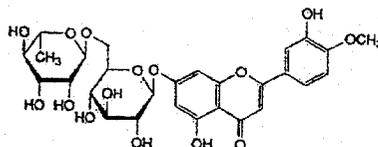


D

DIOSMIN
Diosminum

C₂₈H₃₂O₁₅

M, 609

DEFINITION

7-[[6-O-(6-Deoxy-α-L-mannopyranosyl)-β-D-glucopyranosyl]oxy]-5-hydroxy-2-(3-hydroxy-4-methoxyphenyl)-4H-1-benzopyran-4-one.

Substance obtained through iodine-assisted oxidation of (2S)-7-[[6-O-(6-deoxy-α-L-mannopyranosyl)-β-D-glucopyranosyl]oxy]-5-hydroxy-2-(3-hydroxy-4-methoxyphenyl)-2,3-dihydro-4H-1-benzopyran-4-one (hesperidin) of natural origin.

Content: 90.0 per cent to 102.0 per cent (anhydrous substance).

CHARACTERS

Appearance: greyish-yellow or light yellow hygroscopic powder.

Solubility: practically insoluble in water, soluble in dimethyl sulphoxide, practically insoluble in alcohol. It dissolves in dilute solutions of alkali hydroxides.

IDENTIFICATION

A. Infrared absorption spectrophotometry (2.2.24).

Comparison: *diosmin CRS*.

B. Examine the chromatograms obtained in the assay.

Results: the principal peak in the chromatogram obtained with test solution is similar in retention time and size to the principal peak in the chromatogram obtained with reference solution (a).

TESTS

Iodine: maximum 0.1 per cent.

Determine the total content of iodine by potentiometry, using an iodide-selective electrode (2.2.36), after oxygen combustion (2.5.10).

Test solution. Wrap 0.100 g of the substance to be examined in a piece of filter paper and place it in a sample carrier. Introduce into the flask 50 ml of a 0.2 g/l solution of *hydrazine R*. Flush the flask with oxygen for 10 min. Ignite the filter paper. Stir the contents of the flask immediately after the end of the combustion to dissolve completely the combustion products. Continue stirring for 1 h.

Reference solution. Dilute 2.0 ml of a 16.6 g/l solution of potassium iodide R to 100.0 ml with water R. Dilute 10.0 ml of the solution to 100.0 ml with water R.

Introduce into a beaker 30 ml of a 200 g/l solution of potassium nitrate R in 0.1 M nitric acid. Immerse the electrodes and stir for 10 min. The potential of the solution (nT_1) must remain stable. Add 1 ml of the test solution and measure the potential (nT_2).

Introduce into a beaker 30 ml of a 200 g/l solution of potassium nitrate R in 0.1 M nitric acid. Immerse the electrodes and stir for 10 min. The potential of the solution must remain stable (nR_1). Add 80 μ l of the reference solution and measure the potential (nR_2).

The absolute value $|nT_2 - nT_1|$ is not higher than the absolute value $|nR_2 - nR_1|$.

Related substances. Liquid chromatography (2.2.29).

Test solution. Dissolve 25.0 mg of the substance to be examined in dimethyl sulphoxide R and dilute to 25.0 ml with the same solvent.

Reference solution (a). Dissolve 25.0 mg of diosmin CRS in dimethyl sulphoxide R and dilute to 25.0 ml with the same solvent.

Reference solution (b). Dilute 5.0 ml of reference solution (a) to 100.0 ml with dimethyl sulphoxide R.

Reference solution (c). Dissolve 25.0 mg of diosmin for system suitability CRS in dimethyl sulphoxide R and dilute to 25.0 ml with the same solvent.

Column:

- size: $l = 0.10$ m, $\phi = 4.6$ mm.
- stationary phase: octadecylsilyl silica gel for chromatography R (3 μ m).
- temperature: 40 °C.

Mobile phase: acetonitrile R, glacial acetic acid R, methanol R, water R (2.6:28:66 V/V/V/V).

Flow rate: 1.5 ml/min.

Detection: spectrophotometer at 275 nm.

Injection: 10 μ l loop injector; inject the test solution and reference solutions (b) and (c).

Run time: 6 times the retention time of diosmin.

