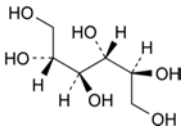
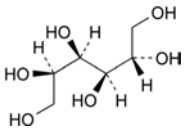


D. galactitol (dulcitol),

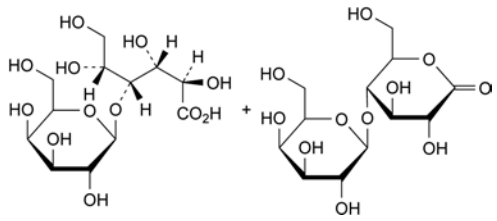


E. D-glucitol (D-sorbitol).

01/2008:1647

LACTOBIONIC ACID

Acidum lactobionicum



$C_{12}H_{22}O_{12}$ (acid form) M_r 358.3
[96-82-2]

$C_{12}H_{20}O_{11}$ (δ-lactone) M_r 340.3
[5965-65-1]

DEFINITION

Mixture in variable proportions of 4-*O*-β-D-galactopyranosyl-D-gluconic acid and 4-*O*-β-D-galactopyranosyl-D-glucono-1,5-lactone.

Content: 98.0 per cent to 102.0 per cent (anhydrous substance).

CHARACTERS

Appearance: white or almost white powder.

Solubility: freely soluble in water, slightly soluble in glacial acetic acid, in anhydrous ethanol and in methanol.

mp: about 125 °C with decomposition.

IDENTIFICATION

A. Infrared absorption spectrophotometry (*2.2.24*).

Comparison: *lactobionic acid CRS*.

If the spectra obtained show differences, dissolve the substance to be examined and the reference substance separately in *water R*, dry at 105 °C and record new spectra using the residues.

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

Test solution. Dissolve 10 mg of the substance to be examined in *water R* and dilute to 1 mL with the same solvent.

Reference solution. Dissolve 10 mg of *lactobionic acid CRS* in *water R* and dilute to 1 mL with the same solvent.

Plate: *TLC silica gel plate R*.

Mobile phase: *concentrated ammonia R1, ethyl acetate R, water R, methanol R* (2:2:2:4 *V/V/V/V*).

Application: 5 µL.

Development: over 3/4 of the plate.

Detection: spray 3 times with *ammonium molybdate solution R6* and heat in an oven at 110 °C for 15 min.

Results: the principal spot in the chromatogram obtained with the test solution is similar in position and colour to the principal spot in the chromatogram obtained with the reference solution.

TESTS

Appearance of solution. The solution is clear (*2.2.1*) and not more intensely coloured than reference solution Y_5 (*2.2.2, Method II*).

Dissolve 3.0 g in 25 mL of *water R*.

Specific optical rotation (*2.2.7*): + 23.0 to + 29.0 (anhydrous substance).

Dissolve 1.0 g in 80 mL of *water R* and dilute to 100.0 mL with the same solvent. Allow to stand for 24 h.

Reducing sugars: maximum 0.2 per cent, calculated as glucose.

Dissolve 5.0 g in 25 mL of *water R* with the aid of gentle heat. Cool and add 20 mL of *cupri-citric solution R* and a few glass beads. Heat so that boiling begins after 4 min and maintain boiling for 3 min. Cool rapidly and add 100 mL of a 2.4 per cent *V/V* solution of *glacial acetic acid R* and 20.0 mL of *0.025 M iodine*. With continuous shaking, add 25 mL of a mixture of 6 volumes of *hydrochloric acid R* and 94 volumes of *water R* and, when the precipitate has dissolved, titrate the excess of iodine with *0.05 M sodium thiosulfate* using 1 mL of *starch solution R*, added towards the end of the titration, as indicator. Not less than 12.8 mL of *0.05 M sodium thiosulfate* is required.

Heavy metals (*2.4.8*): maximum 20 ppm.

1.0 g complies with limit test E. Prepare the reference solution using 2 mL of *lead standard solution (10 ppm Pb) R*.

Water (*2.5.12*): maximum 5.0 per cent, determined on 0.50 g.

Use a mixture of 1 volume of *formamide R* and 2 volumes of *methanol R* as solvent.

Total ash (*2.4.16*): maximum 0.2 per cent.

