Artesunate for injection (Artesunati ad injectionem)

2016-01

Category. Antimalarial.

Storage. Artesunate for injection should be kept in a hermetically closed container.

Additional information. Strength in the current WHO Model List of Essential Medicines (EML): 60 mg.

Strength in the current WHO EML for children: 60 mg.

The reconstituted injection is a sterile solution of artesunate in 5% sodium bicarbonate intravenous infusion. It is prepared by dissolving Artesunate for injection in the requisite amount of 5% sodium bicarbonate intravenous infusion immediately before use. This solution is diluted further with a suitable diluent for injection in accordance with the manufacturer's instructions.

The reconstituted injection should be used immediately after preparation.

Requirements

The powder for injection and the reconstituted injection comply with the monograph for *Parenteral preparations*.

Definition. Artesunate for injection is a sterile powder containing Artesunate. It contains not less than 90.0% and not more than 110.0% of the amount of artesunate ($C_{19}H_{28}O_8$)stated on the label.

Manufacture. The manufacturing process and the product packaging are designed and controlled so as to minimize the moisture content of the powder. They ensure that, if tested, the powder would comply with a water limit of not more than 5 mg/g when determined as described under *2.8 Determination of water by the Karl Fischer method*, method A.

Identity tests

-Either test A alone or tests B, C, and D may be applied.

A. Carry out the examination as described under *1.7 Spectrophotometry in the infrared region*. The infrared absorption spectrum is concordant with the spectrum obtained from artesunate RS or with the *reference spectrum* of artesunate.

B. Carry out the test as described under *1.14.1 Chromatography*, Thin-layer chromatography, using silica gel R5 as the coating substance and a mixture of 70 volumes of ethanol R, 30 volumes of toluene R and 1.5 volumes of ammonia (~260 g/L) TS as the mobile phase. Apply separately to the plate 1 µL of the following two solutions in methanol R. For solution (A) dissolve a quantity of the powder to obtain a solution containing 1.0 mg of Artesunate per mL. For solution (B) use 1.0 mg of artesunate RS per mL. After removing the plate from the chromatographic chamber allow it to dry in air or in a current of cool air. Spray with anisaldehyde/methanol TS and heat the plate to 120 °C for 5 minutes. Examine the chromatogram in daylight.

The principal spot obtained with solution A corresponds in position, appearance and intensity with that obtained with solution B.

C. Dissolve a quantity of the powder containing 0.1 g of Artesunate in 40 mL of dehydrated ethanol R, shake and filter. To half of the filtrate (keep the remaining filtrate for test D) add about 0.5 mL of hydroxylamine hydrochloride TS2 and 0.25 mL of sodium hydroxide (~80 g/L) TS. Heat the mixture in a water-bath to boiling, cool, add 2 drops of hydrochloric acid (~70 g/L) TS and 2 drops of ferric chloride (50 g/L) TS; a reddish-brown colour is produced.

D. Evaporate the remaining filtrate from test C on a water-bath to a volume of about 5 mL. Place a few drops of the mixture on a white porcelain dish, add one drop of vanillin/sulfuric acid TS1; a red colour is produced.

Bacterial endotoxins. Carry out the test as described under *3.4 Test for bacterial endotoxins*; contains not more than 2.5 IU of endotoxin per mg of artesunate.

Related substances

Carry out the test as described under *1.14.1 Chromatography*, High-performance liquid chromatography, using the conditions given below under “Assay”, method A.

Use solutions (1) and (3) as described under “Assay”, method A. For solution (4) dilute 1 mL of solution (1) to 100 mL with acetonitrile R.

Inject separately 20 µL each of solutions (1), (3) and (4). Record the chromatograms for about 4 times the retention time of artesunate. In the chromatogram obtained with solution (3) the following peaks are eluted at the following relative retention with reference to artesunate (retention time about 9 minutes): 10-*epi*-artenimol about 0.58; artenimol about 0.91; and impurity B (artemisinin) about 1.30. The test is not valid unless the peak-to-valley ratio (Hp/Hv) is at least 5.0, where Hp is the height above

the extrapolated baseline of the peak due to artenimol and Hv is the height above the extrapolated baseline at the lowest point of the curve separating the peak due to artenimol from the peak due to artesunate. The chromatogram obtained with solution (1) may show a peak due to impurity C eluting at a relative retention of about 2.7 with reference to artesunate.

