Regulation (EC) No 648/2004 of the European Parliament and of the Council of 31 March 2004 on detergents (Text with EEA relevance)

ANNEX VIII

TEST METHODS AND ANALYTICAL METHODS

3. Determination of non-ionic surfactants in biodegradation test liquors

3.1. Principle

Surface active agents are concentrated and isolated by gas stripping. In the sample used, the quantity of non-ionic surfactant should be in the range 250-800 g.

The stripped surfactant is dissolved in ethyl acetate.

After phase separation and evaporation of the solvent, the non-ionic surfactant is precipitated in aqueous solution with modified Dragendorff reagent ($KBiI_4$ + $BaCl_2$ + glacial acetic acid).

The precipitate is filtered, washed with glacial acetic acid and dissolved in ammonium tartrate solution. The bismuth in the solution is titrated potentiometrically with pyrrolidinedithiocarbamate solution at pH 4-5 using a bright platinum indicator electrode and a calomel or silver/silver chloride reference electrode. The method is applicable to non-ionic surfactants containing 6-30 alkylene oxide groups.

The titration result is multiplied by the empirical factor of 54 for conversion to the reference substance nonylphenol condensed with 10 mols ethylene oxide (NP 10).

3.2. Reagents and Equipment

Reagents are to be made up in deionised water.

3.2.1. Pure ethyl acetate, freshly distilled.

3.2.2. Sodium bicarbonate, $NaHCO_3$ AR.

3.2.3. Dilute hydrochloric acid [20 ml concentrated acid (HCl) diluted to 1 000 ml with water]

3.2.4. Methanol AR, freshly distilled, stored in a glass bottle.

3.2.5. Bromocresol purple, 0,1 g in 100 ml methanol.

3.2.6. Precipitating agent: the precipitating agent is a mixture of two volumes of solution A and one volume of solution B. The mixture is stored in a brown bottle and can be used for up to one week after mixing.

3.2.6.1. Solution A

Dissolve 1,7 g bismuth nitrate, $BiONO_3.H_2O$ AR, in 20 ml glacial acetic acid, and make up to 100 ml with water. Then dissolve 65 g potassium iodide AR in 200 ml water. Mix these two solutions in a 1 000 ml measuring flask, add 200 ml glacial acetic acid (3.2.7) and make up to 1 000 ml with water.

3.2.6.2. Solution B

Dissolve 290 g barium chloride, $BaCl_2.2H_2O$ AR, in 1 000 ml of water.

3.2.7. Glacial acetic acid 99-100 % (lower concentrations are unsuitable).

3.2.8. Ammonium tartrate solution: mix 12,4 g tartaric acid AR and 12,4 ml of ammonia solution AR (d = 0,910 g/ml) and make up to 1 000 ml with water (or use the equivalent amount of ammonium tartrate AR).

3.2.9. Dilute ammonia solution: 40 ml ammonia solution AR (d = 0,910 g/ml) diluted to 1 000 ml with water.

3.2.10. Standard acetate buffer: dissolve 40 g solid sodium hydroxide AR, in 500 ml water in a beaker and allow to cool. Add 120 ml glacial acetic acid (3.2.7). Mix thoroughly, cool and transfer to a 1 000 ml volumetric flask. Make up to the mark with water.

3.2.11. Pyrrolidinedithiocarbamate solution (known as 'carbate solution'): dissolve 103 mg sodium pyrrolidinedithiocarbamate, $C_5H_8NNaS_2.2H_2O$, in about 500 ml water, add 10 ml of n-amyl alcohol AR and 0,5 g $NaHCO_3$ AR, and make up to 1 000 ml with water.

3.2.12. Copper sulphate solution (for standardisation of 3.2.11).
STOCK SOLUTION

Mix 1,249 g copper sulphate, $CuSO_4.5H_2O$ AR, with 50 ml 0,5 M sulphuric acid and make up to 1 000 ml with water.
STANDARDSOLUTION

Mix 50 ml stock solution with 10 ml 0,5 M H_2SO_4 and make up to 1 000 ml with water.

3.2.13. Sodium chloride AR.

3.2.14. Gas-stripping apparatus (see Figure 5).

The diameter of the sintered disc must be the same as the internal diameter of the cylinder.

