Reference solution (a). Dilute 5.0 ml of the stock solution to 100.0 ml with an 8 g/l solution of *ammonia R*.

Reference solution (b). Dilute 10.0 ml of the stock solution to 100.0 ml with an 8 g/l solution of *ammonia R*.

Reference solution (c). Dilute 15.0 ml of the stock solution to 100.0 ml with an 8 g/l solution of *ammonia R*.

The chromatographic procedure may be carried out using:

- a stainless steel column 0.10 m long and 4 mm in internal diameter packed with *octadecylsilyl silica gel for chromatography R* (5 µm),
- as mobile phase at a flow rate of 1.5 ml/min a mixture of 6 volumes of *glacial acetic acid R*, 30 volumes of *acetonitrile R* and 64 volumes of *water R*,
- as detector a spectrophotometer set at 254 nm,
- a 10 µl loop injector.

Inject reference solution (c). Adjust the sensitivity of the system so that the height of the peaks are at least 50 per cent of the full scale of the recorder. Inject each reference solution and determine the peak areas.

Establish a calibration curve with the concentration of the reference solutions (g/100 ml) as the abscissa and the corresponding areas as the ordinate.

Inject the test solution. Using the retention time and the peak area determined from the chromatograms obtained with the reference solutions, locate and integrate the peak due to glycyrrhizic acid in the chromatogram obtained with the test solution.

Calculate the percentage content of glycyrrhizic acid from the expression:

$$A \times \frac{5}{m} \times B \times \frac{822}{840}$$

A = concentration of monoammonium glycyrrhizate in the test solution determined from the calibration curve, in g/100 ml,

B = declared percentage content of *monoammonium glycyrrhizate CRS*,

m = mass of the drug, in grams,

822 = molecular weight of glycyrrhizic acid,

840 = molecular weight of the monoammonium glycyrrhizate (without any water of crystallisation).

STORAGE

Store protected from light.

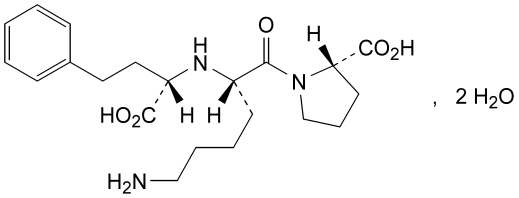
LABELLING

The label states whether the drug is peeled or unpeeled.

01/2005:1120
corrected

LISINOPRIL DIHYDRATE

Lisinoprilum dihydricum



$C_{21}H_{31}N_3O_5,2H_2O$ M_r 441.5

DEFINITION

Lisinopril dihydrate contains not less than 98.5 per cent and not more than the equivalent of 101.5 per cent of (2*S*)-1-[(2*S*)-6-amino-2-[[(1*S*)-1-carboxy-3-phenylpropyl]amino]hexanoyl]pyrrole-2-carboxylic acid, calculated with reference to the anhydrous substance.

CHARACTERS

A white or almost white, crystalline powder, soluble in water, sparingly soluble in methanol, practically insoluble in acetone and in ethanol.

IDENTIFICATION

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

TESTS

Specific optical rotation (*2.2.7*). Dissolve 0.5 g in *zinc acetate solution R* and dilute to 50.0 ml with the same solvent. The specific optical rotation is −43 to −47, calculated with reference to the anhydrous substance.

Related substances. Examine by liquid chromatography (*2.2.29*).

Test solution. Dissolve 20.0 mg of the substance to be examined in mobile phase A and dilute to 10.0 ml with the same mobile phase.

Reference solution (a). Dissolve the contents of 1 vial of *lisinopril dihydrate for performance test CRS* with 1.0 ml of mobile phase A.

Reference solution (b). Dilute 0.5 ml of the test solution to 50.0 ml with mobile phase A.

The chromatographic procedure may be carried out using:

- a stainless steel column 0.25 m long and 4.6 mm in internal diameter packed with *octylsilyl silica gel for chromatography R*,
- as mobile phase at a flow rate of 1.8 ml/min:

  Mobile phase A. Prepare a mixture of 30 volumes of *acetonitrile R* and 970 volumes of a 3.12 g/l *sodium dihydrogen phosphate R* solution adjusted to pH 5.0 with a 50 g/l solution of *sodium hydroxide R*,

  Mobile phase B. Prepare a mixture of 200 ml of *acetonitrile R* and 800 ml of a 3.12 g/l *sodium dihydrogen phosphate R* solution adjusted to pH 5.0 with a 50 g/l solution of *sodium hydroxide R*,

Time (min)	Mobile phase A (per cent *V/V*)	Mobile phase B (per cent *V/V*)	Comment
0 - 35	100 → 70	0 → 30	linear gradient
35 - 45	70	30	isocratic
45 - 50	70 → 100	30 → 0	switch to initial eluent composition
50 = 0	100	0	restart gradient

- as detector a spectrophotometer set at 210 nm,

maintaining the temperature of the column at 50 °C.

Equilibrate the column with mobile phase A for at least 30 min. Adjust the sensitivity of the system so that the height of the principal peak in the chromatogram obtained with 20 µl of reference solution (b) is at least 50 per cent of the full scale of the recorder.