ASSAY

Dissolve 0.350 g in 50 mL of *carbon dioxide-free water R*, previously heated to 30 °C. Immediately titrate with *0.1 M sodium hydroxide* and determine the 2 equivalence points potentiometrically (*2.2.20*).

The first equivalence point (V_1) corresponds to the acid form of lactobionic acid and the second equivalence point ($V_2 - V_1$) corresponds to the δ-lactone form.

1 mL of *0.1 M sodium hydroxide* is equivalent to 35.83 mg of $C_{12}H_{22}O_{12}$.

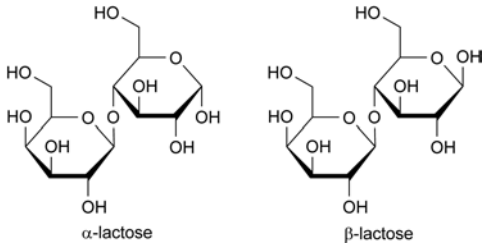
1 mL of *0.1 M sodium hydroxide* is equivalent to 34.03 mg of $C_{12}H_{20}O_{11}$.

The sum of the 2 results is expressed as a percentage content of lactobionic acid.

07/2009:1061

LACTOSE, ANHYDROUS

Lactosum anhydricum



$C_{12}H_{22}O_{11}$ M_r 342.3

DEFINITION

O-β-D-Galactopyranosyl-(1→4)-β-D-glucopyranose or mixture of *O*-β-D-galactopyranosyl-(1→4)-α-D-glucopyranose and *O*-β-D-galactopyranosyl-(1→4)-β-D-glucopyranose.

CHARACTERS

Appearance: white or almost white, crystalline powder.

Solubility: freely but slowly soluble in water, practically insoluble in ethanol (96 per cent).

IDENTIFICATION

First identification: A, D.

Second identification: B, C, D.

A. Infrared absorption spectrophotometry (*2.2.24*).

Comparison: *anhydrous lactose CRS.*

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

Solvent mixture: *water R, methanol R* (2:3 *V/V*).

Test solution. Dissolve 10 mg of the substance to be examined in the solvent mixture and dilute to 20 mL with the solvent mixture.

Reference solution (a). Dissolve 10 mg of *anhydrous lactose CRS* in the solvent mixture and dilute to 20 mL with the solvent mixture.

Reference solution (b). Dissolve 10 mg of *anhydrous lactose CRS*, 10 mg of *fructose CRS*, 10 mg of *glucose CRS* and 10 mg of *sucrose CRS* in the solvent mixture and dilute to 20 mL with the solvent mixture.

Plate: *TLC silica gel G plate R.*

Mobile phase: *water R, methanol R, glacial acetic acid R, ethylene chloride R* (10:15:25:50 *V/V/V/V*); measure the volumes accurately, as a slight excess of water produces cloudiness.

Application: 2 µL; thoroughly dry the points of application.

Development A: over a path of 15 cm.

Drying A: in a current of warm air.

Development B: immediately, over a path of 15 cm, after renewing the mobile phase.

Drying B: in a current of warm air.

Detection: spray with a solution of 0.5 g of *thymol R* in a mixture of 5 mL of *sulfuric acid R* and 95 mL of *ethanol (96 per cent) R*; heat at 130 °C for 10 min.

System suitability: reference solution (b):

– the chromatogram shows 4 clearly separated spots.

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

C. Dissolve 0.25 g in 5 mL of *water R*. Add 5 mL of *ammonia R* and heat in a water-bath at 80 °C for 10 min. A red colour develops.

D. Water (see Tests).

TESTS

Appearance of solution. The solution is clear (*2.2.1*) and not more intensely coloured than reference solution BY_7 (*2.2.2, Method II*).

Dissolve 1.0 g in boiling *water R* and dilute to 10 mL with the same solvent.

Acidity or alkalinity. Dissolve 6.0 g by heating in 25 mL of *carbon dioxide-free water R*, cool and add 0.3 mL of *phenolphthalein solution R1*. The solution is colourless. Not more than 0.4 mL of *0.1 M sodium hydroxide* is required to change the colour of the indicator to pink or red.

