

Test solution (a). Dissolve 0.250 g of the substance to be examined in *water R* and dilute to 5.0 ml with the same solvent.

Test solution (b). Dissolve 0.50 g of the substance to be examined in the internal standard solution and dilute to 10.0 ml with the internal standard solution.

Reference solution. Dilute 0.50 ml of *2-propanol R* to 100.0 ml with the internal standard solution.

Column:

- size: $l = 1.5$ m, $\varnothing = 4$ mm;
- stationary phase: *ethylvinylbenzene-divinylbenzene copolymer R* (125-150 μ m).

Carrier gas: nitrogen for chromatography *R*.

Flow rate: 40 ml/min.

Temperature:

- column: 150 °C;
- injection port: 180 °C;
- detector: 200 °C.

Detection: flame ionisation.

Injection: the chosen volume of the test solutions and the reference solution.

Calculate the content of propan-2-ol taking its density at 20 °C to be 0.785 g/ml.

Limit:

- *propan-2-ol:* 8.0 per cent to 8.5 per cent.

Water (2.5.12): maximum 0.3 per cent, determined on 2.500 g.

ASSAY

Dissolve 0.100 g in 0.01 M sodium hydroxide and dilute to 100.0 ml with the same solvent. Dilute 10.0 ml of the solution to 100.0 ml with 0.01 M sodium hydroxide. Dilute 10.0 ml of this solution to 100.0 ml with 0.01 M sodium hydroxide. Measure the absorbance (2.2.25) at the absorption maximum at 308 nm.

Calculate the percentage content of warfarin sodium ($C_{19}H_{15}NaO_4$) taking the specific absorbance to be 431.

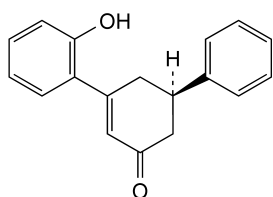
STORAGE

In an airtight container, protected from light.

IMPURITIES

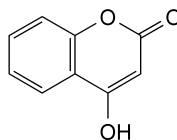
Specified impurities: B, C.

Other detectable impurities (the following substances would, if present at a sufficient level, be detected by one or other of the tests in the monograph. They are limited by the general acceptance criterion for other/unspecified impurities and/or by the general monograph *Substances for pharmaceutical use (2034)*. It is therefore not necessary to identify these impurities for demonstration of compliance. See also 5.10. *Control of impurities in substances for pharmaceutical use*): A.

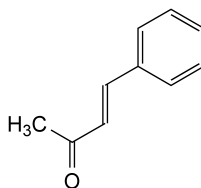


and enantiomer

A. (5*RS*)-3-(2-hydroxyphenyl)-5-phenylcyclohex-2-enone,



B. 4-hydroxy-2*H*-1-benzopyran-2-one (4-hydroxycoumarin),



C. (3*E*)-4-phenylbut-3-en-2-one (benzalacetone).

01/2008:0169

WATER FOR INJECTIONS

Aqua ad iniectabilia

H₂O
[7732-18-5]

M_r 18.02

DEFINITION

Water for the preparation of medicines for parenteral administration when water is used as vehicle (water for injections in bulk) and for dissolving or diluting substances or preparations for parenteral administration (sterilised water for injections).

Water for injections in bulk

PRODUCTION

Water for injections in bulk is obtained from water that complies with the regulations on water intended for human consumption laid down by the competent authority or from purified water by distillation in an apparatus of which the parts in contact with the water are of neutral glass, quartz or suitable metal and which is fitted with an effective device to prevent the entrainment of droplets. The correct maintenance of the apparatus is essential. The first portion of the distillate obtained when the apparatus begins to function is discarded and the distillate is collected.

During production and subsequent storage, appropriate measures are taken to ensure that the total viable aerobic count is adequately controlled and monitored. Appropriate alert and action limits are set so as to detect adverse trends. Under normal conditions, an appropriate action limit is a total viable aerobic count (2.6.12) of 10 micro-organisms per 100 ml when determined by membrane filtration, using agar medium S, using at least 200 ml of water for injections in bulk and incubating at 30-35 °C for 5 days. For aseptic processing, stricter alert limits may need to be applied.

Total organic carbon (2.2.44): maximum 0.5 mg/l.

Conductivity. Determine the conductivity off-line or in-line under the following conditions.

EQUIPMENT

Conductivity cell:

- electrodes of a suitable material such as stainless steel;
- cell constant: within 2 per cent of the given value determined using a certified reference solution with a conductivity less than 1500 μ S \cdot cm⁻¹.

Conductometer: resolution 0.1 μ S \cdot cm⁻¹ on the lowest range.