Inject 20 µl of reference solution (a). The resulting chromatogram resembles that of the specimen chromatogram supplied with *lisinopril dihydrate for performance test CRS* in that the peaks due to impurity A and impurity E fall on either side of the peak due to lisinopril. Measure the heights *A1* and *A2* above the baseline of the

peaks due to impurity A and impurity E and the heights *B1* and *B2* above the baseline of the lowest points of the curve separating these peaks from the peak due to lisinopril. The test is not valid unless *A1* is greater than nine times *B1* and *A2* is greater than nine times *B2*.

If necessary, adjust the pH of the mobile phase to 4.5 with *phosphoric acid R* and repeat the chromatography. A further adjustment to pH 4.0 may be necessary with some columns before satisfactory separation of impurity A, lisinopril and impurity E is obtained. If, after adjustment, the retention time of the peak due to impurities C and D becomes extended to the point where integration becomes difficult, increase the content of mobile phase B from 30 per cent to 40 per cent over the interval from 35 min to 45 min from the start of the chromatogram. Maintain this concentration for a further 10 min. Return the concentration of mobile phase A to 100 per cent over the next 10 min prior to the next injection.

Inject 20 µl of the test solution and 20 µl of reference solution (b). In the chromatogram obtained with the test solution: the area of any peak due to impurity E is not greater than 0.3 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.3 per cent); the area of any peak, apart from the principal peak and any peak due to impurity E, is not greater than 0.3 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.3 per cent) and the sum of the areas of all such peaks is not greater than half the area of the principal peak in the chromatogram obtained with reference solution (b) (0.5 per cent). Disregard any peak due to the solvent, any peak occurring in the first 3 minutes and any peak with an area less than 0.05 times the area of the principal peak in the chromatogram obtained with reference solution (b).

Water (*2.5.12*): 8.0 to 9.5 per cent, determined on 0.200 g by the semi-micro determination of water.

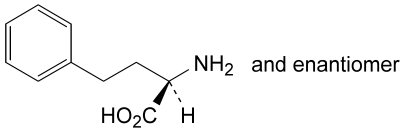
Sulphated ash (*2.4.14*). Not more than 0.1 per cent, determined on 1.0 g.

ASSAY

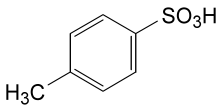
Dissolve 0.350 g in 50 ml of *distilled water R*. Titrate with *0.1 M sodium hydroxide*, determining the end-point potentiometrically (*2.2.20*).

1 ml of *0.1 M sodium hydroxide* is equivalent to 40.55 mg of $C_{21}H_{31}N_3O_5$.

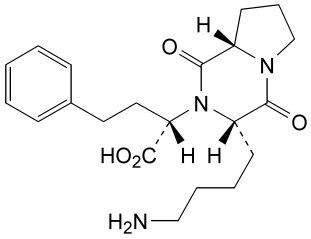
IMPURITIES



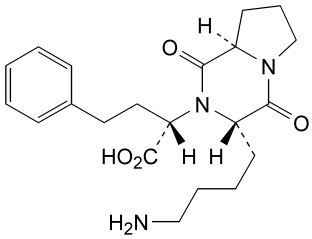
A. (2*RS*)-2-amino-4-phenylbutanoic acid,



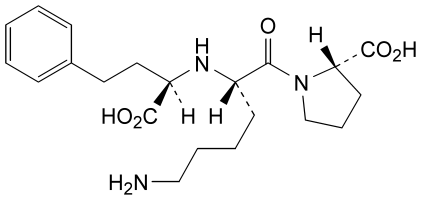
B. 4-methylbenzenesulphonic acid,



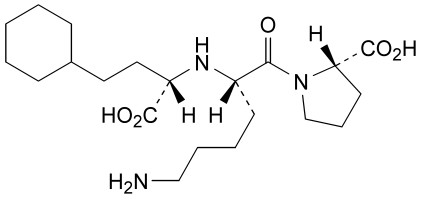
C. (2*S*)-2-[(3*S*,8a*S*)-3-(4-aminobutyl)-1,4-dioxohexahydropyrrolo[1,2-*a*]pyrazin-2(1*H*)-yl]-4-phenylbutanoic acid (*S*,*S*,*S*-diketopiperazine),



D. (2*S*)-2-[(3*S*,8a*R*)-3-(4-aminobutyl)-1,4-dioxohexahydropyrrolo[1,2-*a*]pyrazin-2(1*H*)-yl]-4-phenylbutanoic acid (*R*,*S*,*S*-diketopiperazine),



E. (2*S*)-1-[(2*S*)-6-amino-2-[[(1*R*)-1-carboxy-3-phenylpropyl]amino]hexanoyl]pyrrole-2-carboxylic acid (lisinopril *R*,*S*,*S*-isomer),



F. (2*S*)-1-[(2*S*)-6-amino-2-[[(1*S*)-1-carboxy-3-cyclohexylpropyl]amino]hexanoyl]pyrrole-2-carboxylic acid (cyclohexyl analogue).

01/2005:0228

LITHIUM CARBONATE

Lithii carbonas

Li_2CO_3 M_r 73.9

DEFINITION

Lithium carbonate contains not less than 98.5 per cent and not more than the equivalent of 100.5 per cent of Li_2CO_3.

CHARACTERS

A white powder, slightly soluble in water, practically insoluble in alcohol.

IDENTIFICATION

A. When moistened with *hydrochloric acid R*, it gives a red colour to a non-luminous flame.