UPLC-MS based metabolic profiling of the phenotypes of *Acanthopanax senticosus* reveals the changes in active metabolites distinguishing the diversities of the plant grown in northeast area of China

SUN Hui 1, HAN Ying 1, ZHANG Ai-Hua 1, MENG Xiang-Cai 1*, WANG Zhen-Yue 2, SUN Wen-Jun 1, SUN Hai-Feng 2, WANG Xi-Jun 1*

1National TCM Key Laboratory of Serum Pharmaco-Chemistry, Heilongjiang University of Chinese Medicine, and Key Pharmacometabolomics Platform of Chinese Medicines, Harbin 150040, China; 2Department of Chinese Herbal Medicinal Resource, Heilongjiang University of Chinese Medicine, Harbin 150040, China

Available online 20 May 2012

[ABSTRACT] *Acanthopanax senticosus* (Rupr.et Maxim.) Harms. (AS) is a medicinal plant which mainly distributes in Northeastern area of China, due to the widely utilization in clinical and pharmaceutical industry, the wild resource of the plant is limited, the cultivation of the plant is in needed currently. Therefore it is of importance to select a medicinal priority for cultivating from two different sexual type of AS. In the present study, UPLC-MS technology-based metabolic profiles was used to discriminate phenotypes. By processing the metabolomic data with principal component analysis (PCA), partial least squares-discriminant analysis (PLS-DA) and orthogonal projection to latent structures (OPLS) analysis, eleven secondary metabolites were found to be responsible for classing the two varieties of *Acanthopanax senticosus*, they might be taken as potential biological markers of sexual type. These marker metabolites include flavone, coumarin, terpenes and organic acids, which are the main active constituents related to the therapeutic effects of *Acanthopanax senticosus*. Among these valuable secondary metabolites, flavone, anthraquinone and organic acids are highly expressed in short capillament *Acanthopanax senticosus*, while coumarin and terpenoid are highly expressed in the long capillament variety. As a result, by monitoring these highly expressed metabolites and through selective cultivation, an *Acanthopanax senticosus* with higher amounts of medicinally relevant constituents could be created.

[KEY WORDS] *Acanthopanax senticosus*; Araliaceae; Phenotype; Metabolomics; UPLC-MS; Secondary metabolite


1 Introduction

Metabolomics is the comprehensive assessment of endogenous metabolites of a biological system. It is able to provide a “holistic view” of the metabolites and allows the simultaneous detection of a wide range of compounds, providing an immediate image of the metabolome of a plant. The analysis of the molecular phenotype using metabolomics will reveal a novel understanding of plant metabolism and its interaction with the environment as well distinguishing the diversities of the plant. Technological advances in metabolomics have facilitated to diversification in plant secondary metabolism. Metabonomic analysis is an important method for characterising plant ecotypic variations; hence, it may become a powerful tool for plant phenotypes.

*Acanthopanax senticosus* (Rupr.et Maxim.) Harms. (*AS, Ciwujia* in Chinese) belongs to the *Araliaceae* family and is a medicinal plant distributed mainly in northeast China. Its roots and leaves have been used as crude drugs that have a...
wide range of bioactivities such as antioxidant [4], fatigue reduction [5], anti-inflammation [6], immune system regulation [7-8], myocardium protection [9], etc. In China and some other Asia countries, many patented medicines such as *Ciwujia Injecta* and *Ciwujia Tablet* contain AS, the consumption of AS has dramatically increased recently. By the end of 2008, the yearly consumption of AS was nearly 5 000 tons, which is far beyond the wild resource supply [10]. Further more, AS is a rare and endangered species [11], wild resource is being protected from collecting, AS must be cultivated to meet the increasing need of AS originated crude drug in the market. However AS has phenotypes in its habits, it should be of importance to select a medicinal priority variety from the phenotypes to be cultivated.

AS has mainly two different sexual types, that is AS with long capillament which length is between 3.96 cm and 4.44 cm, and AS with short capillament which length between 0.45 cm and 0.93 cm (Fig. 1). The genetic diversity of the phenotypes has been assessed previously [12-13], but the differentiate in secondary metabolites between the two varieties is still unclear, which quality is better for medicinal use is unknown either. The present study is to discriminate the phenotypes of AS by analyzing metabolic profiles based on the UPLC-MS technology to select a priority variety to cultivate for medical purpose.

