Traditional Eastern Medication
Application guide

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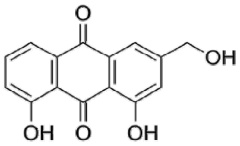
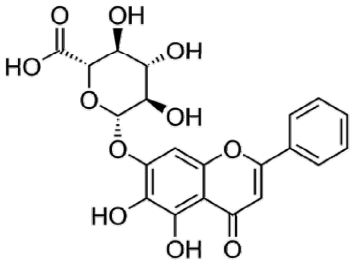
Content

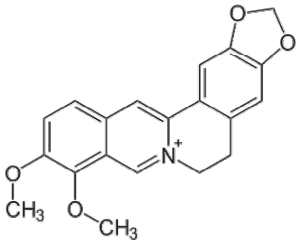
Molecular Structures



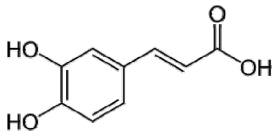
Aloe Emodin
Mw: 270.237 g/mol
LogP: 1.8



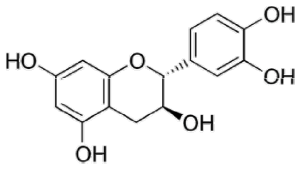
Baicalin
Mw: 446.36 g/mol
LogP: 1.1



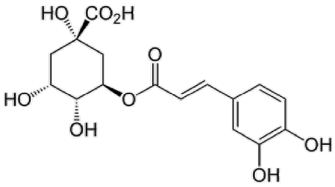
Berberine
Mw: 336.361 g/mol
LogP: 3.6



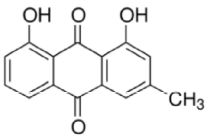
Caffeic acid
Mw:180.157 g/mol
LogP: 1.2



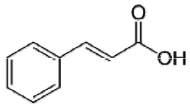
Catechin
Mw: 290.268 g/mol
LogP: 0.4



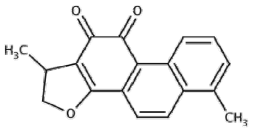
Chlorogenic acid
Mw: 354.31 g/mol
LogP: -0.4



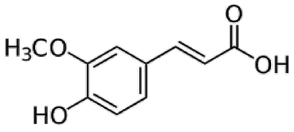
Chrysophanol
Mw: 254.237 g/mol
LogP: 3.5



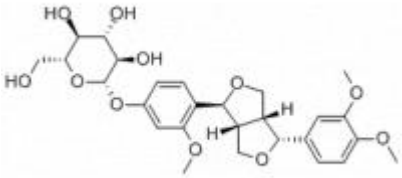
Cinnamic acid
Mw: 148.159 g/mol
LogP: 2.1



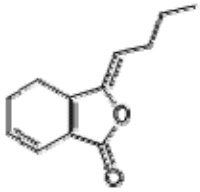
Dihydrotanshinone
Mw: 278.30 g/mol
LogP: 3.2



Ferulic acid
Mw: 194.184 g/mol
LogP: 1.5

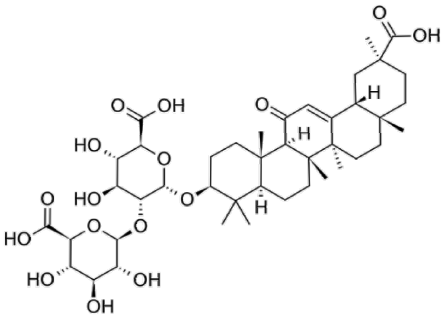


Forsythin
Mw:534.55 g/mol
LogP: 0

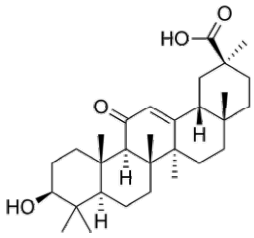


Ligustilide
Mw:190.238 g/mol
LogP: 2.7

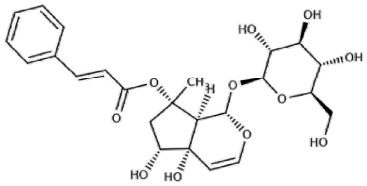
Molecular Structures



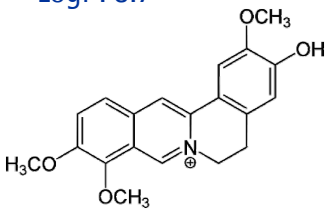
Glycyrrhizic acid
Mw: 822.932 g/mol
LogP: 3.7



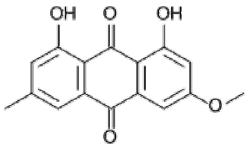
Glycyrrhetinic Acid
Mw: 470.684 g/mol
LogP: 6.4



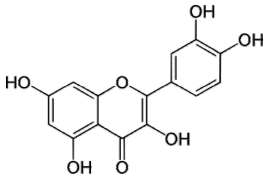
Harpagoside
Mw:494.489 g/mol
LogP: -0.6



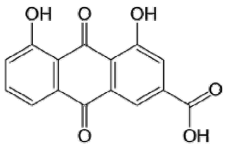
Jatrorrhizine
Mw: 338.377 g/mol
LogP: 3.4



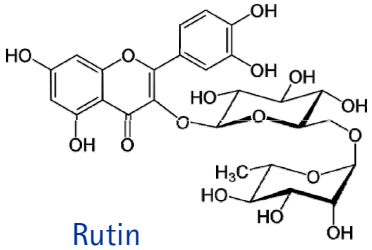
Physcion
Mw: 284.263 g/mol
LogP: 3



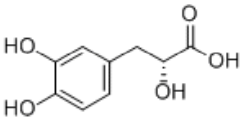
Quercetin
Mw: 302.236 g/mol
LogP: 1.5



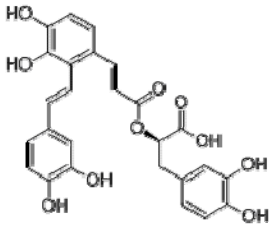
Rhein
Mw: 284.22 g/mol
LogP: 2.2



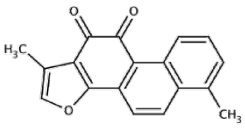
Rutin
Mw: 610.5175 g/mol
LogP: -1.3



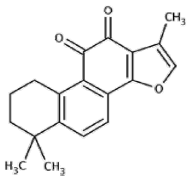
Salvianic Acid
Mw: 198.173 g/mol
LogP: -0.2



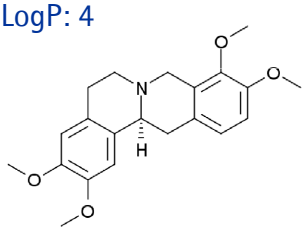
Salvianolic Acid B
Mw: 718.614 g/mol
LogP: 4



Tanshinone I
Mw: 276.29 g/mol
LogP: 3.7



Tanshinone IIA
Mw: 276.29 g/mol
LogP: 3.7



Tetrahydropalmatine
Mw: 355.4275 g/mol
LogP: 3.2

Application Index

Introduction

What is Traditional Eastern Medication?

Traditional Eastern Medicine, or Traditional Chinese Medicine (TCM), encompass a broad range of medicine practices sharing common concepts which have been developed in China and the Far East and are based on a tradition of more than 2,000 years, including various forms of herbal medicine, acupuncture, massage, exercise, and dietary therapy. In traditional Chinese herbal medicine, plant elements are by far the most commonly, but not solely, used substances; animal and mineral products are also utilized.

Ever since TCM was introduced in the treatment and prevention of illnesses, there have now and then been problems with the authenticity and the quality of certain medications. The methods, techniques and theories of assessing the quality of Chinese medicine have also gone through the phases of formation, development and continuous improvement. Today chromatographic fingerprint analysis is a requirement.

The methods included in this application guide show separations of different type of molecules; mainly hydrophobic but also slightly hydrophilic in different plant materials. Due to the nature of the molecules of interest we have used reversed phase columns, RP-18 endcapped stationary phases on either particulate silica particles or monolithic silica columns (Chromolith® columns). All methods are shown with complete experimental details to ease the implementation in your laboratory.

Merck Millipore can offer virtually everything, beside the HPLC instrument, for your needs!

