Reference solution (a). Dissolve 50 mg of *levamisole hydrochloride for system suitability CRS* in *methanol R*, add 0.5 ml of *concentrated ammonia R* and dilute to 5.0 ml with *methanol R*.

Reference solution (b). Dilute 1.0 ml of the test solution to 100.0 ml with *methanol R*. Dilute 5.0 ml of the solution to 25.0 ml with *methanol R*.

Column:

– *size*: *l* = 0.10 m, Ø = 4.6 mm,

– *stationary phase*: *base-deactivated octadecylsilyl silica gel for chromatography R* (3 µm).

Mobile phase:

– *mobile phase A*: dissolve 0.5 g of *ammonium dihydrogen phosphate R* in 90 ml of *water R*; adjust to pH 6.5 with a 40 g/l solution of *sodium hydroxide R* and dilute to 100 ml with *water R*,

– *mobile phase B*: *acetonitrile R*,

Time (min)	Mobile phase A (per cent *V/V*)	Mobile phase B (per cent *V/V*)
0 - 8	90 → 30	10 → 70
8 - 10	30	70
10 - 11	30 → 90	70 → 10

Flow rate: 1.5 ml/min.

Detection: spectrophotometer at 215 nm.

Equilibration: at least 4 min with the mobile phase at the initial composition.

Injection: 10 µl.

Relative retention with reference to levamisole (retention time = about 3 min): impurity A = about 0.9; impurity B = about 1.4; impurity C = about 1.5; impurity D = about 1.6; impurity E = about 2.0.

System suitability: reference solution (a):

– the chromatogram obtained is similar to the chromatogram supplied with *levamisole hydrochloride for system suitability CRS*.

Limits:

– *correction factors*: for the calculation of contents, multiply the peak areas of the following impurities by the corresponding correction factor: impurity A = 2.0; impurity B = 1.7; impurity C = 2.9; impurity D = 1.3; impurity E = 2.7;

– *impurities A, B, C, D, E*: for each impurity, not more than the area of the principal peak in the chromatogram obtained with reference solution (b) (0.2 per cent);

– *any other impurity*: not more than half the area of the principal peak in the chromatogram obtained with reference solution (b) (0.1 per cent);

– *total*: not more than 1.5 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.3 per cent);

– *disregard limit*: 0.25 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.05 per cent).

Heavy metals (*2.4.8*): maximum 20 ppm.

12 ml of solution S complies with limit test A. Prepare the standard using *lead standard solution (1 ppm Pb) R*.

Loss on drying (*2.2.32*): maximum 0.5 per cent, determined on 1.000 g by drying in an oven at 105 °C for 4 h.

Sulphated ash (*2.4.14*): maximum 0.1 per cent, determined on 1.0 g.

ASSAY

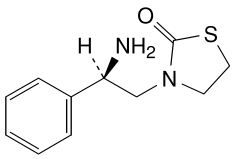
Dissolve 0.200 g in 30 ml of *alcohol R* and add 5.0 ml of *0.01 M hydrochloric acid*. Carry out a potentiometric titration (*2.2.20*), using *0.1 M sodium hydroxide*. Read the volume added between the 2 points of inflexion.

1 ml of *0.1 M sodium hydroxide* is equivalent to 24.08 mg of $C_{11}H_{13}ClN_2S$.

STORAGE

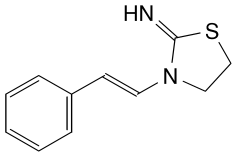
Protected from light.

IMPURITIES

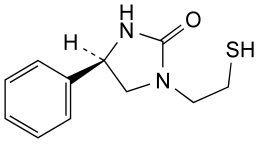


and enantiomer

A. 3-[(2*RS*)-2-amino-2-phenylethyl]thiazolidin-2-one,

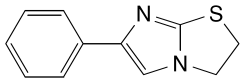


B. 3-[(*E*)-2-phenylethenyl]thiazolidin-2-imine,

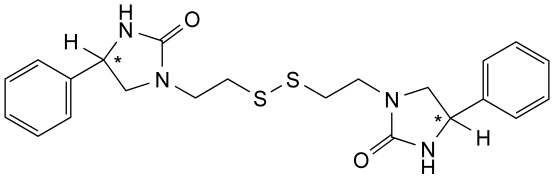


and enantiomer

C. (4*RS*)-4-phenyl-1-(2-sulphanylethyl)imidazolidin-2-one,



D. 6-phenyl-2,3-dihydroimidazo[2,1-*b*]thiazole,

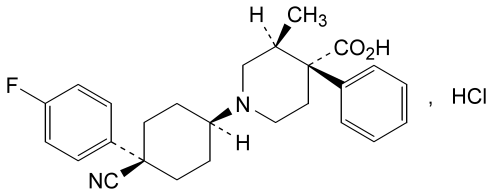


E. 1,1′-[(disulphane-1,2-diyl)bis(ethylene)]bis[(4*RS*)-4-phenylimidazolidin-2-one].

01/2008:1484
corrected 6.0

LEVOCABASTINE HYDROCHLORIDE

Levocabastini hydrochloridum



$C_{26}H_{30}ClFN_2O_2$ M_r 457.0
[79547-78-7]

DEFINITION

(3*S*,4*R*)-1-[*cis*-4-Cyano-4-(4-fluorophenyl)cyclohexyl]-3-methyl-4-phenylpiperidine-4-carboxylic acid monohydrochloride.

