Reference solution (a). Dissolve 6.0 mg of *oxitropium bromide impurity D CRS* in the mobile phase and dilute to 50.0 ml with the mobile phase.

Reference solution (b). Dilute 5.0 ml of reference solution (a) to 200.0 ml with the mobile phase. Dilute 5.0 ml of this solution to 50.0 ml with the mobile phase.

Reference solution (c). To 5.0 ml of the test solution add 5.0 ml of reference solution (a).

Column:

– *size*: *l* = 0.125 m, Ø = 4.0 mm;

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

Mobile phase: mix 185 volumes of *acetonitrile for chromatography R* with 1000 volumes of a 7.8 g/l solution of *sodium dihydrogen phosphate R*.

Flow rate: 2.0 ml/min.

Detection: spectrophotometer at 210 nm.

Injection: 50 µl of the test solution and reference solutions (b) and (c).

System suitability: reference solution (c):

– *resolution*: minimum 3.0 between the peaks due to impurity D and oxitropium.

Limit:

– *impurity D*: not more than the area of the corresponding peak in the chromatogram obtained with reference solution (b) (0.2 per cent).

Loss on drying (*2.2.32*): maximum 0.5 per cent, determined on 1.000 g by drying in an oven at 105 °C.

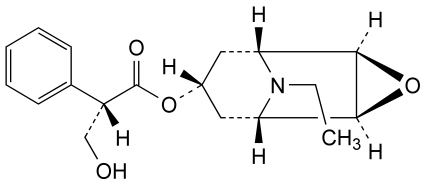
ASSAY

Dissolve 0.350 g in 100 ml of *water R* and add 5.0 ml of *dilute nitric acid R*. Titrate with *0.1 M silver nitrate*. Determine the end-point potentiometrically (*2.2.20*) using a silver indicator electrode and a silver-silver chloride reference electrode.

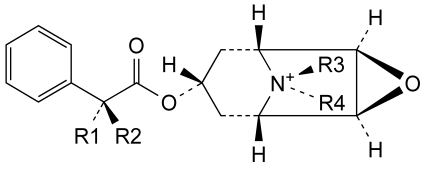
1 ml of *0.1 M silver nitrate* is equivalent to 41.23 mg of $C_{19}H_{26}BrNO_4$.

IMPURITIES

Specified impurities: A, B, C, D.



A. (1*R*,2*R*,4*S*,5*S*,7*s*)-9-ethyl-3-oxa-9-azatricyclo-[3.3.1.$0^{2,4}$]nonan-7yl (2*S*)-3-hydroxy-2-phenylpropanoate (*N*-ethylnorhyoscine),



B. R1 = CH_2OH, R2 = H, R3 = R4 = CH_3: (1*R*,2*R*,4*S*,5*S*,7*s*)-7-[[(2*S*)-3-hydroxy-2-phenylpropanoyl]oxy]-9,9-dimethyl-3-oxa-9-azoniatricyclo[3.3.1.$0^{2,4}$]nonane (methylhyoscine),

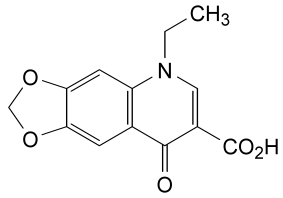
C. R1 = CH_2OH, R2 = H, R3 = C_2H_5, R4 = CH_3: (1*R*,2*R*,4*S*,5*S*,7*s*,9*s*)-9-ethyl-7-[[(2*S*)-3-hydroxy-2-phenylpropanoyl]oxy]-9-methyl-3-oxa-9-azoniatricyclo[3.3.1.$0^{2,4}$]nonane (pseudo-isomer),

D. R1 + R2 = CH_2, R3 = CH_3, R4 = C_2H_5: (1*R*,2*R*,4*S*,5*S*,7*s*,9*r*)-9-ethyl-9-methyl-7-[(2-phenylprop-2-enoyl)oxy]-3-oxa-9-azoniatricyclo[3.3.1.$0^{2,4}$]nonane (apo-*N*-ethylhyoscine).

