– *stationary phase*: 6 per cent polycyanopropylphenyl siloxane and 94 per cent of polydimethylsiloxane.

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 *alcohol 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 *alcohol 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 sulphuric acid R* and heat on a water-bath for 5 min. Add 3 ml of carbonate-free *dilute sodium hydroxide solution R* (prepared by the method described for carbonate-free *1 M sodium hydroxide* (*4.2.2*)), mix and add dropwise 1 ml of freshly prepared *copper sulphate 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.

1 ml of solution S diluted to 15 ml with *water R* complies with the limit test for chlorides. Prepare the standard using 1 ml of *chloride standard solution (5 ppm Cl) R* diluted to 15 ml with *water R*.

Heavy metals (*2.4.8*): maximum 5 ppm.

Dilute 8 ml of solution S to 20 ml with *water R*. 12 ml of the solution complies with limit test A. Prepare the standard using *lead standard solution (1 ppm Pb) R*.

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

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

ASSAY

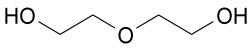
Thoroughly mix 0.075 g with 45 ml of *water R*. Add 25.0 ml of a mixture of 1 volume of *0.1 M sulphuric acid* and 20 volumes of *0.1 M sodium periodate*. Allow to stand protected from light for 15 min. Add 5.0 ml of a 500 g/l solution of *ethylene glycol R* and allow to stand protected from light for 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 $C_3H_8O_3$.

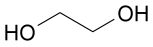
STORAGE

In an airtight container.

IMPURITIES



A. 2,2'-oxydiethanol (diethylene glycol),



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

C. propylene glycol.

01/2008:0497

GLYCEROL (85 PER CENT)

Glycerolum (85 per centum)

DEFINITION

Aqueous solution of propane-1,2,3-triol.

Content: 83.5 per cent *m/m* to 88.5 per cent *m/m* of propane-1,2,3-triol ($C_3H_8O_3$; M_r 92.1).

CHARACTERS

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

Solubility: miscible with water and with alcohol, slightly soluble in acetone, practically insoluble in fatty oils and in essential oils.

IDENTIFICATION

First identification: A, B.

Second identification: A, C, D.

A. It complies with the test for refractive index (see Tests).

B. Infrared absorption spectrophotometry (*2.2.24*).
Comparison: *Ph. Eur. reference spectrum of glycerol (85 per cent).*

C. Mix 1 ml with 0.5 ml of *nitric acid R*. Superimpose 0.5 ml of *potassium dichromate solution R*. A blue ring develops at the interface of the liquids. Within 10 min, the blue colour does not diffuse into the lower layer.

D. Heat 1 ml with 2 g of *potassium hydrogen sulphate R* in an evaporating dish. Vapours (acrolein) are evolved which blacken filter paper impregnated with *alkaline potassium tetraiodomercurate solution R*.

TESTS

Solution S. Dilute 117.6 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*): 1.449 to 1.455.

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 11.8 g of *glycerol (85 per cent) 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*: 6 per cent polycyanolpropylphenyl siloxane and 94 per cent of polydimethylsiloxane.

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 30 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 *alcohol 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 *alcohol 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 sulphuric acid R* and heat on a water-bath for 5 min. Add 3 ml of carbonate-free *dilute sodium hydroxide solution R* (prepared by the method described for carbonate-free *1 M sodium hydroxide* (*4.2.2*)), mix and add dropwise 1 ml of freshly prepared *copper sulphate 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.

1 ml of solution S diluted to 15 ml with *water R* complies with the limit test for chlorides. Prepare the standard using 1 ml of *chloride standard solution (5 ppm Cl) R* diluted to 15 ml with *water R*.

Heavy metals (*2.4.8*): maximum 5 ppm.

Dilute 8 ml of solution S to 20 ml with *water R*. 12 ml of the solution complies with limit test A. Prepare the standard using *lead standard solution (1 ppm Pb) R*.

Water (*2.5.12*): 12.0 per cent to 16.0 per cent, determined on 0.200 g.

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

ASSAY

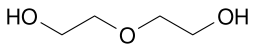
Thoroughly mix 0.075 g with 45 ml of *water R*. Add 25.0 ml of a mixture of 1 volume of *0.1 M sulphuric acid* and 20 volumes of *0.1 M sodium periodate*. Allow to stand protected from light for 15 min. Add 5.0 ml of a 500 g/l solution of *ethylene glycol R* and allow to stand protected from light for 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 $C_3H_8O_3$.

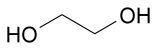
STORAGE

In an airtight container.

