– *stationary phase*: *propylsilyl silica gel for chromatography R* (5 µm);

– *temperature*: 40 °C.

Mobile phase:

– *mobile phase A*: dissolve 4 g of *potassium dihydrogen phosphate R* in 1000 ml of *water for chromatography R*, and adjust to pH 4.0 with *phosphoric acid R*;

– *mobile phase B*: mobile phase A, *acetonitrile R1* (25:75 *V/V*);

Time (min)	Mobile phase A (per cent *V/V*)	Mobile phase B (per cent *V/V*)
0	90	10
0 - 15	90 → 30	10 → 70
15 - 15.1	30 → 90	70 → 10
15.1 - 20	90	10

Flow rate: 1.5 ml/min.

Detection: spectrophotometer at 210 nm.

Injection: 5 µl.

System suitability: reference solution (b):

– *resolution*: minimum 5 between the peaks due to clonidine and impurity B.

Limits:

– *unspecified impurities*: for each impurity, not more than the area of the principal peak in the chromatogram obtained with reference solution (a) (0.10 per cent);

– *total*: not more than twice the area of the principal peak in the chromatogram obtained with reference solution (a) (0.2 per cent);

– *disregard limit*: 0.5 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.05 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.

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

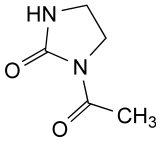
ASSAY

Dissolve 0.200 g in 70 ml of *ethanol (96 per cent) R*. Titrate with *0.1 M ethanolic sodium hydroxide* determining the end-point potentiometrically (*2.2.20*).

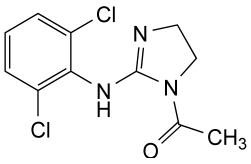
1 ml of *0.1 M sodium hydroxide* is equivalent to 26.66 mg of $C_9H_{10}Cl_3N_3$.

IMPURITIES

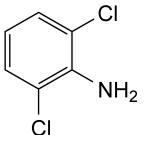
Other detectable impurities (the following substances would, if present at a sufficient level, be detected by one or other of the tests in the monograph. They are limited by the general acceptance criterion for other/unspecified impurities and/or by the general monograph *Substances for pharmaceutical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



A. 1-acetylimidazolidin-2-one,



B. 1-acetyl-2-[(2,6-dichlorophenyl)amino]-4,5-dihydro-1*H*-imidazole,

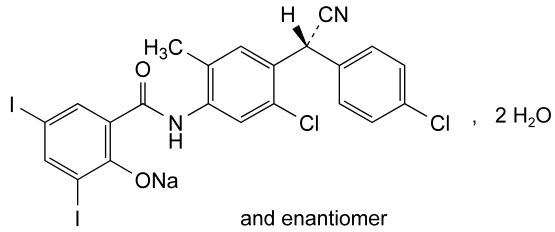


C. 2,6-dichloroaniline.

01/2008:1716

CLOSANTEL SODIUM DIHYDRATE FOR VETERINARY USE

Closantelum natricum dihydricum ad usum veterinarium



$C_{22}H_{13}Cl_2I_2N_2NaO_2,2H_2O$ M_r 721

[61438-64-0]

DEFINITION

N-[5-Chloro-4-[(*RS*)-(4-chlorophenyl)cyanomethyl]-2-methylphenyl]-2-hydroxy-3,5-diiodobenzamide sodium salt dihydrate.

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

CHARACTERS

Appearance: yellow powder, slightly hygroscopic.

Solubility: very slightly soluble in water, freely soluble in ethanol (96 per cent), soluble in methanol.

It shows polymorphism (*5.9*).

IDENTIFICATION

A. Infrared absorption spectrophotometry (*2.2.24*).

Preparation: discs without recrystallisation.

Comparison: *closantel sodium dihydrate CRS*.

B. Dissolve 0.1 g in 2 ml of *ethanol (96 per cent) R*. The solution gives reaction (a) of sodium (*2.3.1*).

TESTS

Appearance of solution. The solution is clear (*2.2.1*) and not more intensely coloured than reference solution GY_4 (*2.2.2, Method II*).

Dissolve 0.50 g in *ethanol (96 per cent) R* and dilute to 50 ml with the same solvent.

Related substances. Liquid chromatography (*2.2.29*). *Prepare the solutions immediately before use and protect from light.*

Test solution. Dissolve 0.100 g of the substance to be examined in *methanol R* and dilute to 10.0 ml with the same solvent.

Reference solution (a). Dissolve 10 mg of *closantel for system suitability CRS* (containing impurities A to J) in *methanol R* and dilute to 1.0 ml with the same solvent.

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

– *temperature*: 35 °C.

