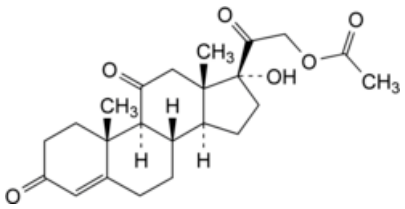
Edition: BP 2025 (Ph. Eur. 11.6 update)

Cortisone Acetate

General Notices

(Ph. Eur. monograph 0321)



$C_{23}H_{30}O_6$ 402.5 *50-04-4*

Action and use

Corticosteroid.

Preparation

Cortisone Tablets

Ph Eur

DEFINITION

17-Hydroxy-3,11,20-trioxopregn-4-en-21-yl acetate.

Content

97.0 per cent to 103.0 per cent (dried substance).

CHARACTERS

Appearance

White or almost white, crystalline powder.

Solubility

Practically insoluble in water, freely soluble in methylene chloride, soluble in dioxan, sparingly soluble in acetone, slightly soluble in ethanol (96 per cent) and in methanol.

It shows polymorphism (*5.9*).

IDENTIFICATION

First identification: A, B.

Second identification: C, D, E.

A. Infrared absorption spectrophotometry (2.2.24).

Comparison *cortisone acetate CRS*.

If the spectra obtained in the solid state show differences, record new spectra using 50 g/L solutions in *methylene chloride R* in a 0.2 mm cell.

B. Thin-layer chromatography (2.2.27).

Solvent mixture *methanol R*, *methylene chloride R* (1:9 *V/V*).

Test solution Dissolve 10 mg of the substance to be examined in the solvent mixture and dilute to 10 mL with the solvent mixture.

Reference solution (a) Dissolve 20 mg of *cortisone acetate CRS* in the solvent mixture and dilute to 20 mL with the solvent mixture.

Reference solution (b) Dissolve 10 mg of *hydrocortisone acetate R* in reference solution (a) and dilute to 10 mL with reference solution (a).

Plate *TLC silica gel F_{254} plate R*.

Mobile phase Add a mixture of 1.2 volumes of *water R* and 8 volumes of *methanol R* to a mixture of 15 volumes of *ether R* and 77 volumes of *methylene chloride R*.

Application 5 µL.

Development Over a path of 15 cm.

Drying In air.

Detection A Examine in ultraviolet light at 254 nm.

Results A The principal spot in the chromatogram obtained with the test solution is similar in position and size to the principal spot in the chromatogram obtained with reference solution (a).

Detection B Spray with *alcoholic solution of sulfuric acid R*. Heat at 120 °C for 10 min or until the spots appear. Allow to cool. Examine in daylight and in ultraviolet light at 365 nm.

Results B The principal spot in the chromatogram obtained with the test solution is similar in position, colour in daylight, fluorescence in ultraviolet light at 365 nm and size to the principal spot in the chromatogram obtained with reference solution (a).

System suitability Reference solution (b):

— the chromatogram shows 2 clearly separated spots.

C. Thin-layer chromatography (2.2.27).

Test solution (a) Dissolve 25 mg of the substance to be examined in *methanol R* with gentle heating and dilute to 5 mL with the same solvent (solution A). Dilute 2 mL of this solution to 10 mL with *methylene chloride R*.

Test solution (b) Transfer 2 mL of solution A to a 15 mL glass tube with a ground-glass stopper or a polytetrafluoroethylene cap. Add 10 mL of *saturated methanolic potassium hydrogen carbonate solution R* and immediately pass a stream of *nitrogen R* briskly through the solution for 5 min. Stopper the tube. Heat in a water-bath at 45 °C protected from light for 2.5 h. Allow to cool.

Reference solution (a) Dissolve 25 mg of *cortisone acetate CRS* in *methanol R* with gentle heating and dilute to 5 mL with the same solvent (solution B). Dilute 2 mL of this solution to 10 mL with *methylene chloride R*.

Reference solution (b) Transfer 2 mL of solution B to a 15 mL glass tube with a ground-glass stopper or a polytetrafluoroethylene cap. Add 10 mL of *saturated methanolic potassium hydrogen carbonate solution R* and

immediately pass a stream of *nitrogen R* briskly through the solution for 5 min. Stopper the tube. Heat in a water-bath at 45 °C protected from light for 2.5 h. Allow to cool.