Fig. 1 Flower of *Acanthopanax senticosus* with different length capillament. These two varieties were collected from Xiangyang Farm of Tieli Forest Enterprise in Heilongjiang Province at July, 2008. Each varieties were collected in 9 stocks with space between of 5 meters. (a) long capillament variety: capillament length between 3.96 cm and 4.44 cm; (b) short capillament variety: capillament length between 0.45 cm and 0.93 cm

2 Experimental

2.1 Chemicals

Acetonitrile, HPLC grade, was purchased from Merck Company, Inc. (Darmstadt, Germany). Methanol, HPLC grade, was purchased from Fisher Company, Inc. (New Jersey, USA). Distilled water was purchased from Watsons Water Company, Inc. (Guangzhou, China). Formic acid of analytical grade was purchased from Ke Oumi Chemical Reagent Company (Tianjin, China). Leucine enkephalin was purchased from Sigma Aldrich (MO, USA).

2.2 Plant samples

Samples of different phenotype of AS were collected from Xiang Yang Forest Farm of Tieli Forest Enterprise in Heilongjiang Province at July, 2008; long and short capillament AS were collected in 9 stocks respectively with space between of 5 meters. All samples were identified by Prof. WANG Xi-Jun, Pharmacognosy Department, Heilongjiang University of Chinese Medicine. The samples were immediately frozed in liquid nitrogen prior to stored at –70 °C until further processing.

2.3 Analysis sample preparation

About 1g fresh leaves of AS was homogenized with 20 mL of methanol, after ultrasonic treatment for 30 min, the homogenate was centrifuged for 5 min at 13 000 r·min⁻¹, the supernatant was filtered with micropore film (0.22 μm) for analysis.

2.4 Chromatography condition

Chromatographic separation was performed on an Acquity UPLC BEH C₁₈ column (1.7 μm, 2.1 mm × 50 mm, Waters Corporation, Milford, USA) using an Acquity ultra performance liquid chromatography system equipped with an Acquity photodiode array detector (Waters, Milford, USA). The composition of mobile phase and its gradient program was showed in Table 1. The total flow rate was 0.5 mL·min⁻¹, the column temperature was maintained at 35 °C and the elute was directed to the mass spectrometer without split.

2.5 Mass spectrometry

Mass spectrometry was performed on a Waters Synapt™ High Definition MS (Waters MS Technologies, Manchester, UK) with an electrospary ionization source (ESI) operating in positive and negative ion mode. The desolvation gas rate was set to 550 L·h⁻¹ at a temperature of 250 °C, the capillary voltage and cone voltage were set at 2.3 KV, 45 V in negative ion mode, and the desolvation gas rate was set to 600 L·h⁻¹ at a temperature of 300 °C, the capillary voltage and cone volt-
Table 1 Solvent gradient program for UPLC analysis

<table>
<thead>
<tr>
<th>t/min</th>
<th>A/%</th>
<th>B/%</th>
<th>curve</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial</td>
<td>99</td>
<td>1</td>
<td>Initial</td>
</tr>
<tr>
<td>0.5</td>
<td>90</td>
<td>10</td>
<td>6</td>
</tr>
<tr>
<td>1.0</td>
<td>86</td>
<td>14</td>
<td>6</td>
</tr>
<tr>
<td>1.5</td>
<td>82</td>
<td>18</td>
<td>6</td>
</tr>
<tr>
<td>2.0</td>
<td>66</td>
<td>34</td>
<td>6</td>
</tr>
<tr>
<td>2.8</td>
<td>52</td>
<td>48</td>
<td>6</td>
</tr>
<tr>
<td>3.8</td>
<td>32</td>
<td>68</td>
<td>6</td>
</tr>
<tr>
<td>7.5</td>
<td>1</td>
<td>99</td>
<td>6</td>
</tr>
</tbody>
</table>

Note: A. 0.1% formic acid-distilled water; B. 0.1% formic acid-acetonitrile.

Fig. 2 BPI chromatograms for methanol extracting solution of Acanthopanax senticosus scanned by negative ion mode (A) and positive ion mode (B). Chromatographic separation were performed on a BEH C18 column (1.7 μm, 2.1 mm × 50 mm) with linear gradient from 1% to 99% acetonitrile (0.1% formic acid) in 7 min at 0.5 mL·min⁻¹ flow rate. In both positive and negative ion mode, the m/z range was scanned from 100–1 000. Mass spectrum condition in positive ion mode: capillary voltage is 2.5 KV, cone voltage is 35 V, desolvation gas rate was set to 600 L·h⁻¹ at a temperature of 300 °C. Mass spectrum condition in negative ion mode: capillary voltage is 2.3 KV, cone voltage is 45 V, desolvation gas rate was set to 550 L·h⁻¹ at a temperature of 250 °C.

