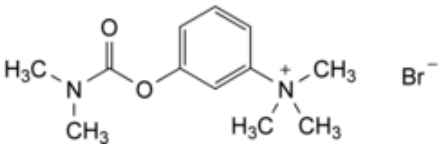
Edition: BP 2025 (Ph. Eur. 11.6 update)

Neostigmine Bromide

General Notices

(Ph. Eur. monograph 0046)



$C_{12}H_{19}BrN_2O_2$ 303.2 *114-80-7*

Action and use

Cholinesterase inhibitor.

Preparation

Neostigmine Tablets

Ph Eur

DEFINITION

3-[(Dimethylcarbamoyl)oxy]-*N*,*N*,*N*-trimethylanilinium bromide.

Content

98.5 per cent to 101.0 per cent (dried substance).

CHARACTERS

Appearance

White or almost white, crystalline powder or colourless crystals, hygroscopic.

Solubility

Very soluble in water, freely soluble in ethanol (96 per cent).

IDENTIFICATION

First identification: B, D.

Second identification: A, C, D.

A. Ultraviolet and visible absorption spectrophotometry (2.2.25).

Test solution Dissolve 20 mg in *0.5 M sulfuric acid* and dilute to 100 mL with the same acid.

Spectral range 230-350 nm.

Absorption maxima 260 nm and 266 nm.

Specific absorbance at the absorption maxima:

— 260 nm: about 16;

— 266 nm: about 14.

B. Infrared absorption spectrophotometry (2.2.24).

Comparison *neostigmine bromide CRS*.

C. To 50 mg add 0.4 g of *potassium hydroxide R* and 2 mL of *ethanol (96 per cent) R* and heat on a water-bath for 3 min, replacing the evaporated ethanol (96 per cent). Cool and add 2 mL of *water R* and 2 mL of *diazobenzenesulfonic acid solution R1*. An orange-red colour develops.

D. It gives reaction (a) of bromides (2.3.1).

TESTS

Solution S

Dissolve 2.5 g in *distilled water R* and dilute to 50 mL with the same solvent.

Appearance of solution

Solution S is clear (2.2.1) and colourless (2.2.2, *Method II*).

Related substances

Liquid chromatography (2.2.29).

Test solution Dissolve 50 mg of the substance to be examined in the mobile phase and dilute to 50.0 mL with the mobile phase.

Reference solution (a) Dilute 1.0 mL of the test solution to 100.0 mL with the mobile phase. Dilute 1.0 mL of this solution to 10.0 mL with the mobile phase.

Reference solution (b) Dissolve 4 mg of *3-dimethylaminophenol R* (impurity B) in 50 mL of the mobile phase. Dilute 1 mL of the solution to 200 mL with the mobile phase.

Reference solution (c) Dissolve the contents of a vial of *neostigmine impurity A CRS* in 1 mL of reference solution (b).

Reference solution (d) Mix 1 mL of the mobile phase and 1 mL of reference solution (a).

Column:

— *size*: *l* = 0.25 m, Ø = 4.0 mm;

— *stationary phase*: *base-deactivated octylsilyl silica gel for chromatography R* (5 µm);

— *temperature*: 30 °C.

Mobile phase To 710 mL of a 3.6 g/L solution of *sodium dihydrogen phosphate R* previously adjusted to pH 3.2 with *phosphoric acid R*, add 4.3 g of *sodium dodecyl sulfate R* and 290 mL of *acetonitrile for chromatography R*.

Flow rate 1.6 mL/min.

Detection Spectrophotometer at 220 nm.

Injection 50 µL of the test solution and reference solutions (a), (c) and (d).

Run time Twice the retention time of neostigmine.

Identification of impurities Use the chromatogram obtained with reference solution (c) to identify the peaks due to impurities A and B.

Relative retention With reference to neostigmine (retention time = about 20 min): impurity B = about 0.56; impurity A = about 0.61.

System suitability:

— *resolution*: minimum 1.5 between the peaks due to impurities B and A in the chromatogram obtained with reference solution (c);

— *signal-to-noise ratio*: minimum 25 for the principal peak in the chromatogram obtained with reference solution (d).

Calculation of percentage contents:

— for each impurity, use the concentration of neostigmine bromide in reference solution (a);

— *correction factor*: multiply the peak area of impurity B by 0.5.

Limits:

— *impurity B*: maximum 0.01 per cent;

— *unspecified impurities*: for each impurity, maximum 0.10 per cent;

— *total*: maximum 0.3 per cent;

— *reporting threshold*: 0.05 per cent; disregard the peak due to impurity B.

Sulfates (*2.4.13*)

Maximum 200 ppm, determined on solution S.

Loss on drying (*2.2.32*)

Maximum 1.0 per cent, determined on 1.000 g by drying in an oven at 105 °C.

Sulfated ash (*2.4.14*)

Maximum 0.1 per cent, determined on 1.0 g.

ASSAY

Dissolve 0.225 g in 2 mL of *formic acid R*. Add 50 mL of *acetic anhydride R*. Titrate with *0.1 M perchloric acid*, determining the end-point potentiometrically (*2.2.20*).

1 mL of *0.1 M perchloric acid* is equivalent to 30.32 mg of $C_{12}H_{19}BrN_2O_2$.

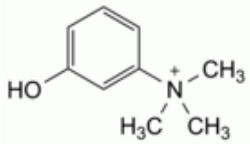
STORAGE

In an airtight container, protected from light.

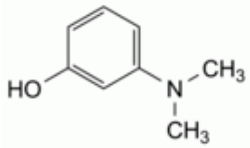
IMPURITIES

Specified impurities B.

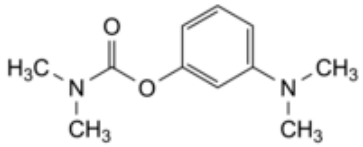
Other detectable impurities (the following substances would, if present at a sufficient level, be detected by one or other of the tests in the monograph. They are limited by the general acceptance criterion for other/unspecified impurities and/or by the general monograph Substances for pharmaceutical use (2034). It is therefore not necessary to identify these impurities for demonstration of compliance. See also 5.10. Control of impurities in substances for pharmaceutical use) A, C.



A. 3-hydroxy-*N*,*N*,*N*-trimethylanilinium,



B. 3-(dimethylamino)phenol,



C. 3-(dimethylamino)phenyl dimethylcarbamate.

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