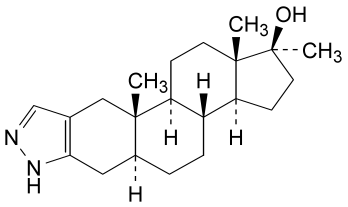
01/2008:1568

STANOZOLOL

Stanozololum



$C_{21}H_{32}N_2O$ M_r 328.5
[10418-03-8]

DEFINITION

Stanozolol contains not less than 98.5 per cent and not more than the equivalent of 101.0 per cent of 17-methyl-2′*H*-5α-androst-2-eno[3,2-*c*]pyrazol-17β-ol, calculated with reference to the dried substance.

CHARACTERS

A white or almost white crystalline powder, hygroscopic, practically insoluble in water, soluble in dimethylformamide, slightly soluble in ethanol (96 per cent), very slightly soluble in methylene chloride.

It shows polymorphism (*5.9*).

IDENTIFICATION

A. Examine by infrared spectrophotometry (*2.2.24*), comparing with the spectrum obtained with *stanozolol CRS*. If the spectra obtained in the solid state show differences, dissolve the substance to be examined and the reference substance separately in the minimum volume of methylene chloride, evaporate to dryness at room temperature under an air-stream and record new spectra using the residues.

B. Examine the chromatograms obtained in the test for related substances. The principal spot in the chromatogram obtained with test solution (b) is similar in position, colour and size to the principal spot in the chromatogram obtained with reference solution (c).

TESTS

Specific optical rotation (*2.2.7*). Dissolve 0.10 g in *chloroform R* and dilute to 10.0 ml with the same solvent. The specific optical rotation is + 34 to + 40, calculated with reference to the dried substance.

Related substances. Examine by thin-layer chromatography (*2.2.27*), using a *TLC silica gel F_{254} plate R*.

Test solution (a). Dissolve 0.10 g of the substance to be examined in a mixture of 1 volume of *methanol R* and 9 volumes of *methylene chloride R* and dilute to 5 ml with the same mixture of solvents.

Test solution (b). Dilute 1 ml of test solution (a) to 10 ml with a mixture of 1 volume of *methanol R* and 9 volumes of *methylene chloride R*.

Reference solution (a). Dilute 1.0 ml of test solution (a) to 200 ml with a mixture of 1 volume of *methanol R* and 9 volumes of *methylene chloride R*.

Reference solution (b). Dissolve 5 mg of *stanozolol impurity A CRS* in reference solution (a) and dilute to 50 ml with the same solution.

Reference solution (c). Dissolve 10 mg of *stanozolol CRS* in a mixture of 1 volume of *methanol R* and 9 volumes of *methylene chloride R* and dilute to 5 ml with the same mixture of solvents.

Apply to the plate 5 µl of each solution. Develop over a path corresponding to two thirds of the plate height using a mixture of 10 volumes of *methanol R* and 90 volumes of *methylene chloride R*. Allow the plate to dry in air, spray with *alcoholic solution of sulphuric acid R*, heat at 105 °C for 15 min and examine under ultraviolet light at 365 nm. Any secondary spot in the chromatogram obtained with test solution (a) is not more intense than the spot in the chromatogram obtained with reference solution (a) (0.5 per cent). The test is not valid unless the chromatogram obtained with reference solution (b) shows two clearly separated spots.

Loss on drying (*2.2.32*). Not more than 1.0 per cent, determined on 1.000 g by drying at 100 °C at a pressure not exceeding 0.7 kPa.

ASSAY

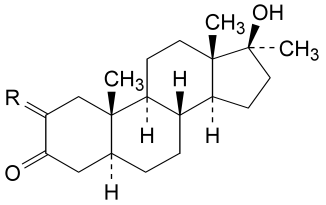
Dissolve 0.250 g in 50 ml of *anhydrous acetic acid R*. Titrate with *0.1 M perchloric acid*, determining the end-point potentiometrically (*2.2.20*).

1 ml of *0.1 M perchloric acid* is equivalent to 32.85 mg of $C_{21}H_{32}N_2O$.

STORAGE

Store in an airtight container, protected from light.

IMPURITIES



A. R = H_2: 17β-hydroxy-17-methyl-5α-androstan-3-one (mestanolone),

B. R = CH-OH: 17β-hydroxy-2-(hydroxymethylene)-17-methyl-5α-androstan-3-one (oxymetholone).

01/2009:1267

STARCH, PREGELATINISED

Amylum pregelificatum

DEFINITION

Pregelatinised starch is prepared from *Maize starch (0344)*, *Potato starch (0355)* or *Rice starch (0349)* by mechanical processing in the presence of water, with or without heat, to rupture all or part of the starch granules, and subsequent drying. It contains no added substances but it may be modified to render it compressible and to improve its flow characteristics.

CHARACTERS

Appearance: white or yellowish-white powder.

It swells in cold water.

IDENTIFICATION

A. Examined under a microscope using a mixture of equal volumes of *glycerol R* and *water R* it presents irregular, translucent, white or yellowish-white flakes or pieces

with an uneven surface. Under polarised light (between crossed nicol prisms), starch granules with a distinct black cross intersecting at the hilum may be seen.

