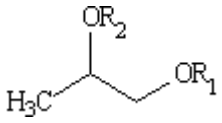
PROPYLENE GLYCOL ESTERS of FATTY ACIDS

Prepared at the 49th JECFA (1997) , published in FNP 52 Add 5 (1997) superseding specifications prepared at the 46th JECFA (1996), published in FNP 52 Add 4 (1996). Metals and arsenic specifications revised at the 55th JECFA (2000). An ADI of 0-25 mg/kg bw was established at the 17th JECFA (1973)

SYNONYMS Propane-1,2-diol esters of fatty acids, INS No. 477

DEFINITION Propylene glycol esters of fatty acids are mixtures of propylene glycol mono- and diesters of saturated and unsaturated fatty acids derived from edible oils and fats. The products are produced either by direct esterification of propylene glycol with fatty acids or by transesterification of propylene glycol with oils or fats. When prepared by transesterification, the product may contain residual mono- and diglycerides and glycerol. The process may be followed by molecular distillation to separate the monoesters.

Structural formula



where R_1 and R_2 represent one fatty acid moiety and hydrogen in the case of mono-esters and two fatty acid moieties in the case of di-esters

Assay Not less than 85% total fatty acid esters

DESCRIPTION White or cream coloured solids of waxy appearance, plastic products or viscous liquids

FUNCTIONAL USES Emulsifier

CHARACTERISTICS

IDENTIFICATION

Solubility (Vol. 4) Insoluble in water, soluble in ethanol and ethyl acetate

Positive test for fatty acids (Vol. 4) Passes test

Positive test for propylene glycol (Vol. 4) Passes test
Glycerol may also be detectable by TLC

PURITY

Sulfated ash (Vol. 4) Not more than 0.5% Test 5 g of the sample (Method I, if the sample is solid; Method II, if liquid)

Acid value (Vol. 4) Not more than 4

Acids (Vol. 4) Acids other than fatty acids shall not be detectable

Dimer and trimer of propylene glycol	Not more than 0.5%
Soap	Not more than 7% (as potassium stearate) See description under TESTS
Free propylene glycol	Not more than 1.5% (soap free) See description under TESTS
Total propylene glycol	Not less than 11% (soap free) See description under TESTS
Lead (Vol. 4)	Not more than 2 mg/kg Determine using an atomic absorption technique appropriate to the specified level. The selection of sample size and method of sample preparation may be based on the principles of the method described in Volume 4, “Instrumental Methods.”

TESTS

PURITY TESTS

Soap

Prepare a solvent mixture consisting of equal parts, by volume, of toluene and methanol, add bromophenol blue TS, and neutralize with 0.5N hydrochloric acid, or use neutralized acetone as the solvent. Weigh accurately about 5 g of the sample, dissolve it in 100 ml of the neutralized solvent mixture, and titrate with 0.5N hydrochloric acid to a definite yellow endpoint. Calculate the percentage of soap in the sample by the formula:

$$\frac{V \times N \times e}{W}$$

where V and N are the volume and normality, respectively, of the hydrochloric acid, W is the weight of the sample, in g, and e is the equivalence factor, e = 31.0.

Free propylene glycol

Reagents and Solutions:

Periodic Acid solution: Dissolve 5.4 g of periodic acid, H_5IO_6 in 100 ml of water, add 1900 ml of glacial acetic acid, and mix. Store in a light-resistant, glass-stoppered bottle or in a clear, glass-stoppered bottle protected from light.

Chloroform:

Use chloroform meeting the following test: To each of three 500-ml flasks add 50.0 ml of Periodic Acid Solution, then add 50 ml of chloroform and 10 ml of water to two of the flasks and 50 ml of water to the third. To each flask add 20 ml of potassium iodide TS, mix gently, and allow to stand at least 1 min, but no longer than 5 min, before titrating. Add 100 ml of water, and titrate with 0.1N sodium thiosulfate, using a magnetic stirrer to keep the solution thoroughly mixed, to the disappearance of the brown iodine colour, then add 2 ml of starch TS and continue the titration to the disappearance of the blue colour. The

difference between the volume of 0.1N sodium thiosulfate required in the titrations with and without the chloroform is not greater than 0.5 ml.

