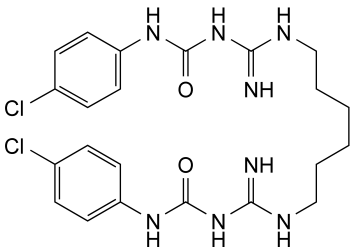
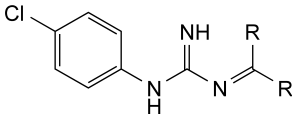


C. 1,1′-[hexane-1,6-diylbis[imino(iminocarbonyl)]]bis[3-(4-chlorophenyl)urea],

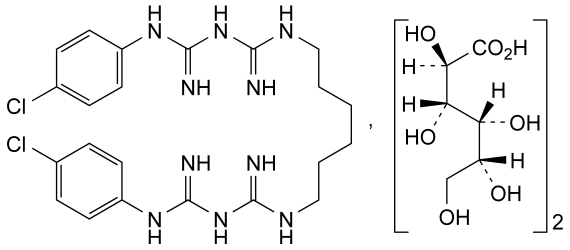


D. 1,1′-[[[[[(4-chlorophenyl)amino]iminomethyl]imino]methylene]bis[imino(hexane-1,6-diyl)]]bis[5-(4-chlorophenyl)biguanide].

01/2005:0658
corrected

CHLORHEXIDINE DIGLUCONATE SOLUTION

Chlorhexidini digluconatis solutio



$C_{34}H_{54}Cl_2N_{10}O_{14}$ M_r 898

DEFINITION

Chlorhexidine digluconate solution is an aqueous solution which contains not less than 190 g/l and not more than 210 g/l of 1,1′-(hexane-1,6-diyl)bis[5-(4-chlorophenyl)biguanide] di-D-gluconate.

CHARACTERS

An almost colourless or pale-yellowish liquid, miscible with water, with not more than 3 parts of acetone and with not more than 5 parts of ethanol (96 per cent).

IDENTIFICATION

First identification: A, B.

Second identification: B, C, D.

A. To 1 ml add 40 ml of *water R*, cool in iced water, make alkaline to *titan yellow paper R* by adding dropwise and with stirring *strong sodium hydroxide solution R* and add 1 ml in excess. Filter, wash the precipitate with *water R* until the washings are free from alkali and recrystallise from *alcohol (70 per cent V/V) R*. Dry at 100 °C to 105 °C. Examine the residue by infrared absorption spectrophotometry (*2.2.24*), comparing with the spectrum obtained with *chlorhexidine CRS*.

B. Examine by thin-layer chromatography (*2.2.27*), using *silica gel G R* as the coating substance.

Test solution. Dilute 10.0 ml of the solution to be examined to 50 ml with *water R*.

Reference solution. Dissolve 25 mg of *calcium gluconate CRS* in 1 ml of *water R*.

Apply separately to the plate 5 µl of each solution. Develop over a path of 10 cm using a mixture of 10 volumes of *ethyl acetate R*, 10 volumes of *concentrated ammonia R*, 30 volumes of *water R* and 50 volumes of *alcohol R*. Dry the plate at 100 °C for 20 min, allow to cool and spray with a 50 g/l solution of *potassium dichromate R* in a 40 per cent *m/m* solution of *sulphuric acid R*. After 5 min, the principal spot in the chromatogram obtained with the test solution is similar in position, colour and size to the principal spot in the chromatogram obtained with the reference solution.

C. To 1 ml add 40 ml of *water R*, cool in iced water, make alkaline to *titan yellow paper R* by adding dropwise and with stirring *strong sodium hydroxide solution R* and add 1 ml in excess. Filter, wash the precipitate with *water R* until the washings are free from alkali and recrystallise from *alcohol (70 per cent V/V) R*. Dry at 100 °C to 105 °C. The residue melts (*2.2.14*) at 132 °C to 136 °C.

D. To 0.05 ml add 5 ml of a 10 g/l solution of *cetrimide R*, 1 ml of *strong sodium hydroxide solution R* and 1 ml of *bromine water R*; a deep red colour is produced.

TESTS

Relative density (*2.2.5*): 1.06 to 1.07.

pH (*2.2.3*). Dilute 5.0 ml to 100 ml with *carbon dioxide-free water R*. The pH of the solution is 5.5 to 7.0.

Chloroaniline. Dilute 2.0 ml to 100 ml with *water R*. To 10 ml of the solution add 2.5 ml of *dilute hydrochloric acid R* and dilute to 20 ml with *water R*. Add rapidly and with thorough mixing after each addition: 0.35 ml of *sodium nitrite solution R*, 2 ml of a 50 g/l solution of *ammonium sulphamate R*, 5 ml of a 1 g/l solution of *naphthyle thylenediamine dihydrochloride R*, 1 ml of *alcohol R*, dilute to 50.0 ml with *water R* and allow to stand for 30 min. Any reddish-blue colour in the solution is not greater than that in a standard prepared at the same time in the same manner using a mixture of 10.0 ml of a 0.010 g/l solution of *chloroaniline R* in *dilute hydrochloric acid R* and 10 ml of *water R* instead of the dilution of the solution to be examined (0.25 per cent with reference to chlorhexidine digluconate at a nominal concentration of 200 g/l).

Related substances. Examined by liquid chromatography (*2.2.29*).

Test solution. Dilute 5.0 ml of the sample to be examined to 50.0 ml with the mobile phase. Dilute 5.0 ml of this solution to 50.0 ml with the mobile phase.

Reference solution (a). Dissolve 15 mg of *chlorhexidine for performance test CRS* in the mobile phase and dilute to 10.0 ml with the mobile phase.

Reference solution (b). Dilute 3.0 ml of the test solution to 100 ml with the mobile phase.

