*Run time*: 3 times the retention time of acesulfame.

*Relative retention* with reference to acesulfame (retention time = about 5.3 min): impurity B = about 1.6.

*System suitability*:

– *peak-to-valley ratio*: minimum 1.2, where $H_p$ = height above the baseline of the peak due to impurity B and $H_v$ = height above the baseline of the lowest point of the curve separating this peak from the peak due to acesulfame in the chromatogram obtained with reference solution (b).

*Limits*:

– *impurity B*: not more than the area of the principal peak in the chromatogram obtained with reference solution (a) (20 ppm),

– *unspecified impurities*: for each impurity, not more than the area of the principal peak in the chromatogram obtained with reference solution (c) (0.1 per cent),

– *total*: not more than the area of the principal peak in the chromatogram obtained with reference solution (c) (0.1 per cent),

– *disregard limit*: 0.5 times the area of the principal peak in the chromatogram obtained with reference solution (c) except for the peak due to impurity B (0.05 per cent).

**Fluorides**: maximum 3 ppm.

Potentiometry (*2.2.36, Method I*).

*Test solution*. Dissolve 3.000 g of the substance to be examined in *distilled water R*, add 15.0 mL of *total-ionic-strength-adjustment buffer R1* and dilute to 50.0 mL with *distilled water R*.

*Reference solutions*. To 0.5 mL, 1.0 mL, 1.5 mL and 3.0 mL of *fluoride standard solution (10 ppm F) R* add 15.0 mL of *total-ionic-strength-adjustment buffer R1* and dilute to 50.0 mL with *distilled water R*.

*Indicator electrode*: fluoride-selective.

*Reference electrode*: silver-silver chloride.

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

12 mL of solution S complies with test A. Prepare the reference solution using *lead standard solution (1 ppm Pb) R*.

**Loss on drying** (*2.2.32*): maximum 1.0 per cent, determined on 1.000 g by drying in an oven at 105 °C for 3 h.

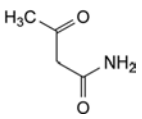
ASSAY

Dissolve 0.150 g in 50 mL of *anhydrous acetic acid R*. Titrate with *0.1 M perchloric acid*, determining the end-point potentiometrically (*2.2.20*).

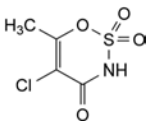
1 mL of *0.1 M perchloric acid* is equivalent to 20.12 mg of $C_4H_4KNO_4S$.

IMPURITIES

*Specified impurities: A, B.*



A. 3-oxobutanamide (acetylacetamide),

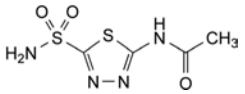


B. 5-chloro-6-methyl-1,2,3-oxathiazin-4(3*H*)-one 2,2-dioxide.

**04/2009:0454**

# ACETAZOLAMIDE

## Acetazolamidum



$C_4H_6N_4O_3S_2$ $M_r$ 222.2
[59-66-5]

DEFINITION

*N*-(5-Sulfamoyl-1,3,4-thiadiazol-2-yl)acetamide.

*Content*: 98.5 per cent to 101.0 per cent (dried substance).

CHARACTERS

*Appearance*: white or almost white, crystalline powder.

*Solubility*: very slightly soluble in water, slightly soluble in ethanol (96 per cent). It dissolves in dilute solutions of alkali hydroxides.

It shows polymorphism (*5.9*).

IDENTIFICATION

*First identification: A, B.*

*Second identification: A, C, D.*

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

*Solution A*. Dissolve 30.0 mg 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*.

*Solution B*. Dilute 25.0 mL of solution A to 100.0 mL with *0.01 M sodium hydroxide*.

*Spectral range*: 230-260 nm for solution A; 260-350 nm for solution B.

*Absorption maximum*: at 240 nm for solution A; at 292 nm for solution B.

*Specific absorbance at the absorption maximum*: 162 to 176 for solution A; 570 to 620 for solution B.

B. Infrared absorption spectrophotometry (*2.2.24*).

*Comparison*: *acetazolamide CRS*.

If the spectra obtained in the solid state show differences, dissolve the substance to be examined and the reference substance separately in *ethanol (96 per cent) R*, evaporate to dryness and record new spectra using the residues.

C. Introduce about 20 mg into a test-tube and add 4 mL of *dilute hydrochloric acid R* and 0.2 g of *zinc powder R*. Immediately place a piece of *lead acetate paper R* over the mouth of the tube. The paper shows a brownish-black colour.

D. Dissolve about 25 mg in a mixture of 0.1 mL of *dilute sodium hydroxide solution R* and 5 mL of *water R*. Add 0.1 mL of *copper sulfate solution R*. A greenish-blue precipitate is formed.

TESTS

**Appearance of solution**. The solution is not more opalescent than reference suspension II (*2.2.1*) and not more intensely coloured than reference solution $Y_5$ or $BY_5$ (*2.2.2, Method II*).

Dissolve 1.0 g in 10 mL of *1 M sodium hydroxide*.

**Related substances**. Liquid chromatography (*2.2.29*).

*Test solution*. Dissolve 40 mg of the substance to be examined in the mobile phase and dilute to 100.0 mL with the mobile phase.

*Reference solution (a)*. Dilute 1.0 mL of the test solution to 100.0 mL with the mobile phase. Dilute 1.0 mL of this solution to 10.0 mL with the mobile phase.

*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

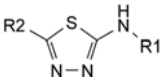
Dissolve 0.200 g in 25 mL of *dimethylformamide R*. Titrate with *0.1 M ethanolic sodium hydroxide*, determining the end-point potentiometrically (*2.2.20*).

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.*



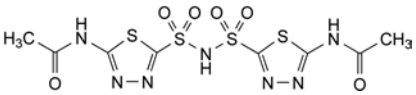
A. R1 = CO-$CH_3$, R2 = Cl: *N*-(5-chloro-1,3,4-thiadiazol-2-yl)acetamide,

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

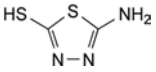
C. R1 = CO-$CH_3$, R2 = SH: *N*-(5-sulfanyl-1,3,4-thiadiazol-2-yl)acetamide,

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,

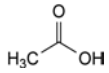


G. 5-amino-1,3,4-thiadiazole-2-thiol.

**01/2008:0590**

# 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.