Fig. 3 BPI chromatograms obtained from different traits of Acanthopanax senticosus in m/z range of 100–1 000 scanned by positive ion mode. (A) long capillament A. c. (B) short capillament A. c.

Fig. 4 BPI chromatogram obtained from different traits of Acanthopanax senticosus in m/z range of 100–1 000 scanned by negative ion mode. (A) long capillament A. c. (B) short capillament A. c.

Note: 198 Chin J Nat Med May 2012 Vol. 10 No. 3
Fig. 5 Scores plot (A) and Loadings plot (B) by PCA processed data acquired from long capillamnet *Acanthopanax senticosus* (▲) and short capillament *Acanthopanax senticosus* (■) scanned by negative ion mode. Each of the blue triangle represented for a cluster of long capillament *Acanthopanax senticosus*, the red square triangle represented for a cluster of long capillament ones. The green dots in (B) pointed the ions who may have a larger contribution to the classification of two groups of sample. Original data classified by components 1 and 2 after Pareto scaling with mean centering.

The preferential distribution of marker ions represented by green dot in the second and third quadrant accounted primarily for the long capillament AS; alternatively, those in the first and fourth quadrants indicated the short capillament one. Secondly, the selected initial data were further processed by PLS to make data purposely analyzed by strengthening inter-class difference and reducing or eliminating inner-class difference (Fig. 6a). PLS loading plot reflected ion information correlating with grouping information. Two sample groups were located at different position, the ions which distributed at same or similar position with sample group may be offered by corresponding sample (Fig. 6b). Furthermore, the data were processed by OPLS, and two sample groups were clearly separated by PC1 (Fig. 7a). The parent ions from the origin of Axis in first quadrant indicated the “existing” or “with a higher content” in short capillament AS samples, while the parent ions from the origin of Axis in third quadrant accounted for the characters of long capillament AS (Fig. 7b). Referencing VIP value of ion dedication to classification (Fig. 7c), seven ions were selected as potential biological markers in negative ion mode. Data of positive ion mode were analyzed in the same manner, in which four ions were picked out. Information of all potential markers was showed in Table 2.

Fig. 6 Scores plot (A) and Loadings plot (B) by PLS obtained from long capillamnet *Acanthopanax senticosus* (▲) and short capillament ones (■) scanned by negative ion mode. Each of the blue triangle represented for a cluster of long capillament *Acanthopanax senticosus*, the red square triangle represented for a cluster of long capillament ones. Every black triangle in (B) represented an ion specially in short or long capillament *Acanthopanax senticosus*. Original data classified by components 1 and 2 after Pareto scaling with mean centering.

3.3 Characterization of potential biological markers

The ions of potential biological markers for each type of AS were characterized by mass fragments coupled with UV spectrum analysis. For example, the ion at 2.10 min (Fig. 8a) with accurate molecular mass of 895.193 3 has a major contribution to the classification. Ultraviolet spectra showed that there are two absorption bands at 252.8 and 332.8 nm, which corresponds to structure of flavone or flavonol. In determinate error of 1 × 10⁻⁶, the molecular formula is C₄₂H₄₀O₂₂, and the degree of unsaturation is 24, which suggested that the compound has multiple rings. MS/MS data showed the main secondary fragment ions at 447, 300, 284, 271, 255, 243, 227 (Fig. 8c). Furthermore, 2D-MS/MS analyzed by ion mobility provided ascription relationship among secondary fragment ions: 895→447→300, 300→284→271→255→243→227, 227→191→151→135 (Fig. 8b). The ion 895 may be the dimer of fragment ion 447, the elemental composition of ion 447 may be C₁₂H₁₀O₁₁, fragment ion 447 ([M – H]⁺) lost a rhamnose to form the fragment ion 300, which continuously lost [O]²⁻ or [CO] to afford the other ions (Fig. 9). Taken
Fig. 7 Scores plot (A), S-plot (B) and VIP Plot (C) by OPLS obtained from long capillament Acanthopanax senticosus (▲) and short capillament ones (■) scanned by negative ion mode. In (A), each of the blue triangle represented for a cluster of long capillament Acanthopanax senticosus, the red square triangle represented for a cluster of long capillament ones. The blue triangles in (B) pointed the ions both have a higher content in long capillament Acanthopanax senticosus responsible for the separation of two groups of sample. The red squares in (B) pointed the ions both have a higher content in short capillament Acanthopanax senticosus responsible for the separation of two groups of sample. VIP Plot arranged from high contribution ions to low ones which lead to the separation, values are $\bar{x} \pm s$.