TCM Fingerprint Analysis

Herbs and other natural products are used worldwide for preventive and therapeutic purposes. Identification and quality control of products of vegetal origin is thus required. Since the 1990s, the Chinese Pharmacopeia (ChP) has established quality control standards for the Chinese chemicals and herbs. Currently in China the regulations clearly indicate that TCM herbal injections must be quality controlled by chromatographic fingerprint technology.
In addition, the technique is also widely used in standardizing the production of herbs.
The chromatographic fingerprint approach is accepted by the World Health Organization (WHO) as a quality and identification evaluation technique.

When developing a new method for fingerprinting, generally the first step is to create conditions maximizing the peak capacity. A rule of thumb in chromatography is that the longer the column – the higher peak capacity (more theoretical plates are available). Therefore long columns have traditionally been used. On the other hand, with a long column packed with small particles the backpressure will be high, and ultimately that constitutes a practical problem and limits the use of more viscous solvents in the mobile phase, as well as increasing the wear on the HPLC system.

Monolithic columns are a suitable alternative to packed particle columns for maximizing the peak capacity. The low column backpressure offers the possibility of connecting several columns in series while still maintaining a low backpressure. One publication has demonstrated that more than 10 columns can be connected in series, making an HPLC column of more than one meter in length with over 100,000 theoretical plates, yet with a relatively low backpressure for use with a standard HPLC system. The technique of coupling two, three and up to four Chromolith® columns in series for fingerprint analysis of herbal material has been used and published by Vander Heyden et al. in two papers.

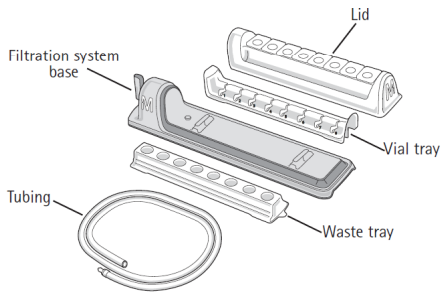
Journal of Chromatography A, 1172 (2007) 1-8
Journal of Pharmaceutical and Biomedical Analysis 95 (2014) 34-46.

In the 2014 paper it was concluded from experimental design and multivariate analysis that a methanol and water mixture (80:20 (v/v)) is the best extraction solvent for pulverized plant material using a shaking bath for 30 minutes as extraction method followed by filtration through 0.20 µm filters. The highest number of separated peaks in the fingerprint (for Artemisia species) was acquired with four coupled Chromolith® columns (100×4.6 mm). Trifluoroacetic acid 0.05% (v/v) was used as mobile-phase additive in a stepwise linear methanol/water gradient, going from 5 to 95% /v/v) methanol in 51 minutes and then keeping the last mobile phase composition isocratic until 60 minutes.

Robustness and resistance towards sample matrix is a major advantage with monolithic columns over particle packed columns for the analysis of herbal extracts of roots, dried berries or leaves. Due to the nature of it's structure, the tendency to clog is much lower for a monolithic column as compared to particle packed columns. This extends column lifetime substantially when analyzing alcohol extracts of plants that only undergo filtration as sample preparation step.
The unique pore structure of monolithic silica HPLC columns, comprising macro- and mesopores, provides lower column backpressure compared to particle packed columns. Even at elevated flow rates and even with more viscous mobile phase solvents, they offer high peak capacity and increased sample throughput.

To make the total analysis easier and less time consuming, it is possible to use a Samplicity® system for parallel filtration of up to eight samples. The Samplicity® provides a convenient, high throughput alternative to syringe-tip filters when preparing samples for chromatography.
The easy to use Samplicity® system is the first vacuum-driven system with the designed in flexibility to filter 1 to 8 samples directly into standard HPLC vials. Just attach a vacuum pump, load samples with a standard pipettor, and flip the lever to recover particulate-free samples—even those with high viscosity or particulates—in seconds. In addition, the system has a low hold-up volume, which allows processing of samples as small as 300 µL.

Components Supplied



Thus combining the Samplicity® system for filtration of alcohol extracted plant materials and analysis with monolithic Chromolith® columns, both sample throughput and peak capacity can be optimized.
On the following pages two applications are shown for the comparative analysis of **Panax Notoginseng** and **Pueraria** using both monolithic and particle packed columns. In addition there are a 8 more TCM's analyzed with Chromolith® columns only as we believe this column is the most suitable alternative for the analysis of alcohol extracted plant materials.

Panax Notoginseng

田七 (Tiánqī), Tienchi Ginseng, San Qi (三七) or Sanchi

Panax notoginseng belong to the genus Panax, and it is most commonly referred to as notoginseng in English. In Chinese it is called 田七 (tiánqī), tienchi ginseng, san qi (三七) or sanchi, three-seven root, and mountain paint. Notoginseng belongs to the same scientific genus as Asian ginseng. In Latin, the word panax means "cure-all", and the family of ginseng plants is one of the most well-known herbs. Notoginseng grows naturally in China and Japan.

We have analysed the same alcohol extracted sample with both a traditional particle packed column (page 10) and a monolithic Chromolith® HighResolution column (page 11).
Both columns were re-equilibrated after each analysis with five column volumes each to provide good reproducibility. Despite the fact that latter column is only 100 mm and the particle packed column was 250 mm long, better resolution of the four target molecules could be attained with the Chromolith® column.

The total analysis time is 30 min shorter with the Chromolith® column.

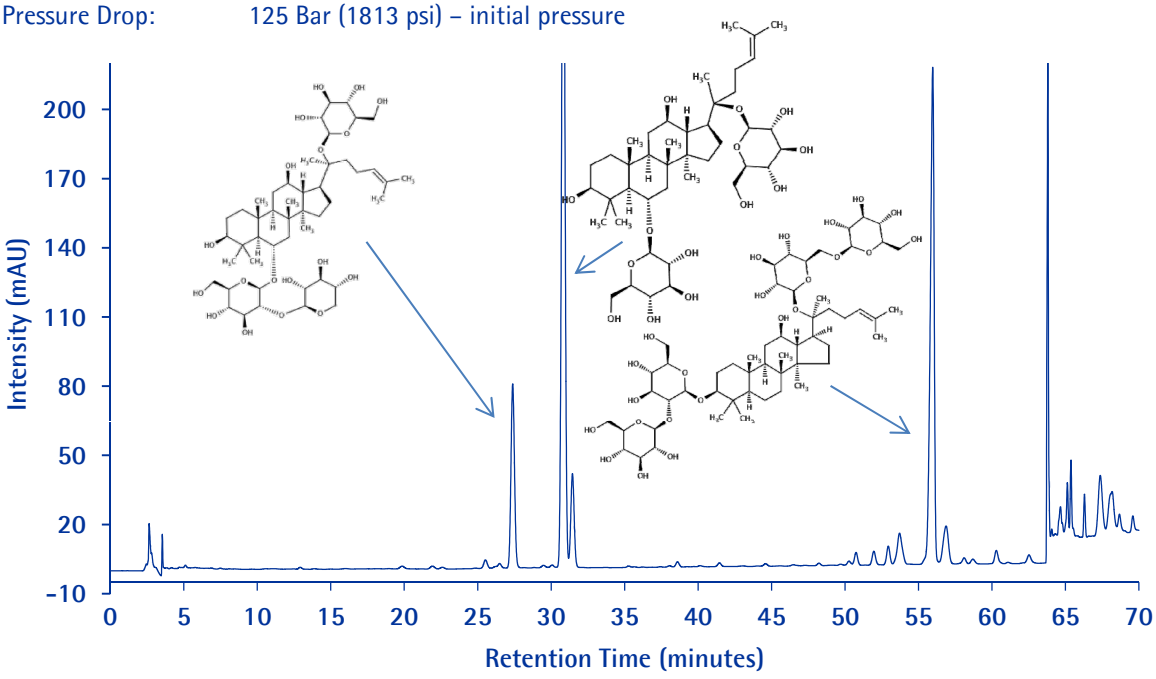
Panax Notoginseng

Particle packed column RP-18 endcapped

Chromatographic Conditions

Column:	Particle packed column, RP-18 endcapped (5µm), 250x4.6mm
Injection:	10 µl
Detection:	UV 203 nm
Flow Rate:	1.5 mL/min
Mobile Phase :	A: Milli-Q water
	B: Acetonitrile
Gradient:	See table.
Temperature:	25°C
Diluent	Mobile phase
Sample:	The extract of panax notoginseng dissolved in methanol.
Pressure Drop:	125 Bar (1813 psi) – initial pressure