IMPURITIES



A. 2,2′-oxydiethanol (diethylene glycol),



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

C. propylene glycol.

01/2008:1427

GLYCEROL DIBEHENATE

Glyceroli dibehenas

DEFINITION

Mixture of diacylglycerols, mainly dibehenylglycerol, together with variable quantities of mono- and triacylglycerols, obtained by esterification of *glycerol (0496)* with behenic (docosanoic) acid.

Content:

– *monoacylglycerols*: 15.0 per cent to 23.0 per cent,
– *diacylglycerols*: 40.0 per cent to 60.0 per cent,
– *triacylglycerols*: 21.0 per cent to 35.0 per cent.

CHARACTERS

Appearance: hard, waxy mass or powder or white or almost white, unctuous flakes.

Solubility: practically insoluble in water, soluble in methylene chloride, partly soluble in hot ethanol (96 per cent).

IDENTIFICATION

A. Melting point (*2.2.14*): 65 °C to 77 °C.

B. Thin-layer chromatography (*2.2.27*).

Test solution. Dissolve 1.0 g of the substance to be examined in *toluene R* with gentle heating and dilute to 20 ml with the same solvent.

Reference solution. Dissolve 1.0 g of *glycerol dibehenate CRS* in *toluene R* with gentle heating and dilute to 20 ml with the same solvent.

Plate: *TLC silica gel plate R*.

Mobile phase: *hexane R, ether R* (30:70 *V/V*).

Application: 10 µl.

Development: over a path of 15 cm.

Drying: in air.

Detection: spray with a 0.1 g/l solution of *rhodamine B R* in *ethanol (96 per cent) R*; examine in ultraviolet light at 365 nm.

Results: the spots in the chromatogram obtained with the test solution are similar in position to the spots in the chromatogram obtained with the reference solution.

C. Composition of fatty acids (see Tests).

TESTS

Acid value (*2.5.1*): maximum 4.0, determined on 1.0 g, using a mixture of equal volumes of *ethanol (96 per cent) R* and *toluene R* as solvent and with gentle heating.

Iodine value (*2.5.4, Method A*): maximum 3.0.

Saponification value (*2.5.6*): 145 to 165.

Carry out the titration with heating.

Free glycerol: maximum 1.0 per cent, determined as described under Assay.

Composition of fatty acids (*2.4.22, Method C*). Raise the temperature of the column to 240 °C and use the mixture of calibrating substances in Table 2.4.22.-3.

Composition of the fatty acid fraction of the substance:

– *palmitic acid*: maximum 3.0 per cent;
– *stearic acid*: maximum 5.0 per cent;
– *arachidic acid*: maximum 10.0 per cent;
– *behenic acid*: minimum 83.0 per cent;
– *erucic acid*: maximum 3.0 per cent;
– *lignoceric acid*: maximum 3.0 per cent.

Nickel (*2.4.31*): maximum 1 ppm.

Water (*2.5.12*): maximum 1.0 per cent, determined on 1.00 g. Use *pyridine R* as the solvent.

Total ash (*2.4.16*): maximum 0.1 per cent, determined on 1.00 g.

ASSAY

Size-exclusion chromatography (*2.2.30*).

Stock solution. Place 0.100 g of *glycerol R* in a flask and dilute to 25.0 ml with *tetrahydrofuran R*.

Test solution. In a 15 ml flask, weigh 0.2 g (*m*) of the substance to be examined. Add 5.0 ml of *tetrahydrofuran R* and shake to dissolve. Heat gently, at about 35 °C. Reweigh the flask and calculate the total mass of solvent and substance (*M*).

Reference solutions. Into four 15 ml flasks, introduce respectively 0.25 ml, 0.5 ml, 1.0 ml and 2.5 ml of the stock solution and add 5.0 ml of *tetrahydrofuran R*. Weigh each flask and calculate the concentration of glycerol in milligrams per gram of each reference solution.

Column:

– *size*: *l* = 0.6 m, Ø = 7 mm;
– *stationary phase*: *styrene-divinylbenzene copolymer R* (5 µm) with a pore size of 10 nm.

Mobile phase: *tetrahydrofuran R*.

Flow rate: 1 ml/min.

Detection: differential refractive index.

Injection: 40 µl; when injecting the test solution, maintain the flask at about 35 °C to avoid precipitation.

Relative retention with reference to glycerol (retention time = about 15 min): triacylglycerols = about 0.73; diacylglycerols = about 0.76; monoacylglycerols = about 0.82.