Reference solution (b). Dissolve the contents of a vial of *acetazolamide for system suitability CRS* (containing impurities A, B, C, D, E and F) in 1.0 mL of the mobile phase.

Column:

– *size*: l = 0.15 m, Ø = 4.6 mm;
– *stationary phase*: *end-capped propoxybenzene silica gel for chromatography R* (4 μm).

Mobile phase: *acetonitrile for chromatography R*, 6.8 g/L solution of *potassium dihydrogen phosphate R* (10:90 *V/V*).

Flow rate: 1.0 mL/min.

Detection: spectrophotometer at 265 nm.

Injection: 25 μl.

Run time: 3.5 times the retention time of acetazolamide.

Identification of impurities: use the chromatogram supplied with *acetazolamide for system suitability CRS* and the chromatogram obtained with reference solution (b) to identify the peaks due to impurities A, B, C, D, E and F.

Relative retention with reference to acetazolamide (retention time = about 8 min): impurity E = about 0.3; impurity D = about 0.4; impurity B = about 0.6; impurity C = about 1.4; impurity A = about 2.1; impurity F = about 2.6.

System suitability: reference solution (b):

– *resolution*: minimum 2.0 between the peaks due to impurities E and D.

Limits:

– *correction factors*: for the calculation of content, multiply the peak areas of the following impurities by the corresponding correction factor: impurity B = 2.3; impurity C = 2.6; impurity D = 1.6;
– *impurities A, B, C, D, E, F*: for each impurity, not more than 1.5 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.15 per cent);
– *unspecified impurities*: for each impurity, not more than the area of the principal peak in the chromatogram obtained with reference solution (a) (0.10 per cent);
– *total*: not more than 6 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.6 per cent);
– *disregard limit*: 0.5 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.05 per cent).

Sulfates (*2.4.13*): maximum 500 ppm.

To 0.4 g add 20 mL of *distilled water R* and dissolve by heating to boiling. Allow to cool with frequent shaking and filter.

Heavy metals (*2.4.8*): maximum 20 ppm.

1.0 g complies with test C. Prepare the reference solution using 2 mL of *lead standard solution (10 ppm Pb) R*.

Loss on drying (*2.2.32*): maximum 0.5 per cent, determined on 1.000 g by drying in an oven at 105 °C.

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

ASSAY

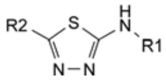
Dissolve 0.200 g in 25 mL of *dimethylformamide R*. Titrate with *0.1 M ethanolic sodium hydroxide*, determining the end-point potentiometrically (*2.2.20*).

1 mL of *0.1 M ethanolic sodium hydroxide* is equivalent to 22.22 mg of $C_4H_6N_4O_3S_2$.

IMPURITIES

Specified impurities: *A, B, C, D, E, F.*

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*): *G.*



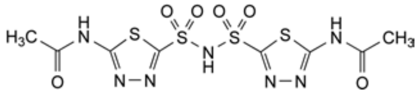
A. R1 = CO-CH_3, R2 = Cl: *N*-(5-chloro-1,3,4-thiadiazol-2-yl)acetamide,

B. R1 = CO-CH_3, R2 = H: *N*-(1,3,4-thiadiazol-2-yl)acetamide,

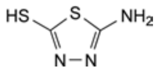
C. R1 = CO-CH_3, R2 = SH: *N*-(5-sulfanyl-1,3,4-thiadiazol-2-yl)acetamide,

D. R1 = H, R2 = SO_2-NH_2: 5-amino-1,3,4-thiadiazole-2-sulfonamide,

E. R1 = CO-CH_3, R2 = SO_2-OH: 5-acetamido-1,3,4-thiadiazole-2-sulfonic acid,



F. *N*-[5-[(5-acetamido-1,3,4-thiadiazol-2-yl)sulfonyl]sulfamoyl-1,3,4-thiadiazol-2-yl]acetamide,



G. 5-amino-1,3,4-thiadiazole-2-thiol.

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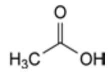
ACETIC ACID, GLACIAL

Acidum aceticum glaciale



$C_2H_4O_2$ M_r 60.1

[64-19-7]

DEFINITION

Content: 99.0 per cent *m/m* to 100.5 per cent *m/m*.

CHARACTERS

Appearance: crystalline mass or clear, colourless, volatile liquid.

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

IDENTIFICATION

A. A 100 g/L solution is strongly acid (*2.2.4*).

B. To 0.03 mL add 3 mL of *water R* and neutralise with *dilute sodium hydroxide solution R*. The solution gives reaction (b) of acetates (*2.3.1*).

TESTS

Solution S. Dilute 20 mL to 100 mL with *distilled water R*.

Appearance. The substance to be examined is clear (*2.2.1*) and colourless (*2.2.2, Method II*).

Freezing point (*2.2.18*): minimum 14.8 °C.

Reducing substances. To 5.0 mL add 10.0 mL of *water R* and mix. To 5.0 mL of this solution add 6 mL of *sulfuric acid R*, cool and add 2.0 mL of *0.0167 M potassium dichromate*. Allow to stand for 1 min and add 25 mL of *water R* and 1 mL of a freshly prepared 100 g/L solution of *potassium iodide R*. Titrate with *0.1 M sodium thiosulfate*, using 1.0 mL of *starch solution R* as indicator. Not less than 1.0 mL of *0.1 M sodium thiosulfate* solution is required.

Chlorides (*2.4.4*): maximum 25 mg/L.

Dilute 10 mL of solution S to 15 mL with *water R*.

Sulfates (*2.4.13*): maximum 50 mg/L, determined on solution S.