3.2.15. Separating funnel, 250 ml.

3.2.16. Magnetic stirrer with magnet 25-30 mm.

3.2.17. Gooch crucible, diameter of the perforated base = 25 mm, Type G4.

3.2.18. Circular glass-fibre filter papers, 27 mm diameter with fibre diameter 0,3-1,5 m.

3.2.19. Two filter flasks with adapters and rubber collars, 500 and 250 ml respectively.

3.2.20. Recording potentiometer fitted with a bright platinum indicator electrode and a calomel or silver/silver chloride reference electrode with a 250 mV range, with automatic burette of 20-25 ml capacity, or alternative manual equipment.

3.3. Method

3.3.1. Concentration and separation of the surfactant

Filter the aqueous sample through a qualitative filter paper. Discard the first 100 ml of the filtrate.

Into the stripping apparatus, previously rinsed with ethyl acetate, place a measured quantity of the sample, such that it contains between 250-800 g non-ionic surfactant.

To improve the separation add 100 g sodium chloride and 5 g sodium bicarbonate.

If the volume of the sample exceeds 500 ml, add these salts to the stripping apparatus in solid form, and dissolve by passing nitrogen or air through.

If a smaller-sized sample is used, dissolve the salts in 400 ml water and then add to the stripping apparatus.

Add water to bring the level to the upper stopcock.

Cautiously add 100 ml ethyl acetate on top of the water.

Fill the wash-bottle in the gas-line (nitrogen or air) two-thirds full with ethyl acetate.

Pass a gas stream of 30-60 l/h through the apparatus; the use of a flowmeter is recommended. The rate of aeration must be increased gradually at the beginning. The gas rate must be so adjusted that the phases remain noticeably separate to minimise the mixing of the phases and the solution of the ethyl acetate in the water. Stop the gas flow after five minutes.

If there is a reduction of more than 20 % in the volume of the organic phase through solution in water, the sublation must be repeated paying special attention to the rate of gas flow.

Run off the organic phase into a separating funnel. Return any water in the separating funnel from the aqueous phase — it should only be a few ml — to the stripping apparatus. Filter the ethyl acetate phase through a dry qualitative filter paper into a 250 ml beaker.

Put a further 100 ml ethyl acetate into the stripping apparatus and again pass nitrogen or air through for five minutes. Draw off the organic phase into the separating funnel used for the first separation, reject the aqueous phase and run the organic phase through the same filter as the first ethyl acetate portion. Rinse both the separating funnel and the filter with about 20 ml ethyl acetate.

Evaporate the ethyl acetate extract to dryness using a water-bath (fume cupboard). Direct a gentle stream of air over the surface of the solution to accelerate the evaporation.

3.3.2. Precipitation and filtration

Dissolve the dry residue from 3.3.1 in 5 ml methanol, add 40 ml water and 0,5 ml dilute HCl (3.2.3) and stir the mixture with a magnetic stirrer.

To this solution add 30 ml of precipitating agent (3.2.6) from a measuring cylinder. The precipitate forms after repeated stirring. After stirring for ten minutes leave the mixture to stand for at least five minutes.

Filter the mixture through a Gooch crucible, the base of which is covered with a glass-fibre filter paper. First wash the filter under suction with about 2 ml glacial acetic acid. Then thoroughly wash the beaker, magnet, and crucible with glacial acetic acid, of which about 40-50 ml is necessary. It is not necessary to quantitatively transfer the precipitate adhering to the sides of the beaker, to the filter, because the solution of the precipitate for the titration is returned to the precipitating beaker, and the remaining precipitate will then be dissolved.

3.3.3. Dissolution of the precipitate

Dissolve the precipitate in the filter crucible by the addition of hot ammonium tartrate solution (about 80 ° C) (3.2.8) in three portions of 10 ml each. Allow each portion to stand in the crucible for some minutes before being sucked through the filter into the flask.

Put the contents of the filter flask into the beaker used for the precipitation. Rinse the sides of the beaker with a further 20 ml of tartrate solution to dissolve the rest of the precipitate.

Carefully wash the crucible, adapter and filter flask with 150-200 ml water, and return the rinsing water to the beaker used for the precipitation.

3.3.4. The titration

Stir the solution using a magnetic stirrer (3.2.16), add a few drops of bromocresol purple (3.2.5) and add the dilute ammonia solution (3.2.9) until the colour turns violet (the solution is initially weakly acid from the residue of acetic acid used for rinsing).

Then add 10 ml standard acetate buffer (3.2.10), immerse the electrodes in the solution, and titrate potentiometrically with standard 'carbate solution' (3.2.11), the burette tip being immersed in the solution.

The titration rate should not exceed 2 ml/min.