Together, ion 447 may be quercetin-3-rhamnoside (C$_{21}$H$_{20}$O$_{11}$, $m/z$ 448.100 6) and ion 895 may be the dimer of quercetin-3-rhamnoside.

The structures of other potential markers were identified in the same manner as above. The structures and the content variation of the identified metabolites in two type AS were shown in Figs.10 and 11.

4 Discussion

The earlier period of sexual differentiation refers to several special macromolecular markers, such as isoenzyme, specific tRNA and transcribed mRNA. Of all macromolecular markers, protein is the most important one [14]. Sun reported that AS with different length of capillament showed distinctions in six isozymogram, such as acid phosphatase, malic dehydrogenase, alcohol dehydrogenase, isocitrate dehydrogenase, cytochrome oxidase and peroxides oxidase [12], correspondingly the secondary metabolism conducted by the above enzymes should be different. As a result, the quality of crude drug originated from different phenotype of the plant is certainly diverse. By using metabolomic theory and technique, the present discriminated the AS with long and short capillament according to their metabolic profiles, and found the potential marker metabolites for each phenotype of AS. The categories of metabolites, such as flavone, coumarin, terpenes, amino acid and organic acid etc., showed obvious difference between two phenotypes. The results demonstrated that during the differentiating of AS from monoecious bisexual flower to dioecious unisexual flower, the whole internal metabolism network is adjusted obviously. Meanwhile, by analyzing distribution of those potential markers, we found that like flavone, anthraquinone and organic acid have a high level of expression in short capillament AS, while coumarin and terpenes are highly expressed in long capillament AS. Other kinds of compound like amino acid have similar level of expression in both varieties. Falconoid plays an important role in the growth, development and fructification of plants. From ecological point of view, flavonoid can not only protect plants from injury from highlight radiation, but also promote breeding of plant. Therefore, flavonoid plays an important role in maintaining species to continue in the differentiating processes of AS [15].

Phenolic compounds including flavonoids, phenols and chinone are mainly synthesized in phenylpropanoid route, in which phenylalanine is converted to diverse phenols by enzymatic reaction [16]. In the process of phenylpropanoid synthetic route for flavone synthesis, the key enzyme at the first branchpoint is isoflavone synthase, a cytochrome oxidase depending on NADPH; the key enzyme at the second branchpoint is flavone synthase, a dioxygenase depending on oxoglutarate and the key enzyme at the third branchpoint is flavanone-3-hydroxylase, a dioxygenase depending on ferri...
<table>
<thead>
<tr>
<th>Sample group</th>
<th>Ion mode</th>
<th>( t_R/\text{min} )</th>
<th>( m/z )</th>
<th>Molecular formula</th>
<th>Name</th>
<th>Structural formula</th>
</tr>
</thead>
<tbody>
<tr>
<td>LC</td>
<td>Negative ion mode</td>
<td>5.34</td>
<td>339.231 6</td>
<td>( C_{18}H_{32}N_2O_4 )</td>
<td>meso-N, N'-di-tert-butoxycarbonyl-2', 2'-bipyridoline</td>
<td><img src="image1" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td></td>
<td>3.36</td>
<td>733.415 2</td>
<td>( C_{60}H_{62}O_{12} )</td>
<td>akebonoic acid-10-O-arabinopyranosyl-(2\rightarrow 1)-glucopyranoside</td>
<td><img src="image2" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td>Positive ion mode</td>
<td>0.23</td>
<td>200.972 8</td>
<td>( C_7H_{12}NO_5S )</td>
<td>3, 6, 7-trioxo-4-thia-1-azabicyclo[3.2.0] heptane-2-carboxylate</td>
<td><img src="image3" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.30</td>
<td>223.060 1</td>
<td>( C_{11}H_{10}O_3 )</td>
<td>isofraxidin</td>
<td><img src="image4" alt="Structural formula" /></td>
</tr>
<tr>
<td>SC</td>
<td>Negative ion mode</td>
<td>2.10</td>
<td>895.193 3</td>
<td>( [C_{21}H_{30}O_{11}]_2 )</td>
<td>dimer-quercetin-3-rhamnoside</td>
<td><img src="image5" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.41</td>
<td>353.085 3</td>
<td>( C_{16}H_{18}O_9 )</td>
<td>chlorogenic acid</td>
<td><img src="image6" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td>Positive ion mode</td>
<td>2.21</td>
<td>431.095 8</td>
<td>( C_{21}H_{30}O_{10} )</td>
<td>kaempferol rhamnoside</td>
<td><img src="image7" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td></td>
<td>1.16</td>
<td>707.180 0</td>
<td>( [C_{16}H_{18}O_9]_2 )</td>
<td>dimer-chlorogenic acid</td>
<td><img src="image8" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.09</td>
<td>515.116 1</td>
<td>( C_{25}H_{24}O_{12} )</td>
<td>cynarin</td>
<td><img src="image9" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td>Positive ion mode</td>
<td>1.04</td>
<td>188.069 7</td>
<td>( C_{7}H_{14}N_2O_4 )</td>
<td>3-(2, 4-dioximidazolidin-1-yl) alanine</td>
<td><img src="image10" alt="Structural formula" /></td>
</tr>
<tr>
<td></td>
<td></td>
<td>2.21</td>
<td>287.053 9</td>
<td>( C_{13}H_{10}O_6 )</td>
<td>1, 2, 5, 7-tetrahydroxy-4-methyl-anthracene-9, 10-dione</td>
<td><img src="image11" alt="Structural formula" /></td>
</tr>
</tbody>
</table>