Mobile phase:

– *mobile phase A*: to 100 ml of a 7.7 g/l solution of *ammonium acetate R* previously adjusted to pH 4.3 with *acetic acid R*, add 50 ml of *acetonitrile R* and 850 ml of *water R*;

– *mobile phase B*: to 100 ml of a 7.7 g/l solution of *ammonium acetate R* previously adjusted to pH 4.3 with *acetic acid R*, add 50 ml of *water R* and 850 ml of *acetonitrile R*;

Time (min)	Mobile phase A (per cent *V/V*)	Mobile phase B (per cent *V/V*)
0 - 2	50	50
2 - 22	50 → 20	50 → 80
22 - 27	20	80
27 - 28	20 → 50	80 → 50
28 - 32	50	50

Flow rate: 1.5 ml/min.

Detection: spectrophotometer at 240 nm.

Injection: 10 µl.

Relative retention with reference to closantel (retention time = about 16 min): impurity A = about 0.07; impurity B = about 0.48; impurity C = about 0.62; impurity D = about 0.65; impurity E = about 0.82; impurity F = about 0.89; impurity G = about 0.93; impurity H = about 1.13; impurity I = about 1.16; impurity J = about 1.55.

System suitability: reference solution (a):

– *resolution*: baseline separation between the peaks due to impurity G and closantel,

– the chromatogram obtained is similar to the chromatogram supplied with *closantel 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 = 1.5; impurity B = 1.3;

– *impurity G*: not more than 2.5 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.5 per cent);

– *impurities F, H, I*: for each impurity, not more than 1.5 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.3 per cent);

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

– *total*: not more than 7.5 times the area of the principal peak in the chromatogram obtained with reference solution (b) (1.5 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).

Water (*2.5.12*): 4.8 per cent to 5.8 per cent, determined on 0.250 g.

Use a mixture of 1 volume of *dimethylformamide R* and 4 volumes of *methanol R* as the solvent.

ASSAY

Dissolve 0.500 g in 50 ml of a mixture of 1 volume of *anhydrous acetic acid R* and 7 volumes of *methyl ethyl ketone R*. Titrate with *0.1 M perchloric acid*, determining the end-point potentiometrically (*2.2.20*).

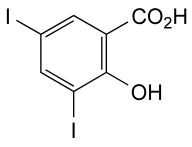
1 ml of *0.1 M perchloric acid* is equivalent to 68.5 mg of $C_{22}H_{13}Cl_2I_2N_2NaO_2$.

STORAGE

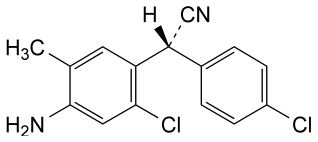
In an airtight container, protected from light.

IMPURITIES

Specified impurities: A, B, C, D, E, F, G, H, I, J.

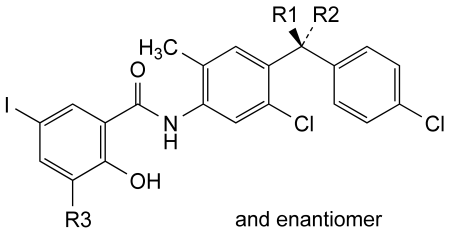


A. 2-hydroxy-3,5-diiodobenzoic acid,



and enantiomer

B. (2*RS*)-(4-amino-2-chloro-5-methylphenyl)(4-chlorophenyl)ethanenitrile,



C. R1 = H, R2 = CO_2H, R3 = I: (2*RS*)-[2-chloro-4-[(2-hydroxy-3,5-diiodobenzoyl)amino]-5-methylphenyl](4-chlorophenyl)acetic acid,

D. R1 = H, R2 = $CONH_2$, R3 = I: *N*-[4-[(1*RS*)-2-amino-1-(4-chlorophenyl)-2-oxoethyl]-5-chloro-2-methylphenyl]-2-hydroxy-3,5-diiodobenzamide,

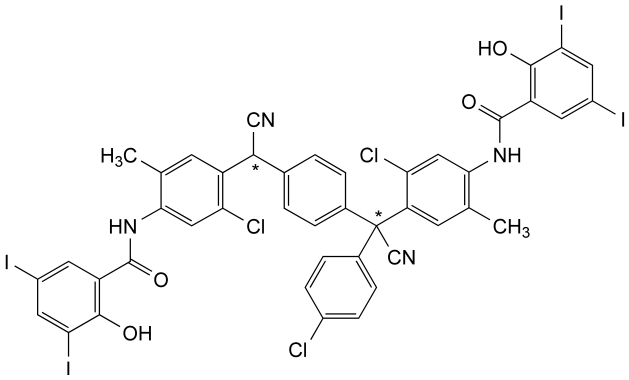
E. R1 = H, R2 = CN, R3 = Cl: 3-chloro-*N*-[5-chloro-4-[(*RS*)-(4-chlorophenyl)cyanomethyl]-2-methylphenyl]-2-hydroxy-5-iodobenzamide,

