acid R and 5 mL of *water R*. Filter. To the filtrate add 1 mL of *strong sodium hydroxide solution R* and filter. To this filtrate add 3 mL of *ammonium chloride solution R*. A gelatinous white precipitate is formed.

B. Add 2.0 g in 20 portions to 100 mL of a 10 g/L solution of *sodium laurilsulfate R* in a 100 mL graduated cylinder about 30 mm in diameter. Allow 2 min between additions for each portion to settle. Allow to stand for 2 h. The apparent volume of the sediment is not less than 22 mL.

C. 0.25 g gives the reaction of silicates (*2.3.1*).

TESTS

Alkalinity. To 2 g add 100 mL of *carbon dioxide-free water R* and shake for 5 min. To 5 mL of this suspension add 0.1 mL of *thymolphthalein solution R*. The liquid becomes bluish. Add 0.1 mL of *0.1 M hydrochloric acid*. The liquid is decolourised within 5 min.

Coarse particles: maximum 0.5 per cent.

To 20 g add 1000 mL of *water R* and mix for 15 min using a high-speed mixer capable of operating at not less than 5000 r/min. Transfer the suspension to a wet sieve (75), tared after drying at 100-105 °C. Wash with 3 quantities, each of 500 mL, of *water R*, ensuring that any agglomerates have been dispersed. Dry the sieve at 100-105 °C and weigh. The particles on the sieve weigh a maximum of 0.1 g.

Heavy metals (*2.4.8*): maximum 50 ppm.

To 5.0 g add 7.5 mL of *dilute hydrochloric acid R* and 27.5 mL of *water R*. Boil for 5 min. Centrifuge and filter the supernatant liquid. Wash the centrifugation residue with *water R* and filter. Dilute the combined filtrates to 50.0 mL with *water R*. To 5 mL of this solution add 5 mL of *water R*, 10 mL of *hydrochloric acid R* and 25 mL of *methyl isobutyl ketone R* and shake for 2 min. Separate the layers. Evaporate the aqueous layer to dryness on a water-bath. Dissolve the residue in 1 mL of *acetic acid R*, dilute to 25 mL with *water R* and filter. 12 mL of the filtrate complies with test A. Prepare the reference solution using *lead standard solution (1 ppm Pb) R*.

Loss on drying (*2.2.32*): maximum 15 per cent, determined on 1.000 g by drying in an oven at 105 °C.

Microbial contamination

TAMC: acceptance criterion 10^3 CFU/g (*2.6.12*).

FUNCTIONALITY-RELATED CHARACTERISTICS

This section provides information on characteristics that are recognised as being relevant control parameters for one or more functions of the substance when used as an excipient (see chapter 5.15). This section is a non-mandatory part of the monograph and it is not necessary to verify the characteristics to demonstrate compliance. Control of these characteristics can however contribute to the quality of a medicinal product by improving the consistency of the manufacturing process and the performance of the medicinal product during use. Where control methods are cited, they are recognised as being suitable for the purpose, but other methods can also be used. Wherever results for a particular characteristic are reported, the control method must be indicated.

The following characteristics may be relevant for bentonite used as viscosity-increasing agent or suspending agent.

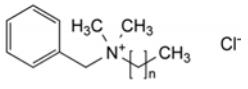
Sedimentation volume. To 6.0 g add 200 mL of *water R* and mix for 20 min using a high-speed mixer capable of operating at 10 000 r/min. Transfer 100 mL of this suspension to a graduated cylinder. Allow to stand for 24 h. The volume of the clear supernatant liquid is not greater than 2 mL.

Swelling power with water: see Identification B.

04/2009:0372
corrected 7.0

BENZALKONIUM CHLORIDE

Benzalkonii chloridum



[8001-54-5]

DEFINITION

Mixture of alkylbenzyldimethylammonium chlorides, the alkyl groups mainly having chain lengths of C_{12}, C_{14} and C_{16}.

Content: 95.0 per cent to 104.0 per cent of alkylbenzyldimethylammonium chlorides (anhydrous substance) calculated using the average relative molecular mass (see Tests).

CHARACTERS

Appearance: white or yellowish-white powder or gelatinous, yellowish-white fragments, hygroscopic. On heating it forms a clear molten mass.

Solubility: very soluble in water and in ethanol (96 per cent). An aqueous solution froths copiously when shaken.

IDENTIFICATION

First identification: B, E.

Second identification: A, C, D, E.

A. Ultraviolet and visible absorption spectrophotometry (*2.2.25*).

Test solution. Dissolve 80 mg in *water R* and dilute to 100.0 mL with the same solvent.

Spectral range: 220-350 nm.

