Technical documentation

Product name:	**qRE Plantago lanceolata L., leaves**
Substance:	Plantago lanceolata L., leaves dry extract
Plant source common names:	en: Narrowleaf plantain; fr: Plantain lancéolé
Reference:	E0012
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, 1976, Vol 4, p 42

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: 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 verbascoside and aucubin

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	100
H_2O	27
acetic acid	11
formic acid	11

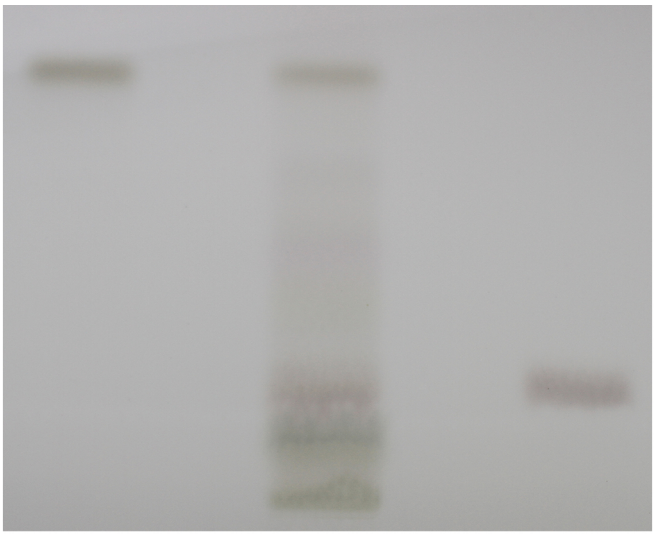
Developing distance: 70 mm from the lower edge

Initial spot volume and concentration:

verbascoside:	3 µl of a 0.2 % (w/v) solution in 50 % (v/v) aqueous ethanol
qRE:	5 µl of a 1.5 % (w/v) solution in 50 % (v/v) aqueous ethanol
aucubin:	1 µl of a 0.2 % (w/v) solution in 50 % (v/v) aqueous ethanol

Reagent mixture: Anisaldehyde reagent

Preparation: Slowly mix 85 mL of ice-cooled methanol with 10 mL of glacial acetic acid and 5 mL of sulfuric acid. Allow the mixture to cool to room temperature, then add 0.5 mL of anisaldehyde (p-methoxy benzaldehyde) Dip the plate in the reagent mixture and dry for 10 minutes at 110 °C. Expose to visible light.



HPTLC

Detection of luteolin

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)
toluene	60
ethyl acetate	25
hexane	20
formic acid	4

Developing distance: 70 mm from the lower edge

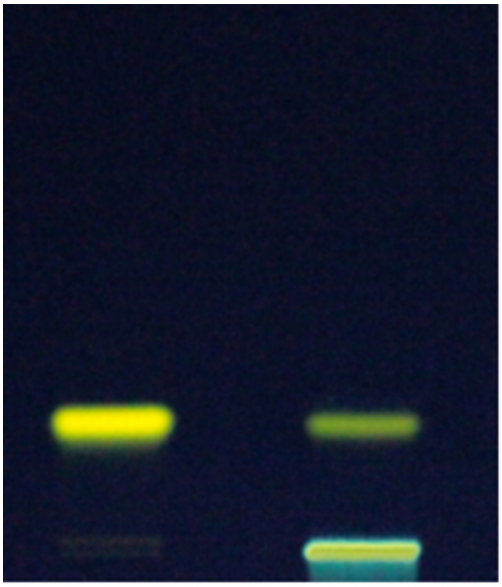
Initial spot volume and concentration:

luteolin: 1 µl of 0.15 % (w/v) solution in 50 % (v/v) aqueous ethanol

qRE: 5 µl of 1.5 % (w/v) solution in 50 % (v/v) aqueous ethanol

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

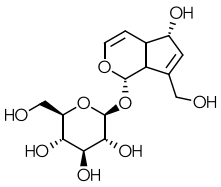
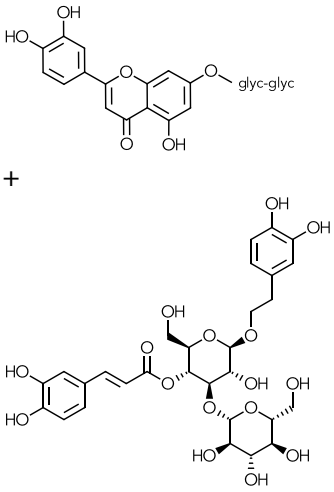
Precolumn: Ascentis® Express C18 0.5 cm × 3.0 mm 2.7 µm
Column: Ascentis® Express C18 15 cm × 3.0 mm 2.7 µm

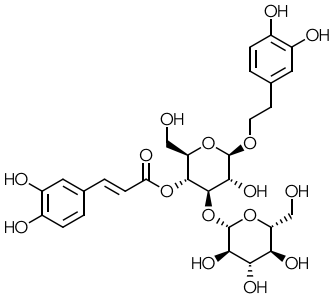
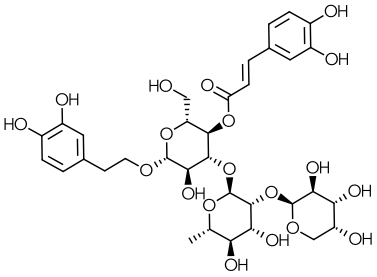
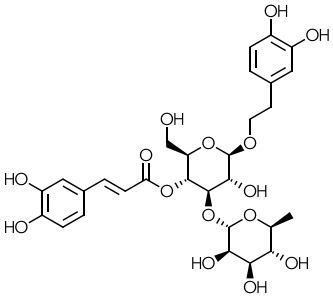
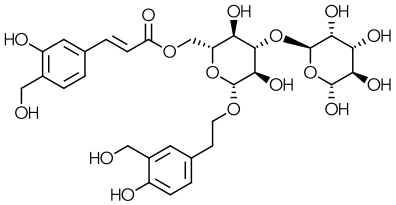
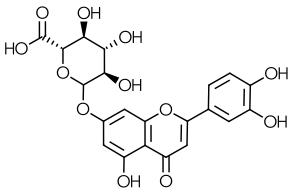
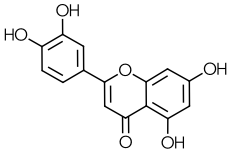
1st sample: 10 µl 3.10 % qRE (w/v) solution in 25 % (v/v) aqueous methanol
2nd sample: 8 µl 3.756 % qRE (w/v) solution in 50 % (v/v) aqueous ethanol

Flow: 0.45 ml/min
Temperature: 25 °C
Mobile phase: 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)	A %	B %
0	97	3
8	97	3
60	74	26

Quantified substances

Compound	CAS No	2D Structure	Peak No
Aucubin	479-98-1		1
Luteolin diheteroside + Plantamajoside or isomer	NA		2

Compound	CAS No	2D Structure	Peak No
Plantamajoside or isomer	NA		3, 4
Lavandulifolioside	NA		5
Verbascoside	61276-17-3		6
Plantainoside E or F	NA		7, 8
Luteolin-7-O-glucuronide	29741-10-4		9
Luteolin	491-70-3		10