Reference solution (b). Dilute 1.0 ml of the test solution to 100.0 ml with the mobile phase.

Column:

– *size*: *l* = 0.25 m, Ø = 4.6 mm,

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

Mobile phase: *acetonitrile R, water R* (50:50 *V/V*).

Flow rate: 1 ml/min.

Detection: spectrophotometer at 210 nm.

Injection: 20 µl.

Run time: 3 times the retention time of the principal peak.

System suitability: reference solution (a):

– *resolution*: minimum 2.0 between the peaks due to glyceryl trinitrate and to pentaerythrityl tetranitrate.

Limits:

– *any impurity*: not more than the area of the principal peak in the chromatogram obtained with reference solution (b) (1 per cent, calculated as glyceryl trinitrate),

– *total*: not more than 3 times the area of the principal peak in the chromatogram obtained with reference solution (b) (3 per cent, calculated as glyceryl trinitrate),

– *disregard limit*: 0.1 times the area of the principal peak in the chromatogram obtained with reference solution (b) (0.1 per cent).

ASSAY

Test solution. Prepare a solution containing 1.0 mg of glyceryl trinitrate in 250.0 ml of *methanol R*.

Reference solution. Dissolve 70.0 mg of *sodium nitrite R* in *methanol R* and dilute to 250.0 ml with the same solvent. Dilute 5.0 ml of the solution to 500.0 ml with *methanol R*.

Into three 50 ml volumetric flasks introduce 10.0 ml of the test solution, 10.0 ml of the reference solution and 10 ml of *methanol R* as a blank. To each flask add 5 ml of *dilute sodium hydroxide solution R*, close the flask, mix and allow to stand at room temperature for 30 min. Add 10 ml of *sulphanilic acid solution R* and 10 ml of *dilute hydrochloric acid R* and mix. After exactly 4 min, add 10 ml of *naphthylethylenediamine dihydrochloride solution R*, dilute to volume with *water R* and mix. After 10 min read the absorbance (*2.2.25*) of the test solution and the reference solution at 540 nm using the blank solution as the compensation liquid.

Calculate the amount of glyceryl trinitrate in milligrams in the test solution from the following expression:

$$\frac{A_T \times m_S \times C}{A_R \times m_T \times 60.8 \times 100}$$

A_T = absorption of the test solution,

m_T = mass of the substance to be examined, in milligrams,

C = percentage content of sodium nitrite used as reference,

A_R = absorption of the reference solution,

m_S = mass of sodium nitrite, in milligrams.

STORAGE

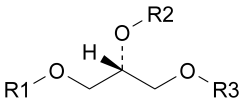
Store diluted solutions (10 g/l) protected from light, at a temperature of 2 °C to 15 °C. Store more concentrated solutions protected from light, at a temperature of 15 °C to 20 °C.

LABELLING

The label states the declared content of glyceryl trinitrate.

IMPURITIES

A. inorganic nitrates,



and enantiomer

B. R1 = NO_2, R2 = R3 = H: (2*RS*)-2,3-dihydroxypropyl nitrate,

C. R1 = R3 = H, R2 = NO_2: 2-hydroxy-1-(hydroxymethyl)ethyl nitrate,

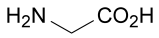
D. R1 = R2 = NO_2, R3 = H: (2*RS*)-3-hydroxypropane-1,2-diyl dinitrate,

E. R1 = R3 = NO_2, R2 = H: 2-hydroxypropane-1,3-diyl dinitrate.

01/2008:0614
corrected 6.0

GLYCINE

Glycinum



$C_2H_5NO_2$ M_r 75.1
[56-40-6]

DEFINITION

2-Aminoacetic acid.

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

CHARACTERS

Appearance: white or almost white, crystalline powder.

Solubility: freely soluble in water, very slightly soluble in ethanol (96 per cent).

It shows polymorphism (*5.9*).

IDENTIFICATION

First identification: A.

Second identification: B, C.

A. Infrared absorption spectrophotometry (*2.2.24*).

Comparison: *glycine 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 *ethanol (60 per cent V/V) R*, evaporate to dryness and record the spectra again.

B. Examine the chromatograms obtained in the test for ninhydrin-positive substances.

Results: 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 (a).

C. Dissolve 50 mg in 5 ml of *water R*, add 1 ml of *strong sodium hypochlorite solution R* and boil for 2 min. Add 1 ml of *hydrochloric acid R* and boil for 4-5 min. Add 2 ml of *hydrochloric acid R* and 1 ml of a 20 g/l solution of *resorcinol R*, boil for 1 min and cool. Add 10 ml of *water R* and mix. To 5 ml of the solution add 6 ml of *dilute sodium hydroxide solution R*. The solution is violet with greenish-yellow fluorescence. After a few minutes, the colour becomes orange and then yellow and an intense fluorescence remains.