Time (min)	% A	% B
0-12	19	81
60	36	64
70	70	30
70.1	19	81
90	19	81



Chromatographic Data

No.	Compound	Retention Time (min)	Theoretical Plates*	Asymmetry
1	R1	27.4	59396	1.1
2	Rg1	30.8	80584	1.0
3	Re	31.4	85373	1.0
4	Rb1	56.0	214486	0.6

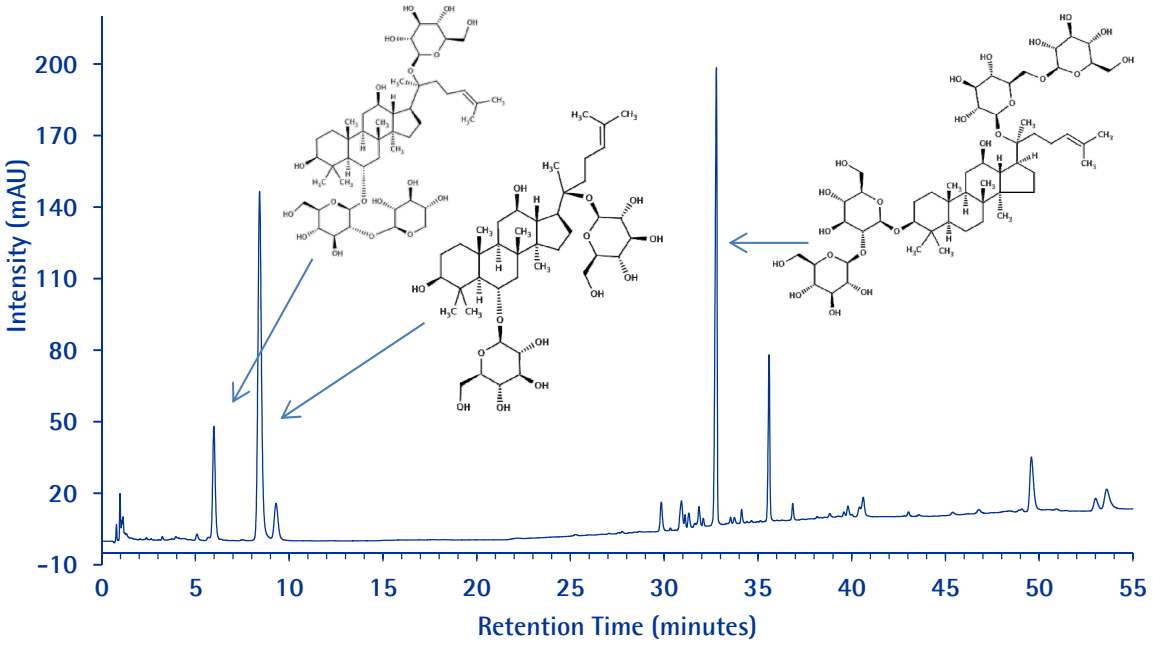
Panax Notoginseng

Chromolith® HighResolution RP-18 endcapped

Chromatographic Conditions

Column:	Chromolith® HighResolution RP-18 endcapped, 100x4.6mm	1.52022.0001
Injection:	10 µl	
Detection:	UV 203 nm	
Flow Rate:	1.5 mL/min	
Mobile Phase :	A: Milli-Q water	
	B: Acetonitrile	
Gradient:	See table	
Temperature:	25°C	
Diluent	Mobile phase	
Sample:	The extract of panax notoginseng dissolved in methanol.	
Pressure Drop:	77 Bar (1165 psi) – initial pressure	

Time (min)	% A	% B
0-20	19	81
45	45	55
55	55	45
55.1	19	81
60	19	81



Chromatographic Data

No.	Compound	Retention Time (min)	Theoretical Plates*	Asymmetry
1	R1	6.0	9105	1.1
2	Rg1	8.5	9853	1.2
3	Re	9.3	9058	1.2
4	Rb1	32.8	425154	0.5

Radix Puerariae

Ge Gen, 葛根, Gan Ge, 干葛

Pueraria is a genus of 15–20 species of plants native to Asia. Pueraria flower is used in traditional Chinese medicine to reduce reactions to alcohol consumption. Pueraria lobata (wild.) Ohwi. is mainly produced in Hunan, Henan, Guangdong provinces of China, while Pueraria thomsonii Benth. is cultivated artificially, mainly produced in Guangxi and Guangdong provinces of China. Pueraria mirifica, also known as กวาวเครือ Kwao Krua (but not exclusively), is a plant found in northern and north eastern Thailand and Myanmar.

We have analysed the same alcohol extracted sample for Puerarin with both a traditional particle packed column (page 13) and a monolithic Chromolith® HighResolution column (page 14). Both columns were re-equilibrated after each analysis with five column volumes each to provide good reproducibility.

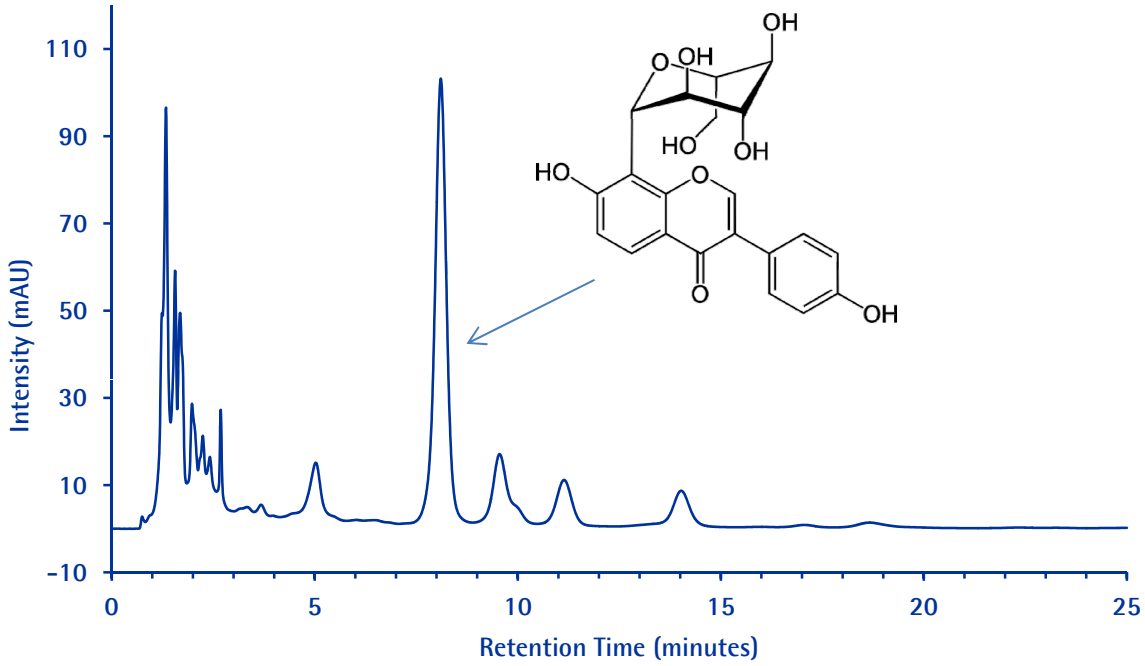
Much higher separation efficiency was attained with the Chromolith® column, and maintaining sufficient resolution between puerarin and the other matrix components. The sensitivity was also higher when using the monolithic column because of faster elution and better peak compression. The total analysis time is 15 min shorter with the Chromolith® column and the column backpressure is much lower.