Absorption maxima: at 257 nm, 263 nm and 269 nm.

Shoulder: at about 250 nm.

B. Examine the chromatograms obtained in the test for average relative molecular mass and ratio of alkyl components.

Results: the principal peaks in the chromatogram obtained with the test solution are similar in retention time to the principal peaks in the chromatogram obtained with the reference solution.

C. To 2 mL of solution S (see Tests) add 0.1 mL of *glacial acetic acid R* and, dropwise, 1 mL of *sodium tetraphenylborate solution R*. A white precipitate is formed. Filter. Dissolve the precipitate in a mixture of 1 mL of *acetone R* and 5 mL of *ethanol (96 per cent) R*, heating to not more than 70 °C. Add *water R* dropwise to the warm solution until a slight opalescence forms. Heat gently until the solution is clear and allow to cool. White crystals separate. Filter, wash with 3 quantities, each of 10 mL, of *water R* and dry *in vacuo* over *diphosphorus pentoxide R* or *anhydrous silica gel R* at a temperature not exceeding 50 °C. The crystals melt (*2.2.14*) at 127 °C to 133 °C.

D. To 5 mL of *dilute sodium hydroxide solution R* add 0.1 mL of *bromophenol blue solution R1* and 5 mL of *methylene chloride R* and shake. The methylene chloride layer is colourless. Add 0.1 mL of solution S and shake. The methylene chloride layer becomes blue.

E. To 2 mL of solution S add 1 mL of *dilute nitric acid R*. A white precipitate is formed which dissolves on the addition of 5 mL of *ethanol (96 per cent) R*. The solution gives reaction (a) of chlorides (*2.3.1*).

TESTS

Solution S. Dissolve 1.0 g in *carbon dioxide-free water R* and dilute to 100 mL with the same solvent.

Appearance of solution. Solution S is clear (*2.2.1*) and not more intensely coloured than reference solution Y_6 (*2.2.2, Method II*).

Acidity or alkalinity. To 50 mL of solution S add 0.1 mL of *bromocresol purple solution R*. Not more than 0.1 mL of *0.1 M hydrochloric acid* or *0.1 M sodium hydroxide* is required to change the colour of the indicator.

Average relative molecular mass and ratio of alkyl components. Liquid chromatography (*2.2.29*).

Test solution. Dissolve 0.400 g of the substance to be examined in *water R* and dilute to 100.0 mL with the same solvent.

Reference solution. Dissolve 40 mg of *benzalkonium chloride for system suitability CRS* in *water R* and dilute to 10.0 mL with the same solvent.

Column:

- *size*: *l* = 0.25 m, Ø = 4.6 mm;
- *stationary phase*: *end-capped nitrile silica gel for chromatography R* (5 µm).

Mobile phase: mix 45 volumes of *acetonitrile R* and 55 volumes of a 13.6 g/L solution of *sodium acetate R* previously adjusted to pH 5.0 with *glacial acetic acid R*.

Flow rate: 2.0 mL/min.

Detection: spectrophotometer at 254 nm.

Injection: 10 µL.

Identification of homologues: use the chromatogram supplied with *benzalkonium chloride for system suitability CRS* and the chromatogram obtained with the reference solution to identify the peaks due to C_{12}, C_{14} and C_{16}.

Relative retention with reference to C_{12} homologue (retention time = about 6 min): C_{14} homologue = about 1.1; C_{16} homologue = about 1.3.

System suitability: reference solution:

- *resolution*: minimum 1.5 between the peaks due to the C_{12} and C_{14} homologues.

Calculate the average relative molecular mass of the sample by summing the products for each homologue, using the following expression:

$$W\left(\frac{A}{B}\right)$$

A = area of the peak due to the given homologue in the chromatogram obtained with the test solution;

B = sum of the areas of the peaks due to all homologues in the chromatogram obtained with the test solution;

W = relative molecular mass for the given homologue: 340, 368 and 396 for the C_{12}, C_{14} and C_{16} homologues, respectively.

Calculate the percentage of each homologue, using the following expression:

$$100\left(\frac{C}{D}\right)$$

C = product of the relative molecular mass of the given homologue and the area of the corresponding peak in the chromatogram obtained with the test solution;

D = sum of the C values for all homologues quantified.

Limits:

- *C_{12} homologue*: minimum 40 per cent;
- *C_{14} homologue*: minimum 20 per cent;
- *sum of C_{12} and C_{14} homologues*: minimum 70 per cent.

Impurities A, B and C. Liquid chromatography (*2.2.29*). *Prepare the solutions immediately before use.*

Test solution. Dissolve 0.50 g of the substance to be examined in *methanol R1* and dilute to 10.0 mL with the same solvent.