B. Disperse 0.5 g in 2 ml of *water R* without heating and add 0.05 ml of *iodine solution R1*. A reddish-violet to blue colour is produced.

TESTS

pH (*2.2.3*): 4.5 to 7.0.

Progressively add 3.0 g to 100.0 ml of *carbon dioxide-free water R*, stirring continuously. Determine the pH when a homogeneous solution is obtained.

Oxidising substances (*2.5.30*). It complies with the test for oxidising substances. Use a mixture of equal volumes of *water R* and *methanol R* as solvent.

Sulphur dioxide (*2.5.29*): maximum 50 ppm.

Iron (*2.4.9*): maximum 20 ppm.

Dissolve the residue obtained in the test for sulphated ash in 20 ml of *dilute hydrochloric acid R*. Filter. The filtrate complies with the test.

Foreign matter. Examined under a microscope using a mixture of equal volumes of *glycerol R* and *water R*, not more than traces of matter other than starch granules are present.

Loss on drying (*2.2.32*): maximum 15.0 per cent, determined on 1.000 g by drying in an oven at 130 °C for 90 min.

Sulphated ash (*2.4.14*): maximum 0.6 per cent, determined on 1.0 g.

Microbial contamination

TAMC: acceptance criterion 10^3 CFU/g (*2.6.12*).

TYMC: acceptance criterion 10^2 CFU/g (*2.6.12*).

Absence of *Escherichia coli* (*2.6.13*).

Absence of *Salmonella* (*2.6.13*).

LABELLING

The type of starch used as starting material is stated.

07/2008:1874
corrected 6.3

ST. JOHN'S WORT DRY EXTRACT, QUANTIFIED

Hyperici herbae extractum siccum quantificatum

DEFINITION

Quantified dry extract obtained from *St. John's wort (1438)*.

Content:

– *total hypericins, expressed as hypericin* ($C_{30}H_{16}O_8$; M_r 504.5): 0.10 per cent to 0.30 per cent (dried extract);
– *flavonoids, expressed as rutin* ($C_{27}H_{30}O_{16}$; M_r 610.5): minimum 6.0 per cent (dried extract);
– *hyperforin* ($C_{35}H_{52}O_4$; M_r 536.8): maximum 6.0 per cent (dried extract) and not more than the content stated on the label.

PRODUCTION

The extract is produced from the herbal drug by a suitable procedure using ethanol (50-80 per cent *V/V*) or methanol (50-80 per cent *V/V*).

CHARACTERS

Appearance: brownish-grey powder.

IDENTIFICATION

Thin-layer chromatography (*2.2.27*).

Test solution. Disperse 0.25 g of the extract to be examined in 5 ml of *methanol R*.

Reference solution. Dissolve 5 mg of *rutin R* and 5 mg of *hyperoside R* in *methanol R* and dilute to 10 ml with the same solvent.

Plate: *TLC silica gel plate R* (5-40 µm) [or *TLC silica gel plate R* (2-10 µm)].

Mobile phase: *anhydrous formic acid R, water R, ethyl acetate R* (6:9:90 *V/V/V*).

Application: 10 µl [or 5 µl] as bands of 10 mm [or 8 mm].

Development: over a path of 10 cm [or 7.5 cm] .

Drying: at 100-105 °C for 10 min.

Detection: spray with a 10 g/l solution of *diphenylboric acid aminoethyl ester R* in *methanol R* and then with a 50 g/l solution of *macrogol 400 R* in *methanol R*. Examine after about 30 min in ultraviolet light at 365 nm.

Results: see below the sequence of the zones present in the chromatograms obtained with the reference solution and the test solution. Furthermore, other fluorescent zones may be present in the chromatogram obtained with the test solution.

Top of the plate	
	A yellowish-orange fluorescent zone
	2 red fluorescent zones (hypericin and pseudohypericin)
	3 yellowish-orange fluorescent zones
Hyperoside: a yellowish-orange fluorescent zone	A yellowish-orange fluorescent zone (hyperoside)
	Yellow and blue possibly superimposed fluorescent zones
Rutin: a yellowish-orange fluorescent zone	A yellowish-orange fluorescent zone (rutin)
Reference solution	**Test solution**

ASSAY

Total hypericins. Liquid chromatography (*2.2.29*).

Test solution. Dissolve 70.0 mg of the extract to be examined in 25.0 ml of *methanol R*. Sonicate and centrifuge the solution. Expose the solution to a xenon lamp at about 765 W/m^2 for 8 min.

Reference solution. Dissolve a quantity of *St. John's wort standardised dry extract CRS* corresponding to 0.15 mg of hypericin in 25.0 ml of *methanol R*. Sonicate and centrifuge. Expose the solution to a xenon lamp at about 765 W/m^2 for 8 min.

Column:

– *size*: *l* = 0.15 m, Ø = 4.6 mm;
– *stationary phase*: *octadecylsilyl silica gel for chromatography R* (5 µm);
– *temperature*: 40 °C.

Mobile phase: mix 39 volumes of *ethyl acetate R*, 41 volumes of a 15.6 g/l solution of *sodium dihydrogen phosphate R* adjusted to pH 2 with *phosphoric acid R* and 160 volumes of *methanol R*.

Flow rate: 1.0 ml/min.