Edition: BP 2025 (Ph. Eur. 11.6 update)

Dexamethasone and Neomycin Ear Spray

General Notices

Action and use

Glucocorticoid.

DEFINITION

Dexamethasone and Neomycin Ear Spray is an emulsion containing Dexamethasone in *microfine powder* and Neomycin Sulfate in a suitable vehicle in a suitable metered-dose container. It may contain acetic acid.

The ear spray complies with the requirements stated under Ear Preparations and with the following requirements.

Content of dexamethasone, $C_{22}H_{29}FO_5$

80.0 to 120.0% of the amount stated to be delivered by actuation of the valve.

Shake the ear spray vigorously before carrying out the following tests.

IDENTIFICATION

A. In the Assay for dexamethasone, the chromatogram obtained with solution (1) shows a peak with the same retention time as the principal peak in the chromatogram obtained with solution (2).
B. Carry out the method for *thin-layer chromatography*, Appendix III A, using the following solutions.
(1) Discharge the container a sufficient number of times to obtain a suitable quantity and dilute with *methanol*, if necessary, to produce a solution containing 0.1% w/v of Dexamethasone.
(2) 0.1% w/v of *dexamethasone BPCRS* in *methanol*.
(3) 0.1% w/v of each of *dexamethasone BPCRS* and *betamethasone BPCRS* in *methanol*.

CHROMATOGRAPHIC CONDITIONS

(a) Use a *TLC silica gel F_{254} plate* (Merck silica gel 60 F_{254} plates are suitable).
(b) Use the mobile phase as described below.
(c) Apply 5 µL of each solution.
(d) Develop the plate to 15 cm.
(e) After removal of the plate, allow it to dry in air and examine under *ultraviolet light (254 nm)* (detection method A). Spray with *alcoholic solution of sulfuric acid*. Heat at 120° for 10 minutes or until the spots appear. Allow to cool. Examine the chromatograms in daylight and under *ultraviolet light (365 nm)* (detection method B).

MOBILE PHASE

5 volumes of *butan-2-ol* saturated with *water*, 10 volumes of *toluene* and 85 volumes of *ether*.

SYSTEM SUITABILITY

The test is not valid unless the chromatogram obtained with solution (3) shows two spots which may, however, not be completely separated.

CONFIRMATION

Method A The principal spot in the chromatogram obtained with solution (1) is similar in position and size to the principal spot in the chromatogram obtained with solution (2).

Method B The principal spot in the chromatogram obtained with solution (1) is similar in position, colour in daylight, fluorescence in ultraviolet light at 365 nm and size to the principal spot in the chromatogram obtained with solution (2).

C. In the test for Neomycin C, the principal spot in the chromatogram obtained with solution (1) is similar in position, colour and size to the principal spot in the chromatogram obtained with solution (4).

Dexamethasone and Neomycin Ear Spray containing acetic acid complies with the following additional test.

D. Discharge the container a sufficient number of times to produce 0.2 g. The solution yields reaction A characteristic of *acetates*, Appendix VI.

TESTS

Acidity

pH, 2.0 to 3.0, Appendix V L.

Neamine

Carry out the method for *thin-layer chromatography*, Appendix III A, using the following solutions.

(1) Discharge the container a sufficient number of times to obtain a suitable quantity (0.5% w/w of neomycin sulfate is suitable).
(2) 0.01% w/v of *neamine EPCRS* in *water*.
(3) Mix 1 volume of solution (1) and 1 volume of solution (2).

CHROMATOGRAPHIC CONDITIONS

(a) Use a *TLC silica gel plate*.
(b) Use the mobile phase as described below.
(c) Apply 5 µL of each solution, as 5-mm bands.
(d) Develop the plate to 15 cm.
(e) After removal of the plate, dry it at 100° to 105° for 10 minutes, spray with *ninhydrin and stannous chloride reagent*, heat at 110° for 15 minutes, spray with the same reagent and heat at 110° for 15 minutes.

MOBILE PHASE

10 volumes of *dichloromethane*, 20 volumes of 13.5M *ammonia* and 30 volumes of *methanol*.

SYSTEM SUITABILITY

The test is not valid unless the chromatogram obtained with solution (3) shows two clearly separated principal spots.

LIMITS

In the chromatogram obtained with solution (1) any spot corresponding to neamine is not more intense than the spot in the chromatogram obtained with solution (2) (2%).

Neomycin C

Carry out the method for *thin-layer chromatography*, Appendix III A, using the following solutions.

(1) Discharge the container a sufficient number of times to obtain a suitable quantity (0.5% w/w of neomycin sulfate is suitable).
(2) 0.075% w/v of *framycetin sulfate EPCRS* in *water*.
(3) Dilute 1 volume of solution (2) to 5 volumes with *water*.
(4) 0.5% w/v of *neomycin sulfate EPCRS* in *water*.

CHROMATOGRAPHIC CONDITIONS

(a) Use a *TLC silica gel plate*.
(b) Use the mobile phase as described below.
(c) Apply 5 µL of each solution, as 5-mm bands.
(d) Develop the plate to 15 cm.
(e) After removal of the plate, dry it at 100° to 105° for 10 minutes, spray with *ninhydrin solution R1* and heat at 100° to 105° for 10 minutes.

MOBILE PHASE

20 volumes of *methanol* and 80 volumes of a 20% w/v solution of *sodium chloride*.