01/2008:1353
corrected 6.0

OXOLINIC ACID

Acidum oxolinicum



$C_{13}H_{11}NO_5$ M_r 261.2
[14698-29-4]

DEFINITION

Oxolinic acid contains not less than 98.0 per cent and not more than the equivalent of 102.0 per cent of 5-ethyl-8-oxo-5,8-dihydro-1,3-dioxolo[4,5-*g*]quinoline-7-carboxylic acid, calculated with reference to the dried substance.

CHARACTERS

An almost white or pale yellow, crystalline powder, practically insoluble in water, very slightly soluble in methylene chloride, practically insoluble in alcohol. It dissolves in dilute solutions of alkali hydroxides.

IDENTIFICATION

First identification: B.

Second identification: A, C.

A. Dissolve 25.0 mg in 5 ml of *0.1 M sodium hydroxide*, heating on a water-bath. Allow to cool and dilute to 100.0 ml with *methanol R*. Dilute 2.0 ml of the solution to 100.0 ml with *0.1 M hydrochloric acid*. Examined between 220 nm and 350 nm (*2.2.25*), the solution shows three absorption maxima, at 260 nm, 322 nm and 336 nm respectively. The ratio of the absorbance measured at the maximum at 260 nm to that measured at the maximum at 336 nm is 4.9 to 5.2.

B. Examine by infrared absorption spectrophotometry (*2.2.24*), comparing with the spectrum obtained with *oxolinic acid CRS*. Examine the substances prepared as discs.

C. Examine by thin-layer chromatography (*2.2.27*), using a suitable silica gel as the coating substance.

Test solution. Dissolve 10 mg of the substance to be examined in 3 ml of *dilute sodium hydroxide solution R* and dilute to 20 ml with *alcohol R*.

Reference solution (a). Dissolve 10 mg of *oxolinic acid CRS* in 3 ml of *dilute sodium hydroxide solution R* and dilute to 20 ml with *alcohol R*.

Reference solution (b). Dissolve 5 mg of *ciprofloxacin hydrochloride CRS* in *methanol R* and dilute to 10 ml with the same solvent. Dilute 1 ml of the solution to 2 ml with reference solution (a).

Apply separately to the plate 10 µl of each solution. At the bottom of a chromatographic tank, place an evaporating disk containing 50 ml of *concentrated ammonia R*. Close the tank and expose the plate to the ammonia vapour for 15 min. Withdraw the plate and transfer

to a chromatographic tank and develop over a path of 15 cm using a mixture of 10 volumes of *acetonitrile R*, 20 volumes of *concentrated ammonia R*, 40 volumes of *methanol R* and 40 volumes of *methylene chloride R*. Allow the plate to dry in air. Examine in ultraviolet light at 254 nm. The principal spot in the chromatogram obtained with the test solution is similar in position, fluorescence and size to the principal spot in the chromatogram obtained with reference solution (a). The identification is not valid unless the chromatogram obtained with reference solution (b) shows two clearly separated spots.

TESTS

Solution S. Dissolve 0.6 g in 20 ml of a 40 g/l solution of *sodium hydroxide R*.

Appearance of solution. Solution S is clear (*2.2.1*) and not more intensely coloured than reference solution B_7 (*2.2.2, Method II*).

Related substances. Examine by thin-layer chromatography (*2.2.27*), using as the coating substance a suitable cellulose with a particle size of narrow distribution.

Test solution. Dissolve 50 mg in 3 ml of *dilute sodium hydroxide solution R* and dilute to 10 ml with *alcohol R*.

Reference solution (a). Dilute 1 ml of the test solution to 50.0 ml with *alcohol R*. Dilute 1.0 ml of the solution to 5.0 ml with *alcohol R*.

Reference solution (b). Dissolve 2 mg of *oxolinic acid impurity B CRS* in *alcohol R* and dilute to 10 ml with the same solvent. Dilute 0.5 ml of the solution to 10 ml with *alcohol R*.

