Journal of Medicinal Plants Research Vol. 5(17), pp. 4001-4008, 9 September, 2011
Available online at http://www.academicjournals.org/JMPR
ISSN 1996-0875 ©2011 Academic Journals

Review

Application of chemometrics in quality evaluation of medicinal plants

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Accepted 7 June, 2011

Plant medicines are gaining more and more attention all over the world. Many modern analytical approaches have been introduced to evaluate the quality of medicinal plants, and significant amount of measurement data has been produced. Therefore, the application of chemometrics in the field of medicinal plants is spontaneous and necessary. Comprehensive methods and hyphenated techniques associated with chemometrics used for extracting useful information and supplying various methods of data processing are now more and more widely used in medicinal plants. This review focuses on the applications of chemometrics in quality evaluation of medicinal plants in the authenticity, efficacy and safety evaluation.

Key words: Chemometrics, application, medicinal plants, chromatographic fingerprint, quality evaluation.

INTRODUCTION

Natural plant medicines are gaining more and more attention all over the world. The establishment of advanced and effective quality evaluation system of medicinal plants is the premise for authenticity, safety and efficacy of medicinal plants (Chen, 2006; Yang and Gao, 1999). Correct identification of the species, accurate determination of the content of heavy metal, pesticide residual and mycotoxin are indispensable for safety of medicinal plants. Determination of trace Elements and active ingredients are essential for reflecting intrinsic quality of medicinal plants, their consistency and efficacy (Ernst, 2002). With the introduction of comprehensive methods and highly selective, sensitive and versatile analytical techniques including thin-layer chromatography (TLC), gas chromatography (GC), high performance liquid chromatography (HPLC), capillary electrophoresis (CE), gas chromatography-mass spectrometry (GC-MS), liquid chromatography-mass/mass spectrometry (LC-MS/MS), the quality evaluation of medicinal plants has been promoted obviously (Wong et al., 2002; Liang et al., 2004; Dirk and markus, 2011). For instance, Devi (2008) used proton induced X-ray emission and proton induced γ-ray emission techniques to estimate essential and trace elements in some medicinal plants. Barthwal et al. (2008) determined heavy metal and its accumulation in medicinal plants collected from environmentally different sites by inductively coupled plasma emission spectrometry. Qing et al. (2009) simultaneous determined 26 pesticide residues in 5 Chinese medicinal materials using solid-phase extraction and gas-chromatographic-

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Abbreviations: ANN, Artificial neural networks; **CA,** cluster analysis; **CDA,** clustering discriminate analysis; **CE,** capillary electrophoresis; **DA**, discriminate analysis; **GC,** gas chromatography; **GC-MS,** gas chromatography-mass spectrometry; **HCA,** hierarchical clustering analysis; **HPLC,** high performance liquid chromatography; **HPLC-DAD,** high performance liquid chromatography-diode-array-detector; **LC-MS/MS,** liquid chromatography-mass/mass spectrometry; **MSC,** multiplicative scattering correction; **PCA**, principal component analysis; **SA,** similarity analysis; **TCM,** traditional Chinese medicine; **TLC,** thin-layer chromatography; **TPA**, target peak alignment.

electronic capture detector method. The authentication of medicinal plants is really a challenging task in cases where botanical and classical microscopy identification is impossible for drying and processing procedure. The analysis of active ingredients in medicinal plants is a challenging task also, because of their chemical diversity, low availability and variability. Chromatographic fingerprinting, which adapts well to the condition where most active components in medicinal plants are unclear, offers a comprehensive, integrated and qualitative profile and is being increasingly used for the authentication and quality evaluation of medicinal plants (Xie and Albert, 2009; Yi et al., 2008). However, the use of modern analytical apparatus produces a significant amount of measurement data. Medicinal plants researchers now face a new challenge of extracting sufficient chemical information and analyzing the result (Daniel and Mok, 2010). These challenges necessitate the use of chemometrics. Chromatographic fingerprint, associated with chemometrics, is typically used to evaluate the quality of medicinal plants by extracting useful information and supplying various methods of data processing. Its analysis has made great progress (Zhang et al., 2008; Yu et al., 2010; Song et al., 2010; Valentina et al., 2010). Computer-based software, SPSS, supports pretreatment methods such as background and retention time correction, peak alignment, identification and matching, translating processed data into information accessible for objective analysis, and can also simultaneously evaluate many different samples (Yang et al.,2007; Fang et al., 2002).

