

Technical documentation

Product name:	qRE <i>Melissa officinalis</i> L., leaves
Substance:	<i>Melissa officinalis</i> L., leaves dry extract
Plant source common names:	en: Common balm; fr: Mélisse
Reference:	E0009
Packaging:	100 mg in a 1.5 ml borosilicate amber vial
Storage conditions:	Keep container closed. Protect from light and moisture. Keep at -18 °C
Retest:	12 months

Botanical identification of plant source

Plants in our botanical garden are identified and a herbal voucher is prepared by an expert botanist. Each batch collected for extraction is verified and identified.

Reference: Flora Europaea, Cambridge University Press, 1972, Vol 3, p 162

Method of production of dry extract

Whole plant or plant parts are collected, freeze-dried and coarsely ground. Extraction is performed by maceration in 50 % (v/v) aqueous ethanol for 48 hours at room temperature. Ethanol is then evaporated under reduced pressure at less than 40 °C and the aqueous residue is freeze-dried.

Organoleptic characteristics of dry extract

Colour: Yellow beige

Odour: Non characteristic

Form: Fine powder

Recommended methods for use

Weight a precise weight of qRE and solubilise in the recommended solvent at the concentration indicated in the HPLC or HPTLC method described in this document.

Sonicate for 90 seconds (70 W). Filter on a 0.45 µm PVDF membrane and put the resulting solution into HPLC dispenser or apply on the HPTLC plate.

Dose and analyse your extract with qRExtract using the HPLC / HPTLC methods described in this document or using your own methods.

HPTLC

Detection of rosmarinic acid

Layer: 10 × 10 cm HPTLC Nano-Sil-20 UV 254 (Carl Roth ref. N084.1)

Thin layer conditionnement: 1 h at room temperature and 33 % relative humidity

Elution solvent:

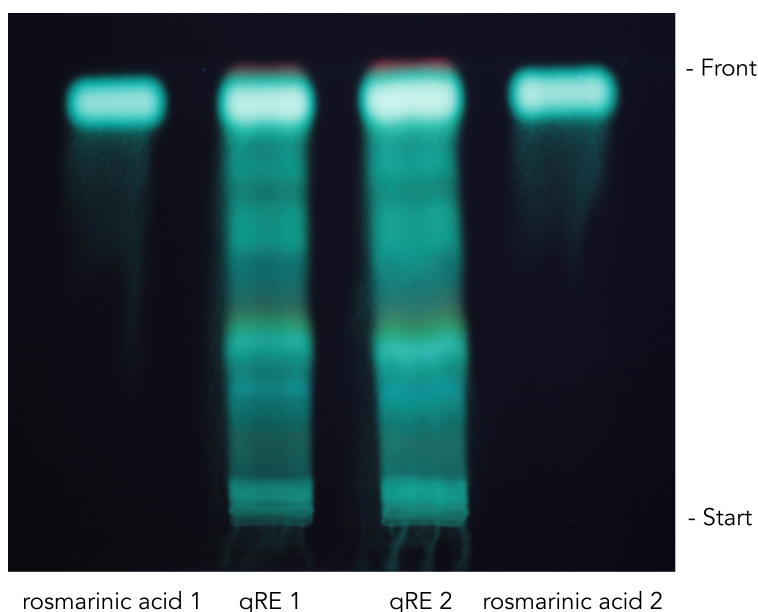
Elution solvent compound	Volume (ml)
ethyl acetate	15
formic acid	1
H ₂ O	1

Developing distance: 70 mm from the lower edge

Initial spot volume and concentration:

rosmarinic acid 1:	2 µl of a 0.2 % (w/v) solution in ethanol 96 %
qRE 1:	7 µl of a 1.5 % (w/v) solution in 50 % (v/v) aqueous ethanol
qRE 2:	10 µl of a 1.5 % (w/v) solution in 50 % (v/v) aqueous ethanol
rosmarinic acid 2:	2.5 µl of a 0.2 % (w/v) solution in ethanol 96 %

Reagent mixture: Natural products - polyethylene glycol reagent (NP/PEG)
 Preparation: Dissolve 0.25 g of diphenylboric acid 2-aminoethylester and 1.25 g of polyethylene glycol 400 in 25 mL of methanol.
 Dip the plate in the reagent mixture and dry for 15 minutes at room temperature.
 Expose to UV light at 365 nm.

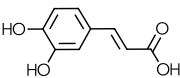
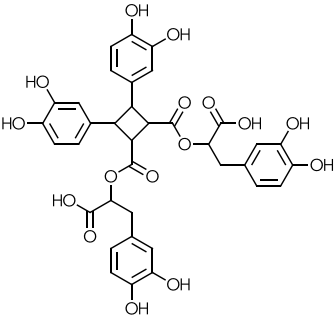
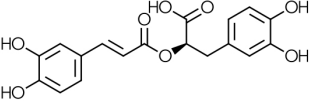
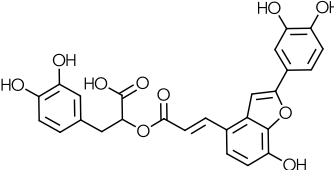
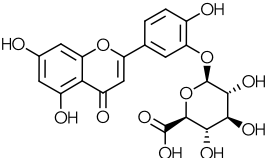


HPLC

Precolumn: Ascentis® Express C18 0.5 cm × 3.0 mm 2.7 μm
Column: Ascentis® Express C18 15 cm × 3.0 mm 2.7 μm
1st sample: 8 μl 0.6378 % qRE (w/v) solution in 50 % (v/v) aqueous ethanol
2nd sample: 10 μl 0.5111 % qRE (w/v) solution in 25 % (v/v) aqueous ethanol
Flow: 0.40 ml/min
Temperature: 25 °C
Mobile phase: A: 0.1 % formic acid (v/v) in water
 B: methanol
Detection: Diode Array Detector, 330 nm
Gradient:

Time (mn)	A %	B %
0	100	0
40	60	40
70	24	76

Quantified substances

Compound	CAS No	2D Structure	Peak No
Caffeic acid	331-39-5		1
Unknown	NA	NA	2
Sagerinic acid	NA		3
Rosmarinic acid	20283-92-5		4
Salvianolic acid C (exists as derivatives in qRE)	NA		5, 7
Luteolin 3'-O-glucuronide	53527-42-7		6