– *any other impurity*: for each impurity, not more than 0.2 times the ratio of the area of the peak due to glutathione to the area of the peak due to the internal standard in the electropherogram obtained with reference solution (b) (0.2 per cent);
– *total*: not more than 2.5 times the ratio of the area of the peak due to glutathione to the area of the peak due to the internal standard in the electropherogram obtained with reference solution (b) (2.5 per cent);
– *disregard limit*: 0.05 times the ratio of the area of the peak due to glutathione to the area of the peak due to the internal standard in the electropherogram obtained with reference solution (b) (0.05 per cent).

Chlorides: maximum 200 ppm.
Dissolve 0.5 g in 5 mL of *dilute nitric acid R* and dilute to 10 mL with the same solvent. Add 10 mL of *strong hydrogen peroxide solution R* and heat on a water-bath for 30 min. Cool and dilute to 50 mL with *water R*. Add 1 mL of *silver nitrate solution R2* and mix. Allow to stand for 5 min protected from light. Any opalescence in the solution is not more intense than that in a standard prepared at the same time and in the same manner using 2 mL of *chloride standard solution (50 ppm Cl) R*. Examine the tubes laterally against a black background.

Sulfates (*2.4.13*): maximum 300 ppm.

Dilute 5 mL of solution S to 15 mL with *distilled water R*.

Ammonium (*2.4.1, Method B*): maximum 200 ppm, determined on 50 mg.

Prepare the standard using 0.1 mL of *ammonium standard solution (100 ppm NH_4) R*.

Iron (*2.4.9*): maximum 10 ppm.

In a separating funnel, dissolve 1.0 g in 10 mL of *dilute hydrochloric acid R*. Shake with 3 quantities, each of 10 mL, of *methyl isobutyl ketone R1*, shaking for 3 min each time. To the combined organic layers, add 10 mL of *water R* and shake for 3 min. The aqueous layer complies with the test.

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 3 h.

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

ASSAY

In a ground-glass-stoppered flask, dissolve 0.500 g of the substance to be examined and 2 g of *potassium iodide R* in 50 mL of *water R*. Cool the solution in iced water and add 10 mL of *hydrochloric acid R1* and 20.0 mL of *0.05 M iodine*. Stopper the flask and allow to stand in the dark for 15 min. Titrate with *0.1 M sodium thiosulfate* using 1 mL of *starch solution R*, added towards the end of the titration, as indicator. Carry out a blank titration.

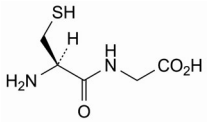
1 mL of *0.05 M iodine* is equivalent to 30.73 mg of $C_{10}H_{17}N_3O_6S$.

STORAGE

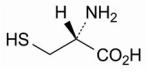
Protected from light.

IMPURITIES

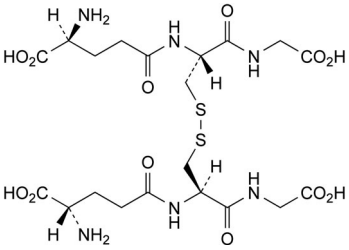
Specified impurities: A, B, C, D, E.



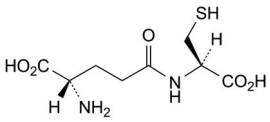
A. L-cysteinylglycine,



B. (2*R*)-2-amino-3-sulfanylpropanoic acid (cysteine),



C. bis(L-γ-glutamyl-L-cysteinylglycine) disulfide (L-glutathione oxidised),



D. L-γ-glutamyl-L-cysteine,

E. unknown structure (product of degradation).

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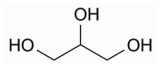
GLYCEROL

Glycerolum



$C_3H_8O_3$ M_r 92.1
[56-81-5]

DEFINITION

Propane-1,2,3-triol.

Content: 98.0 per cent *m/m* to 101.0 per cent *m/m* (anhydrous substance).

CHARACTERS

Appearance: clear, colourless or almost colourless, very hygroscopic, syrupy liquid, unctuous to the touch.

Solubility: miscible with water and with ethanol (96 per cent), slightly soluble in acetone, practically insoluble in fatty oils and in essential oils.

IDENTIFICATION

First identification: A, B.

Second identification: A, C.

A. Refractive index (see Tests).

B. Infrared absorption spectrophotometry (*2.2.24*).
Preparation: to 5 mL add 1 mL of *water R* and mix carefully.
Comparison: *Ph. Eur. reference spectrum of glycerol (85 per cent)*.

C. Relative density (*2.2.5*): 1.258 to 1.268.

TESTS

Solution S. Dilute 100.0 g to 200.0 mL with *carbon dioxide-free water R*.

Appearance of solution. Solution S is clear (*2.2.1*). Dilute 10 mL of solution S to 25 mL with *water R*. The solution is colourless (*2.2.2, Method II*).

Acidity or alkalinity. To 50 mL of solution S add 0.5 mL of *phenolphthalein solution R*. The solution is colourless. Not more than 0.2 mL of *0.1 M sodium hydroxide* is required to change the colour of the indicator to pink.

Refractive index (*2.2.6*): 1.470 to 1.475.

Aldehydes: maximum 10 ppm.

Place 7.5 mL of solution S in a ground-glass-stoppered flask and add 7.5 mL of *water R* and 1.0 mL of *decolorised pararosaniline solution R*. Close the flask and allow to stand for 1 h at a temperature of 25 ± 1 °C. The absorbance (*2.2.25*) of the solution measured at 552 nm is not greater than that of a standard prepared at the same time and in the same manner using 7.5 mL of *formaldehyde standard solution (5 ppm CH_2O) R* and 7.5 mL of *water R*. The test is not valid unless the standard is pink.