System calibration (conductivity cell and conductometer): Stage 2

- against one or more suitable certified standard solutions;
- accuracy: within 3 per cent of the measured conductivity plus $0.1 \mu\text{S}\cdot\text{cm}^{-1}$.

Conductometer calibration: by means of precision resistors or equivalent devices, after disconnecting the conductivity cell, for all ranges used for conductivity measurement and cell calibration (with an accuracy within 0.1 per cent of the stated value, traceable to the official standard).

If in-line conductivity cells cannot be dismantled, system calibration may be performed against a calibrated conductivity cell placed close to the cell to be calibrated in the water flow.

PROCEDURE*Stage 1*

1. Measure the conductivity without temperature compensation, recording simultaneously the temperature. Temperature-compensated measurement may be performed after suitable validation.
2. Using Table 0169.-1, find the closest temperature value that is not greater than the measured temperature. The corresponding conductivity value is the limit at that temperature.
3. If the measured conductivity is not greater than the value in Table 0169.-1, the water to be examined meets the requirements of the test for conductivity. If the conductivity is higher than the value in Table 0169.-1, proceed with stage 2.

Table 0169.-1. – *Stage 1 - Temperature and conductivity requirements (for non-temperature-compensated conductivity measurements)*

Temperature (°C)	Conductivity ($\mu\text{S}\cdot\text{cm}^{-1}$)
0	0.6
5	0.8
10	0.9
15	1.0
20	1.1
25	1.3
30	1.4
35	1.5
40	1.7
45	1.8
50	1.9
55	2.1
60	2.2
65	2.4
70	2.5
75	2.7
80	2.7
85	2.7
90	2.7
95	2.9
100	3.1

4. Transfer a sufficient amount of the water to be examined (100 ml or more) to a suitable container, and stir the test sample. Adjust the temperature, if necessary, and while maintaining it at 25 ± 1 °C, begin vigorously agitating the test sample while periodically observing the conductivity. When the change in conductivity (due to uptake of atmospheric carbon dioxide) is less than $0.1 \mu\text{S}\cdot\text{cm}^{-1}$ per 5 min, note the conductivity.
5. If the conductivity is not greater than $2.1 \mu\text{S}\cdot\text{cm}^{-1}$, the water to be examined meets the requirements of the test for conductivity. If the conductivity is greater than $2.1 \mu\text{S}\cdot\text{cm}^{-1}$, proceed with stage 3.

Stage 3

6. Perform this test within approximately 5 min of the conductivity determination in step 5 under stage 2, while maintaining the sample temperature at 25 ± 1 °C. Add a recently prepared saturated solution of *potassium chloride R* to the test sample (0.3 ml per 100 ml of the test sample), and determine the pH (2.2.3) to the nearest 0.1 pH unit.
7. Using Table 0169.-2, determine the conductivity limit at the measured pH value in step 6. If the measured conductivity in step 4 under stage 2 is not greater than the conductivity requirements for the pH determined, the water to be examined meets the requirements of the test for conductivity. If either the measured conductivity is greater than this value or the pH is outside the range of 5.0-7.0, the water to be examined does not meet the requirements of the test for conductivity.

Table 0169.-2. – *Stage 3 - pH and conductivity requirements (for atmosphere and temperature equilibrated samples)*

pH	Conductivity ($\mu\text{S}\cdot\text{cm}^{-1}$)
5.0	4.7
5.1	4.1
5.2	3.6
5.3	3.3
5.4	3.0
5.5	2.8
5.6	2.6
5.7	2.5
5.8	2.4
5.9	2.4
6.0	2.4
6.1	2.4
6.2	2.5
6.3	2.4
6.4	2.3
6.5	2.2
6.6	2.1
6.7	2.6
6.8	3.1
6.9	3.8
7.0	4.6

In order to ensure the appropriate quality of the water, validated procedures and in-process-monitoring of the electrical conductivity and regular microbial monitoring are applied.

Water for injections in bulk is stored and distributed in conditions designed to prevent growth of micro-organisms and to avoid any other contamination.

CHARACTERS

Appearance: clear and colourless liquid.

TESTS

Nitrates: maximum 0.2 ppm.

Place 5 ml in a test-tube immersed in iced water, add 0.4 ml of a 100 g/l solution of *potassium chloride R*, 0.1 ml of *diphenylamine solution R* and, dropwise with shaking, 5 ml of *nitrogen-free sulphuric acid R*. Transfer the tube to a water-bath at 50 °C. After 15 min, any blue colour in the solution is not more intense than that in a reference solution prepared at the same time in the same manner using a mixture of 4.5 ml of *nitrate-free water R* and 0.5 ml of *nitrate standard solution (2 ppm NO₃) R*.

Aluminium (2.4.17): maximum 10 ppb, if intended for use in the manufacture of dialysis solutions.