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

CHARACTERS

Appearance: white or almost white powder.

Solubility: practically insoluble in water, sparingly soluble in methanol, slightly soluble in ethanol (96 per cent) and in a 2 g/l solution of sodium hydroxide.

IDENTIFICATION

A. Infrared absorption spectrophotometry (*2.2.24*).

Preparation: discs.

Comparison: *levocabastine hydrochloride CRS*.

B. Dissolve 50 mg in a mixture of 0.4 ml of *ammonia R* and 2 ml of *water R*. Mix, allow to stand for 5 min and filter. Acidify the filtrate with *dilute nitric acid R*. It gives reaction (a) of chlorides (*2.3.1*).

C. Specific optical rotation (see Tests).

TESTS

Solution S. Dissolve 0.250 g in *methanol R* and dilute to 25.0 ml with the same solvent.

Appearance of solution. Solution S is clear (*2.2.1*) and not more intensely coloured than reference solution Y_7 (*2.2.2, Method II*).

Specific optical rotation (*2.2.7*): – 102 to – 106 (dried substance), determined on solution S.

Related substances. Capillary electrophoresis (*2.2.47*). *Prepare the solutions immediately before use.*

Test solution. Dissolve 25.0 mg of the substance to be examined in a 2 g/l solution of *sodium hydroxide R* and dilute to 10.0 ml with the same solution.

Reference solution (a). Dissolve 2.5 mg of *levocabastine hydrochloride CRS* and 2.5 mg of *levocabastine impurity D CRS* in a 2 g/l solution of *sodium hydroxide R* and dilute to 200.0 ml with the same solution.

Reference solution (b). Dilute 5.0 ml of this test solution to 100.0 ml with a 2 g/l solution of *sodium hydroxide R*. Dilute 1.0 ml of this solution to 10.0 ml with a 2 g/l solution of *sodium hydroxide R*.

Blank solution. A 2 g/l solution of *sodium hydroxide R*.

Capillary:

– *material*: uncoated fused silica;

– *size*: effective length = 0.5 m, Ø = 75 µm.

Temperature: 50 °C.

Electrolyte solution: dissolve 1.08 g of *sodium dodecyl sulphate R* and 0.650 g of *hydroxypropyl-β-cyclodextrin R* in 5 ml of *2-propanol R* and dilute to 50.0 ml with buffer solution pH 9.0 prepared as follows: dissolve 1.39 g of *boric acid R* in *water R* and adjust to pH 9.0 with *1 M sodium hydroxide* (about 9 ml). Dilute to 100.0 ml with *water R*.

Detection: spectrophotometer at 214 nm.

Preconditioning of the capillary: rinse the capillary for 2 min with a 2 g/l solution of *sodium hydroxide R* and for at least 5 min with the electrolyte solution.

Injection: under pressure (3,45 kPa) for 5 s.

Migration:

Time (min)	Current (µA)
0 - 0.17	0 → 75
0.17 - 15	75 → 130
15 - 40	130
40 - 60	130 → 200

Migration times: levocabastine = about 28 min; impurity D = about 30 min.

System suitability: reference solution (a):

– *resolution*: minimum 4 between the peaks due to levocabastine and impurity D; if necessary adjust the current gradient.

Limits:

– *impurities A, B, C, D, E*: for each impurity, not more than the area of the principal peak in the electropherogram obtained with reference solution (b) (0.5 per cent);

– *total*: not more than twice the area of the principal peak in the electropherogram obtained with reference solution (b) (1.0 per cent);

– *disregard limit*: 0.1 times the area of the principal peak in the electropherogram obtained with reference solution (b) (0.05 per cent); disregard any peak due to the blank.

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

Sulphated ash (*2.4.14*): maximum 0.1 per cent, determined on 1.0 g in a platinum crucible.

ASSAY

Dissolve 0.175 g in 50 ml of *ethanol (96 per cent) R* and add 5.0 ml of *0.01 M hydrochloric acid*. Carry out a potentiometric titration (*2.2.20*), using *0.1 M sodium hydroxide*. Read the volume added between the 1st and 3rd point of inflexion.

1 ml of *0.1 M sodium hydroxide* is equivalent to 22.85 mg of $C_{26}H_{30}ClFN_2O_2$.

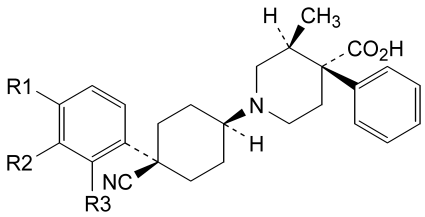
STORAGE

Protected from light.

IMPURITIES

Specified impurities: A, B, C, D, E.