Note: LC = long capillament; SC = short capillament.
Fig. 8 Flowchart of structure identification of potential chemical marker, typically for the ion with \([M - H]^- = 895\) at 2.10 min of RT. (A) Ion mobility spectrum of potential biological marker. The ions in chromatographic peak were separated two clustering by drift time, denoted by green fluorescence dots. (B) Ion mobility spectrum and corresponding MS/MS data of potential chemical marker. The ions which denoted by green fluorescence line in the same column have a similar drift time, means they were generated at the same time, the “parent ion” of this column is the highest \(m/z\) ion. (C) Total MS/MS spectrum of potential biological marker which accumulated with three mass spectrum in (B)

Fig. 9 MS-MS spectrum and corresponding ion fragment for the structure of potential chemical marker with \([M - H]^- 895\) at 2.10 min
Fig. 10  Chemical constitution, variance analysis and content trending plot of seven marker compounds obtained from short capillament Acanthopanax senticosus. Arrow demonstrates the place of potential biological marker in ES’ or ES’ chromatogram, corresponding content trending plot in long capillament Acanthopanax senticosus (LC) and short capillament Acanthopanax senticosus (SC) is showed. In content trending plot, each point represented for a group of Acanthopanax senticosus, values are \( \bar{x} \pm s \)

Terpenoid is a metabolite widely distributed in plant that plays an important role in growth and development processes. There are two synthetic routes for terpenoid synthesis, mevalonic acid route and erythritol route. The synthesis process of terpenoid can be divided into three stages including intermediates formation, terpenoid synthesis and final modification. In stage one, the enzymes such as 3-hydroxide-3-methyl-glu-
Fig. 11 Chemical constitution, variance analysis and content trending plot of seven marker compounds obtained from long capillament Acanthopanax senticosus. Arrow demonstrates the place of potential biological marker in ES+ or ES− chromatogram, corresponding content trending plot in long capillament Acanthopanax senticosus (LC) and short capillament Acanthopanax senticosus (SC) is. In content trending plot, each point represented for a group of Acanthopanax senticosus, values are $x \pm s$.

taryl-coenzyme A reductase and 5-phospho-deoxy-lyxoketose synthetase catalyze the formation of opentenylpyrophosphate (IPP) and DMAPP. In stage two, the enzymes, such as prenyltransferase and terpenoid synthetase, mainly catalyze IPP and DMAPP to form intermediates or end products of various kinds of terpenoid. In stage three, in order to produce numerous natural products, the structure of the end products must be complicated modified with the needs of enzymes including cytochrome P450 hydroxylase and phytoene dehydrogenase. Up to date, in all studies about AS, there has been no report referring to enzyme differences in terpenoid synthetic route in sexual varieties. Our present work demonstrated that akebonoic acid-10-O-arabinopyranosyl-(2→1)-glucopyranoside, a terpenoid compound, ex-
pressed differently in the varieties. This suggested that there are definitely differences in corresponding to the functional enzyme and coenzyme among the varieties of AS, which may reflect the real difference in terpenoid synthetic route.