TESTS

Solution S. Dissolve 5.0 g in *carbon dioxide-free water R* and dilute to 50 ml with the same solvent.

Appearance of solution. Solution S is clear (*2.2.1*) and not more intensely coloured than reference solution Y_7 (*2.2.2, Method II*).

pH (*2.2.3*): 5.9 to 6.4.

Dilute 10 ml of solution S to 20 ml with *carbon dioxide-free water R*.

Ninhydrin-positive substances. Thin-layer chromatography (*2.2.27*).

Test solution (a). Dissolve 0.10 g of the substance to be examined in *water R* and dilute to 10.0 ml with the same solvent.

Test solution (b). Dilute 1.0 ml of test solution (a) to 10.0 ml with *water R*.

Reference solution (a). Dissolve 10 mg of *glycine CRS* in *water R* and dilute to 10.0 ml with the same solvent.

Reference solution (b). Dilute 1.0 ml of test solution (a) to 200 ml with *water R*.

Reference solution (c). Dissolve 10 mg of *glycine CRS* and 10 mg of *alanine CRS* in *water R* and dilute to 25 ml with the same solvent.

Plate: *cellulose for chromatography R* as the coating substance.

Mobile phase: *glacial acetic acid R, water R, butanol R* (20:20:60 *V/V/V*).

Application: 5 µl.

Development: over 2/3 of the plate.

Drying: at 80 °C for 30 min.

Detection: spray with *ninhydrin solution R* and dry at 100-105 °C for 15 min.

System suitability: the chromatogram obtained with reference solution (c) shows 2 clearly separated spots.

Limits: in the chromatogram obtained with test solution (a):

– *any impurity*: any spots, apart from the principal spot, are not more intense than the principal spot in the chromatogram obtained with reference solution (b) (0.5 per cent).

Chlorides (*2.4.4*): maximum 75 ppm.

Dissolve 0.67 g in *water R* and dilute to 15 ml with the same solvent.

Heavy metals (*2.4.8*): maximum 10 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 0.5 per cent, determined on 1.000 g by drying in an oven at 105 °C for 2 h.

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

ASSAY

Dissolve 70.0 mg in 3 ml of *anhydrous formic acid R* and add 30 ml of *anhydrous acetic acid R*. Immediately after dissolution, 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 7.51 mg of $C_2H_5NO_2$.

01/2008:1892
corrected 6.0

GOLDENROD

Solidaginis herba

DEFINITION

Whole or cut, dried, flowering aerial parts of *Solidago gigantea* Ait or *Solidago canadensis* L., their varieties or hybrids and/or mixtures of these.

Content: minimum 2.5 per cent of flavonoids, expressed as hyperoside ($C_{21}H_{20}O_{12}$; M_r 464.4) (dried drug).

IDENTIFICATION

A. The stems are greenish-yellow or greenish-brown, partly tinted reddish, roundish, more or less conspicuously grooved, glabrous and smooth in the lower part, slightly or densely pubescent in the upper part. They are solid with a whitish pith.

The leaves are green, sessile, lanceolate, with a serrate margin, 8-12 cm long and about 1-3 cm wide, the upper surface is green and more or less glabrous, the lower surface is greyish-green and pubescent, especially on the veins. The inflorescence consists of a number of unilateral, curved racemes which together form a pyramidal panicle at the end of the stems.

Each capitulum has an involucre composed of linear-lanceolate, imbricated yellowish-green bracts, surrounding a single row of yellow ligulate florets about the same length as the involucre; yellow, radially arranged tubular florets, as long as, or longer, than the ligulate florets; a brownish inferior ovary surmounted by a white pappus of silky hairs.

B. Reduce to a powder (355) (*2.9.12*). The powder is greyish-green. Examine under a microscope using *chloral hydrate solution R*. The powder shows pappus bristles and their fragments, consisting of multiseriate trichomes composed of elongated cells with the tips free from the surface and forming pointed projections over the entire length; fragments of the leaf mesophyll with vascular bundles accompanied by secretory cells; fragments of the leaf epidermis with sinuous to wavy-walled cells and stomata of the anomocytic type (*2.8.3*); uniseriate covering trichomes with up to 5 or 6 cells, some whip-like with a thicker-walled terminal cell; fragments of the style with long, slender papillae; fragments of the stem with reticulate and spiral vessels; pollen grains, with 3 germinal pores and a spiny exine; numerous whisk-shaped hairs, a few isolated twin-hairs from the ovary, absence of multicellular trichomes with a terminal cell bent at a right angle.

C. Thin-layer chromatography (*2.2.27*).

Test solution. To 0.75 g of the powdered drug (355) (*2.9.12*) add 5 ml of *methanol R* and boil in a water-bath under a reflux condenser for 10 min. Cool and filter.