

## JABORANDI FOR HOMOEOPATHIC PREPARATIONS JABORANDI FOR HOMOEOPATHIC PREPARATIONS

### ***Pilocarpus ad praeparationes homoeopathicas***

#### DEFINITION

Dried leaf or leaflet of *Pilocarpus microphyllus* Stappf., *Pilocarpus jaborandi* Holmes or *Pilocarpus pennatifolius* Lem.

*Content*: minimum 0.5 per cent total alkaloids expressed as pilocarpine ( $C_{11}H_{16}N_2O_2$ ;  $M_r$  208.3), determined on the dried drug.

#### CHARACTERS

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

#### IDENTIFICATION

- A. Jaborandi leaf is yellow-green to pale green-grey and compound-imparipinnate with 3-5 pairs of leaflets. The commercialised drug often consists of the leaflets separated from the rachis. The leaflet of *P. microphyllus* is 2-4 cm long and 0.5-2 cm wide, attached to a winged, pubescent rachis. The leaflet of *P. jaborandi* is 5-15 cm long. The rachis, which may grow up to 25 cm in length, is slightly bulbous at the base. The leaf of *P. pennatifolius* is 8-12 cm long and 2.5-5 cm wide. The cylindrical rachis is slightly bulbous at the base, channelled at the top and measures 20-30 cm. Jaborandi leaflet is leathery and subsessile, except for the terminal leaflet which is petiolate. It is marked with brown dots corresponding to the oil glands. The lamina is entire and the edges more or less curled towards the inside. The numerous pinnate secondary nerves anastomose near the edge of the leaflet to form a fine network of tertiary nerves. The leaflet of *P. microphyllus* is obovate, more or less rounded and clearly notched at the top. The lateral leaflet is asymmetrical at the base. The leaflet of *P. jaborandi* is oval, elliptical, very slightly pubescent, almost always notched at the top and slightly cordate at the base. The leaflet of *P. pennatifolius* is glabrous, oval, oblong, often slightly asymmetrical, notched or slightly acute at the top and, with a short petiole excepted on the terminal leaflet.

---

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

**2002.**

B. Reduce the leaf to a powder (355). The powder is yellow-green. When examined under a microscope using *chloral hydrate solution R*, the following are observed: fragments of epidermis covered in a thick cuticle, with secretory trichomes sunk in the depressions (*P. pennatifolius* and *P. jaborandi*) or exerted secretory trichomes (*P. microphyllus*); fragments of heterogeneous, asymmetrical mesophyll consisting of a single row of palisade cells containing schizolysigenous oil glands and numerous cells with calcium oxalate crystals; fragments of vein consisting of spiral- or ring-shaped vessels and pericyclic fibres.

C. Thin-layer chromatography (2.2.27).

*Test solution.* Add 30 ml of *ethanol (65 per cent V/V) R* to 3 g of powdered drug (355). Cover. Heat for 15 min on a water-bath at 60 °C. Allow to cool. Filter.

*Reference solution.* Dissolve 5 mg of *pilocarpine nitrate R* and 5 mg of *papaverine R* in 10 ml of *ethanol (96 per cent) R*.

*Plate:* TLC silica gel plate *R*.

*Mobile phase:* concentrated ammonia *R*, acetone *R*, methylene chloride *R* (2:48:50 V/V/V).

*Application:* 40 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 of 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	
Papaverine: a pale yellow zone	A blue zone A blue zone A green-blue zone A blue zone
<b>Reference solution</b>	<b>Test solution</b>

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

*Results B:* see below the sequence of zones present in the chromatograms of the reference solution and the test solution. Furthermore other 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	
Papaverine: an orange zone Pilocarpine: an orange zone	An orange zone  An orange zone (pilocarpine)
<b>Reference solution</b>	<b>Test solution</b>

## TESTS

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

**Loss on drying** (2.2.32): maximum 12.0 per cent, determined on 1.0 g of powdered drug (355), by drying in an oven at 100-105 °C for 2 h.