Radix Puerariae

Particle packed column RP-18 endcapped

Chromatographic Conditions

Column:	Particle packed column, RP-18 endcapped (5µm), 250x4.6mm
Injection:	10 µl
Detection:	UV 250 nm
Flow Rate:	1.0 mL/min
Mobile Phase :	Methanol and Water 25:75 (v/v)
Temperature:	40°C
Diluent	Mobile phase
Sample:	The extract of radix pueraria sample dissolved in methanol.
Pressure Drop:	100 Bar (1440 psi)



Chromatographic Data

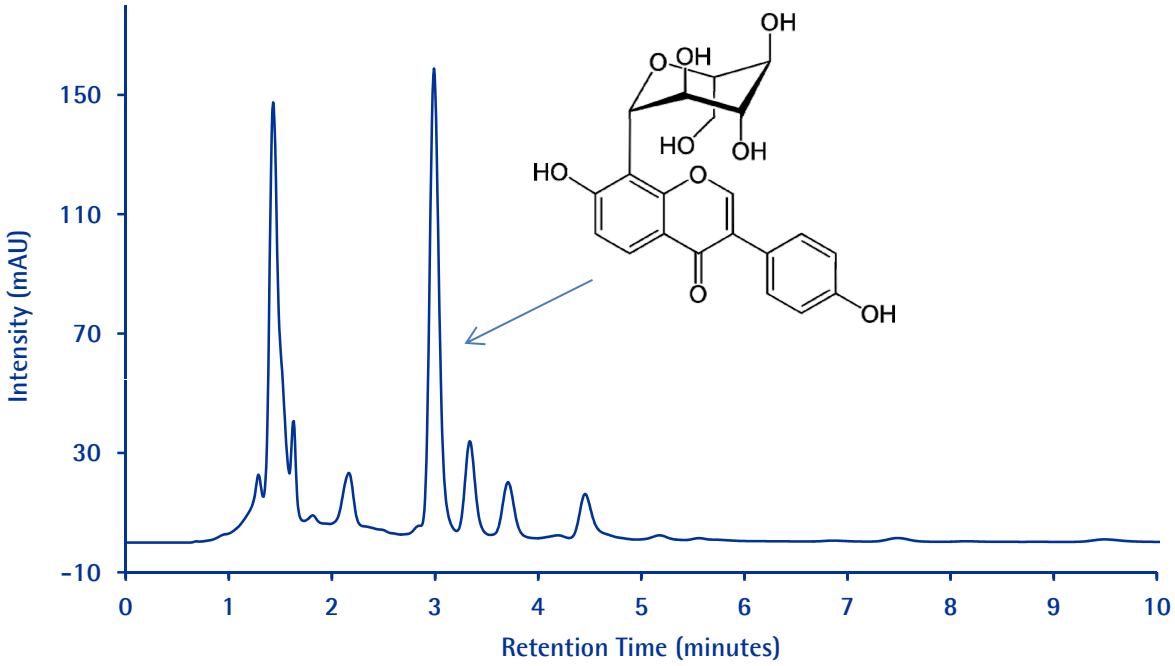
No.	Compound	Retention Time (min)	Theoretical Plates*	Asymmetry
1	Puerarin	8.1	4120	1.0

Radix Puerariae

Chromolith® HighResolution RP-18 endcapped

Chromatographic Conditions

Column:	Chromolith® HighResolution RP-18 endcapped, 100x4.6mm	1.52022.0001
Injection:	5 µl	
Detection:	UV 250 nm	
Flow Rate:	1.0 mL/min	
Mobile Phase :	Methanol and Water 25:75 (v/v)	
Temperature:	25°C	
Diluent	Mobile phase	
Sample:	The extract of panax notoginseng dissolved in methanol.	
Pressure Drop:	43 Bar (619 psi)	



Chromatographic Data

No.	Compound	Retention Time (min)	Theoretical Plates*	Asymmetry
1	Puerarin	3.0	5086	1.2

Radix Angelica Sinensis

Dang Gui or 当归

Angelica is a type of about 60 species of tall biennial and perennial herbs in the Apiaceae family. It can be found in temperate and subarctic regions of the Northern Hemisphere, reaching as far north as Iceland and Lapland. They grow to 1–3 m tall, with large bipinnate leaves and large compound umbels of white or greenish-white flowers. Some species can be found in purple moor and rush pastures. It has a strong fragrance, being sweet, pungent and slightly bitter.

Ferulic acid or hydroxycinnamic acid is an abundant phenolic phytochemical found in plant cell wall components such as arabinoxylans as covalent side chains. It is related to trans-cinnamic acid. As a component of lignin, ferulic acid is a precursor in the manufacture of other aromatic compounds.

Ligustilide is an abundant bioactive ingredient believed to have anti-oxidant properties.

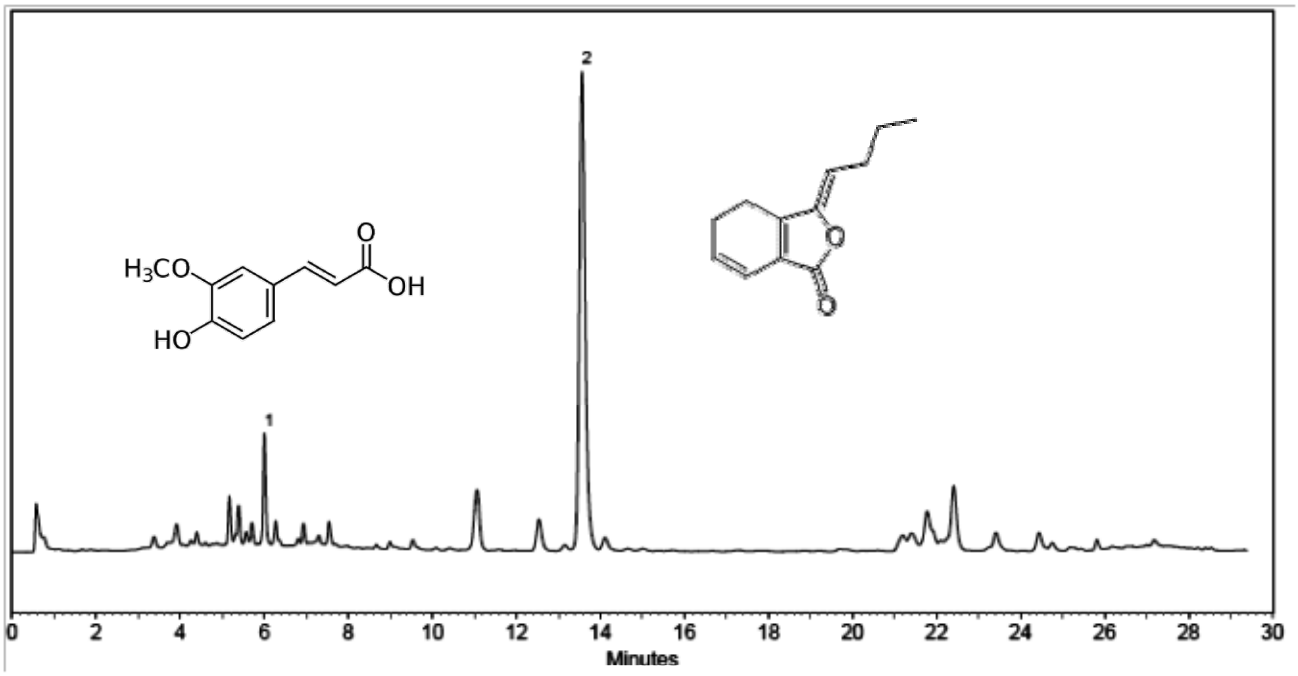
Radix Angelica

Chromolith® RP-18 endcapped

Chromatographic Conditions

Column:	Chromolith® Performance RP-18 endcapped 100x4.6mm	1.02129.0001
Injection:	5 µL	
Detection:	UV 278 nm	
Flow Rate:	3.0 mL/min	
Mobile Phase:	A: Acetonitrile	
	B:0.1 % phosphoric acid	
Temperature:	Ambient	
Diluent	Mobile phase	
Sample:	Alcohol extract of roots	

Time (min)	% A	% B
0	5	95
1	5	95
6	32	68
16	43	57
24	60	40
26	90	10



Chromatographic Data

No.	Compound	Time (min)	T_{USP}	Resolution
1	Ferulic acid	6.1	1.1	-
2	Ligustilide	13.7	1.2	-

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http://www.merckmillipore.com/chromatography
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Coptis Chinensis (Chinese Goldthread)

Duǎn è Huánglián，短萼黄连

Coptis chinensis or Chinese goldthread is one of the 50 fundamental herbs used in traditional Chinese medicine, and is a source for the isoquinoline alkaloids; berberine, palmatine, hydrastine, and coptisine among others. Because of the strong coloring from berberine, it has also been used as a dye, especially for wool and other fibers.

