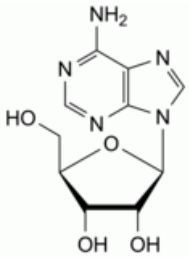
Edition: BP 2025 (Ph. Eur. 11.6 update)

Adenosine

General Notices

(Ph. Eur. monograph 1486)



$C_{10}H_{13}N_5O_4$ 267.2 *58-61-7*

Action and use

Antiarrhythmic.

Ph Eur

DEFINITION

9-β-D-Ribofuranosyl-9*H*-purin-6-amine.

Content

99.0 per cent to 101.0 per cent (dried substance).

CHARACTERS

Appearance

White or almost white, crystalline powder.

Solubility

Slightly soluble in water, soluble in hot water, practically insoluble in ethanol (96 per cent) and in methylene chloride. It dissolves in dilute mineral acids.

mp

About 234 °C.

IDENTIFICATION

Infrared absorption spectrophotometry (*2.2.24*).

Comparison *adenosine CRS*.

TESTS

Solution S

Suspend 5.0 g in 100 mL of *distilled water R* and heat to boiling. Allow to cool, filter with the aid of vacuum and dilute to 100 mL with *distilled water R*.

Appearance of solution

Solution S is colourless (*2.2.2, Method II*).

Acidity or alkalinity

To 10 mL of solution S, add 0.1 mL of *bromocresol purple solution R* and 0.1 mL of *0.01 M hydrochloric acid*. The solution is yellow. Add 0.4 mL of *0.01 M sodium hydroxide*. The solution is violet-blue.

Specific optical rotation (*2.2.7*)

-45 to -49 (dried substance).

Dissolve 1.25 g in *1 M hydrochloric acid* and dilute to 50.0 mL with the same acid. Examine within 10 min of preparing the solution.

Related substances

Liquid chromatography (*2.2.29*).

Solvent mixture Dissolve 6.8 g of *potassium hydrogen sulfate R* and 3.4 g of *tetrabutylammonium hydrogen sulfate R* in *water R*, adjust to pH 6.5 with a 60 g/L solution of *potassium hydroxide R* and dilute to 1000 mL with the same solvent. Use a freshly prepared solvent mixture.

Test solution Dissolve 20 mg of the substance to be examined in the mobile phase and dilute to 20 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 5 mg of *adenine R* (impurity A) and 5 mg of *inosine R* (impurity G) in the mobile phase and dilute to 50 mL with the mobile phase. Dilute 4 mL of this solution to 100 mL with the mobile phase.

Column:

— *size*: *l* = 0.25 m, Ø = 4.6 mm;

— *stationary phase*: *end-capped octadecylsilyl silica gel for chromatography R* (5 µm).

Mobile phase *water R*, solvent mixture (40:60 *V/V*).

Flow rate 1.5 mL/min.

Detection Spectrophotometer at 254 nm.

Injection 20 µL.

Run time 1.5 times the retention time of adenosine.

Relative retention With reference to adenosine (retention time = about 13 min): impurity A = about 0.3; impurity G = about 0.4.

System suitability Reference solution (b):

— *resolution*: minimum 1.5 between the peaks due to impurities A and G.

Limits:

— *correction factors*: for the calculation of content, multiply the peak areas of the following impurities by the corresponding correction factor: impurity A = 0.6; impurity G = 1.4;

— *impurity A*: not more than twice the area of the principal peak in the chromatogram obtained with reference solution (a) (0.2 per cent);

— *impurity G*: not more than the area of the principal peak in the chromatogram obtained with reference solution (a) (0.1 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 5 times the area of the principal peak in the chromatogram obtained with reference solution (a) (0.5 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).

Chlorides (*2.4.4*)

Maximum 100 ppm.

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

Sulfates (*2.4.13*)

Maximum 200 ppm, determined on solution S.

Ammonium (*2.4.1, Method B*)

Maximum 10 ppm, determined on 0.5 g.

Prepare the standard using 5 mL of *ammonium standard solution (1 ppm NH_4) 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

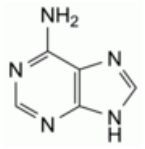
Dissolve 0.200 g, warming slightly if necessary, in a mixture of 20 mL of *acetic anhydride R* and 30 mL of *anhydrous acetic acid R*. Titrate with *0.1 M perchloric acid*, determining the end-point potentiometrically (*2.2.20*).

1 mL of *0.1 M perchloric acid* is equivalent to 26.72 mg of $C_{10}H_{13}N_5O_4$.

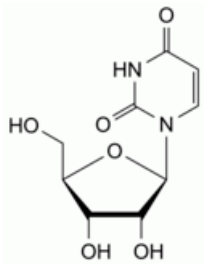
IMPURITIES

Specified impurities *A, G.*

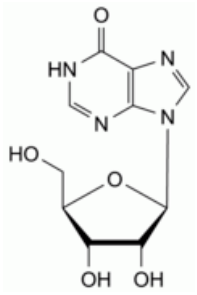
Other detectable impurities (the following substances would, if present at a sufficient level, be detected by one or other of the tests in the monograph. They are limited by the general acceptance criterion for other/unspecified impurities and/or by the general monograph Substances for pharmaceutical 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) F, H.



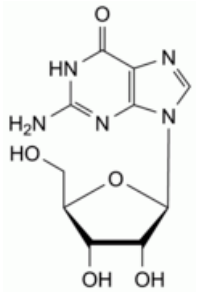
A. 7*H*-purin-6-amine (adenine),



F. 1-β-D-ribofuranosylpyrimidine-2,4(1*H*,3*H*)-dione (uridine),



G. 9-β-D-ribofuranosyl-1,9-dihydro-6*H*-purin-6-one (inosine),



H. 2-amino-9-β-D-ribofuranosyl-1,9-dihydro-6*H*-purin-6-one (guanosine).

Ph Eur