In this review, we introduced the origin and development of chemometrics in the first part. Then we described its several common methods such as hierarchical clustering analysis (HCA), principal component analysis (PCA), similar analysis (SA) applied in the quality evaluation of medicinal plants. In the final part, we discussed its application with regard to authenticity, determination of chemical component, including trace elements, heavy metal, pesticide residual and mycotoxin by analytical techniques, especially on the use of chemometrics combined with chromatographic fingerprint for quality evaluation based on active ingredients.

ORIGIN AND DEVELOPMENT OF CHEMOMETRICS

The name "chemometrics," first coined by a Swedish scientist Svante Wold in 1971 (Wold and Sjöström, 1998), is a discipline of chemicals. Chemometrics integrates mathematics, statistics, and formal logic which can offer theories and methods for chemical measurement, provide new approaches for the analysis of various types of spectroscopy and chemical measurement data. Moreover, chemometrics can be implemented in chemistry to optimize experimental procedures and provide maximum relevant chemical information (Kateman, 1983; Jurgen, 2004). Chemometrics has developed rapidly since the early twentieth century. The driving forces for chemometrics are twofold: (1) the flood of data produced by modern multi-element and multi-component apparatus, which requires the application of chemometric methods to extract essential information; and (2) the popularity of computers, which makes processing complex data possible (Yu, 1991; Daniel and Mok, 2008).

Chemometrics is a breakthrough for the software-subjectivation development of analysis apparatus in the twenty-first century (Li and Zhu, 2005; Wu et al., 1999; Liang and Yu, 1999). It promotes equipment intellectualization and offers new ideas and methods for the construction of new and high-dimensional and hyphenated equipment. Furthermore, with the rapid development of the microcomputer (that is, establishment and retrieval of chemical spectrum library), the analysis of high-dimensional data, artificial neural networks, research of artificial intelligence of chemistry, and expert systems has made great progress. With the development of analytical chemistry, chemometrics is also being developed vigorously (Yu, 1992; Wold, 1991).

COMMON CHEMOMETRIC METHODS IN QUALITY EVALUATION OF MEDICINAL PLANTS

Among the different kinds of sensitive and versatile analytical techniques, high performance liquid chromatography (HPLC) is extensively applied for the quality evaluation of medicinal plants due to its superior precision, high resolution and extensive applicability (Marston, 2007; Xie, 2007). Liquid chromatography mass spectrometry (LC-MS) has played an increasingly important role in complex chemical identification and toxic determination of herbal medicines (Ganzera et al., 2003). The improved instrumentation is able to record small differences between samples, which might have large implications for the discrimination of medicinal plants. At the same time, they generate enormous amounts of data.

Taking these into consideration, we discussed the chemometrics applied in the handling of fingerprints in this part. Before analyzing the fingerprint, the pretreatment is essential for overlapped peaks and sifted baseline caused by the unknown components and unclear interferences. Then we discussed the commonly methods applied in quality evaluation of medicinal plants, such as similar analysis (SA), principle component analysis (PCA), cluster analysis (CA), discriminate analysis (DA) and pattern recognition. These methods play a vital role in the discrimination and classification of medicinal plants.

Pretreatment of chromatogram

To obtain accurate quantitative results from the finger print, pretreatment of individual datum collected from the apparatus via peak alignment and data transformation is necessary. Many approaches in peak alignment have been suggested including target peak alignment (TPA) and correlation optimized warping (COW) (Xu et al., 2006; Gong et al., 2006). Gong et al. (2004) developed HPLC-DAD fingerprints of cortex Cinnamomi, herba Menthae, rhizoma Chuanxiong and radix Angelicae. The data sets were analyzed prior and after warping and the beneficial effects of aligning fingerprints in the classification of the species were clearly shown. Another important pretreatment step is the transformation of the data. By putting weight on different parts of the fingerprints, the variability of the desired information is maximized while that of the unwanted information is minimized. In gray and black systems of unknown components, factor analysis is widely used to clarify the overlapped chromatography (Wiberg et al., 2004). Evolutionary factor analysis (EFA), alternative moving window factor analysis (AMWFA), and fixed size moving window evolving factor analysis (FSMWETA) are representative methods (Zeng et al., 2006). Under the method of fixed size and moving direction of local window, the computation time of the matrix is significantly reduced, peak purity is determined, and information of component number is shown (Zeng et al., 2003). Zhao et al. (2006) determined the volatile oil content in different samples of Rhododendron with GC-MS, and identified 128 volatile components based on retention factor and sub-window factor analysis. Gong et al. (2006) sifted the representative variables from all the variables in groups, analyzed samples of 79 Chinese herbal medicines, and verified the feasibility of the method.