Nitrogen compound metabolic pathway in plant is a dynamic complex processes, which not only is regulated by plant genetic and growth development processes, but also is stimulated by inducers like biogeocoenosis, nutritional level and nutrient modality. In addition, it is controlled by idio-enzyme of space, area and substrate. In our previous studies about primer SBSA8’s RAPD-PCR amplification in long and short capillament AS, we identified an idio-fragment, whose size is 947 bp, specific detected in short capillament AS. The result of sequence homology analyzing showed that this idio-fragment has a higher homology with a member of glutamine synthetase protein families, which is one of the key enzymes in plants nitrogen metabolism. The main pathway of nitrogen metabolism in AS is to reduce nitrates into ammonium, then participates in synthesis and transformation of amine acid. Glutamine synthetase plays an important role in this process. The present study demonstrated that the different expression of the enzyme resulted in the corresponding different nitrogen compounds, such as 3-(2, 4-dioxoimidazo-1yl) alamine and meso-N,N,N-diter- butyoxycarbonyl-2, 2-bipyrrolidine in the sexual variety of AS. This result would provide important information to the biosynthesis of these compounds.

As showed in the present study, flavone, anthraquinone and organic acid have high level of expressions in secondary metabolism of short capillament AS, while coumarin and terpenes are expressed highly in long capillament AS. The difference in secondary metabolites, which are the basic chemicals of crude drug for showing clinical efficacy, will definitely result from the changes of bioactivity of phenotypes. The previous pharmacological study indicated that phenol glycoside is the essential component of radix and rhizome of AS, thereinto chlorogenic acid are anti-fatigue compounds. Flavonoid glycosides, whose total content can reach 37.25% in leaf of AS, such kaempferol rhamnose and the dimer of quercetin-3-rhamnose, show effects of preventing and curing degenerative disease such as neoplasms, aging and cardiovascualr disease. Isofraxidin has apparent effect of tranquilizing and allaying excitement that related to the main clinical efficacy of AS. Anthraquinone have anti-septicis, antibitis and hepatoprotective activity. Triterpenoid saponins, fifteen kinds of which had been detected from AS, also have antiviris and anti-tumor effects.

5 Conclusion

Metabolomics has provided a potential strategy and technology for better understanding the differences in metabolic profiles of secondary metabolites of plant. The present study revealed the potential marker metabolites of two different sexual type of AS, and the distribution of those potential markers in the two phenotypes. The short capillament AS is rich in the constituents which active on anti-fatigue and preventing and curing degenerative disease. Alternatively, the long capillament variety contains a larger amount of constituents with the activity of tranquilizing and allaying excitement clinically. By analyzing the metabolic profiles, a higher quality phenotype of medicinal plant with better expression of active compounds can be selected to cultivate on particular purpose.

References


基于 UPLC-MS 的代谢谱区分生长在中国东北地区植物刺五加多样性

孙晖1，韩莹1，张爱华1，孟祥才1*，王振月2，孙文军1，孙海峰2，王喜军1*

1黑龙江中医药大学中药血清药物化学国家重点实验室，哈尔滨 150040；
2黑龙江中医药大学中药学院中药资源教研室，哈尔滨 150040

【摘要】 刺五加主要分布于中国东北地区，在临床和制药行业具有广泛应用。由于受到野生资源量的限制，必须加快刺五加的品系繁育和人工栽培。本研究采用主成分分析、偏最小二乘-判别分析及代谢标识物鉴别相结合的最新研究技术，对不同性别类型的中药刺五加进行区分，力求解决中药材刺五加遗传背景复杂、不同性别类型药材的鉴别困难的问题。结果发现黄酮、香豆素、萜类和有机酸类在短花丝类型刺五加中高表达，而香豆素、萜类在长花丝类型中高表达。因此，通过监测这些高表达的代谢物，可以区分不同性别类型的刺五加。

【关键词】 刺五加；五加科；表型；代谢组学；超高效液相色谱-质谱；次生代谢产物

【基金项目】 国家十五科技攻关计划重大项目(No.2004BA721A19)；国家自然科学基金(No.90709019)；国家重大新药创制项目(No.2009ZX090502-005)；国家重点基础研究计划(No.2005CB523406)