Jatrorrhizine is a protoberberine alkaloidt is believed to have antiinflammatory effects, and to improve blood flow, as well as having antimicrobial and antifungal activity.

Berberine is a quaternary ammonium salt from the protoberberine group of isoquinoline alkaloids. It has a strong yellow color and in earlier days it was used to dye wool, leather and wood. It is considered antibiotic and with some activity to fungal infections.

Tetrahydropalmatine (THP) is an alkaloid found in several different plant species, mainly in the Corydalis genus (Yan Hu Suo). The pharmaceutical industry has synthetically produced the more potent enantiomer Levo-tetrahydropalmatine (Levo-THP), which has been marketed worldwide under different brand names as an alternative to anxiolytic and sedative drugs of the benzodiazepine group and analgesics such as opiates. It is also sold as a dietary supplement.

Coptis Chinensis (Chinese Goldthread)

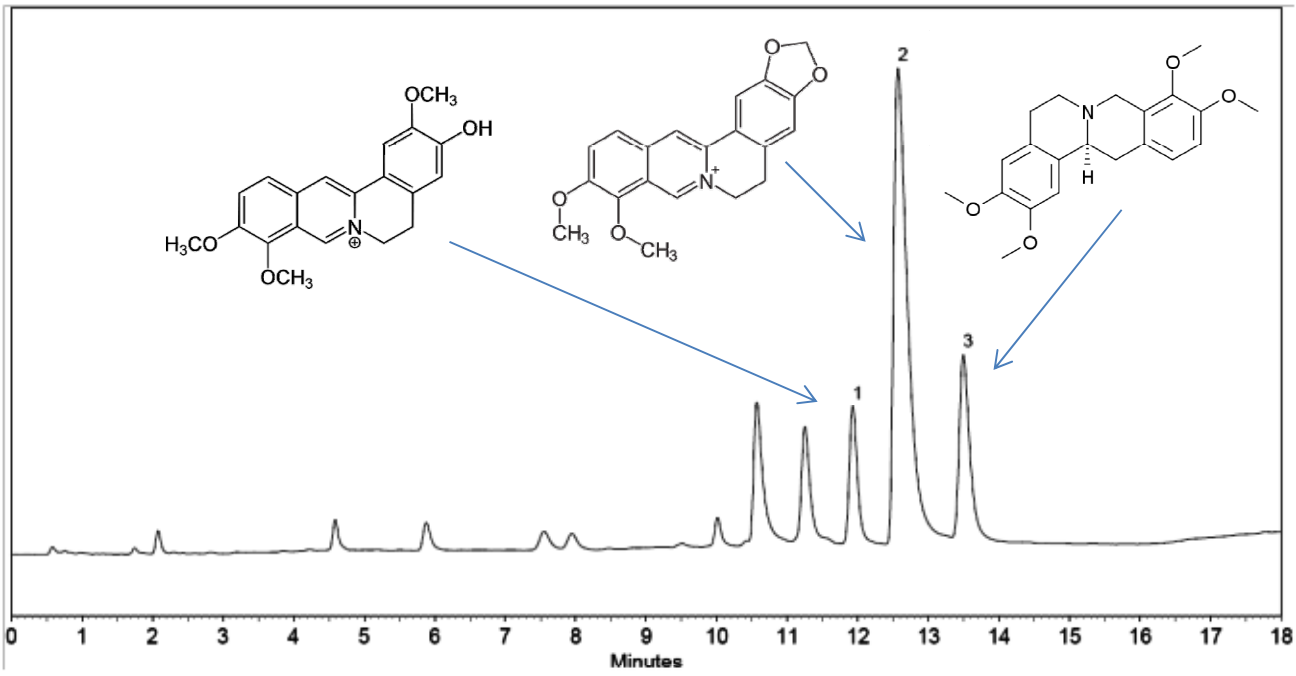
Chromolith® RP-18 endcapped

Chromatographic Conditions

Column:	Chromolith® Performance RP-18 endcapped 100x4.6mm
Injection:	5 µL
Detection:	UV 280 nm
Flow Rate:	3.0 mL/min
Mobile Phase:	A: Acetonitrile
	B:0.1 % phosphoric acid
Temperature:	30°C
Diluent	Mobile phase
Sample:	Alcohol extract of roots

1.02129.0001

Time (min)	% A	% B
0	7	93
2	10	90
3	17	83
7	20	80
8	27	73
15	35	65



Chromatographic Data

No.	Compound	Time (min)	T_{USP}	Resolution
1	Jatrorrhizine	11.9	1.4	
2	Berberine	12.6	2.8	-
3	Tetrahydropalmatine (THP)	13.6	1.6	-

Fructus Forsythiae

Lian Qiao , 连翘

Forsythia is a genus of flowering plants in the family Oleaceae (olive family).
There are about 11 species, mostly native to eastern Asia, but one native to southeastern Europe. They are shrubs typically growing to a height of 1–3 m with rough grey-brown bark. The leaves are usually simple but sometimes trifoliate and range from 2–10 cm in length. The actual fruit is a dry capsule, containing several winged seeds. It has slightly fragrant smell and bitter taste.

Caffeic acid is also known as hydroxycinnamic acid.
It is found in all plants because it is a key intermediate in the biosynthesis of lignin, one of the principal components of plant biomass and its residues.

Rutin or rutoside, quercetin-3-O-rutinoside and sophorin, is the glycoside between the flavonol quercetin and the disaccharide rutinose (α-L-rhamnopyranosyl-(1→6))-β-D-glucopyranose). Rutin is a citrus flavonoid glycoside found in many plants.

Forsythin is believed to have antibacterial effect.

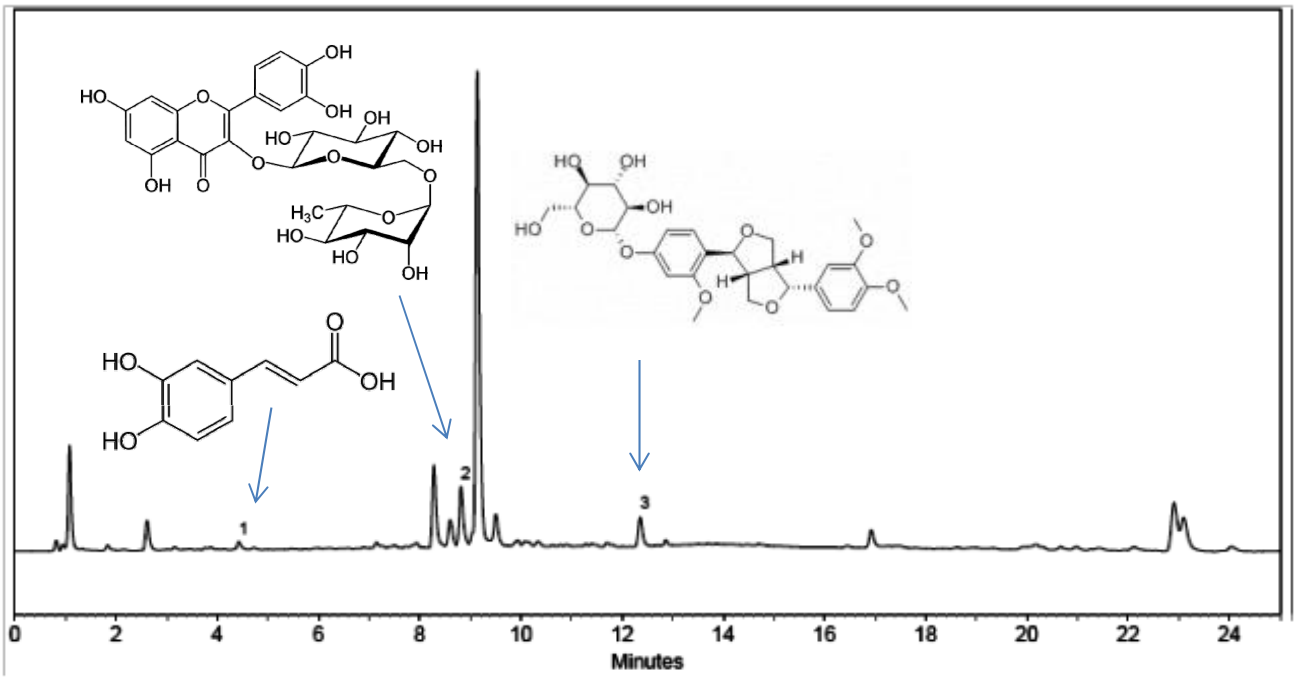
Fructus Forsythiae

Chromolith® RP-18 endcapped

Chromatographic Conditions

Column: Chromolith® Performance RP-18 endcapped 100x4.6mm 1.02129.0001
Injection: 5 µL
Detection: UV 265 nm
Flow Rate: 2.0 mL/min
Mobile Phase: A: Acetonitrile
B:0.1 % phosphoric acid
Temperature: Ambient
Diluent Mobile phase
Sample: Alcohol extract of seeds