Procedure:
Melt the sample, if not liquid, at a temperature not higher than 100º above its melting point, and mix thoroughly. Transfer an accurately weighed portion of the sample, equivalent to about 30 mg of propylene glycol into a 100 ml beaker, and dissolve in 25 ml of chloroform. Transfer the solution with the aid of an additional 25 ml of chloroform, into a separator, wash the beaker with 25 ml of water, and add the washing to the separator. Stopper the separator tightly, shake vigorously for 30 to 60 sec, and allow the layers to separate. (Add 1 to 2 ml of glacial acetic acid to break emulsions formed due to the presence of soap.) Collect the aqueous layer in a 500-ml glass-stoppered Erlenmeyer flask, and extract the chloroform solution again using two 25-ml portions of water. To the combined aqueous extracts add 50.0 ml of Periodic Acid Solution. Run two blanks by adding 50.0 ml of this reagent solution to two 500 ml glass-stoppered Erlenmeyer flasks, each containing 75 ml of water and allow to stand for at least 30 min, but no longer than 90 min. To each flask, add 20 ml of potassium iodide TS, and allow to stand at least 1 min, but no longer than 5 min, before titrating. Add 100 ml of water, and titrate with 0.1N sodium thiosulfate, using a magnetic stirrer to keep the solution thoroughly mixed, to the disappearance of the brown iodine colour, then add 2 ml of starch TS and continue the titration to the disappearance of the blue colour.

Calculation:
Calculate the percentage of free propylene glycol by the formula:

$$\frac{(b - S) \times N \times 3.81}{W}$$

where "b" is the number of ml of sodium thiosulfate consumed in the blank determination; "S" is the number of ml required in the titration of the aqueous extracts from the sample; "N" is the exact normality of the sodium thiosulfate; "W" is the weight, in g, of the original sample taken and 3.81 is the molecular weight of propylene glycol divided by 20.

Note:
If the aqueous extract contains more than 30 mg of propylene glycol, dilute the extract in a volumetric flask and transfer a suitable aliquot into a 500 ml glass-stoppered Erlenmeyer flask before proceeding with the test. The weight of the sample should be corrected in the calculation.

Total propylene glycol and glycerol

Sample preparation
Transfer about 15 g of sample, accurately weighed, into a 500-ml flask, add 250 ml of ethanol and 7.5 g of potassium hydroxide and mix. Reflux the solution for 2 h, transfer into an 800-ml beaker rinsing the flask with about 100 ml of water and adding the rinse water to the beaker. Heat on a steam or water bath, adding water occasionally to replace the ethanol and evaporate until the odour of ethanol can no longer be detected.

Adjust the volume to about 250 ml with hot water, neutralize with diluted sulfuric acid (1 in 2), add a slight excess of acid, heat with gentle stirring until the fatty acid layer separates. Transfer the fatty acids into a warm 500-ml separatory funnel, wash with four 20-ml portions of hot water and combine the washings with the original aqueous layer from the saponification. Extract the combined aqueous layer with three 20 ml portions of petroleum ether. Neutralize the aqueous layer with sodium hydroxide TS to pH 7. Transfer the solution to a 500-ml volumetric flask and dilute to the mark with water.

Determination of apparent propylene glycol

Pipette 5.0 ml of the solution into a 125 ml Erlenmeyer flask, add 5.0 ml of 1M periodic acid, swirl and let stand 15 min. Add 10 ml of a saturated solution of sodium bicarbonate, followed by 15.0 ml of 0.1N sodium arsenite and 1 ml of potassium iodide solution (1 in 20) and mix. Add enough sodium bicarbonate so that at the end point some remains undissolved, and titrate with 0.1N iodine, using a 10-ml microburette and continuing the titration to a faint yellow colour. Perform a blank determination and make the appropriate corrections. Each ml of 0.1N iodine is equivalent to 3.805 mg of propylene glycol.

Calculate the apparent propylene glycol content (g /100 g of esters) from the formula:

$$\frac{38.05 \times \text{ml } 0.1\text{N iodine solution}}{\text{sample weight (g)}}$$

If the qualitative test for glycerol included under Identification Test (Positive test for propylene glycol) showed the product to contain glycerol, it becomes necessary to correct for the glycerol content of the polyol solution obtained after saponification and separation of liberated fatty acids.

Determination of glycerol content

Pipette 50 ml of the solution prepared in “A. Sample preparation” into a 600-ml beaker, add bromothymol blue TS and acidify with 0.2N sulfuric acid to a definite greenish-yellow colour. Neutralize with 0.05N sodium hydroxide to a definite blue end point free of green colour. Prepare a blank containing 50 ml of water and neutralize in the same manner. Pipette 50 ml of sodium periodate TS into each beaker, mix by swirling, cover with a watch glass and allow to stand for 30 min at room temperature (not above 35^{o}) in the dark or in subdued light. Add 10 ml of a mixture of equal volumes of ethylene glycol and water and allow to stand 20 min. Dilute each solution to about 300 ml and titrate with 0.1N sodium hydroxide to pH 8.1±0.1 using a calibrated pH meter. Each ml of 0.1N sodium hydroxide, after correction for the blank, is equivalent to 9.210 mg of glycerol.

