphenylacetic acid in NH₄Cl-NH₃·H₂O buffer solution (pH=10.5). The back pressure of the post-column derivative is 500psi~1000psi or the flow rate is 0.2mL/min. The temperature of the reaction tube is 35°C.

7.2 Preparation of standard working solution

Accurately pipette 1.0mL, 2.0mL, 4.0mL, 8.0mL, 10.0mL of standard stock solution (4.6) in a 100mL volumetric flask. Use acetonitrile to dilute to the scale mark. Shake well. Filter it by 0.45μm filter membrane for future use. At this time, the mass concentration of the standard series solution is 10.0 mg/L, 20.0 mg/L, 40.0 mg/L, 80.0 mg/L, 100.0 mg/L. Under the set chromatographic conditions (7.1), respectively take 10μL for analysis. Take the mass concentration of the standard series solution as the abscissa (mg/L), the peak area of the reference substance as the ordinate to draw the calibration curve (linear correlation coefficient $R \geq 0.9990$). Refer to Figure A.1 in Annex A for the liquid chromatogram of benzoyl peroxide standard sample.

7.3 Determination steps

After the sample is processed according to the steps in Chapter 6, perform on-board testing according to the instrument conditions of 7.1. The response value of benzoyl peroxide in the test sample solution shall be within the linear range of the standard curve. If it exceeds the linear range, it shall be diluted before injection for determination.

7.4 Blank test

Except not adding specimen, all are tested in accordance with the processing steps in Chapter 6 and the instrument conditions in 7.1.

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