Relative retention with reference to diosmin (retention time = about 4.6 min): impurity A = about 0.5, impurity B = about 0.6, impurity C = about 0.8, impurity D = about 2.2, impurity E = about 2.6, impurity F = about 4.5.

System suitability: reference solution (c):

- resolution: minimum of 2.5 between the peaks due to impurities B and C.

Limits:

- correction factors: for the calculation of contents, multiply the peak areas of the following impurities by the corresponding correction factor: impurity A = 0.38; impurity F = 0.61.
- impurity A: not more than 0.2 times the area of the principal peak in the chromatogram obtained with reference solution (b) (1 per cent).
- impurity B: not more than the area of the principal peak in the chromatogram obtained with reference solution (b) (5 per cent).

- impurity C: not more than 0.6 times the area of the principal peak in the chromatogram obtained with reference solution (b) (3 per cent).
- impurity E: not more than 0.6 times the area of the principal peak in the chromatogram obtained with reference solution (b) (3 per cent).
- impurity F: not more than 0.6 times the area of the principal peak in the chromatogram obtained with reference solution (b) (3 per cent).
- any other impurity: not more than 0.2 times the area of the principal peak in the chromatogram obtained with reference solution (b) (1 per cent).
- total of other impurities and impurity A: not more than 0.2 times the area of the principal peak in the chromatogram obtained with reference solution (b) (1 per cent).
- total: not more than twice the area of the principal peak in the chromatogram obtained with reference solution (b) (10 per cent).
- disregard limit: 0.02 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.1 per cent).

Heavy metals (2.4.8): maximum 20 ppm.

2.0 g complies with limit test C. Prepare the standard using 4.0 ml of lead standard solution (10 ppm Pb) R.

Water (2.5.12): maximum 6.0 per cent, determined on 0.300 g.

Sulphated ash (2.4.14): maximum 0.2 per cent, determined on 1.0 g.

ASSAY

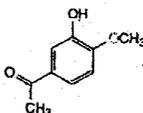
Liquid chromatography (2.2.29), as described in the test for related substances.

Injection: test solution and reference solution (a).

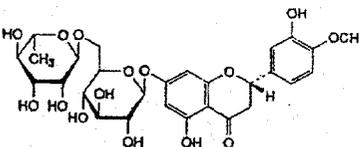
STORAGE

In an airtight container.

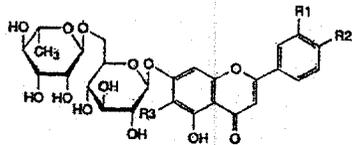
IMPURITIES



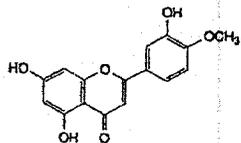
A. 1-(3-hydroxy-4-methoxyphenyl)ethanone (acetovanillone).



B. (2S)-7-[[6-O-(6-deoxy- α -L-mannopyranosyl)- β -D-glucopyranosyl]oxy]-5-hydroxy-2-(3-hydroxy-4-methoxyphenyl)-2,3-dihydro-4H-1-benzopyran-4-one (hesperidin).



- C. R1 = R3 = H, R2 = OH: 7-[[6-O-(6-deoxy-α-L-mannopyranosyl)-β-D-glucopyranosyl]oxy]-5-hydroxy-2-(4-hydroxyphenyl)-4*H*-1-benzopyran-4-one (isorhaifin),
- D. R1 = OH, R2 = OCH₃, R3 = I: 7-[[6-O-(6-deoxy-α-L-mannopyranosyl)-β-D-glucopyranosyl]oxy]-5-hydroxy-2-(3-hydroxy-4-methoxyphenyl)-6-iodo-4*H*-1-benzopyran-4-one (6-iododiosmin),
- E. R1 = R3 = H, R2 = OCH₃: 7-[[6-O-(6-deoxy-α-L-mannopyranosyl)-β-D-glucopyranosyl]oxy]-5-hydroxy-2-(4-methoxyphenyl)-4*H*-1-benzopyran-4-one (linarin),



- F. 5,7-dihydroxy-2-(3-hydroxy-4-methoxyphenyl)-4*H*-1-benzopyran-4-one (diosmetin).