LEMON BALM FOR HOMOEOPATHIC PREPARATIONS MELISSA OFFICINALIS FOR HOMOEOPATHIC PREPARATIONS

Melissa officinalis ad praeparationes homoeopathicas

DEFINITION

Fresh aerial parts of *Melissa officinalis* L., harvested before flowering.

CHARACTERS

Macroscopic and microscopic characters described under identifications tests A and B.

Lemon-like odour.

IDENTIFICATION

A. Lemon balm is a perennial plant, 30-80 cm high, with an erect, ramified, more or less pubescent stem. The leaves are opposite, oval-cordiform with long petioles. The lamina is thin 3-4 cm long, with roughly dentate or crenulate margins, bright green on the top, paler on the underside. The veins form a network between the meshes of which the lamina protrudes, giving the underside of the leaf a characteristic waffle-like appearance.

B. Examine a fragment of epidermis from the leaf under a microscope, using *chloral hydrate solution R*. The epidermis presents: fragments with sinuous walls; short, straight, finely striated, conical unicellular covering trichomes; multicellular covering trichomes with pointed tip and thick verrucous walls; 8-celled Labiateae-type secretory trichomes; secretory trichomes with 1-3 celled stalk and 1- (or more rarely 2-) celled head; diacytic stomata on the underside only (*2.8.3*).

TESTS

Foreign matter (*2.8.2*): maximum 5 per cent.

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2003.

Loss on drying (*2.2.32*): minimum 60.0 per cent, determined on 5.0 g of finely-cut drug, by drying in an oven at 100-105 °C for 2 h.

STOCK

DEFINITION

Lemon balm mother tincture complies with the requirements of the general technique for the preparation of mother tinctures (see *Homoeopathic Preparations (1038)* and French Pharmacopoeia Authority Supplement). The mother tincture is prepared with ethanol (65 per cent *V/V)*, using the fresh aerial parts of *Melissa officinalis* L.

Content: minimum 0.025 per cent *m/m* of total hydroxycinnamic derivatives, expressed as rosmarinic acid ($C_{18}H_{16}O_8$; M_r 360.3).

CHARACTERS

Green-brown liquid.

IDENTIFICATION

Thin-layer chromatography (*2.2.27*).

Test solution. Mother tincture.

Reference solution. Dissolve 5 mg of *caffeic acid R* and 5 mg of *rosmarinic acid R* in 20 ml of *ethanol (96 per cent) R*.

Plate: *TLC silica gel plate R*.

Mobile phase: *water R, methanol R, glacial acetic acid R, methylene chloride R* (2:3:8:15 *V/V/V/V*).

Application: 20 µl, as bands.

Development: over a path of 10 cm.

Drying: in air.

Detection A: examine in ultraviolet light at 365 nm.

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

The General Chapters and General Monographs of the European Pharmacopoeia and Preamble of the French Pharmacopoeia apply.

Top of the plate	
Caffeic acid: a blue zone Rosmarinic acid: a blue zone	A blue zone (caffeic acid) A blue zone (rosmarinic acid)
	A blue-grey zone Two blue zones
Reference solution	**Test solution**

Detection B: first spray with a 10 g/l solution of *diphenylboric acid aminoethyl ester R* in *methanol R* then spray with a 50 g/l solution of *macrogol 400 R* in *methanol R*. Allow the plate to dry in air for about 30 min. Examine in ultraviolet light at 365 nm.

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

Top of the plate	
Caffeic acid: a green-blue zone Rosmarinic acid: a green-zone	A green-blue zone (caffeic acid) A green zone (rosmarinic acid)
	A green zone
Reference solution	**Test solution**

TESTS

Ethanol content (*2.9.10*): 60 per cent *V*/*V* to 70 per cent *V*/*V*.

Methanol and 2-propanol (*2.9.11*): maximum 0.05 per cent *V*/*V*; maximum 0.05 per cent *V*/*V*.

Dry residue (*2.8.16*): minimum 1.1 per cent *m*/*m*.

ASSAY

Ultraviolet and visible absorption spectrophotometry (*2.2.25*).

Test solution. In a 100.0 ml volumetric flask, introduce 5.00 g of mother tincture and dilute to 100.0 ml with *ethanol* (50 per cent *V*/*V*) *R* (solution A). Introduce 1.0 ml of this solution in a 100.0 ml volumetric flask, add 2 ml of *0.5 M hydrochloric acid*, 2 ml of a solution prepared by dissolving 10 g of *sodium nitrite R* and 10 g of *sodium molybdate R* in 100 ml of *water R*, then

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add 2 ml of *dilute sodium hydroxide solution R*. Dilute to 10.0 ml with *water R* and mix.

Compensation liquid. In a 10.0 ml volumetric flask, introduce 1.0 ml of solution A, 2 ml of *0.5 M hydrochloric acid R*, 2 ml of *dilute sodium hydroxide solution R* and dilute to 10.0 ml with *water R*.

Detection: 505 nm.

Calculate the percentage content *m*/*m* of total hydroxycinnamic derivatives, expressed as rosmarinic acid, from the expression:

$$\frac{A \times 2.5}{m}$$

i.e. taking the specific absorbance of rosmarinic acid to be 400 at 505 nm.

A = absorbance at 505 nm,

m = mass of the sample, in grams.

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