# MISTLETOE FROM THE APPLETREE FOR HOMOEOPATHIC PREPARATIONS VISCUM ALBUM FOR HOMOEOPATHIC PREPARATIONS

Viscum album ad praeparationes homoeopathicas
Other Latin name used in homoeopathy: Viscum mali

#### DEFINITION

Fresh, aerial, fructiferous part of *Viscum album* L. collected on the apple tree *Malus domestica* Borkh.

## **IDENTIFICATION**

Aerial parts consisting of big, rounded clumps. Articulated stems, glabrous, yellowish-green, with a circular section, abundantly branching out in a false dichotomy. Opposite leaves, sessile, simple, entire, oblong, obtuse at the apex and narrowed at the base. Coriaceous lamina, green to yellowish-green, showing 3-5 parallel ribs. Whitish, globular, sessile fruit, about 8 mm in diameter, topped by remains of stigmas and containing an albuminate seed, embedded within a very glutinous and translucent pulp.

## **TESTS**

**Loss on drying** (2.2.32): minimum 50.0 per cent, determined on 5.0 g of finely-cut drug by drying in an oven at 105 °C for 2 h.

# **STOCK**

#### DEFINITION

The mother tincture of mistletoe from the appletree is prepared with ethanol (45 per cent V/V), using the fresh, aerial, fructiferous part of *Viscum album* L. collected on the apple tree *Malus domestica* Borkh.

Content: minimum 0.006 per cent m/m of lignans, expressed as syringic acid (C<sub>9</sub>H<sub>10</sub>O<sub>5</sub>;  $M_r$  198.2).

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

## **PRODUCTION**

Method 4c (2371). Drug fragmented into segments, smaller than 5 cm long. Maceration time: about 3 weeks.

### CHARACTERS

Brown liquid more or less orange.

### IDENTIFICATION

Thin-layer chromatography (2.2.27).

Test solution. Mother tincture.

Reference solution. Dissolve 2 mg of chlorogenic acid R and 2 mg of caffeic acid R in 20 ml of methanol R.

*Plate*: TLC silica gel plate R (5-40  $\mu$ m) [or TLC silica gel plate R (2-10  $\mu$ m)].

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

Application: 20 µl [or 5 µl] as bands.

Development: over a path of 10 cm [or 7 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.

Top of the plate		
Caffeic acid: a greenish-blue zone		
Chlorogenic acid: a greenish-blue zone	A blue zone A greenish-blue zone	
Reference solution	Test solution	

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

### **TESTS**

Ethanol content (2.9.10): 40 per cent V/V to 50 per cent V/V.

**Dry residue** (2.8.16): minimum 2.5 per cent m/m.

### ASSAY

Liquid chromatography (2.2.29).

Test solution. In a 20.0 ml volumetric flask, place 8.000 g of mother tincture and dilute to 20.0 ml with a mixture of 10 volumes of acetonitrile R1 and 90 volumes of trifluoroacetic acid (0.05 per cent V/V) R.

Reference solution. In a 100.0 ml volumetric flask, dissolve 10.0 mg of syringic acid R in water R and dilute to 100.0 ml with the same solvent. Place 10.0 ml of this solution into a 20.0 ml volumetric flask and dilute to 20.0 ml with a mixture of 10 volumes of acetonitrile R1 and 90 volumes of trifluoroacetic acid (0.05 per cent V/V) R.

#### Column:

- size:  $l = 0.25 \text{ m}, \emptyset = 4.6 \text{ mm},$
- stationary phase: octadecylsilyl silica gel for chromatography R (5 μm),
- temperature: 30 °C.

# Mobile phase:

- mobile phase A: trifluoroacetic acid (0.05 per cent V/V) R,
- mobile phase B: acetonitrile R1.

Time (min)	Mobile phase A (per cent V/V)	Mobile phase B (per cent V/V)
0-20 20-25 25-45 45-50 50-55	$ \begin{array}{c} 90 \\ 90 \rightarrow 85 \\ 85 \\ 85 \rightarrow 0 \\ 0 \end{array} $	$   \begin{array}{c}     10 \\     10 \to 15 \\     15 \\     15 \to 100 \\     100   \end{array} $

Flow rate: 1.0 ml/min.

Detection: spectrophotometer at 220 nm.

*Injection* : 20 μl.

Determine the peaks of lignans 1, 2, 3, 4 with the chromatogram of the mother tincture.

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Relative retention with reference to syringic acid (retention time = about 22 min.): lignan 1 = about 0.6; lignan 2 = about 0.7; lignan 3 = about 1.5; lignan 4 = about 1.6.

Calculate the percentage content m/m of lignans, expressed as syringic acid, from the expression:

$$\frac{\sum A_1 \times m_2}{A_2 \times m_1} \times 0.1 \times p$$

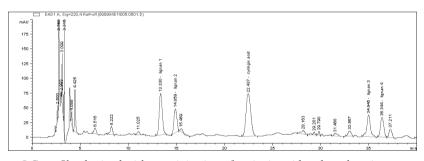
 $\sum A_1$  = sum of the peak areas due to lignans 1, 2, 3, and 4 in the chromatogram obtained with the test solution,

 $A_2$  = area of the peak due to syringic acid in the chromatogram obtained with the reference solution,

 $m_1 = \text{mass of the mother tincture sample, in grams,}$ 

 $m_2$  = mass of syringic acid R sample, in grams,

p = percentage content of syringic acid in syringic acid R.



LC profile obtained with a co-injection of syringic acid and mother tincture

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