Time (min)	% A	% B
0	5	95
10	23	77
12	35	65
22	60	40
25	95	5



Chromatographic Data

No.	Compound	Time (min)	T_{USP}	Resolution
1	Caffeic acid	4.4	1.3	
2	Rutin	8.8	1.2	-
3	Forsythin	12.4	1.2	-

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Radix Glycyrrhizae (Liquorice)

Gan Cao, 甘草

Liquorice or licorice is the root of Glycyrrhiza glabra from which a sweet flavor can be extracted. The liquorice plant is a legume that is native to southern Europe and parts of Asia. It is not botanically related to anise, star anise, or fennel, which are sources of similar flavoring compounds. In traditional Chinese medicine, liquorice is commonly used in herbal formulae to "harmonize" the other ingredients in the formula.

Glycyrrhetinic acid or Enoxolone is a pentacyclic triterpenoid derivative of the beta-amyrin type obtained from the hydrolysis of glycyrrhizic acid. It is used in flavoring and it masks the bitter taste of drugs like aloe and quinine. It is believed to have antiviral, antifungal, antiprotozoal, and antibacterial activities.

Glycyrrhizin (or glycyrrhizic acid or glycyrrhizinic acid) is the main sweet-tasting constituent of the liquorice root. It has been used intravenously in Japan as a treatment for hepatitis C and as an emulsifier and gel-forming agent in foodstuff and cosmetics.

Radix Glycyrrhizae (Liquorice)

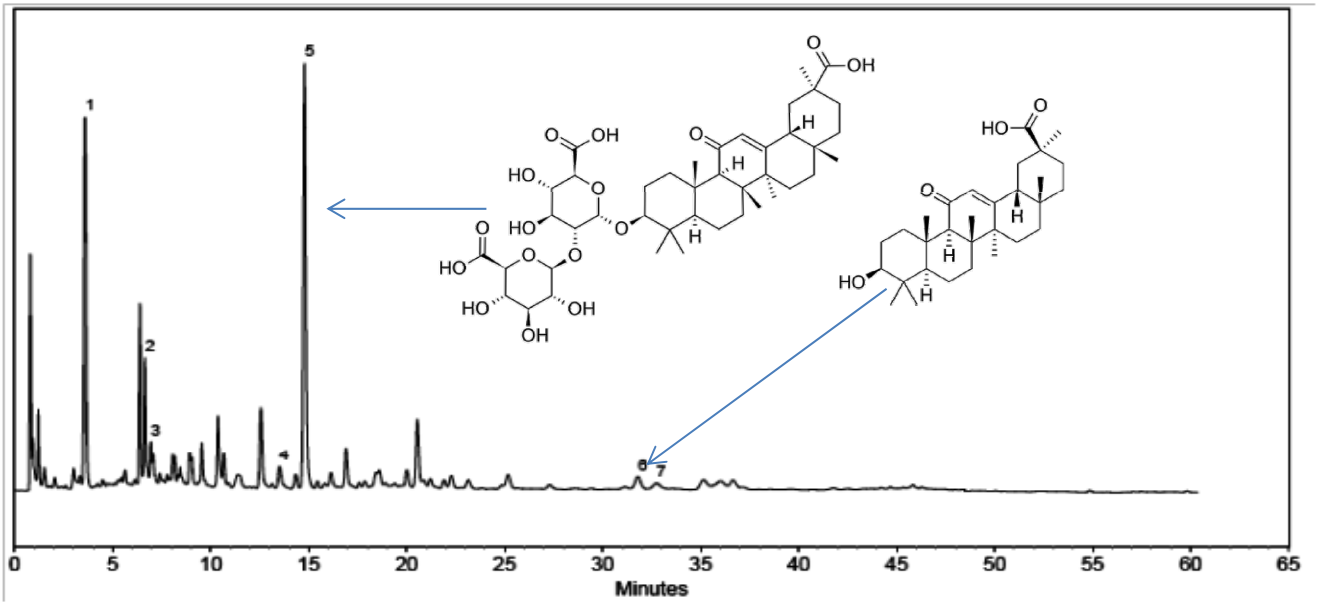
Chromolith® RP-18 endcapped

Chromatographic Conditions

Column:	Chromolith® Performance RP-18 endcapped 100x4.6mm
Injection:	5 µL
Detection:	UV 250 and 276 nm
Flow Rate:	2.0 mL/min
Mobile Phase:	A: Acetonitrile
	B:Milli-Q water
Temperature:	30 °C
Diluent	Mobile phase
Sample:	Alcohol extract of roots

1.02129.0001

Time (min)	% A	% B
0	15	85
1	15	85
6	28	72
15	35	65
18	40	60
40	50	50
50	80	20



Chromatographic Data

No.	Compound	Time (min)	T_{USP}	Resolution
1	Liquiritin	3.6	-	-
2	Isoliquiritin	6.7	-	-
3	Liquiritigenin	7.0	-	-
4	Iquiritigenin	13.5	2.3	-
5	Glycyrrhizic Acid	15.0	1.3	-
6	Glycyrrhetinic acid	31.8	1.2	-
7	ß-Glycyrrhetinic Acid	32.8	1.3	-

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Radix Scrophulariae

Xuan Shen, 玄参, 元参, Yuan Shen

Radix Scrophulariae originates from China's Yangtze River basin and other areas like Shaanxi, Fujian Province. Another herb belonging to the same genus scrophularia is named Scrophularia buergeriana and it is also used as herb medicine. It has a special smell similar to burnt sugar; bitter and slightly sweet.

Cinnamic acid is an unsaturated carboxylic acid, it occurs naturally in a number of plants. It exists as both as cis- and trans-isomer, although the latter is more common. Cinnamic acid has a honey-like odor.

Harpagoside is a glycoside and believed to have anti-inflammatory effects.