Specific optical rotation (*2.2.7*): + 54.4 to + 55.9 (anhydrous substance).

Dissolve 10.0 g in 80 mL of *water R*, heating to 50 °C. Allow to cool and add 0.2 mL of *dilute ammonia R1*. Allow to stand for 30 min and dilute to 100.0 mL with *water R*.

Absorbance (*2.2.25*).

Test solution (a). Dissolve 1.0 g in boiling *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*.

Spectral range: 400 nm for test solution (a) and 210-300 nm for test solution (b).

Results:

– at 400 nm: maximum 0.04 for test solution (a);

– from 210 nm to 220 nm: maximum 0.25 for test solution (b);

– from 270 nm to 300 nm: maximum 0.07 for test solution (b).

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

2.0 g complies with test C. Prepare the reference solution using 1.0 mL of *lead standard solution (10 ppm Pb) R*.

Water (*2.5.12*): maximum 1.0 per cent, determined on 0.50 g, using a mixture of 1 volume of *formamide R* and 2 volumes of *methanol R* as the solvent.

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

Microbial contamination

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

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

FUNCTIONALITY-RELATED CHARACTERISTICS

This section provides information on characteristics that are recognised as being relevant control parameters for one or more functions of the substance when used as an excipient (see chapter 5.15). This section is a non-mandatory part of the monograph and it is not necessary to verify the characteristics to demonstrate compliance. Control of these characteristics can however contribute to the quality of a medicinal product by improving the consistency of the manufacturing process and the performance of the medicinal product during use. Where control methods are cited, they are recognised as being suitable for the purpose, but other methods can also be used. Wherever results for a particular characteristic are reported, the control method must be indicated.

The following characteristics may be relevant for anhydrous lactose used as a filler/diluent in solid dosage forms (compressed and powder).

Particle size distribution (*2.9.31* or *2.9.38*).

Bulk and tapped density (*2.9.34*). Determine the bulk density and the tapped density. Calculate the Hausner index using the following expression:

$$\frac{V_0}{V_f}$$

V_0 = volume of bulk substance;

V_f = volume of tapped substance.

α-Lactose and β-lactose. Gas chromatography (*2.2.28*).

Silylation reagent. Mix 28 volumes of *N-trimethylsilyl-imidazole R* and 72 volumes of *pyridine R*.

Test solution. Dissolve about 1 mg of the substance to be examined in 0.45 mL of *dimethyl sulfoxide R*. Add 1.8 mL of the silylation reagent. Mix gently and allow to stand for 20 min.

Reference solution. Prepare a mixture of *α-lactose monohydrate R* and *β-lactose R* having an anomeric ratio of about 1:1 based on the labelled anomeric contents of the α-lactose monohydrate and β-lactose. Dissolve about 1 mg of this mixture in 0.45 mL of *dimethyl sulfoxide R*. Add 1.8 mL of the silylation reagent. Mix gently and allow to stand for 20 min.

Column:

– *material*: glass;

– *size*: *l* = 0.9 m, Ø = 4 mm;

– *stationary phase*: *silanised diatomaceous earth for gas chromatography R* impregnated with 3 per cent *m/m* of *poly[(cyanopropyl)(methyl)][(phenyl)(methyl)] siloxane R*.

Carrier gas: *helium for chromatography R.*

Flow rate: 40 mL/min.

Temperature:

– *column*: 215 °C;

– *injection port and detector*: 275 °C.

Detection: flame ionisation.

Injection: 2 µL.

System suitability: reference solution:

– *relative retention* with reference to β-lactose: α-lactose = about 0.7;

– *resolution*: minimum 3.0 between the peaks due to α-lactose and β-lactose.

Calculate the percentage content of α-lactose from the following expression:

$$\frac{100 S_a}{S_a + S_b}$$

Calculate the percentage content of β-lactose from the following expression:

$$\frac{100 S_b}{S_a + S_b}$$

S_a = area of the peak due to α-lactose;

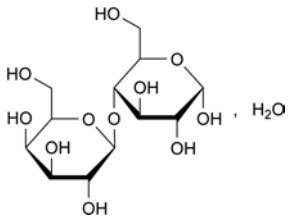
S_b = area of the peak due to β-lactose.