The endpoint is the intersection of the tangents to the two branches of the potential curve.

It will be observed occasionally that the inflection in the potential curve becomes flattened; this can be eliminated by carefully cleaning the platinum electrode (by polishing with emery paper).

3.3.5. Blank determinations

At the same time run a blank determination through the whole procedure with 5 ml methanol and 40 ml water, according to the instructions in 3.3.2. The blank titration should be below 1 ml, otherwise the purity of the reagents (3.2.3, 3.2.7, 3.2.8, 3.2.9, 3.2.10) is suspect, especially their content of heavy metals, and they must be replaced. The blank must be taken into account in the calculation of the results.

3.3.6. Control of the factor of the 'carbate solution'

Determine the factor for the carbate solution on the day of use. To do this, titrate 10 ml of the copper sulphate solution (3.2.12) with 'carbate solution' after the addition of 100 ml water and 10 ml standard acetate buffer (3.2.10). If the amount used is a ml, the factor f is:

$$f = \frac{10}{a}$$

and all the results of the titration are multiplied by this factor.

3.4. Calculation of results

Every non-ionic surfactant has its own factor, depending on its composition, particularly on the length of the alkene oxide chain. The concentration of non-ionic surfactant is expressed in relation to a standard substance — a nonyl phenol with ten ethylene oxide units (NP 10) — for which the conversion factor is 0,054.

Using this factor the amount of surfactant present in the sample is found expressed as mg of NP 10 equivalent, as follows:

(b — c) xfx 0,054 = mg non-ionic surfactant as NP 10

where:

b = volume of 'carbate solution' used by the sample (ml),
c = volume of 'carbate solution' used by the blank (ml),
f = factor of the 'carbate solution'.

3.5. Expression of results

Express the results in mg/l as NP 10 to the nearest 0,1.

Changes to legislation:
There are outstanding changes not yet made to Regulation (EC) No 648/2004 of the European Parliament and of the Council. Any changes that have already been made to the legislation appear in the content and are referenced with annotations.
View outstanding changes

Changes and effects yet to be applied to the whole legislation item and associated provisions

- Signature words omitted by S.I. 2019/672 reg. 22
- Annex 1 para. 1 words substituted by S.I. 2019/672 reg. 23(2)(a)
- Annex 1 para. 1 words substituted by S.I. 2019/672 reg. 23(2)(b)
- Annex 1 para. 1 words substituted by S.I. 2019/672 reg. 23(2)(c)
- Annex 1 para. 2 words substituted by S.I. 2019/672 reg. 23(3)(a)
- Annex 1 para. 2 words substituted by S.I. 2019/672 reg. 23(3)(b)
- Annex 2 s. A words substituted by S.I. 2019/672 reg. 24(2)
- Annex 2 s. B words substituted by S.I. 2019/672 reg. 24(2)
- Annex 2 s. D words substituted by S.I. 2019/672 reg. 24(2)
- Annex 2 s. C words substituted by S.I. 2019/672 reg. 24(3)
- Art. 2(9) word omitted by S.I. 2019/672 reg. 6(2)(a)
- Art. 2(9) words substituted by S.I. 2019/672 reg. 6(2)(b)
- Art. 2(9) words substituted by S.I. 2019/672, reg. 6(2) (as substituted) by S.I. 2020/1617 reg. 2(4)(a)
- Art. 2(9a) words substituted by S.I. 2019/672 reg. 6(3)
- Art. 2(9A) words substituted in earlier amending provision S.I. 2019/672, reg. 6(3) by S.I. 2020/1617 reg. 2(4)(b)
- Art. 2(10) words inserted by S.I. 2019/672 reg. 6(4)
- Art. 2(10) words inserted by S.I. 2019/672, reg. 6(4) (as substituted) by S.I. 2020/1617 reg. 2(4)(c)
- Art. 2(13)-(15) inserted by S.I. 2019/672 reg. 6(5)
- Art. 2(16) inserted in earlier amending provision S.I. 2019/672, reg. 6(5) by S.I. 2020/1617 reg. 2(4)(d)
- Annex 3 Pt. B words omitted by S.I. 2019/672 reg. 25(3)(c)
- Annex 3 Pt. A para. 2 words substituted by S.I. 2019/672 reg. 25(2)(a)
- Annex 3 Pt. A para. 3 words substituted by S.I. 2019/672 reg. 25(2)(b)
- Annex 3 Pt. A para. 4 words substituted by S.I. 2019/672 reg. 25(2)(c)
- Annex 3 Pt. A para. 5 words substituted by S.I. 2019/672 reg. 25(2)(d)
- Annex 3 Pt. B para. 1 words substituted by S.I. 2019/672 reg. 25(3)(a)
- Annex 3 Pt. B para. 2 words substituted by S.I. 2019/672 reg. 25(3)(b)
- Art. 3(1)(a) substituted by S.I. 2019/672 reg. 7(2)(b)
- Art. 3(1)(a) words substituted in earlier amending provision S.I. 2019/672, reg. 7(2)(b) by S.I. 2020/1617 reg. 2(5)
- Art. 3(1)(b) words substituted by S.I. 2019/672 reg. 7(2)(c)
- Art. 3(1)(c) words substituted by S.I. 2019/672 reg. 7(2)(d)
- Art. 3A inserted by S.I. 2019/672, reg. 7A (as inserted) by S.I. 2020/1617 reg. 2(6)
- Annex 4 point 3 words inserted by S.I. 2019/672 reg. 26(8)(b)
- Annex 4 words omitted by S.I. 2019/672 reg. 26(6)
- Annex 4 point 1 heading words omitted by S.I. 2019/672 reg. 26(7)
- Annex 4 point 3 words omitted by S.I. 2019/672 reg. 26(8)(a)
- Annex 4 words omitted by S.I. 2019/672 reg. 26(13)
- Annex 4 words substituted by S.I. 2019/672 reg. 26(2)
- Annex 4 words substituted by S.I. 2019/672 reg. 26(3)
- Annex 4 words substituted by S.I. 2019/672 reg. 26(4)
- Annex 4 words substituted by S.I. 2019/672 reg. 26(5)
- Annex 4 point 4.1.2 words substituted by S.I. 2019/672 reg. 26(9)(a)
- Annex 4 point 4.1.2 words substituted by S.I. 2019/672 reg. 26(9)(b)
- Annex 4 point 4.1.3 words substituted by S.I. 2019/672 reg. 26(10)
- Annex 4 point 4.2.2 words substituted by S.I. 2019/672 reg. 26(11)(a)