Classification and discrimination methods

Similarity analysis of chromatographic fingerprint

One of the most common and easiest tools is similarity analysis based on the correlation coefficient. The similarity analysis method of peak height, peak area, and ratio between peaks of the fingerprint has been successfully applied for quality evaluation of medicinal plants (Gong et al., 2003b; Xu et al., 2007). Many means of improving the analysis of fingerprint similarity, such as cosine angle have been proposed (Liu et al., 2004). Gan and Ye (2006) established the HPLC fingerprint, using the peak areas of Chuanxiong from three main producing areas of China. The correlation coefficient and vector cosine offer a better differentiation of the similarity or difference between the fingerprints from the same samples of Chuanxiong.

Principal component analysis (PCA) and cluster analysis (CA)

PCA is the most commonly method applied in the fingerprint and other data sets for exploratory data analysis. It reflects the original measurement by discovering the dominant factors while excluding the relevant interference factors, thereby allowing a more accurate estimate (Liang et al., 2004; Marek, 2003) (Figure 1). CA classifies objects based on quantitative characteristics. Generally, the different clustering techniques are divided into two subtypes: hierarchical and non-hierarchical (example, fuzzy clustering). In the quality evaluation of medicinal plants, the most popular clustering technique is hierarchical clustering analysis (HCA) (Figure 2). The main advantage of hierarchical cluster analysis is the flexibility to alter the similarity measurement criterion and the applied linkage method to suit different applications. Fingerprint based PCA can directly reflect the difference between samples, whereas CA can classify objects based on their quantitative characteristics (Huang et al., 2008). A combination of CA and PCA has been widely used in current quality assessment of medicinal plants.

Pattern recognition

Pattern recognition unlike SA, PCA and HA uses discrete information on the samples in the calibration set. It can establish identification model by analyzing data, find out regular law, obtain accurate analysis of components in medicinal plants and provide a comprehensive evaluation of their quality (Gong et al., 2003a; Xie et al., 2006). Over the years, pattern recognition methods have been intensively used for the classification of samples (Wang et al., 2007; Zhai et al., 2010). Tian et al. (2009) evaluated Chaihu (Bupleuri Radix) by both high-performance liquid chromatography-evaporative light scattering detector (HPLC-ELSD) and high-performance thin-layer chromatography (HPTLC) analyses of its principal bioactive components (saikosaponins). The pretreated data acquired from both HPLC fingerprints and HPTLC fluorescent images were processed by chemometrics for similarity and pattern recognition, including Artificial Neural Networks (ANNs), k-nearest neighbor (k-NN) and an expert's panel. Among these, k-NN classifier was apparent that exhibited good performance with sufficient flexibility for processing HPTLC fingerprint images which were otherwise not easily dealt with by other algorithms due to the shift of Rf values and varying hue/saturation of the band colors between different TLC plates. The results indicated that these two chromatographic fingerprint methods can be considered complementary measure of quality control.

Discriminate analysis (DA)

DA is a type of clear-cut classification that segregates objects through clear indicators; it is different from CA that needs no indicators. In the research of quality