Plate *TLC silica gel F_{254} plate R*.

Mobile phase Add a mixture of 1.2 volumes of *water R* and 8 volumes of *methanol R* to a mixture of 15 volumes of *ether R* and 77 volumes of *methylene chloride R*.

Application 5 µL.

Development Over a path of 15 cm.

Drying In air.

Detection A Examine in ultraviolet light at 254 nm.

Results A The principal spot in each of the chromatograms obtained with the test solutions is similar in position and size to the principal spot in the chromatogram obtained with the corresponding reference solution.

Detection B Spray with *alcoholic solution of sulfuric acid R* and heat at 120 °C for 10 min or until the spots appear. Allow to cool. Examine in daylight and in ultraviolet light at 365 nm.

Results B The principal spot in each of the chromatograms obtained with the test solutions is similar in position, colour in daylight, fluorescence in ultraviolet light at 365 nm and size to the principal spot in the chromatogram obtained with the corresponding reference solution. The principal spots in the chromatograms obtained with test solution (b) and reference solution (b) have an R_F value distinctly lower than that of the principal spots in the chromatograms obtained with test solution (a) and reference solution (a).

D. Add about 2 mg to 2 mL of *sulfuric acid R* and shake to dissolve. Within 5 min, a faint yellow colour develops. Add this solution to 10 mL of *water R* and mix. The colour is discharged and a clear solution remains.
E. About 10 mg gives the reaction of acetyl (*2.3.1*).

TESTS

Specific optical rotation (*2.2.7*)

+ 211 to + 220 (dried substance).

Dissolve 0.250 g in *dioxan R* and dilute to 25.0 mL with the same solvent.

Related substances

Liquid chromatography (*2.2.29*). *Prepare the solutions immediately before use.*

Test solution Dissolve 25.0 mg of the substance to be examined in *acetonitrile R* and dilute to 10.0 mL with the same solvent.

Reference solution (a) Dissolve 2 mg of *cortisone acetate CRS* and 2 mg of *hydrocortisone acetate CRS* (impurity A) in *acetonitrile R* and dilute to 100.0 mL with the same solvent.

Reference solution (b) Dilute 1.0 mL of the test solution to 100.0 mL with *acetonitrile R*.

Column:

— *size*: *l* = 0.25 m, Ø = 4.6 mm;

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

Mobile phase In a 1000 mL volumetric flask mix 400 mL of *acetonitrile R* with 550 mL of *water R* and allow to equilibrate; dilute to 1000 mL with *water R* and mix again.

Flow rate 1 mL/min.

Detection Spectrophotometer at 254 nm.

Equilibration With the mobile phase for about 30 min.

Injection 20 µL; inject *acetonitrile R* as a blank.

Run time Twice the retention time of cortisone acetate.

Retention time Impurity A = about 10 min; cortisone acetate = about 12 min.

System suitability Reference solution (a):

— *resolution*: minimum 4.2 between the peaks due to impurity A and cortisone acetate; if necessary, adjust the concentration of acetonitrile in the mobile phase.

Limits:

— *impurity A*: not more than 0.5 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.5 per cent);

— *total*: not more than 1.5 times the area of the principal peak in the chromatogram obtained with reference solution (b) (1.5 per cent);

— *disregard limit*: 0.05 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.05 per cent).

Loss on drying (*2.2.32*)

Maximum 0.5 per cent, determined on 0.500 g by drying in an oven at 105 °C.

ASSAY

Dissolve 0.100 g in *ethanol (96 per cent) R* and dilute to 100.0 mL with the same solvent. Dilute 2.0 mL of this solution to 100.0 mL with *ethanol (96 per cent) R*. Measure the absorbance (*2.2.25*) at the absorption maximum at 237 nm.

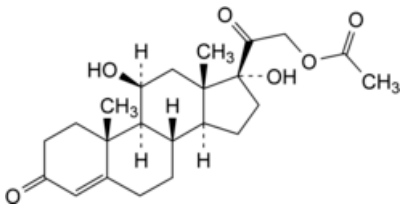
Calculate the content of $C_{23}H_{30}O_6$ taking the specific absorbance to be 395.

STORAGE

Protected from light.

IMPURITIES

Specified impurities *A.*



A. 11β,17-dihydroxy-3,20-dioxopregn-4-en-21-yl acetate (hydrocortisone acetate).