- Annex 4 point 4.2.2 words substituted by S.I. 2019/672 reg. 26(11)(b)
- Annex 4 point 4.2.2 words substituted by S.I. 2019/672 reg. 26(11)(c)
- Annex 4 point 4.2.2 words substituted by S.I. 2019/672 reg. 26(11)(d)
- Annex 4 point 4.2.3 words substituted by S.I. 2019/672 reg. 26(12)(a)
- Annex 4 point 4.2.3 words substituted by S.I. 2019/672 reg. 26(12)(b)
- Annex 5 words omitted by S.I. 2019/672 reg. 27
- Annex 7 Pt. B words inserted by S.I. 2019/672 reg. 28(3)(b)
- Annex 7 Pt. A words substituted by S.I. 2019/672 reg. 28(2)(a)(i)
- Annex 7 Pt. A words substituted by S.I. 2019/672 reg. 28(2)(a)(ii)
- Annex 7 Pt. A words substituted by S.I. 2019/672 reg. 28(2)(b)(i)
- Annex 7 Pt. A words substituted by S.I. 2019/672 reg. 28(2)(b)(ii)
- Annex 7 Pt. B words substituted by S.I. 2019/672 reg. 28(3)(a)
- Annex 7 Pt. C words substituted by S.I. 2019/672 reg. 28(4)
- Annex 7 Pt. D words substituted by S.I. 2019/672 reg. 28(5)
- Annex 8 words substituted by S.I. 2019/672 reg. 29
- Art. 10(3)(4) inserted by S.I. 2019/672 reg. 13(4)
- Art. 15(3)-(8) inserted by S.I. 2019/671 reg. 3(4)
- Art. 15(3) words omitted in earlier amending provision S.I. 2019/671, reg. 3(4) by S.I. 2020/1617 reg. 3(2)(b)(i)
- Art. 15(6)(7) omitted in earlier amending provision S.I. 2019/671, reg. 3(4) by S.I. 2020/1617 reg. 3(2)(b)(ii)
- Art. 18A inserted by S.I. 2019/672 reg. 21
- Art. 18A(1) words omitted in earlier amending provision S.I. 2019/672, reg. 21 by S.I. 2020/1617 reg. 2(9)(a)
- Art. 18A(3) substituted in earlier amending provision S.I. 2019/672, reg. 21 by S.I. 2020/1617 reg. 2(9)(b)