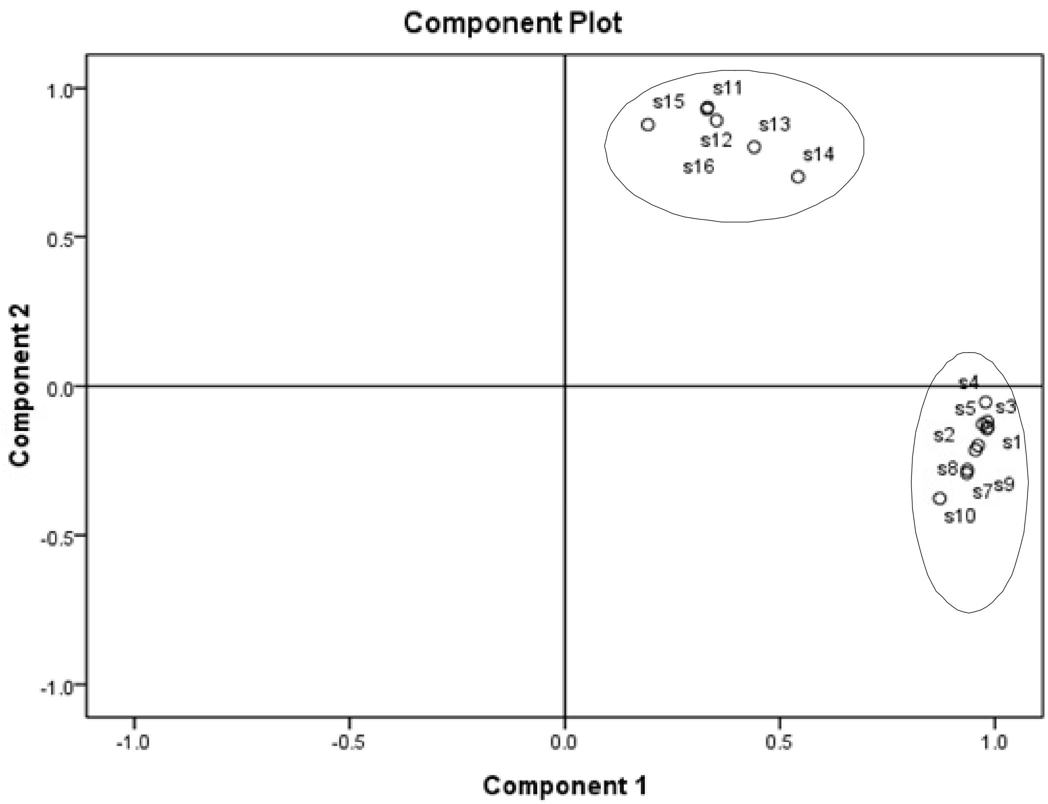

Figure 1. Simulation of a scores plot as generated by hierarchical clustering of fourteen samples.

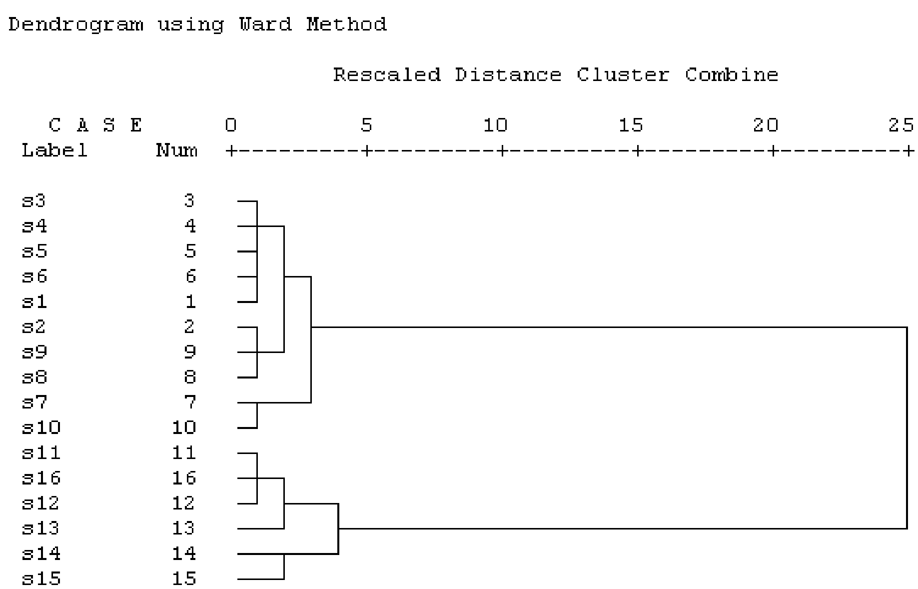

Figure 2. Simulation of a dendrogram as generated by hierarchical clustering of sixteen samples

evaluation of medicinal plants, DA is used to set up a discriminate function. Multiple regressions of a few typical properties are sifted by variance regression testing technology through a recognized index (such as the amount of active ingredient).

Consequently, the unknown sample is identified and classified by observing the information of various types of known samples. Maximum likelihood, Fisher discriminate analysis, Bayes discriminate analysis, and logistic regression are common DA methods (Philip, 2003; Chen et al., 2007).