Reference solution (c). Dissolve 5 mg of the substance to be examined and 5 mg of *oxolinic acid impurity A CRS* in 2 ml of *dilute sodium hydroxide solution R* and dilute to 40 ml with *alcohol R*.

Apply separately to the plate 5 µl of each solution, in sufficiently small portions to obtain small spots. Develop over a path of 6 cm (corresponding to two thirds of the plate height) with a mixture of 15 volumes of *ammonia R*, 30 volumes of *water R* and 55 volumes of *propanol R*. Allow the plate to dry in air and examine in ultraviolet light at 254 nm. In the chromatogram obtained with the test solution: any spot corresponding to oxolinic acid impurity B is not more intense than the spot in the chromatogram obtained with reference solution (b) (0.2 per cent); any spot apart from the principal spot and any spot corresponding to oxolinic acid impurity B is not more intense than the spot in the chromatogram obtained with reference solution (a) (0.4 per cent). The test is not valid unless the chromatogram obtained with reference solution (c) shows two clearly separated principal spots.

Heavy metals (*2.4.8*). 2.0 g complies with limit test D for heavy metals (10 ppm). Prepare the standard using 2 ml of *lead standard solution (10 ppm Pb) R*.

Loss on drying (*2.2.32*). Not more than 0.5 per cent determined on 1.000 g by heating in an oven at 105 °C.

Sulphated ash (*2.4.14*). Not more than 0.1 per cent determined on 1.0 g.

ASSAY

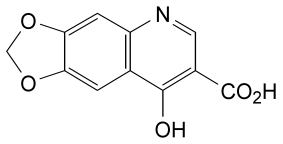
Dissolve 0.200 g in 150 ml of *dimethylformamide R*. Titrate with *0.1 M tetrabutylammonium hydroxide*, determining the end-point potentiometrically (*2.2.20*). Use a glass indicator electrode and a calomel reference electrode containing, as the electrolyte, a saturated solution of *potassium chloride R* in *methanol R*. Carry out a blank titration.

1 ml of *0.1 M tetrabutylammonium hydroxide* is equivalent to 26.12 mg of $C_{13}H_{11}NO_5$.

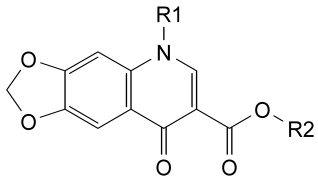
STORAGE

Store protected from light.

IMPURITIES



A. 8-hydroxy-1,3-dioxolo[4,5-*g*]quinoline-7-carboxylic acid,



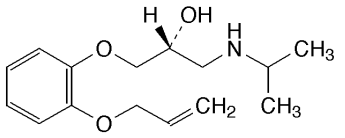
B. R1 = R2 = C_2H_5: ethyl 5-ethyl-8-oxo-5,8-dihydro-1,3-dioxolo[4,5-*g*]quinoline-7-carboxylate,

C. R1 = CH_3, R2 = H: 5-methyl-8-oxo-5,8-dihydro-1,3-dioxolo[4,5-*g*]quinoline-7-carboxylic acid.

01/2008:0628
corrected 6.0

OXPRENOLOL HYDROCHLORIDE

Oxprenololi hydrochloridum



$C_{15}H_{24}ClNO_3$ M_r 301.8
[6452-73-9]

DEFINITION

(2*RS*)-1-[(1-methylethyl)amino]-3-[2-(prop-2-enyloxy)phenoxy]propan-2-ol hydrochloride.

Content: 98.5 per cent to 101.5 per cent (dried substance).

CHARACTERS

Appearance: white or almost white, crystalline powder.

Solubility: very soluble in water, freely soluble in alcohol.

IDENTIFICATION

First identification: B, D.

Second identification: A, C, D.

A. Melting point (*2.2.14*): 107 °C to 110 °C.

B. Infrared absorption spectrophotometry (*2.2.24*).

Comparison: *oxprenolol hydrochloride CRS*.

If the spectra obtained in the solid state show differences, dissolve the substance to be examined and the reference substance separately in *ethyl acetate R*, evaporate to dryness and record new spectra using the residues.