Cephalexin Hydrochloride

$C_{16}H_{17}N_3O_4S \cdot HCl \cdot H_2O$ 401.87

5-Thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid, 7-[(aminophenylacetyl)amino]-3-methyl-8-oxo-, monohydrochloride, monohydrate, [6*R*-[6α,7β (*R**)]]-;

(6*R*,7*R*)-7-[(2*R*)-2-Amino-2-phenylacetamido]-3-methyl-8-oxo-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid, monohydrochloride, monohydrate;

7-(D-2-Amino-2-phenylacetamido)-3-methyl-3-cephem-4-carboxylic acid hydrochloride monohydrate [105879-42-3].

DEFINITION

Cephalexin Hydrochloride contains the equivalent of NLT 800 µg and NMT 880 µg of cephalexin ($C_{16}H_{17}N_3O_4S$) per mg.

IDENTIFICATION

Delete the following:

• **A. THIN-LAYER CHROMATOGRAPHY** •

Standard solution: 25 mg/mL of USP Cephalexin RS in water with the aid of 0.1 N hydrochloric acid

Sample solution: 25 mg/mL in water with the aid of 0.1 N hydrochloric acid

Chromatographic system

(See *Chromatography* ⟨621⟩, *Thin-Layer Chromatography*.)

Mode: TLC

Adsorbent: 0.25-mm layer of chromatographic silica gel mixture

Application volume: 5 µL

Developing solvent system: Ethyl acetate, acetonitrile, glacial acetic acid, and water (21:7:7:9)

Analysis

Samples: *Standard solution* and *Sample solution*

Allow the spots to dry, place the plate in a saturated chamber containing the solvent system and lined with filter paper. Develop the chromatogram until the solvent front has moved three-fourths of the length of the plate. Remove the plate from the developing chamber, mark the solvent front, allow the plate to air-dry, and examine under short-wavelength UV light.

Acceptance criteria: The R_F value of the principal spot of the *Sample solution* corresponds to that of the *Standard solution*.•5•

Add the following:

• **A.** The retention time of the major peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the *Assay*.•5•

Delete the following:

• **B. PROCEDURE** •

Sample solution: 0.02 mg/mL of cephalexin in water

Analysis: The UV absorption spectrum of the *Sample solution* exhibits maxima and minima at the same wavelengths as that of a similar solution of USP Cephalexin RS, concomitantly measured.•5•

Change to read:

• •**B.**•5• **IDENTIFICATION TESTS—GENERAL,** *Chloride* ⟨**191**⟩**:** 10 mg/mL meets the requirements

ASSAY

Change to read:

• **PROCEDURE** •

Mobile phase: 0.985 g/L of sodium 1-pentanesulfonate in a mixture of acetonitrile, methanol, triethylamine, and water (20:10:3:170), adjusted with phosphoric acid to a pH of 3.0 ± 0.1

••5•

Standard stock solution: 1 mg/mL of USP Cephalexin RS in water

Standard solution: •0.4 mg/mL of cephalexin in *Mobile phase* from *Standard stock solution*•5•

Sample stock solution: 1.15 mg/mL of Cephalexin Hydrochloride in water

Sample solution: •0.4 mg/mL of cephalexin in *Mobile phase* from *Sample stock solution*•5•

Chromatographic system

(See *Chromatography* ⟨621⟩, *System Suitability*.)

Mode: LC

Detector: UV 254 nm

Column: 4.6-mm × 25-cm; packing L1 of low acidity

Flow rate: 1.5 mL/min

Injection size: 20 µL

System suitability

Sample: *Standard solution*

••5•

Suitability requirements

••5•

Relative standard deviation: NMT 2.0%

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the quantity, in µg, of $C_{16}H_{17}N_3O_4S$ in each mg of Cephalexin Hydrochloride taken:

$$\text{Result} = (r_U/r_S) \times (C_S/C_U) \times P$$

r_U = peak response from the *Sample solution*

r_S = peak response from the *Standard solution*•5•

C_S = concentration of USP Cephalexin RS in the *Standard stock solution* (mg/mL)

C_U = concentration of Cephalexin Hydrochloride from the *Sample stock solution* (mg/mL)

P = designated content of cephalexin in USP Cephalexin RS (µg/mg)

Acceptance criteria: 800–880 µg/mg

IMPURITIES

Organic Impurities

• **PROCEDURE 1** •

Solution A: 1 g of sodium 1-pentanesulfonate in a mixture of 1000 mL of water and 15 mL of triethylamine. Adjust with phosphoric acid to a pH of 2.5 ± 0.1.

Solution B: 1 g of sodium 1-pentanesulfonate in a mixture of 300 mL of water and 15 mL of triethylamine. Adjust with phosphoric acid to a pH of 2.5 ± 0.1, and add 350 mL of acetonitrile and 350 mL of methanol.

Mobile phase: See the gradient table below.

Time (min)	Solution A (%)	Solution B (%)
0	100	0
1	100	0
33.3	0	100
34.3	0	100

Diluent: 18 mg/mL of monobasic potassium phosphate in water

Standard solutions: 0.08 mg/mL and 0.16 mg/mL of $C_{16}H_{17}N_3O_4S$ from USP Cephalexin RS in *Diluent*, taking into account the stated potency of the USP Cephalexin RS

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Sample solution: 6 mg/mL of Cephalexin Hydrochloride in *Diluent*

Chromatographic system

(See *Chromatography* ⟨621⟩*, System Suitability*.)

Mode: LC

Detector: UV 254 nm

Column: 4.6-mm × 25-cm; packing L1 of low acidity

Flow rate: 1 mL/min

Injection size: 20 µL

Analysis

Samples: *Standard solutions* and *Sample solution*

Plot the responses of the cephalexin peaks of the *Standard solutions* versus their concentrations, calculated on the anhydrous basis, in mg/mL, and draw a straight line through the two points and zero. From the line so obtained and the peak responses of the *Sample solution*, determine the concentration, I, in mg/mL, of each cephalexin-related substance from the *Sample solution* other than the cephalexin peak.

Calculate the percentage of each cephalexin-related substance represented by each peak of the *Sample solution*, other than the cephalexin peak.

$$\text{Result} = (I/C) \times 100$$

I = concentration of each cephalexin-related substance other than cephalexin in the *Sample solution* (mg/mL)

C = concentration mg/mL of cephalexin from the *Sample solution*

Acceptance criteria

Individual impurities: NMT 1.0% of any individual cephalexin-related substance is found.

Total impurities: NMT 5.0%

- **PROCEDURE 2: DIMETHYLANILINE ⟨223⟩:** Meets the requirement

SPECIFIC TESTS

- **CRYSTALLINITY ⟨695⟩:** Meets the requirements
- **pH ⟨791⟩:** 1.5–3.0, in a solution containing 10 mg/mL
- **WATER DETERMINATION,** *Method I* **⟨921⟩:** 3.0%–6.5%

ADDITIONAL REQUIREMENTS

- **PACKAGING AND STORAGE:** Preserve in tight containers.
- **USP REFERENCE STANDARDS ⟨11⟩**

USP Cephalexin RS

In-Process Revision

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