Prescribed solution. To 400 ml of the water to be examined add 10 ml of *acetate buffer solution pH 6.0 R* and 100 ml of *distilled water R*.

Reference solution. Mix 2 ml of *aluminium standard solution (2 ppm Al) R*, 10 ml of *acetate buffer solution pH 6.0 R* and 98 ml of *distilled water R*.

Blank solution. Mix 10 ml of *acetate buffer solution pH 6.0 R* and 100 ml of *distilled water R*.

Heavy metals (2.4.8): maximum 0.1 ppm.

Heat 200 ml in a glass evaporating dish on a water-bath until the volume is reduced to 20 ml. 12 ml of the concentrated solution complies with limit test A. Prepare the standard using 10 ml of *lead standard solution (1 ppm Pb) R*.

Bacterial endotoxins (2.6.14): less than 0.25 IU/ml.

Sterilised water for injections

DEFINITION

Water for injections in bulk that has been distributed into suitable containers, closed and sterilised by heat in conditions which ensure that the product still complies with the test for bacterial endotoxins. Sterilised water for injections is free from any added substances.

Examined in suitable conditions of visibility, it is clear and colourless.

Each container contains a sufficient quantity of water for injections to permit the nominal volume to be withdrawn.

TESTS

Acidity or alkalinity. To 20 ml add 0.05 ml of *phenol red solution R*. If the solution is yellow, it becomes red on the addition of 0.1 ml of *0.01 M sodium hydroxide*; if red, it becomes yellow on the addition of 0.15 ml of *0.01 M hydrochloric acid*.

Conductivity: maximum 25 µS·cm⁻¹ for containers with a nominal volume of 10 ml or less; maximum 5 µS·cm⁻¹ for containers with a nominal volume greater than 10 ml.

Use equipment and the calibration procedure as defined under Water for injections in bulk, maintaining the sample temperature at 25 ± 1 °C.

Oxidisable substances. Boil 100 ml with 10 ml of *dilute sulphuric acid R*. Add 0.2 ml of *0.02 M potassium permanganate* and boil for 5 min. The solution remains faintly pink.

Chlorides (2.4.4): maximum 0.5 ppm for containers with a nominal volume of 100 ml or less.

15 ml complies with the limit test for chlorides. Prepare the standard using a mixture of 1.5 ml of *chloride standard solution (5 ppm Cl) R* and 13.5 ml of *water R*. Examine the solutions down the vertical axes of the tubes.

For containers with a nominal volume greater than 100 ml, use the following test: to 10 ml add 1 ml of *dilute nitric acid R* and 0.2 ml of *silver nitrate solution R2*. The solution shows no change in appearance for at least 15 min.

Nitrates: maximum 0.2 ppm.

Place 5 ml in a test-tube immersed in iced water, add 0.4 ml of a 100 g/l solution of *potassium chloride R*, 0.1 ml of *diphenylamine solution R* and, dropwise with shaking, 5 ml of *nitrogen-free sulphuric acid R*. Transfer the tube to a water-bath at 50 °C. After 15 min, any blue colour in the solution is not more intense than that in a reference solution prepared at the same time in the same manner using a mixture of 4.5 ml of *nitrate-free water R* and 0.5 ml of *nitrate standard solution (2 ppm NO₃) R*.

Sulphates. To 10 ml add 0.1 ml of *dilute hydrochloric acid R* and 0.1 ml of *barium chloride solution R1*. The solution shows no change in appearance for at least 1 h.

Aluminium (2.4.17): maximum 10 ppb, if intended for use in the manufacture of dialysis solutions.

Prescribed solution. To 400 ml of the water to be examined add 10 ml of *acetate buffer solution pH 6.0 R* and 100 ml of *distilled water R*.

Reference solution. Mix 2 ml of *aluminium standard solution (2 ppm Al) R*, 10 ml of *acetate buffer solution pH 6.0 R* and 98 ml of *distilled water R*.

Blank solution. Mix 10 ml of *acetate buffer solution pH 6.0 R* and 100 ml of *distilled water R*.

Ammonium: maximum 0.2 ppm.

To 20 ml add 1 ml of *alkaline potassium tetraiodomercurate solution R*. After 5 min, examine the solution down the vertical axis of the tube. The solution is not more intensely coloured than a standard prepared at the same time by adding 1 ml of *alkaline potassium tetraiodomercurate solution R* to a mixture of 4 ml of *ammonium standard solution (1 ppm NH₄) R* and 16 ml of *ammonium-free water R*.

Calcium and magnesium. To 100 ml add 2 ml of *ammonium chloride buffer solution pH 10.0 R*, 50 mg of *mordant black 11 triturate R* and 0.5 ml of *0.01 M sodium edetate*. A pure blue colour is produced.

Heavy metals (2.4.8): maximum 0.1 ppm.

