

**INDIAN TOBACCO FOR HOMOEOPATHIC
PREPARATIONS
LOBELIA INFLATA FOR HOMOEOPATHIC
PREPARATIONS**

Lobelia inflata ad præparationes homœopathicas

DEFINITION

Fresh, blooming, aerial part of *Lobelia inflata* L.

IDENTIFICATION

- A. Angular, hairy, ramose stem, often purplish at the base, 20-50 cm high. Thick, isolated, sessile, pubescent, oval-lanceolate leaves, unevenly crenated, dentate, 5 cm long on average. Flowers displayed on terminal racemes. Calyx with 5 linear lobes topping the ovary. Pale blue, bilabiate corolla, split at the back, 5 stamens fused by their filaments and anthers; bilocular ovary topped by 2 stigmas.
- B. Take a sample of epidermis from the underside. Examine under a microscope, using *chloral hydrate solution R*: abaxial epidermis composed of epidermic cells deeply lobed, puzzle-shaped, stomatas of anomocytic type (2.8.3), surrounded by 3-5 subsidiary cells. Uniseriate, unicellular covering trichomes, stiff, pointed that may reach 150 µm conspicuous on the ribs and the lamina margin.

TESTS

Foreign matter (2.8.2): maximum 5 per cent.

Loss on drying (2.2.32): minimum 65.0 per cent, determined on 5.0 g of finely-cut drug, by drying in an oven at 100-105 °C for 2 h.

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

2009.

STOCK

DEFINITION

Indian tobacco tincture is prepared with ethanol (65 per cent *V/V*), using the fresh, blooming, aerial part of *Lobelia inflata* L.

Adjusted content: minimum 0.01 per cent and maximum 0.05 per cent of total alkaloids, expressed as lobeline ($C_{22}H_{27}NO_2$; M_r 337.5).

PRODUCTION

Method 4c (2371). Drug fragmented into segments 5 cm long. Maceration time: about 5 weeks.

CHARACTERS

Greenish-brown liquid.

IDENTIFICATION

A. Thin-layer chromatography (2.2.27).

Test solution. Mother tincture.

Reference solution. Dissolve 5 mg of *rutin R* and 5 mg of *quercitrin R* in 20 ml of *ethanol (96 per cent) R*.

Plate: TLC silica gel plate *R*.

Mobile phase: *glacial acetic acid R*, *anhydrous formic acid R*, *water R*, *ethyl acetate R* (11:11:27:100 *V/V/V/V*).

Application: 20 μ l, as bands.

Development: over a path of 10 cm.

Drying: in air.

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

Results: 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	
Quercitrin : an orange zone	A greenish-yellow zone
Rutin : an orange zone	An orange zone
	An orange zone
	A light orange zone
Reference solution	Test solution

B. Thin-layer chromatography (2.2.27).

Test solution. To 20 ml of mother tincture, add 10 ml of *water R* and alkalinize with *concentrated ammonia R*. Shake twice with 15 ml of *ether R* each time. Combine the ether phases. Evaporate under reduced pressure. Dilute the residue in 1 ml of *methanol R*.

Reference solution. Dissolve 5 mg of *lobeline hydrochloride R* and 5 mg of *senecionine R* in 10 ml of *ethanol (96 per cent) R*.

Plate : TLC silica gel plate *R*.

Mobile phase : glacial acetic acid *R*, water *R*, butanol *R* (10:10:40 V/V/V).

Application : 30 µl, as bands.

Development : over a path of 10 cm.

Drying : in air.

Detection : spray with *potassium iodobismuthate solution R*. Examine in daylight.

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

Top of the plate	
	An orange zone
Lobeline (hydrochloride) : an orange zone	An orange zone (lobeline) An orange zone
Senecionine : an orange zone	An orange zone
Reference solution	Test solution

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

2009.

TESTS

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

Dry residue (2.8.16): minimum 0.8 per cent *m/m*.

ASSAY

In a 250 ml volumetric flask, place a sample of mother tincture, close to 70.00 g, accurately weighed. Evaporate to dryness, under reduced pressure at 60 °C. Allow to cool. Dissolve the residue in 10 ml of *ether R*. Add 35 ml of *hydrochloric acid R1* (1 per cent *V/V*). Mix then evaporate the ether under reduced pressure. Transfer and filter the whole quantity into a separating funnel. Rinse the flask and the filter with fractions of 20 ml of *hydrochloric acid R1* (1 per cent *V/V*). Alkalinize with *dilute ammonia R1*. Shake with successive fractions of 20 ml of *ether R* until complete elution of the alkaloids. Collect the ether phases and wash them with 20 ml of *water R* then dry them on *anhydrous sodium sulphate R*. Evaporate to dryness under reduced pressure. Dilute the residue with 15 ml of *anhydrous acetic acid R*. Titrate with *perchloric acid 0.01 M* in presence of *quinaldine red solution R*.

Carry out a blank titration with 15 ml of *anhydrous acetic acid R*.

1 ml of *perchloric acid 0.01 M* corresponds to 0.003375 g of total alkaloids, expressed as lobeline.