Loss on drying (*2.2.32*). Determine on 1.000 g by drying in an oven at 80 °C for 2 h.

07/2009:0187

LACTOSE MONOHYDRATE

Lactosum monohydricum



$C_{12}H_{22}O_{11},H_2O$ M_r 360.3

DEFINITION

O-β-D-Galactopyranosyl-(1→4)-α-D-glucopyranose monohydrate.

CHARACTERS

Appearance: white or almost white, crystalline powder.

Solubility: freely but slowly soluble in water, practically insoluble in ethanol (96 per cent).

IDENTIFICATION

First identification: *A, D.*

Second identification: *B, C, D.*

A. Infrared absorption spectrophotometry (*2.2.24*).

Comparison: *lactose CRS.*

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

Solvent mixture: *water R, methanol R* (2:3 *V/V*).

Test solution. Dissolve 10 mg of the substance to be examined in the solvent mixture and dilute to 20 mL with the solvent mixture.

Reference solution (a). Dissolve 10 mg of *lactose CRS* in the solvent mixture and dilute to 20 mL with the solvent mixture.

Reference solution (b). Dissolve 10 mg of *fructose CRS*, 10 mg of *glucose CRS*, 10 mg of *lactose CRS* and 10 mg of *sucrose CRS* in the solvent mixture and dilute to 20 mL with the solvent mixture.

Plate: *TLC silica gel G plate R.*

Mobile phase: *water R, methanol R, glacial acetic acid R, ethylene chloride R* (10:15:25:50 *V/V/V/V*); measure the volumes accurately, as a slight excess of water produces cloudiness.

Application: 2 µL; thoroughly dry the points of application.

Development A: over a path of 15 cm.

Drying A: in a current of warm air.

Development B: immediately, over a path of 15 cm, after renewing the mobile phase.

Drying B: in a current of warm air.

Detection: spray with a solution of 0.5 g of *thymol R* in a mixture of 5 mL of *sulfuric acid R* and 95 mL of *ethanol (96 per cent) R*; heat at 130 °C for 10 min.

System suitability: reference solution (b):

– the chromatogram shows 4 clearly separated spots.

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

C. Dissolve 0.25 g in 5 mL of *water R*. Add 5 mL of *ammonia R* and heat in a water-bath at 80 °C for 10 min. A red colour develops.

D. Water (see Tests).

TESTS

Appearance of solution. The solution is clear (*2.2.1*) and not more intensely coloured than reference solution BY_7 (*2.2.2, Method II*).

Dissolve 1.0 g in boiling *water R* and dilute to 10 mL with the same solvent.

Acidity or alkalinity. Dissolve 6.0 g by heating in 25 mL of *carbon dioxide-free water R*, cool and add 0.3 mL of *phenolphthalein solution R1*. The solution is colourless. Not more than 0.4 mL of *0.1 M sodium hydroxide* is required to change the colour of the indicator to pink or red.

Specific optical rotation (*2.2.7*): + 54.4 to + 55.9 (anhydrous substance).

Dissolve 10.0 g in 80 mL of *water R*, heating to 50 °C. Allow to cool and add 0.2 mL of *dilute ammonia R1*. Allow to stand for 30 min and dilute to 100.0 mL with *water R*.

Absorbance (*2.2.25*).

Test solution (a). Dissolve 1.0 g in boiling *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*.

Spectral range: 400 nm for test solution (a) and 210-300 nm for test solution (b).

Results:

– at 400 nm: maximum 0.04 for test solution (a);

– from 210 nm to 220 nm: maximum 0.25 for test solution (b);

– from 270 nm to 300 nm: maximum 0.07 for test solution (b).

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

Dissolve 4.0 g in *water R* with warming, add 1 mL of *0.1 M hydrochloric acid* and dilute to 20 mL with *water R*. 12 mL of the solution complies with test A. Prepare the reference solution using *lead standard solution (1 ppm Pb) R*.

Water (*2.5.12*): 4.5 per cent to 5.5 per cent, determined on 0.50 g, using a mixture of 1 volume of *formamide R* and 2 volumes of *methanol R* as the solvent.

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

Microbial contamination

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

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