Reference solution (a). Dissolve 25.0 mg of *benzyl alcohol CRS* (impurity A) in *methanol R1* and dilute to 100.0 mL with the same solvent.

Reference solution (b). Dissolve 75.0 mg of *benzaldehyde CRS* (impurity B) in *methanol R1* and dilute to 100.0 mL with the same solvent. Dilute 1.0 mL of this solution to 10.0 mL with *methanol R1*.

Reference solution (c). Dilute 1.0 mL of reference solution (a) to 10.0 mL with *methanol R1*.

Column:

- *size*: *l* = 0.15 m, Ø = 4.6 mm;
- *stationary phase*: *end-capped octadecylsilyl silica gel for chromatography R* (5 µm);
- *temperature*: 30 °C.

Mobile phase:

- *mobile phase A*: dissolve 1.09 g of *sodium hexanesulfonate R* and 6.9 g of *sodium dihydrogen phosphate monohydrate R* in *water R*; adjust to pH 3.5 with *concentrated phosphoric acid R* and dilute to 1000.0 mL with the same solvent;
- *mobile phase B*: *methanol R1*;

Time (min)	Mobile phase A (per cent *V/V*)	Mobile phase B (per cent *V/V*)
0 - 10	80	20
10 - 14	80 → 50	20 → 50
14 - 35	50	50
35 - 36	50 → 20	50 → 80
36 - 55	20	80

Flow rate: 1.0 mL/min.

Detection: spectrophotometer at 210 nm for impurities A and C, and at 257 nm for impurity B.

Injection: 20 µL.

Relative retention with reference to impurity A (retention time = about 10 min): impurity B = about 1.3; impurity C = 2.4.

System suitability: at 210 nm:

- *signal-to-noise ratio*: minimum 10 for the principal peak in the chromatogram obtained with reference solution (c);
- *symmetry factor*: minimum 0.6 for the peak due to impurity A in the chromatogram obtained with reference solution (a).

Limits:

- *correction factor*: for the calculation of content, multiply the peak area of impurity C by 1.3;
- *impurity A*: not more than the area of the corresponding peak in the chromatogram obtained with reference solution (a) (0.5 per cent);
- *impurity B*: not more than the area of the corresponding peak in the chromatogram obtained with reference solution (b) (0.15 per cent);
- *impurity C*: not more than 0.1 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.05 per cent).

Amines and amine salts. Dissolve 5.0 g with heating in 20 mL of a mixture of 3 volumes of *1 M hydrochloric acid* and 97 volumes of *methanol R* and add 100 mL of *2-propanol R*. Pass a stream of *nitrogen R* slowly through the solution. Titrate with up to 12.0 mL of *0.1 M tetrabutylammonium hydroxide* and record the potentiometric titration curve (*2.2.20*). If the curve shows 2 points of inflexion, the volume of titrant added between the 2 points is not greater than 5.0 mL. If the curve shows no point of inflexion, the substance to be examined does not comply with the test. If the curve shows 1 point of inflexion, repeat the test but add 3.0 mL of a 25.0 g/L solution of *dimethyldecylamine R* in *2-propanol R* before the titration. If the titration curve after addition of 12.0 mL of the titrant shows only 1 point of inflexion, the substance to be examined does not comply with the test.

Water (*2.5.12*): maximum 10 per cent, determined on 0.300 g.

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

ASSAY

Dissolve 2.00 g in *water R* and dilute to 100.0 mL with the same solvent. Transfer 25.0 mL of the solution to a separating funnel, add 25 mL of *methylene chloride R*, 10 mL of *0.1 M sodium hydroxide* and 10.0 mL of a freshly prepared 50 g/L solution of *potassium iodide R*. Shake well, allow to separate and discard the methylene chloride layer. Shake the aqueous layer with 3 quantities, each of 10 mL, of *methylene chloride R* and discard the methylene chloride layers. To the aqueous layer add 40 mL of *hydrochloric acid R*, allow to cool and titrate with *0.05 M potassium iodate* until the deep-brown colour is almost discharged. Add 5 mL of *methylene chloride R* and continue the titration, shaking vigorously, until the methylene chloride layer no longer changes colour. Carry out a blank titration on a mixture of 10.0 mL of the freshly prepared 50 g/L solution of *potassium iodide R*, 20 mL of *water R* and 40 mL of *hydrochloric acid R*.

1 mL of *0.05 M potassium iodate* is equivalent to $\frac{x}{10}$ mg of benzalkonium chloride where x is the average relative molecular mass of the sample.

STORAGE

In an airtight container.