Esters. Add 10.0 mL of *0.1 M sodium hydroxide* to the final solution obtained in the test for acidity or alkalinity. Boil under a reflux condenser for 5 min. Cool. Add 0.5 mL of *phenolphthalein solution R* and titrate with *0.1 M hydrochloric acid*. Not less than 8.0 mL of *0.1 M hydrochloric acid* is required to change the colour of the indicator.

Impurity A and related substances. Gas chromatography (*2.2.28*).

Test solution. Dilute 10.0 mL of solution S to 100.0 mL with *water R*.

Reference solution (a). Dilute 10.0 g of *glycerol R1* to 20.0 mL with *water R*. Dilute 10.0 mL of the solution to 100.0 mL with *water R*.

Reference solution (b). Dissolve 1.000 g of *diethylene glycol R* in *water R* and dilute to 100.0 mL with the same solvent.

Reference solution (c). Dilute 1.0 mL of reference solution (b) to 10.0 mL with reference solution (a). Dilute 1.0 mL of this solution to 20.0 mL with reference solution (a).

Reference solution (d). Mix 1.0 mL of the test solution and 5.0 mL of reference solution (b) and dilute to 100.0 mL with *water R*. Dilute 1.0 mL of this solution to 10.0 mL with *water R*.

Reference solution (e). Dilute 5.0 mL of reference solution (b) to 100.0 mL with *water R*.

Column:

- *size*: *l* = 30 m, Ø = 0.53 mm;
- *stationary phase*: *cyanopropyl(3)phenyl(3)methyl(94)polysiloxane R*.

Carrier gas: *helium for chromatography R*.

Split ratio: 1:10.

Linear velocity: 38 cm/s.

Temperature:

	Time (min)	Temperature (°C)
Column	0	100
	0 - 16	100 → 220
	16 - 20	220
Injection port		220
Detector		250

Detection: flame ionisation.

Injection: 0.5 µL.

Elution order: impurity A, glycerol.

System suitability: reference solution (d):

- *resolution*: minimum 7.0 between the peaks due to impurity A and glycerol.

Limits:

- *impurity A*: not more than the area of the corresponding peak in the chromatogram obtained with reference solution (c) (0.1 per cent);
- *any other impurity with a retention time less than the retention time of glycerol*: not more than the area of the peak due to impurity A in the chromatogram obtained with reference solution (c) (0.1 per cent);
- *total of all impurities with retention times greater than the retention time of glycerol*: not more than 5 times the area of the peak due to impurity A in the chromatogram obtained with reference solution (c) (0.5 per cent);
- *disregard limit*: 0.05 times the area of the peak due to impurity A in the chromatogram obtained with reference solution (e) (0.05 per cent).

Halogenated compounds: maximum 35 ppm.

To 10 mL of solution S add 1 mL of *dilute sodium hydroxide solution R*, 5 mL of *water R* and 50 mg of *halogen-free nickel-aluminium alloy R*. Heat on a water-bath for 10 min, allow to cool and filter. Rinse the flask and the filter with *water R* until 25 mL of filtrate is obtained. To 5 mL of the filtrate add 4 mL of *ethanol (96 per cent) R*, 2.5 mL of *water R*, 0.5 mL of *nitric acid R* and 0.05 mL of *silver nitrate solution R2* and mix. Allow to stand for 2 min. Any opalescence in the solution is not more intense than that in a standard prepared at the same time by mixing 7.0 mL of *chloride standard solution (5 ppm Cl) R*, 4 mL of *ethanol (96 per cent) R*, 0.5 mL of *water R*, 0.5 mL of *nitric acid R* and 0.05 mL of *silver nitrate solution R2*.

Sugars. To 10 mL of solution S add 1 mL of *dilute sulfuric acid R* and heat on a water-bath for 5 min. Add 3 mL of an 85 mg/mL solution of *sodium hydroxide R* in *carbon dioxide-free water R*, mix and add dropwise 1 mL of freshly prepared *copper sulfate solution R*. The solution is clear and blue. Continue heating on the water-bath for 5 min. The solution remains blue and no precipitate is formed.

Chlorides (*2.4.4*): maximum 10 ppm.

Dilute 1 mL of solution S to 15 mL with *water R*. Prepare the standard using 1 mL of *chloride standard solution (5 ppm Cl) R* diluted to 15 mL with *water R*.

Water (*2.5.12*): maximum 2.0 per cent, determined on 1.000 g.

Sulfated ash (*2.4.14*): maximum 0.01 per cent, determined on 5.0 g after heating to boiling and ignition.

ASSAY

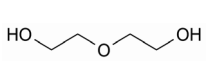
Thoroughly mix 0.075 g with 45 mL of *water R*. Add 25.0 mL of a mixture of 1 volume of *0.1 M sulfuric acid* and 20 volumes of *0.1 M sodium periodate*. Allow to stand protected from light for 15 min. Add 5.0 mL of a 500 g/L solution of *ethylene glycol R* and allow to stand protected from light for 20 min. Using 0.5 mL of *phenolphthalein solution R* as indicator, titrate with *0.1 M sodium hydroxide*. Carry out a blank titration.

1 mL of *0.1 M sodium hydroxide* is equivalent to 9.21 mg of $C_3H_8O_3$.

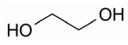
STORAGE

In an airtight container.

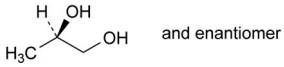
IMPURITIES



A. 2,2′-oxydi(ethan-1-ol) (diethylene glycol),



B. ethane-1,2-diol (ethylene glycol),



C. (2*RS*)-propane-1,2-diol (propylene glycol).