**Total ash** (2.4.16): maximum 10.0 per cent.

## ASSAY

To 10.0 g of powdered drug (355), add 10 ml of *dilute ammonia R2*. Homogenise then allow to stand for 30 min. Transfer completely into the cartridge of a continuous extraction device fitted with a 125 ml extractor tube and a 250 ml round-bottomed flask. Pour 125 ml of *methylene chloride R* into the flask. Extract the powder for 3 h whilst adjusting the heat so that the solvent is renewed in the tube every 5 min. When the solvent is cool, extract several times using about 0.05 M *sulphuric acid*. Alkalinise using *dilute ammonia R2* to pH 9, then extract using *methylene chloride R* until the alkaloids have been totally extracted. Wash the combined organic solutions with 20 ml of *water R*. Evaporate to dryness. Dissolve the residue in 10 ml of *methylene chloride R*, add 20.0 ml of 0.02 M *hydrochloric acid*. Evaporate the methylene chloride then titrate the excess hydrochloric acid using 0.02 M *sodium hydroxide* with *methyl red mixed solution R* as indicator until the colour changes from violet to green.

Calculate the percentage content of total alkaloids expressed as *pilocarpine*, from the expression:

$$\frac{(20 - n) \times 0.4166}{m}$$

$m$  = mass of the sample in grams,

$n$  = volume of 0.02 M *sodium hydroxide* used in millilitres.

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

2002.

## STOCK

### DEFINITION

Jaborandi 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 dried leaf of *Pilocarpus microphyllus* Stappf., *Pilocarpus jaborandi* Holmes or *Pilocarpus pennatifolius* Lem.

*Ajusted content:* minimum 0.02 per cent *m/m* and maximum 0.06 per cent *m/m* of total alkaloids, expressed as pilocarpine ( $C_{11}H_{16}N_2O_2$ ;  $M_r$  208.3).

### CHARACTERS

Yellow-brown liquid.

### IDENTIFICATION

Thin-layer chromatography (2.2.27).

*Test solution.* Mother tincture.

*Reference solution.* Dissolve 5 mg of *pilocarpine nitrate R* and 5 mg of *papaverine R* in 10 ml of *ethanol (96 per cent) R*.

*Plate:* TLC silica gel plate *R*.

*Mobile phase:* concentrated ammonia *R*, acetone *R*, methylene chloride *R* (2:48:50 *V/V/V*).

*Application:* 40  $\mu$ l, 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 of the reference solution and the test solution. Furthermore other 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	
Papaverine: a pale yellow zone	A blue zone A blue zone A green-blue zone A blue zone
<b>Reference solution</b>	<b>Test solution</b>

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

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

Top of the plate	
Papaverine: an orange zone Pilocarpine: an orange zone	An orange zone An orange zone (pilocarpine)
<b>Reference solution</b>	<b>Test solution</b>

## 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:** minimum 0.8 per cent *m/m* (see French Pharmacopoeia Authority Supplement).

## ASSAY

Evaporate 100.0 g of Jaborandi mother tincture at a low temperature until a residue of about 20 g is obtained. Transfer the residue completely to a separating funnel using a few millilitres of *water R*. Add 10 ml of *dilute ammonia R2*. Extract successively with 20 ml fractions of *methylene chloride R* until the alkaloids have been totally extracted. Combine the organic layers then extract several times using about 0.05 *M sulphuric acid*. Alkalinise using *dilute ammonia R2* to pH 9, then extract using *methylene chloride R* until the alkaloids have been totally extracted. Wash the combined organic solutions with 20 ml of *water R*. Evaporate to dryness. Dissolve the residue in 10 ml of *methylene chloride R*, add 20.0 ml of 0.02 *M hydrochloric acid*.

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

2002.

Evaporate the methylene chloride then titrate the excess hydrochloric acid using *0.02 M sodium hydroxide* with *methyl red mixed solution R* as indicator until the colour changes from violet to green.

Calculate the percentage content  $m/m$  of total alkaloids expressed as pilocarpine, from the expression:

$$\frac{(20 - n) \times 0.4166}{m}$$

$m$  = mass of the sample in grams,

$n$  = volume of 0.02 M sodium hydroxide used in millilitres.