Calculate the glycerol content (g/100 g of esters) from the formula:

$$\frac{9.210 \times \text{ml } 0.1\text{N NaOH}}{\text{sample weight (g)}}$$

The true propylene glycol content (in g/100 g of esters) is equal to the apparent propylene glycol content (in g/100 g of esters) minus 1.65 x the glycerol content (in g/100 g of esters).

METHOD OF ASSAY

Determine by gas chromatography using the following: Gas chromatograph, with split injection or on-column injection, oven temperature programming and flame ionisation detector. For split injection an injection port with programmable temperature is preferable. For on-column injection, the reaction mixture is diluted 1:50 with pyridine prior to injection.

Column:
Fused silica capillary column, surface fully deactivated by silylation agent, 12-25 m, 0.25-0.35 mm i.d., coating 95% methyl- 5% phenyl silicone (or other phase with similar polarity), film thickness 0.1-0.2 µm.

Injection:
Volume 1-5µl: split injection (Split ratio 1:10-1:50); direct injection (hold for 1 min)

Temperatures:
Injection port 320º (or 60º for on-column injection); column initial 50º (or 60º for on-column injection); programme rate 10º/ min; final temperature 350º, hold 1 min; detector 400º; carrier gas flow 2-5 ml He/ min (at 80º)
N.B. The precise temperature conditions will be dependent on the details of the equipment used.

Reagents:
N,N - bis(trimethylsilyl)fluoroacetamide (BSTFA)
Trimethylchlorosilane (TMCS)
Pyridine, analytical grade, kept over KOH
n-Heptadecane, analytical grade, 99% minimum

Reference materials:
Propylene glycol, propylene glycol monostearate.
Internal standard solution: Accurately weigh approximately 100 mg internal standard, n-heptadecane into a 100-ml volumetric flask, dilute with pyridine to the mark.

Reference solution:
Accurately weigh approximately 100 mg propylene glycol monostearate into a 25-ml volumetric flask adding internal standard solution to the mark. When pure reference material of other components such as propylene glycol and di-fatty acid esters of propylene glycol are available, the method is suitable for these also.

Procedure:
Accurately weigh approximately 100 mg of the homogenised sample into a 25-ml volumetric flask and dilute to volume with the internal standard solution. Transfer 0.8 ml of the sample solution to a 2.5-ml screw cap vial with Teflon faced septa or 2.0-ml vial for auto sampler. Add 0.3 ml BSTFA and 0.1 ml TMCS. Close the vial and shake vigorously. Heat the

reaction mixture in a heating device at 70º for approximately 20 min, inject 1 to 5 µl of the reaction mixture into the gas chromatograph showing a stable baseline. Do not delay GC analysis. Repeat the reaction with a further 100 mg sample. Make two injections per reaction sample. Transfer 0.8 ml of reference solution to a vial and add the silylating agents, 0.3 ml BSTFA and 0.1 ml TMCS . Heat the reaction mixture and inject into the gas chromatograph as described above.

Identification:
Analyse reference solution using the same operating conditions as for the sample solution. Identify peaks by comparison of retention time with known substances or apply coupled GC/MS.

Calculation and expression of results:
Calculate response factor R_x of the reference substance X vs. internal standard using the reference solution chromatogram. The value of the response factor is given by the formula:

$$R_x = (m_{is}/m_x) \times (A_x/A_{is})$$

where:
m_{is} = mass of internal standard in mg
m_x = mass of reference substance X in mg
A_x = peak area of reference substance X
A_{is} = peak area of internal standard

Calculate percentage of mass content m'_x of component X in the sample by the formula:

$$m'_x = 1/R_x \times (m'_{is}/m'_s) \times (A'_x / A'_{is})\%$$

where:
m'_{is} = mass of internal standard in sample in mg
m'_s = mass of sample in mg
A'_x = peak area of component X in the sample
A'_{is} = peak area of internal standard in sample

When calculating the total content of propylene glycol monoesters in the sample the response factor of propylene monostearate is used for all propylene glycol monoesters in the sample. The FID response of propylene glycol monostearate does not differ significantly from that of other fatty acid monoesters of propylene glycol.