In the chromatogram obtained with solution (1):

- -the combined areas of any peaks corresponding to 10-*epi*-artenimol and artenimol (impurity A) are not greater than the area of the principal peak obtained with solution (4) (1.0%);
- -the area of any peak corresponding to impurity B (artemisinin) is not greater than 0.5 times the area of the principal peak obtained with solution (4) (0.5%);
- -the area of any peak corresponding to impurity C, when multiplied by a correction factor of 0.07, is not greater than 0.3 times the area of the principal peak obtained with solution (4) (0.3%);
- -the area of any other peak, other than the principal peak, is not greater than 0.3 times the area of the principal peak in the chromatogram obtained with solution (4) (0.3%);
- -the sum of the corrected area of any peak corresponding to impurity C and the areas of all other peaks, other than the principal peak, is not greater than twice the area of the principal peak obtained with solution (4) (2.0%). Disregard any peak with an area less than 0.1 times the area of the principal peak in the chromatogram obtained with solution (4) (0.1%).

Assay

-Either method A or method B may be applied.

A. Carry out the test as described under *1.14.1 Chromatography*, High-performance liquid chromatography, using a stainless steel column (10 cm × 4.6 mm) packed with particles of silica gel, the surface of which has been modified with chemically-bonded octadecylsilyl groups (3 µm). As the mobile phase use a mixture of 44 volumes of acetonitrile R and 56 volumes of buffer pH 3.0.

Prepare the buffer pH 3.0 by dissolving 1.36 g of potassium dihydrogen phosphate R in 900 mL of water R, adjust the pH to 3.0 with phosphoric acid (~1440 g/L) TS and dilute to 1000 mL with water R.

Prepare the following solutions in acetonitrile R. For solution (1) determine the weight of the contents of 10 containers. Transfer a quantity of the mixed contents containing about 40 mg of Artesunate, accurately weighed, to a 10 mL volumetric flask, add 7 mL and shake to dissolve. Dilute to volume and filter. For solution (2) dissolve 40 mg of artesunate RS, accurately weighed, and dilute to 10 mL. For solution (3) dissolve about 1 mg of artenimol RS, about 1 mg of artemisinin RS and about 10 mg of artesunate RS in 10 mL.

Operate with a flow rate of 1.0 mL per minute. Maintain the column temperature at 30 °C and use as detector an ultraviolet spectrophotometer set at a wavelength of about 216 nm.

Inject separately 20 µL each of solutions (1), (2) and (3). Record the chromatograms for about 4 times the retention time of artesunate. In the chromatogram obtained with solution (3) the following peaks are eluted at the following relative retention with reference to artesunate (retention time about 9 minutes): 10-*epi*-artenimol about 0.58; artenimol about 0.91; and impurity B (artemisinin) about 1.30. The test is not valid unless the peak-to-valley ratio (Hp/Hv) is at least 5.0, where Hp is the height above the extrapolated baseline of the peak due to artenimol and Hv = the height above the extrapolated baseline at the lowest point of the curve separating the peak due to artenimol from the peak due to artesunate. The chromatogram obtained with solution (1) may show a peak due to impurity C eluting at a relative retention of about 2.7 with reference to artesunate.

Measure the areas of the peak responses obtained in the chromatograms from solutions (1) and (2) and calculate the content of artesunate ($C_{19}H_{28}O_8$) per sealed container, using the declared content of $C_{19}H_{28}O_8$ in artesunate.

B. Determine the weight of the contents of 10 containers. Dissolve a quantity of the mixed contents containing about 0.25 g of Artesunate, accurately weighed, in 25 mL of neutralized ethanol TS and titrate with sodium hydroxide (0.05 mol/L) VS, using 2 drops of phenolphthalein/ethanol TS as indicator.

Each mL of sodium hydroxide (0.05 mol/L) VS is equivalent to 19.22 mg of $C_{19}H_{28}O_8$.

Calculate the content of artesunate per sealed container.

Impurities

The impurities limited by the requirements of this monograph include those listed in the monograph for Artesunate.