Reference solution (c). Dilute 1.0 ml of reference solution (b) to 50 ml with the mobile phase.

The chromatographic procedure may be carried out using:

– a stainless steel column 0.2 m long and 4 mm in internal diameter, packed with *octadecylsilyl silica gel for chromatography R* (5 µm),

– as mobile phase at a flow-rate of 1.0 ml/min, a solution of 2.0 g of *sodium octanesulphonate R* in a mixture of 120 ml of *glacial acetic acid R*, 270 ml of *water R* and 730 ml of *methanol R*,

– as detector a spectrophotometer set at 254 nm.

Equilibrate the column with mobile phase for at least 1 hour. Adjust the sensitivity of the system so that the height of the principal peak in the chromatogram obtained with 10 µl of reference solution (b) is at least 50 per cent of the full scale of the recorder.

Inject 10 µl of reference solution (a). The test is not valid unless the resulting chromatogram is similar to the specimen chromatogram provided with *chlorhexidine for performance test CRS* in that the peaks due to impurity A and impurity B precede that due to chlorhexidine. If necessary, adjust the concentration of acetic acid in the mobile phase (increasing the concentration decreases the retention times).

Inject separately 10 µl of the test solution and 10 µl each of reference solutions (b) and (c). Record the chromatograms of reference solutions (b) and (c) until the peak due to chlorhexidine has been eluted and record the chromatogram of the test solution for six times the retention time of the peak due to chlorhexidine. In the chromatogram obtained with the test solution, the sum of the areas of the peaks, apart from the principal peak is not greater than the area of the principal peak in the chromatogram obtained with reference solution (b) (3.0 per cent). Disregard any peak with a relative retention time of 0.25 or less with respect to the principal peak and any peak whose area is less than that of the principal peak in the chromatogram obtained with reference solution (c).

ASSAY

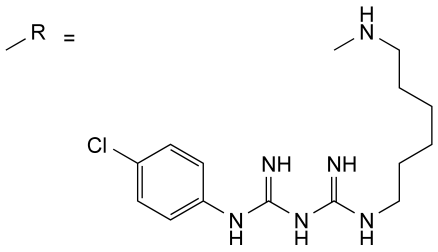
Determine the density (*2.2.5*) of the solution to be examined. Transfer 1.00 g to a 250 ml beaker and add 50 ml of *anhydrous acetic acid R*. Titrate with *0.1 M perchloric acid*. Determine the end-point potentiometrically (*2.2.20*).

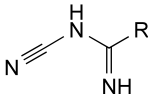
1 ml of *0.1 M perchloric acid* is equivalent to 22.44 mg of $C_{34}H_{54}Cl_2N_{10}O_{14}$.

STORAGE

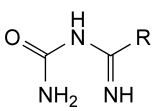
Store protected from light.

IMPURITIES

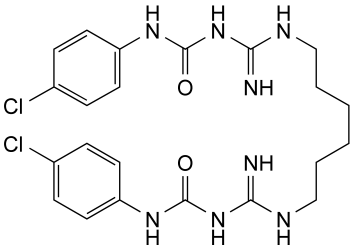




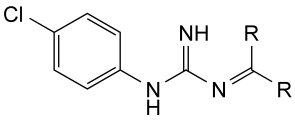
A. 1-(4-chlorophenyl)-5-[6-(3-cyanoguanidino)hexyl]biguanide,



B. [[[6-[5-(4-chlorophenyl)guanidino]hexyl]amino]iminomethyl]urea,



C. 1,1′-[hexane-1,6-diylbis[imino(iminocarbonyl)]]bis[3-(4-chlorophenyl)urea],

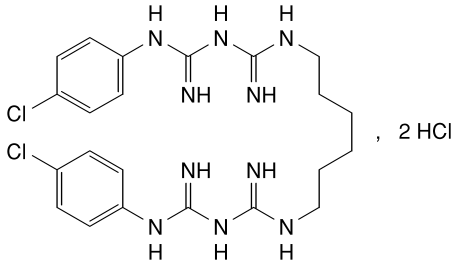


D. 1,1′-[[[[[(4-chlorophenyl)amino]iminomethyl]imino]methylene]bis[imino(hexane-1,6-diyl)]]bis[5-(4-chlorophenyl)biguanide].

01/2005:0659

CHLORHEXIDINE DIHYDROCHLORIDE

Chlorhexidini dihydrochloridum



$C_{22}H_{32}Cl_4N_{10}$ M_r 578.4

DEFINITION

Chlorhexidine dihydrochloride contains not less than 98.0 per cent and not more than the equivalent of 101.0 per cent of 1,1'-(hexane-1,6-diyl)bis[5-(4-chlorophenyl)biguanide] dihydrochloride, calculated with reference to the dried substance.

CHARACTERS

A white or almost white, crystalline powder, sparingly soluble in water and in propylene glycol, very slightly soluble in alcohol.

IDENTIFICATION

First identification: A, D.

Second identification: B, C, D.

A. Examine by infrared absorption spectrophotometry (*2.2.24*), comparing with the spectrum obtained with *chlorhexidine dihydrochloride CRS*.

B. Dissolve about 5 mg in 5 ml of a warm 10 g/l solution of *cetrimide R* and add 1 ml of *strong sodium hydroxide solution R* and 1 ml of *bromine water R*. A dark red colour is produced.

C. Dissolve 0.3 g in 10 ml of a mixture of equal volumes of *hydrochloric acid R* and *water R*. Add 40 ml of *water R*, filter if necessary and cool in iced water. Make alkaline to *titan yellow paper R* by adding dropwise, and with stirring, *strong sodium hydroxide solution R* and add 1 ml in excess. Filter, wash the precipitate with *water R*