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Naloxone Injection

General Notices

Action and use

Opioid receptor antagonist.

DEFINITION

Naloxone Injection is a sterile solution of Naloxone Hydrochloride in Water for Injections.

The injection complies with the requirements stated under Parenteral Preparations and with the following requirements.

Content of anhydrous naloxone hydrochloride, $C_{19}H_{21}NO_4$,HCl

95.0 to 105.0% of the stated amount.

IDENTIFICATION

A. Carry out the method for *thin-layer chromatography*, Appendix III A, using the following solutions.
(1) Dilute a volume of the injection with *water*, if necessary, to produce a solution containing the equivalent of 0.002% w/v of anhydrous naloxone hydrochloride. Add 1 mL of *ammonia buffer pH 10.0* to a volume of the solution containing the equivalent of 0.1 mg of anhydrous naloxone hydrochloride, extract with three 20-mL quantities of a mixture of 1 volume of *propan-2-ol* and 3 volumes of *chloroform*, dry the combined extracts over *anhydrous sodium sulfate*, filter, evaporate the filtrate to dryness and dissolve the residue in 1 mL of *methanol*.
(2) 0.01% w/v of *naloxone hydrochloride BPCRS* in *methanol*.

CHROMATOGRAPHIC CONDITIONS

(a) Use as the coating *silica gel* and heat the plate at 105° for 15 minutes immediately before use.
(b) Use the mobile phase as described below.
(c) Apply 20 µL of each solution.
(d) Protect the plate from light during development and develop to 10 cm.
(e) After removal of the plate, dry in a current of air, spray with a freshly prepared 0.5% w/v solution of *potassium hexacyanoferrate(III)* in *iron(III) chloride solution R1* and examine in daylight.

MOBILE PHASE

5 volumes of *methanol* and 95 volumes of the upper layer from a mixture of 60 mL of 2M *ammonia* and 100 mL of *butan-1-ol*.

CONFIRMATION

The principal spot in the chromatogram obtained with solution (1) corresponds to that in the chromatogram obtained with solution (2).

B. In the Assay, the retention time of the principal peak in the chromatogram obtained with solution (1) has the same retention time as the principal peak in the chromatogram obtained with solution (2).

TESTS

Acidity

pH, 3.0 to 4.5, Appendix V L.

Related substances

Carry out the method for *liquid chromatography*, Appendix III D, using the following solutions in 0.0025M *hydrochloric acid R*.

(1) Dilute a quantity of injection, if necessary, to produce a solution containing the equivalent of 0.002% w/v of anhydrous naloxone hydrochloride.
(2) Dilute 1 volume of solution (1) to 20 volumes. Further dilute 1 volume of this solution to 25 volumes.
(3) 0.005% w/v *naloxone for peak identification EPCRS*.

CHROMATOGRAPHIC CONDITIONS

Use a stainless steel column (12.5 cm × 4.0 mm) packed with *end-capped octylsilyl silica gel for chromatography* (5 µm) (Hypersil C8, 5 µm is suitable).
(b) Use gradient elution and the mobile phase described below.
(c) Use a flow rate of 1.5 mL per minute.
(d) Use a column temperature of 40°.
(e) Use a detection wavelength of 230 nm.
(f) Inject 200 µL of each solution.

MOBILE PHASE

Mobile phase A 94 volumes of 0.11% w/v *sodium octanesulfonate R* in *water R* adjusted to pH 2.0 with 50% v/v solution of *phosphoric acid R* (solution A), 4 volumes *tetrahydrofuran*, 2 volumes *acetonitrile R*.

Mobile phase B 79 volumes solution A, 17 volumes *acetonitrile R*, 4 volumes *tetrahydrofuran*.

Time (Minutes)	Mobile phase A (% v/v)	Mobile phase B (% v/v)	Comment
0-40	100	0	isocratic
40-50	100→0	0→100	linear gradient
50-60	0→100	100→0	linear gradient
60-70	100	0	re-equilibration

When the chromatograms are recorded under the prescribed conditions, the retention times relative to naloxone hydrochloride (retention time about 11 minutes) are: impurity C, about 0.6; impurity A, about 0.8; impurity F, about 0.9; impurity D, about 1.1; impurity E, about 3.0; impurity B, about 3.2.

SYSTEM SUITABILITY

In the chromatogram obtained with solution (3):

The peak-to-valley ratio at least 2.0, where *Hp* is the height above the baseline of the peak due to impurity D and *Hv* is the height above the baseline of the lowest point of the curve separating this peak from the peak due to naloxone.

LIMITS

Identify any peaks in the chromatogram obtained with solution (1) corresponding to impurities A, B, C, D, E and F using solution (3) and multiply the area of the peak corresponding to impurity E using a correction factor of 0.5.

In the chromatogram obtained with solution (1):

the area of any peak corresponding to impurity A, B, C, E and F is not greater than the area of the principal peak in the chromatogram obtained with solution (2) (0.2%);

the area of any other *secondary peak* is not greater than the area of the principal peak in the chromatogram obtained with solution (2) (0.2%);

the sum of the areas of any secondary peaks is not greater than 4 times the area of the principal peak in the chromatogram obtained with solution (2) (0.8 %).

Disregard any peak with an area less than 0.5 times the area of the principal peak in the chromatogram obtained with solution (2) (0.1%).

ASSAY

Carry out the method for *liquid chromatography*, Appendix III D, using the following solutions in a mixture of 1 volume of *orthophosphoric acid*, 450 volumes of *methanol* and 550 volumes of *water*.

(1) Dilute a weighed quantity of injection containing 0.1 mg of anhydrous naloxone hydrochloride to 10 mL.
(2) 0.001% w/v of *naloxone hydrochloride BPCRS*.
(3) 0.001% w/v of *naloxone hydrochloride BPCRS* and 0.0005% w/v of *noroxymorphone*.

CHROMATOGRAPHIC CONDITIONS

(a) Use a stainless steel column (25 cm × 4.6 mm) packed with *end-capped octadecylsilyl silica gel for chromatography* (5 to 10 µm) (Zorbax C18, 7 to 8 µm is suitable).
(b) Use isocratic elution and the mobile phase described below.
(c) Use a flow rate of 1 mL per minute.
(d) Use an ambient column temperature.
(e) Use a detection wavelength of 229 nm.
(f) Inject 20 µL of each solution.

MOBILE PHASE

0.068% w/v of *sodium octanesulfonate* and 0.1% w/v of *sodium chloride* in a mixture of 1 volume of *orthophosphoric acid*, 450 volumes of *methanol* and 550 volumes of *water*.

When the chromatograms are recorded under the prescribed conditions, the retention times of noroxymorphone and naloxone are about 3.1 minutes and 3.8 minutes respectively.

SYSTEM SUITABILITY

The test is not valid unless, in the chromatogram obtained with solution (3), the *resolution* between the peaks due to naloxone and noroxymorphone is at least 1.3.

DETERMINATION OF CONTENT

Calculate the content of $C_{19}H_{21}NO_4$,HCl in the injection using the declared content of $C_{19}H_{21}NO_4$,HCl in *naloxone hydrochloride BPCRS*.

STORAGE

Naloxone Injection should be protected from light.

LABELLING

The quantity of active ingredient is stated in terms of the equivalent amount of anhydrous naloxone hydrochloride.

When Neonatal Naloxone Injection is prescribed or demanded, an injection containing 20 mg/mL of naloxone hydrochloride shall be dispensed or supplied.