Heat 200 ml in a glass evaporating dish on a water-bath until the volume is reduced to 20 ml. 12 ml of the concentrated solution complies with limit test A. Prepare the standard using 10 ml of *lead standard solution (1 ppm Pb) R*.

Residue on evaporation: maximum 4 mg (0.004 per cent) for containers with a nominal volume of 10 ml or less; maximum 3 mg (0.003 per cent) for containers with a nominal volume greater than 10 ml.

Evaporate 100 ml to dryness on a water-bath and dry in an oven at 100-105 °C.

Particulate contamination: sub-visible particles (2.9.19). It complies with test A or test B, as appropriate.

Sterility (2.6.1). It complies with the test for sterility.

Bacterial endotoxins (2.6.14): less than 0.25 IU/ml.

01/2008:1927

WATER, HIGHLY PURIFIED

Aqua valde purificata

H₂O
[7732-18-5]

M_r 18.02

DEFINITION

Water intended for use in the preparation of medicinal products where water of high biological quality is needed, except where *Water for injections* (0169) is required.

PRODUCTION

Highly purified water is obtained from water that complies with the regulations on water intended for human consumption laid down by the competent authority.

Current production methods include for example double-pass reverse osmosis coupled with other suitable techniques such as ultrafiltration and deionisation. Correct operation and maintenance of the system is essential.

During production and subsequent storage, appropriate measures are taken to ensure that the total viable aerobic count is adequately controlled and monitored. Appropriate alert and action limits are set so as to detect adverse trends. Under normal conditions, an appropriate action limit is a total viable aerobic count (2.6.12) of 10 micro-organisms per 100 ml when determined by membrane filtration, using agar medium S, at least 200 ml of highly purified water and incubating at 30-35 °C for 5 days.

Total organic carbon (2.2.44): maximum 0.5 mg/l.

Conductivity. Determine the conductivity off-line or in-line under the following conditions.

EQUIPMENT

Conductivity cell:

- electrodes of a suitable material such as stainless steel;
- cell constant: within 2 per cent of the given value determined using a certified reference solution with a conductivity less than 1500 µS·cm⁻¹.

Conductometer: resolution 0.1 µS·cm⁻¹ on the lowest range.

System calibration (conductivity cell and conductometer):

- against one or more suitable certified standard solutions;
- accuracy: within 3 per cent of the measured conductivity plus 0.1 µS·cm⁻¹.

Conductometer calibration: by means of precision resistors or equivalent devices after disconnecting the conductivity cell, for all ranges used for conductivity measurement and cell calibration (with an accuracy within 0.1 per cent of the stated value, traceable to the official standard).

If in-line conductivity cells cannot be dismantled, system calibration may be performed against a calibrated conductivity cell placed close to the cell to be calibrated in the water flow.

PROCEDURE

Stage 1

1. Measure the conductivity without temperature compensation, recording simultaneously the temperature. Temperature-compensated measurement may be performed after suitable validation.

2. Using Table 1927-1, find the closest temperature value that is not greater than the measured temperature. The corresponding conductivity value is the limit at that temperature.
3. If the measured conductivity is not greater than the value in Table 1927-1, the water to be examined meets the requirements of the test for conductivity. If the conductivity is higher than the value in Table 1927-1, proceed with stage 2.

Table 1927-1. – Stage 1 - Temperature and conductivity requirements (for non-temperature-compensated conductivity measurements)

Temperature (°C)	Conductivity (µS·cm ⁻¹)
0	0.6
5	0.8
10	0.9
15	1.0
20	1.1
25	1.3
30	1.4
35	1.5
40	1.7
45	1.8
50	1.9
55	2.1
60	2.2
65	2.4
70	2.5
75	2.7
80	2.7
85	2.7
90	2.7
95	2.9
100	3.1

Stage 2

4. Transfer a sufficient amount of the water to be examined (100 ml or more) to a suitable container, and stir the test sample. Adjust the temperature, if necessary, and while maintaining it at 25 ± 1 °C, begin vigorously agitating the test sample while periodically observing the conductivity. When the change in conductivity (due to uptake of atmospheric carbon dioxide) is less than 0.1 µS·cm⁻¹ per 5 min, note the conductivity.
5. If the conductivity is not greater than 2.1 µS·cm⁻¹, the water to be examined meets the requirements of the test for conductivity. If the conductivity is greater than 2.1 µS·cm⁻¹, proceed with stage 3.

Stage 3

6. Perform this test within approximately 5 min of the conductivity determination in step 5 under stage 2, while maintaining the sample temperature at 25 ± 1 °C. Add a recently prepared saturated solution of *potassium chloride R* to the test sample (0.3 ml per 100 ml of the test sample), and determine the pH (2.2.3) to the nearest 0.1 pH unit.