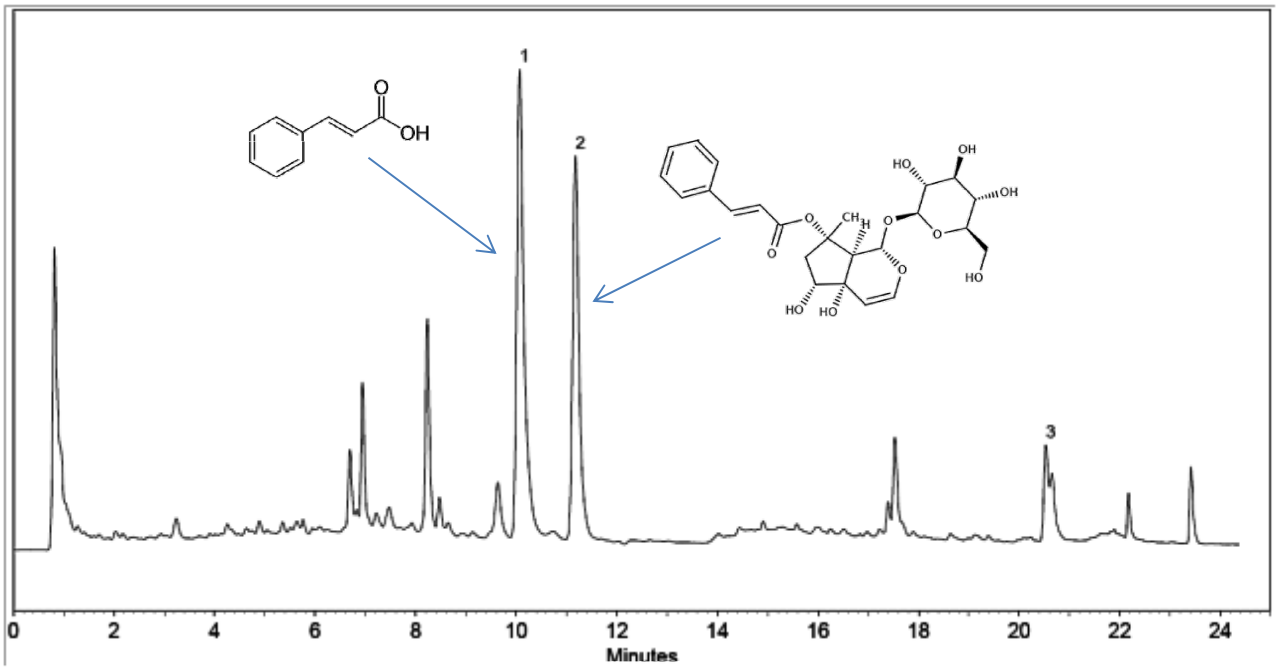
Radix Scrophulariae

Chromolith® RP-18 endcapped

Chromatographic Conditions

Column:	Chromolith® Performance RP-18 endcapped 100x4.6mm	1.02129.0001
Injection:	5 µL	
Detection:	UV 278 nm	
Flow Rate:	2.0 mL/min	
Mobile Phase:	A: Acetonitrile	
	B:0.1 % phosphoric acid	
Temperature:	30°C	
Diluent	Mobile phase	
Sample:	Alcohol extract of roots	

Time (min)	% A	% B
0	5	95
6	22	78
12	25	75
14	44	56
20	66	34
20.1	95	5



Chromatographic Data

No.	Compound	Time (min)	T_{USP}	Resolution
1	Cinnamic acid	10.2	1.6	-
2	Harpagoside	11.2	1.3	-

Rheum Palmatum

Da-Huang, 大黄

Rheum Palmatum or Chinese rhubarb, Turkish rhubarb, Turkey rhubarb,, Indian rhubarb, Russian rhubarb or rhubarb root. Currently it is estimated that more than 800 types of compounds in the Chinese medicines that contain rhubarb. Rheum palmatum has anthraquinone glycosides and dianthrone glycosides, which are the main reason why it is used as a laxative. Anthraquinone glycosides include Chrysophanol-1-monoglucoside or Chrysophaein, Emodin-6-monoglucoside, Aloe-emodin-8-monoglucoside, Physcion monoglucoside, Rhein-8-monoglucoside. Dianthrone glycosides include Sennoside A、B、C、D、E、F. Free anthraquinones include Chrysophanol, Emodin, Physcion, Aloe-emodin, Rhein etc. In addition, rhubarb also contains fatty acid, calcium oxalate, glucose, fructose and starch.

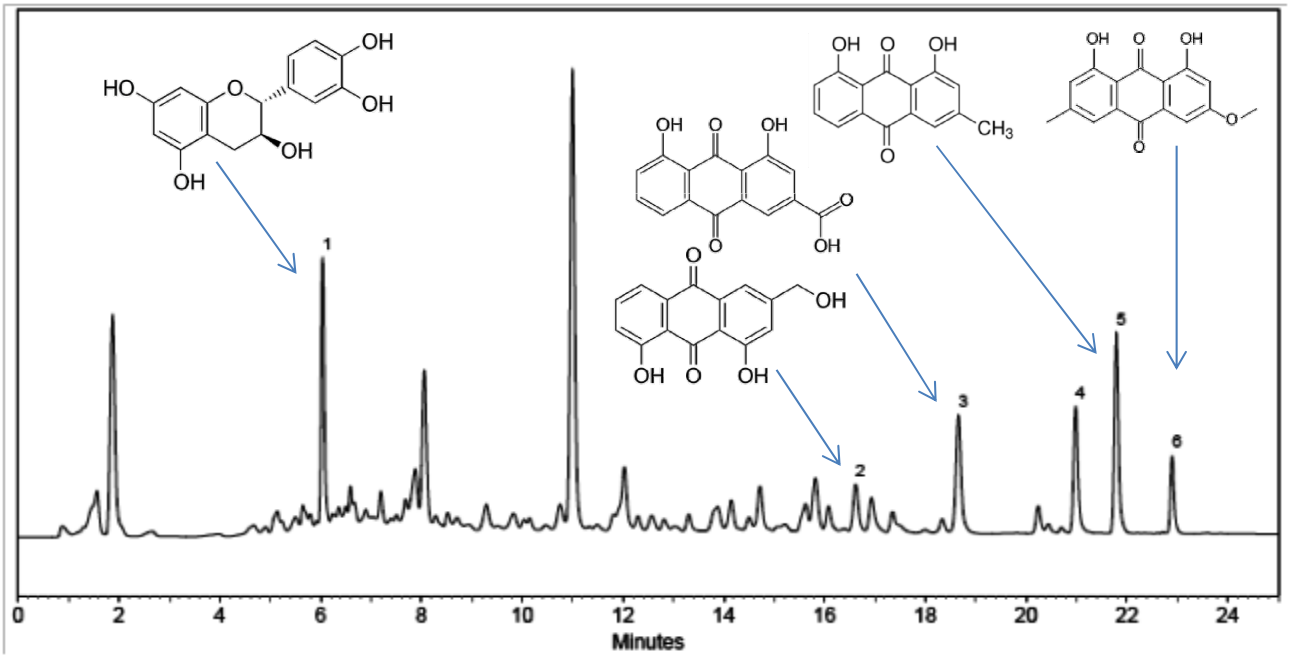
Rheum Palmatum

Chromolith® RP-18 endcapped

Chromatographic Conditions

Column: Chromolith® Performance RP-18 endcapped 100x4.6mm 1.02129.0001
Injection: 5 µL
Detection: UV 254 nm
Flow Rate: 2.0 mL/min
Mobile Phase: A: Methanol
B:Milli-Q water
Temperature: 30 °C
Diluent Mobile phase
Sample: Alcohol extract of roots

Time (min)	% A	% B
0	5	95
2	5	95
4	25	75
18	70	30
22	95	5



Chromatographic Data

No.	Compound	Time (min)	T_{USP}	Resolution
1	Catechin	6.1	1.1	-
2	Aloe Emodin	16.6	1.2	-
3	Rhein	18.6	1.4	-
4	Emodin	21.0	1.4	-
5	Chrysophanol	21.8	1.4	-
6	Physcion (Parietin)	22.8	1.4	-

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Salvia Miltiorrhiza

Dānshēn, 丹参

Salvia miltiorrhiza, also known as red sage, Chinese sage, tan shen, or danshen, is a perennial plant in the Salvia family. The dried root of Salvia miltiorrhiza, is a Chinese medicine used to promote blood flow and treat vascular disease. The main active constituents of salvia are diterpene quinones, known as tanshinones. Most of these compounds are colored, providing the reddish appearance of the roots. The tanshinones are unique chemical constituents, and similar compounds are not found in other Chinese herbs.

