Technical documentation

Product name:	qRE Salvia sclarea L., leaves
Substance:	Salvia sclarea L., leaves dry extract
Plant source common names:	en: Clary sage; fr: Sauge sclarée
Reference:	E0076
Packaging:	100 mg in a 1.5 ml borosilicate amber vial
Storage conditions:	Keep container closed. Protect from light and moisture.
	Keep inferior to -15 °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 190

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 (v/v) 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. Residual water content measurement is done by Karl Fischer titration.

Organoleptic characteristics of dry extract

Colour: Dark brown 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 luteolin-7-glucuronide and rosmarinic acid

Layer: Thin layer conditionnement:	10 × 10 cm HPTLC Nano-Sil-20 UV 254 (Carl Roth ref. N084.1) 1 h at room temperature and 33 % relative humidity		
Elution solvent:	Elution solvent compound	Volume (ml)	
	ethyl acetate	100	
	H ₂ O	27	
	acetic acid	11	
	formic acid	11	

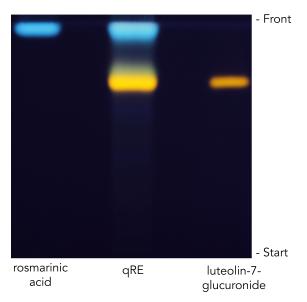
Initial spot volume and concentration:

rosmarinic acid:	1 μl of a 0.2 % (w/v) solution in ethanol 96 %
qRE:	5 μl of a 1 % (w/v) solution in 50 % (v/v) aqueous ethanol
luteolin-7-glucuronide:	1 μl of a 0.03 % (w/v) solution in 50 % (v/v) aqueous ethanol

Reagent mixture:

<u>Natural products - polyethylene glycol reagent (NP/PEG)</u> 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.



QQUANTIFIED Reference Extracts

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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				
Sample:	8 µl 0.63 % qR	8 µl 0.63 % qRE₀(w/v) solution in 50 % (v/v) aqueous ethanol			
Flow:	0.45 ml/min				
Temperature:	25 °C				
Mobile phase:	A: 0.1 % formi	A: 0.1 % formic acid (v/v) in water			
	B: 0.1 % formic acid (v/v) in acetonitrile				
Detection:	Diode Array Detector, 210 nm				
Gradient:	Time (mn)	Α%	<u>B %</u>		
	0	97	3		
	10	82	18		
	15	81	19		
	25	81	19		
	51	32.5	67.5		

Quantified substances

Compound	CAS No	2D Structure	Peak No
Luteolin-7-glucuronide isomer	29741-10-4	HO + C + C + C + C + C + C + C + C + C +	1
Apigenin glucuronide	29741-9-1	HO + CO + CO + OH + OC + CO + OH + OC + CO + OH + OC + CO + OH + OH	3

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Compound	CAS No	2D Structure	Peak No
Hispidulin-7-glucuronide	31105-76-7		4
Rosmarinic acid	20283-92-5	HO CH OH	5
Luteolin-7-glucuronide isomer	29741-10-4		6
Unknown	NA	NA	2, 7, 8, 9, 10, 11