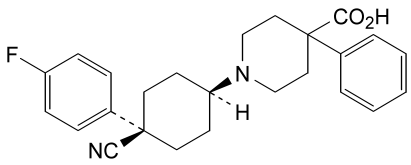
Other detectable impurities (the following substances would, if present at a sufficient level, be detected by one or other of the tests in the monograph. They are limited by the general acceptance criterion for other/unspecified impurities and/or by the general monograph *Substances for pharmaceutical 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*): *F, G, H, I.*



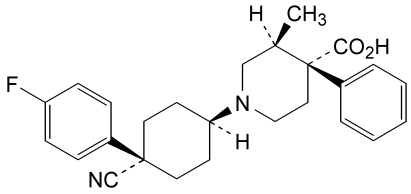
A. R1 = R2 = R3 = H: (3*S*,4*R*)-1-(*cis*-4-cyano-4-phenylcyclohexyl)-3-methyl-4-phenylpiperidine-4-carboxylic acid,

B. R1 = R2 = H, R3 = F: (3*S*,4*R*)-1-[*cis*-4-cyano-4-(2-fluorophenyl)cyclohexyl]-3-methyl-4-phenylpiperidine-4-carboxylic acid,

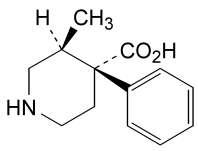
C. R1 = H, R2 = F, R3 = H: (3*S*,4*R*)-1-[*cis*-4-cyano-4-(3-fluorophenyl)cyclohexyl]-3-methyl-4-phenylpiperidine-4-carboxylic acid,



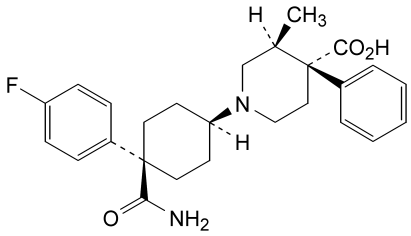
D. 1-[*cis*-4-cyano-4-(4-fluorophenyl)cyclohexyl]-4-phenylpiperidine-4-carboxylic acid,



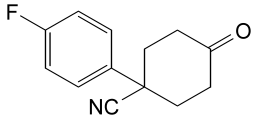
E. (3*S*,4*R*)-1-[*trans*-4-cyano-4-(4-fluorophenyl)cyclohexyl]-3-methyl-4-phenylpiperidine-4-carboxylic acid,



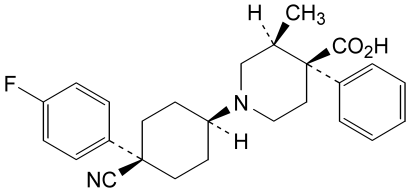
F. (3*S*,4*R*)-3-methyl-4-phenylpiperidine-4-carboxylic acid,



G. (3*S*,4*R*)-1-[*cis*-4-carbamoyl-4-(4-fluorophenyl)cyclohexyl]-3-methyl-4-phenylpiperidine-4-carboxylic acid,



H. 1-(4-fluorophenyl)-4-oxocyclohexanecarbonitrile,

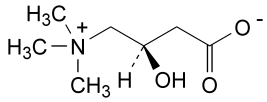


I. (3*S*,4*S*)-1-[*cis*-4-cyano-4-(4-fluorophenyl)cyclohexyl]-3-methyl-4-phenylpiperidine-4-carboxylic acid.

01/2008:1339
corrected 6.0

LEVOCARNITINE

Levocarnitinum



$C_7H_{15}NO_3$ M_r 161.2
[541-15-1]

DEFINITION

(3*R*)-3-Hydroxy-4-(trimethylammonio)butanoate.

Content: 98.0 per cent to 102.0 per cent (anhydrous substance).

CHARACTERS

Appearance: white or almost white, crystalline powder or colourless crystals, hygroscopic.

Solubility: freely soluble in water, soluble in warm ethanol (96 per cent), practically insoluble in acetone.

IDENTIFICATION

First identification: A, B.

Second identification: A, C.

A. Specific optical rotation (see Tests).

B. Infrared absorption spectrophotometry (*2.2.24*).

Preparation: discs, prepared using substance previously dried *in vacuo* at 50 °C for 5 h.

Comparison: *levocarnitine CRS.*

C. To 1 ml of solution S (see Tests) add 9 ml of *water R*, 10 ml of *dilute sulphuric acid R* and 30 ml of *ammonium reineckate solution R*. A pink precipitate is formed. Allow to stand for 30 min. Filter and wash with *water R*, with *ethanol (96 per cent) R* and then with *acetone R* and dry at 80 °C. The precipitate melts (*2.2.14*) at 147 °C to 150 °C.

TESTS

Solution S. Dissolve 5.00 g in *carbon dioxide-free water R* prepared from *distilled water R* and dilute to 50.0 ml with the same solvent.

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

pH (*2.2.3*): 6.5 to 8.5.

Dilute 10 ml of solution S to 20 ml with *carbon dioxide-free water R*.

Specific optical rotation (*2.2.7*): – 29.0 to – 32.0 (anhydrous substance), determined on solution S at 25 °C.