Salvia Miltiorrhiza

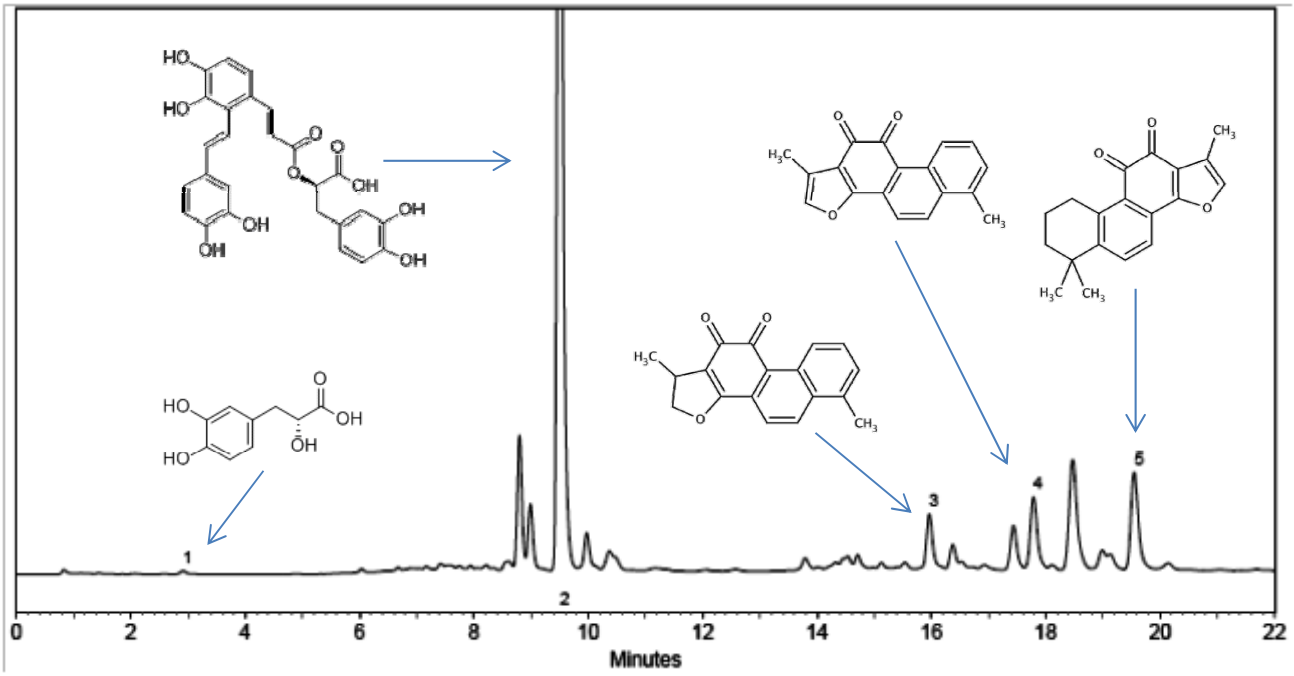
Chromolith® RP-18 endcapped

Chromatographic Conditions

Column:	Chromolith® Performance RP-18 endcapped 100x4.6mm
Injection:	5 µL
Detection:	UV 254 nm
Flow Rate:	2.0 mL/min
Mobile Phase:	A: Acetonitrile
	B::0.1 % phosphoric acid
Temperature:	30 °C
Diluent	Mobile phase
Sample:	Alcohol extract of roots

1.02129.0001

Time (min)	% A	% B
0	3	97
3	3	97
5	20	80
11	33	67
12	46	54
20	70	30
22	95	5



Chromatographic Data

No.	Compound	Time (min)	T_{USP}	Resolution
1	Salvianic acid A	2.9	1.3	-
2	Salvianolic acid B	9.5	1.2	-
3	Dihydrotanshinone	15.9	1.3	-
4	Tanshinone I	17.8	1.4	-
5	Tanshinone IIA	19.6	1.4	-

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Shuanghuanglian Oral Liquid Solution

Shuānghuánglián shuān, 双黄连口服液

Shuang Huang Lian is a Chinese herbal remedy that is used to treat respiratory tract infections. Shuang Huang Lian formulations consist of at least 29 components, including shuang hua (Lonicera japonica flower bud), lian qiao (Forsythia suspensa fruit), and huang qin (Scutellaria baicalensis).

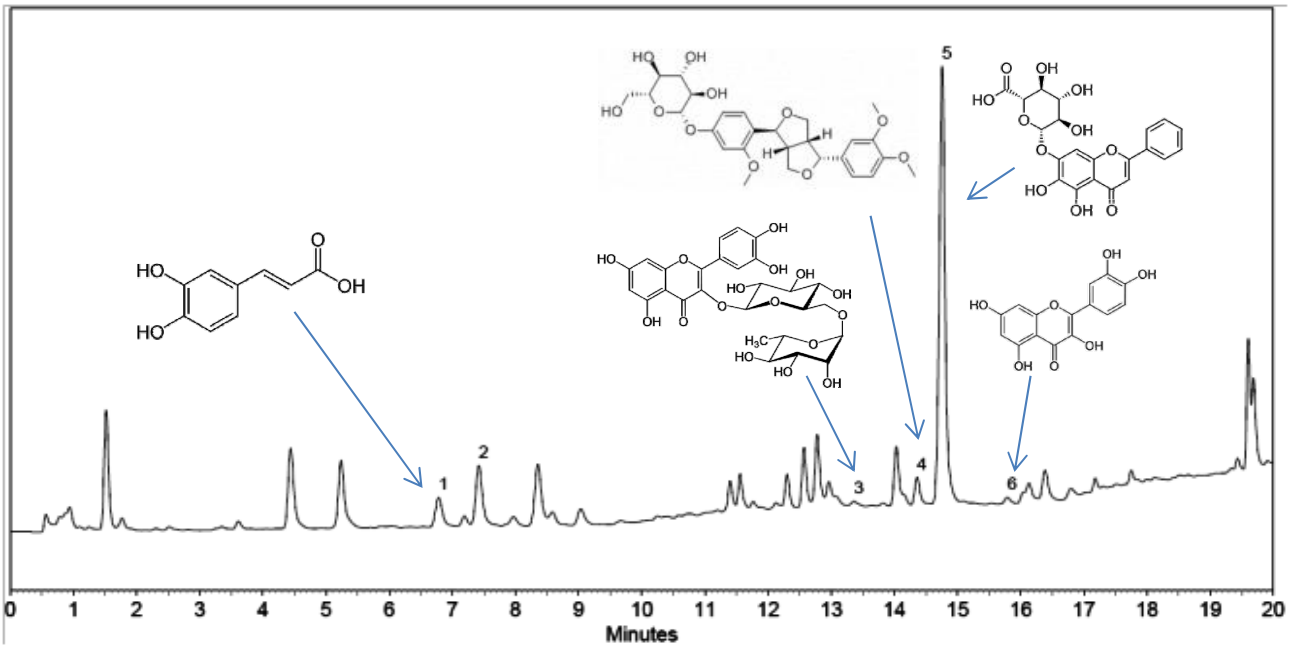
Shuanghuanglian Oral Liquid Solution

Chromolith® RP-18 endcapped

Chromatographic Conditions

Column: Chromolith® Performance RP-18 endcapped 100x4.6mm 1.02129.0001
Injection: 5 µL
Detection: UV 265 nm
Flow Rate: 3.0 mL/min
Mobile Phase: A: Methanol
B:0.1 % phosphoric acid
Temperature: 30 °C
Diluent Mobile phase
Sample: Oral liquid solution diluted 1:1 (v/v) with mobile phase

Time (min)	% A	% B
0	3	97
0.5	3	97
8	17	83
10	30	70
15	45	55
20	95	5



Chromatographic Data

No.	Compound	Time (min)	T_{USP}	Resolution
1	Caffeic acid	6.1	1.2	-
2	Chlorogenic acid	16.6	1.3	-
3	Rutin	18.6	1.4	-
4	Forsythin	21.0	1.4	-
5	Baicalin	21.8	1.4	-
6	Quercetin	22.8	1.4	-

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Solvents and Reagents

Product	P/N
Acetonitrile Gradient Grade for Chromatography	1.00030
Acetonitrile for Chromatography	1.14291
Methanol Gradient Grade for Chromatography	1.06007
ortho-Phosphoric acid 85% for analysis EMSURE® ACS,ISO,Reag. Ph Eur	1.00573
Water for chromatography*	1.15333
* or use a Milli-Q Integral Water Purification System	
Bold blue Samplicity system bundle with 0.45 µm PTFE philic Millex Samplicity filters	SAMPLCRBL
Glossy green Samplicity system bundle with 0.45 µm PTFE philic Millex Samplicity filters	SAMPLCRGR
Bold blue Samplicity system bundle with 0.45 µm PVDF philic Millex Samplicity filters	SAMPHV0BL
Bold blue Samplicity system bundle with 0.2 µm PTFE philic Millex Samplicity filters	SAMPLG0BL
Glossy green Samplicity system bundle with 0.2 µm PTFE philic Millex Samplicity filters	SAMPLG0GR
Glossy green Samplicity system bundle with 0.45 µm PVDF philic Millex Samplicity filters	SAMPHV0GR

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