IMPURITIES

Specified impurities: *A, B, C.*

A. R = CH_2OH: benzyl alcohol,

B. R = CHO: benzaldehyde,

C. R = CH_2Cl: (chloromethyl)benzene.

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corrected 7.0

BENZALKONIUM CHLORIDE SOLUTION

Benzalkonii chloridi solutio

DEFINITION

Aqueous solution of a mixture of alkylbenzyldimethylammonium chlorides, the alkyl groups mainly having chain lengths of C_{12}, C_{14} and C_{16}.

Content: 475 g/L to 525 g/L of alkylbenzyldimethylammonium chlorides, calculated using the average relative molecular mass (see Tests). The solution may contain ethanol (96 per cent).

CHARACTERS

Appearance: clear, colourless or slightly yellowish liquid.

Solubility: miscible with water and with ethanol (96 per cent).

It froths copiously when shaken.

IDENTIFICATION

First identification: *B, E.*

Second identification: *A, C, D, E.*

A. Ultraviolet and visible absorption spectrophotometry (*2.2.25*).

Test solution. Dilute 0.3 mL to 100.0 mL with *water R*.

Spectral range: 220-350 nm.

Absorption maxima: at 257 nm, 263 nm and 269 nm.

Shoulder: at about 250 nm.

B. Examine the chromatograms obtained in the test for average relative molecular mass and ratio of alkyl components.

Results: the principal peaks in the chromatogram obtained with the test solution are similar in retention time to the principal peaks in the chromatogram obtained with the reference solution.

C. To 0.05 mL add 2 mL of *water R*, 0.1 mL of *glacial acetic acid R* and, dropwise, 1 mL of *sodium tetraphenylborate solution R*. A white precipitate is formed. Filter. Dissolve the precipitate in a mixture of 1 mL of *acetone R* and 5 mL of *ethanol (96 per cent) R*, heating to not more than 70 °C. Add *water R* dropwise to the warm solution until a slight opalescence forms. Heat gently until the solution is clear and allow to cool. White crystals separate. Filter, wash with 3 quantities, each of 10 mL, of *water R* and dry *in vacuo* over *diphosphorus pentoxide R* or *anhydrous silica gel R* at a temperature not exceeding 50 °C. The crystals melt (*2.2.14*) at 127 °C to 133 °C.

D. To 5 mL of *dilute sodium hydroxide solution R* add 0.1 mL of *bromophenol blue solution R1* and 5 mL of *methylene chloride R* and shake. The methylene chloride layer is colourless. Add 0.05 mL of the solution to be examined and shake. The methylene chloride layer becomes blue.

E. To 0.05 mL add 1 mL of *dilute nitric acid R*. A white precipitate is formed which dissolves on the addition of 5 mL of *ethanol (96 per cent) R*. The solution gives reaction (a) of chlorides (*2.3.1*).

TESTS

Solution S. Dilute 2.0 g to 100 mL with *carbon dioxide-free water R*.

Appearance of solution. Solution S is clear (*2.2.1*) and not more intensely coloured than reference solution Y_6 (*2.2.2, Method II*).

Acidity or alkalinity. To 50 mL of solution S add 0.1 mL of *bromocresol purple solution R*. Not more than 0.1 mL of *0.1 M hydrochloric acid* or *0.1 M sodium hydroxide* is required to change the colour of the indicator.

Average relative molecular mass and ratio of alkyl components. Liquid chromatography (*2.2.29*).

Test solution. Determine the density (*2.2.5*) of the solution to be examined. Dilute a quantity of the solution to be examined equivalent to about 0.400 g of benzalkonium chloride to 100.0 mL with *water R*.

Reference solution. Dissolve 40 mg of *benzalkonium chloride for system suitability CRS* in *water R* and dilute to 10.0 mL with the same solvent.

Column:

– *size*: *l* = 0.25 m, Ø = 4.6 mm;

– *stationary phase*: *end-capped nitrile silica gel for chromatography R* (5 µm).

Mobile phase: mix 45 volumes of *acetonitrile R* and 55 volumes of a 13.6 g/L solution of *sodium acetate R* previously adjusted to pH 5.0 with *glacial acetic acid R*.

Flow rate: 2.0 mL/min.

Detection: spectrophotometer at 254 nm.

Injection: 10 µL.

Identification of homologues: use the chromatogram supplied with *benzalkonium chloride for system suitability CRS* and the chromatogram obtained with the reference solution to identify the peaks due to homologues C_{12}, C_{14} and C_{16}.

Relative retention with reference to C_{12} homologue (retention time = about 6 min): C_{14} homologue = about 1.1; C_{16} homologue = about 1.3.

System suitability: reference solution:

– *resolution*: minimum 1.5 between the peaks due to the C_{12} and C_{14} homologues.