Iron (*2.4.9*): maximum 5 ppm.

Dilute 5.0 mL of solution A obtained in the test for heavy metals to 10.0 mL with *water R*.

Heavy metals (*2.4.8*): maximum 5 ppm.

Dissolve the residue obtained in the test for residue on evaporation by heating with 2 quantities, each of 15 mL, of *water R* and dilute to 50.0 mL (solution A). 12 mL of solution A complies with test A. Prepare the reference solution using *lead standard solution (2 ppm Pb) R*.

Residue on evaporation: maximum 0.01 per cent.

Evaporate 20 g to dryness on a water-bath and dry at 100-105 °C. The residue weighs a maximum of 2.0 mg.

ASSAY

Weigh accurately a conical flask with a ground-glass stopper containing 25 mL of *water R*. Add 1.0 mL of the substance to be examined and weigh again accurately. Add 0.5 mL of *phenolphthalein solution R* and titrate with *1 M sodium hydroxide*.

1 mL of *1 M sodium hydroxide* is equivalent to 60.1 mg of $C_2H_4O_2$.

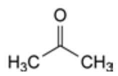
STORAGE

In an airtight container.

01/2008:0872

ACETONE

Acetonum



C_3H_6O M_r 58.08
[67-64-1]

DEFINITION

Propanone.

CHARACTERS

Appearance: volatile, clear, colourless liquid.

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

The vapour is flammable.

IDENTIFICATION

A. Relative density (see Tests).

B. To 1 mL, add 3 mL of *dilute sodium hydroxide solution R* and 0.3 mL of a 25 g/L solution of *sodium nitroprusside R*. An intense red colour is produced which becomes violet with the addition of 3.5 mL of *acetic acid R*.

C. To 10 mL of a 0.1 per cent *V/V* solution of the substance to be examined in *ethanol (50 per cent V/V) R*, add 1 mL of a 10 g/L solution of *nitrobenzaldehyde R* in *ethanol (50 per cent V/V) R* and 0.5 mL of *strong sodium hydroxide solution R*. Allow to stand for about 2 min and acidify with *acetic acid R*. A greenish-blue colour is produced.

TESTS

Appearance of solution. To 10 mL add 10 mL of *water R*. The solution is clear (*2.2.1*) and colourless (*2.2.2, Method II*).

Acidity or alkalinity. To 5 mL add 5 mL of *carbon dioxide-free water R*, 0.15 mL of *phenolphthalein solution R* and 0.5 mL of *0.01 M sodium hydroxide*. The solution is pink. Add 0.7 mL of *0.01 M hydrochloric acid* and 0.05 mL of *methyl red solution R*. The solution is red or orange.

Relative density (*2.2.5*): 0.790 to 0.793.

Reducing substances. To 30 mL add 0.1 mL of *0.02 M potassium permanganate* and allow to stand in the dark for 2 h. The mixture is not completely decolourised.

Related substances. Gas chromatography (*2.2.28*).

Test solution. The substance to be examined.

Reference solution (a). To 0.5 mL of *methanol R* add 0.5 mL of *2-propanol R* and dilute to 100.0 mL with the test solution. Dilute 1.0 mL of this solution to 10.0 mL with the test solution.

Reference solution (b). Dilute 100 µL of *benzene R* to 100.0 mL with the test solution. Dilute 0.20 mL of this solution to 100.0 mL with the test solution.

Column:
- *material*: fused silica,
- *size*: *l* = 50 m, Ø = 0.3 mm,
- *stationary phase*: *macrogol 20 000 R* (film thickness 1 µm).

Carrier gas: *helium for chromatography R*.

Linear velocity: 21 cm/s.

Split ratio: 1:50.

Temperature:

	Time (min)	Temperature (°C)
Column	0 - 11	45 → 100
	11 - 20	100
Injection port		150
Detector		250

Detection: flame ionisation.

Injection: 1 µL.

Retention time: impurity C = about 7.5 min.

System suitability:
- *resolution*: minimum 5.0 between the peak due to impurity A (2nd peak) and the peak due to impurity B (3rd peak) in the chromatogram obtained with reference solution (a),
- *signal-to-noise ratio*: minimum 5 for the peak due to impurity C in the chromatogram obtained with reference solution (b).

Limits:
- *impurities A, B*: for each impurity, not more than the difference between the areas of the corresponding peaks in the chromatogram obtained with reference solution (a) and the areas of the corresponding peaks in the chromatogram obtained with the test solution (0.05 per cent *V/V*),
- *impurity C*: not more than the difference between the area of the peak due to impurity C in the chromatogram obtained with reference solution (b) and the area of the corresponding peak in the chromatogram obtained with the test solution (2 ppm *V/V*),
- *any other impurity*: for each impurity, not more than the difference between the area of the peak due to impurity A in the chromatogram obtained with reference solution (a) and the area of the corresponding peak in the chromatogram obtained with the test solution (0.05 per cent *V/V*).

Matter insoluble in water. Dilute 1.0 mL to 20 mL with *water R*. The solution is clear (*2.2.1*).

Residue on evaporation: maximum 50 ppm.

Evaporate 20.0 g to dryness on a water-bath and dry at 100-105 °C. The residue weighs a maximum of 1 mg.

Water (*2.5.12*): maximum 3 g/L, determined on 10.0 mL. Use 20 mL of *anhydrous pyridine R* as solvent.

STORAGE

Protected from light.

IMPURITIES

Specified impurities: A, B, C.

A. CH_3-OH: methanol,

B. CH_3-CHOH-CH_3: propan-2-ol (isopropanol),

C. C_6H_6: benzene.