

KINKELIBA FOR HOMOEOPATHIC PREPARATIONS COMBRETUM RAIMBAULTII FOR HOMOEOPATHIC PREPARATIONS

Combretum micranthum ad praeparationes homoeopathicas

Other Latin names used in homoeopathy: **Kinkeliba**

Combretum

The herbal drug complies with the requirements of the monograph *Kinkeliba*.

STOCK

DEFINITION

Kinkeliba 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 *Combretum micranthum* G. Don

Content:

- minimum 0.10 per cent *m/m* of total flavonoid derivatives, expressed as vitexin ($C_{21}H_{20}O_{10}$; M_r 432.4),
- minimum 1.10 per cent *m/m* of total polyphenols.

CHARACTERS

Dark red brown liquid.

Cloying odour.

IDENTIFICATION

A. Thin-layer chromatography (2.2.27).

Test solution. Mother tincture.

Reference solution. Dissolve 5 mg of *orientin R*, 5 mg of *isoorientin R*, 5 mg of *vitexin R* and 5 mg of *isovitexin R* in *ethanol (96 per cent) R* and dilute to 20 ml with the same solvent.

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

2002 - 2007.

Plate: TLC silica gel plate R.

Mobile phase: anhydrous formic acid R, water R, methyl ethyl ketone R, ethyl acetate R (10:20:30:50 V/V/V/V).

Application: 20 µl, as bands.

Development: over a path of 15 cm.

Drying: in air (the formic acid odour remains perceptible).

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 in air for about 30 min. Examine in ultraviolet light at 365 nm.

Results: see below the sequence of fluorescent zones present in the chromatograms of the reference solution and the test solution.

Top of the plate	
Vitexin: a green-yellow zone Orientin: an orange-yellow zone Isovitexin: a green-yellow zone Isoorientin: an orange-yellow zone	A blue zone A green-yellow zone A green-yellow zone (vitexin) An orange-yellow zone (orientin) A green-yellow zone (isovitexin) An orange-yellow zone (isoorientin) Two orange-yellow zones
Reference solution	Test solution

B. Thin-layer chromatography (2.2.27).

Test solution. Mother tincture.

Reference solution. Dissolve 10 mg of gallic acid R and 10 mg of pyrogallol R in 100 ml of ethanol (96 per cent) R.

Plate: TLC silica gel plate R.

Mobile phase: water, anhydrous formic acid R, methylene chloride R, ethyl acetate R (2:10:40:50 V/V/V/V).

Application: 20 µl, as bands.

Development: over a path of 15 cm.

Drying: in air.

Detection: spray with solution of fast blue b salt R. Heat the plate to 100-105 °C for 5 min. Examine in daylight.

Results: see below the sequence of zones present in the chromatograms of the reference solution and the test solution.

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

Top of the plate	
Pyrogallol: a brown zone Gallic acid: a pink-brown zone	A brown zone A purple-blue zone A pink-brown zone (gallic acid) A purple-pink zone A purple-pink zone A pink-brown 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: minimum 2.0 per cent *m/m* (see French Pharmacopoeia Authority Supplement).

ASSAY

Total polyphenols. Visible absorption spectrophotometry (2.2.25).

Test solution. In a volumetric flask, place 4.00 g of mother tincture. Add 50 ml of *water R* and dilute to 100.0 ml with the same solvent. Filter the liquid through a paper filter with a diameter of 12 cm. Discard the first 50 ml of filtrate, and use the rest for the assay. Dilute 5.0 ml of filtrate to 25.0 ml with *water R*. To 5.0 ml of this solution, add 1.0 ml of *solution of phosphotungstic acid R*, mix and dilute to 50.0 ml with a 150 g/l solution of *sodium carbonate R*.

Compensation liquid. Water R.

Exactly 2 min later, measure the absorbance of the test solution at 715 nm in comparison with the compensation liquid.

Reference solution. (Carry out the following operations away from light). Dissolve 50.0 mg of *pyrogallol R* in *water R* and dilute to 100.0 ml with *water R*. Dilute 5.0 ml of this solution to 100.0 ml with *water R*. To 5.0 ml of this solution, add 1.0 ml of *solution of phosphotungstic acid R* and dilute to 50.0 ml with a 150 g/l solution of *sodium carbonate R*.

Compensation liquid. Water R.

Measure the absorbance of the reference solution at 715 nm, in comparison with the compensation liquid, exactly 2 min after and within 15 min following the *pyrogallol R* dissolution.

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

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Calculate the percentage content m/m of total polyphenols from the expression:

$$\frac{A_1 \times 13.12}{A_2 \times m}$$

A_1 = absorbance of the test solution at 715 nm,

A_2 = absorbance of the reference solution at 715 nm,

m = mass of the sample in grams.

Total flavonoid derivatives. Visible absorption spectrophotometry (2.2.25).

Test solution. Place 10.00 g of mother tincture in a volumetric flask and dilute to 100.0 ml with *ethanol R* (60 per cent V/V) (stock solution). In a volumetric flask, place 2.0 ml of stock solution, 2 ml of a 2 per cent m/V solution of *aluminium chloride R* in *methanol R* and dilute to 25.0 ml with *methanol R*.

Compensation liquid. In a volumetric flask, place 2.0 ml of stock solution and dilute to 25.0 ml with *methanol R*. After 25 min, measure the absorbance of the test solution at 394 nm, in comparison with the compensation liquid.

Calculate the percentage content m/m of total flavonoid derivatives, expressed as vitexin, from the expression:

$$\frac{A \times 4.63}{m}$$

i.e. taking the specific absorbance to be 270.

A = absorbance of the test solution at 394 nm,

m = mass of the sample in grams.