SYSTEM SUITABILITY

The test is not valid unless, in the chromatogram obtained with solution (4), a spot appears with an Rf value slightly less than that of the principal spot.

LIMITS

In the chromatogram obtained with solution (1) the spot with an Rf value slightly less than that of the principal spot (neomycin C) is not more intense than the spot in the chromatogram obtained with solution (2) (15%) but is more intense than the spot in the chromatogram obtained with solution (3) (3%).

Related substances

Dexamethasone

Carry out the method for *liquid chromatography*, Appendix III D, using the following solutions.

(1) After priming the pump, discharge the container a sufficient number of times to obtain 2.5 mg of Dexamethasone, add 1.5 mL of *acetonitrile R* and 5 mL of mobile phase A. Mix with the aid of ultrasound, add sufficient mobile phase A to produce 10 mL and filter through a 0.45-µm filter.
(2) Dilute 1 mL of solution (1) to 100 mL with mobile phase A.
(3) 0.002% w/v of each of *methylprednisolone BPCRS* and *dexamethasone BPCRS* in mobile phase A.
(4) Dilute 1 mL of solution (2) to 20 mL with mobile phase A.

CHROMATOGRAPHIC CONDITIONS

(a) Use a stainless steel column (25 cm × 4.6 mm) packed with *octadecylsilyl silica gel for chromatography* (5 µm) (Hypersil ODS is suitable).
(b) Use gradient elution and the mobile phase described below.
(c) Use a flow rate of 2.5 mL per minute.
(d) Use a column temperature of 45°.
(e) Use a detection wavelength of 254 nm.
(f) Inject 20 µL of each solution.

MOBILE PHASE

Mobile phase A 25% v/v *acetonitrile*.

Mobile phase B *acetonitrile*.

Time (min)	Mobile phase A (per cent *V/V*)	Mobile phase B (per cent *V/V*)	Comment
0	100	0	isocratic
15	100→0	0→100	begin linear gradient
40	0	100	end chromatogram, return to 100 A
41	100	0	begin equilibration with A
46 = 0	100	0	end equilibration, begin next chromatogram

When the chromatograms are recorded under the prescribed conditions, the retention times are: methylprednisolone, about 12 minutes; dexamethasone, about 14 minutes.

SYSTEM SUITABILITY

The test is not valid unless:

(a) in the chromatogram obtained with solution (3), the *resolution factor* between the peaks corresponding to methylprednisolone and dexamethasone is at least 1.5 (if necessary, adjust the concentration of *acetonitrile* in mobile phase A);

(b) in the chromatogram obtained with solution (4), the *signal to noise ratio* of the peak due to dexamethasone is at least 10.

LIMITS

In the chromatogram obtained with solution (1):

the area of any *secondary peak* is not greater than 0.5 times the area of the principal peak in the chromatogram obtained with solution (2) (0.5%);

the sum of the areas of all the *secondary peaks* is not greater than the area of the principal peak in the chromatogram obtained with solution (2) (1%).

Disregard any peak due to mobile phase A and any peak with an area less than the area of the principal peak in the chromatogram obtained with reference solution (4) (0.05%).

ASSAY

For dexamethasone

Carry out the method for *liquid chromatography*, Appendix III D, using the following solutions protected from light.

(1) After priming the pump, discharge the container a sufficient number of times to obtain 1 mg of Dexamethasone, add 10 mL of *methanol*, place in an ultrasonic bath for 10 minutes, cool, dilute to 25 mL with *water*, mix and filter through a 0.45-µm PTFE filter.
(2) Dilute 10 volumes of a 0.010% w/v solution of *dexamethasone BPCRS* in *methanol* to 25 volumes with *water*.

CHROMATOGRAPHIC CONDITIONS

(a) Use a stainless steel column (15 cm × 4.6 mm) packed with *octadecylsilyl silica gel for chromatography* (5 µm) (Spherisorb ODS 2 is suitable).
(b) Use isocratic elution and the mobile phase described below.
(c) Use a flow rate of 2 mL per minute.
(d) Use an ambient column temperature.
(e) Use a detection wavelength of 254 nm.
(f) Inject 20 µL of each solution.

MOBILE PHASE

Mix 10 volumes of *glacial acetic acid*, 350 volumes of *acetonitrile* and 640 volumes of *water* and filter (Whatman GF/F is suitable).

DETERMINATION OF CONTENT

Calculate the content of $C_{22}H_{29}FO_5$ in the ear spray using the declared content of $C_{22}H_{29}FO_5$ in *dexamethasone BPCRS*

For neomycin sulfate

Prime the pump and discharge the container a sufficient number of times to obtain a quantity of the emulsion containing 3250 IU; dilute to 50 mL with *sterile phosphate buffer pH 8.0*. Dilute 10 mL of the resulting solution to 100 mL with the same solvent and carry out the *microbiological assay of antibiotics*, Appendix XIV A. The precision of the assay is such that the fiducial limits of error are not less than 95% and not more than 105% of the estimated potency. The upper fiducial limit of error is not less than 90.0% and the lower fiducial limit of error is not more than 115.0% of the stated number of IU per mL.

STORAGE

Dexamethasone and Neomycin Ear Spray should not be allowed to freeze.

LABELLING

The strength with respect to Neomycin Sulfate is stated as the number of IU (Units) per mL.