F. R1 + R2 = O, R3 = I: *N*-[5-chloro-4-(4-chlorobenzoyl)-2-methylphenyl]-2-hydroxy-3,5-diiodobenzamide,

G. R1 = H, R2 = C(=NH)OCH_3, R3 = I: methyl (2*RS*)-2-[2-chloro-4-[(2-hydroxy-3,5-diiodobenzoyl)amino]-5-methylphenyl]-2-(4-chlorophenyl)acetimidate,

H. R1 = H, R2 = CO-OCH_3, R3 = I: methyl (2*RS*)-[2-chloro-4-[(2-hydroxy-3,5-diiodobenzoyl)amino]-5-methylphenyl](4-chlorophenyl)acetate,

I. R1 = R3 = H, R2 = CN: *N*-[5-chloro-4-[(*RS*)-(4-chlorophenyl)cyanomethyl]-2-methylphenyl]-2-hydroxy-5-iodobenzamide,

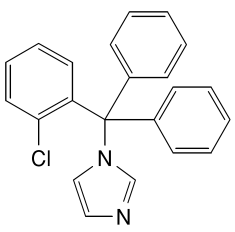


J. *N*-[5-chloro-4-[[4-[[2-chloro-4-[(2-hydroxy-3,5-diiodobenzoyl)amino]-5-methylphenyl]cyanomethyl]phenyl](4-chlorophenyl)cyanomethyl]-2-methylphenyl]-2-hydroxy-3,5-diiodobenzamide.

01/2008:0757
corrected 6.0

CLOTRIMAZOLE

Clotrimazolum



$C_{22}H_{17}ClN_2$ M_r 344.8
[23593-75-1]

DEFINITION

Clotrimazole contains not less than 98.5 per cent and not more than the equivalent of 100.5 per cent of 1-[(2-chlorophenyl)diphenylmethyl]-1*H*-imidazole, calculated with reference to the dried substance.

CHARACTERS

A white or pale yellow, crystalline powder, practically insoluble in water, soluble in alcohol and in methylene chloride.

IDENTIFICATION

First identification: B.

Second identification: A, C, D.

A. Melting point (*2.2.14*): 141 °C to 145 °C.

B. Examine by infrared absorption spectrophotometry (*2.2.24*), comparing with the spectrum obtained with *clotrimazole CRS*.

C. Examine before spraying in ultraviolet light at 254 nm, the chromatograms obtained in the test for (2-chlorophenyl)diphenylmethanol. The principal spot in the chromatogram obtained with test solution (b) is similar in position and size to the principal spot in the chromatogram obtained with reference solution (a).

D. Dissolve about 10 mg in 3 ml of *sulphuric acid R*. The solution is pale yellow. Add 10 mg of *mercuric oxide R* and 20 mg of *sodium nitrite R*. Allow to stand with occasional shaking. An orange colour develops, becoming orange-brown.

TESTS

Appearance of solution. Dissolve 1.25 g in *alcohol R* and dilute to 25 ml with the same solvent. The solution is clear (*2.2.1*) and not more intensely coloured than reference solution BY_6 (*2.2.2, Method II*).

(2-Chlorophenyl)diphenylmethanol. Examine by thin-layer chromatography (*2.2.27*), using *silica gel* GF_{254} *R* as the coating substance.

Test solution (a). Dissolve 0.50 g of the substance to be examined in *alcohol R* and dilute to 5 ml with the same solvent.

Test solution (b). Dilute 1 ml of test solution (a) to 10 ml with *alcohol R*.

Reference solution (a). Dissolve 50 mg of *clotrimazole CRS* in *alcohol R* and dilute to 5 ml with the same solvent.

Reference solution (b). Dissolve 10 mg of *(2-chlorophenyl)diphenylmethanol CRS* in *alcohol R* and dilute to 5 ml with the same solvent. Dilute 1 ml of the solution to 10 ml with *alcohol R*.

Apply separately to the plate 10 µl of each solution. Develop over a path of 15 cm using a mixture of 0.5 volumes of *concentrated ammonia R1*, 10 volumes of *propanol R* and 90 volumes of *toluene R*. Allow the plate to dry in air. Spray with a 10 per cent *V/V* solution of *sulphuric acid R* in *alcohol R* and heat at 100 °C to 105 °C for 30 min. Any spot corresponding to (2-chlorophenyl)diphenylmethanol in the chromatogram obtained with test solution (a) is not more intense than the spot in the chromatogram obtained with reference solution (b) (0.2 per cent).

Imidazole. Examine by thin-layer chromatography (*2.2.27*), using *silica gel G R* as the coating substance.

Test solution. Dissolve 0.50 g of the substance to be examined in *alcohol R* and dilute to 10 ml with the same solvent.