Clustering discriminate analysis (CDA) is a combination of CA and DA, and can be used to induce and extract signals from complicated data and set up feature models. CDA has been increasingly used in the quality evaluation of medicinal plants for its comprehensive and objective advantages (Gong et al., 2004; Zhang, 1994; Yu et al., 2002).

APPLICATION OF CHEMOMETRICS IN QUALITY EVALUATION OF MEDICINAL PLANTS

After introducing the origin and development of chemometrics, the commonly applied methods in handling procedure of data generated from hypnated analytical instruments, we will describe the application of chemometrics in quality evaluation by giving detailed examples in the field of authenticity, efficacy, consistency and safety evaluation of medicinal plants, respectively.

Authenticity

Ascertaining authenticity is the first important step to assess the quality of plant medicine. Each medicinal plant contains certain characteristic constituents. Therefore, constituents including their respective chemical ratios can be analyzed to identify medicinal plants and distinguish the fakes.

Lai et al. (2010) analyzed fingerprint of Cassia seed, generated by HPLC-UV at two wavelengths, with chemometrics methods. Samples were clustered into four groups in accordance with the plant sources and preparation procedures by PCA analyzed. And partial least squares (PLS), back propagation artificial neural network (BP-ANN), and radial basis function artificial neural network (RBF-ANN) were effectively applied to predict the category of the four different samples in the test set. Guo et al. (2009) established HPLC-DAD method to simultaneously determine 10 triterpenoid acids, and used HCA and PCA to differentiate and classify the samples based on the contents of the 10 triterpenoid acids. The presented HPLC-DAD method, combined with chemometrics, was demonstrated to be very helpful in searching *Ziziphus jujuba* var. inermis resources, and to be possibly useful in chemotaxonomic characterization.

Furthermore, not only can authenticity be identified but substitutes can also be searched according to the theory that "herbs containing the same properties have similar potency". For instance, comparing fingerprints of species under the monograph in Chinese Pharmacopoeia, and the nonofficial species by similarity analysis, one can investigate the possibility of using these nonofficial species as alternatives to the official species (Xu et al., 2003; Xu et al., 2009; Zhu et al., 2010).

Taking herbal *Cistanches* for instance, Jiang et al. (2009a) developed HPLC-DAD-MS method, and compared the fingerprints of another non-official species *Cistanche salsa* and *Cistanche sinensis* with the official one *Cistanche deserticola*. They found that *C. salsa* has high similarity with the standard while *C. sinensis* varies, suggesting that *C. salsa* may be used as alternative species.

Efficacy and consistency

The efficacy of plant medicines is closely related to their chemical constituents and their concentration, and consistency might vary slightly according to differences in climate, cultivating or wilding, harvest time, possessing procedure, and storage.

When evaluating the recorded fingerprints, minor differences in concentrations might influence the quality of the plant medicines, while small differences between the fingerprints can discriminate between species. PCA, SA and HCA can classify and discriminate medicinal plants fingerprints effectively.

Lin et al. (2011) developed enhanced fingerprints of various *Artemisia selengensis* Turcz by means of HPLC-DAD. The results were analyzed by SA, HCA and PCA. Each method highlighted different properties of the data matrix according to the fingerprints from different types of *A. selengensis* Turcz. It provided comprehensive information for matching and discrimination of the fingerprints, and appeared to be suited for quality assurance purposes for medicinal plants.

Ni et al. (2009) developed HPLC-DAD fingerprint of Cassia seed, and analyzed the results with a series of chemometrics methods. Consequently, the preference ranking organization method for enrichment evaluations (PROMETHEE) and geometrical analysis for interactive assistance (GAIA) and multi-criteria decision making (MCDM) methods provided the most comprehensive information for matching and discrimination of the fingerprints, and appeared to be best suited for quality assurance purposes for these and similar types of sample.

Kong et al. (2009) developed ultra performance liquid chromatography with photodiode array detector (UPLC-PAD) method for quantitative analysis of five active alkaloids and chemical fingerprint analysis. The data were analyzed by chemometrics methods such as SA, PCA and HCA to classify the samples. The presented method, combined with chemometrics, demonstrated to be very helpful in the quality control and evaluation of *Rhizoma Coptidis*.

Safety

The safety issue of plant medicines has been reported to contain heavy metals, pesticide residual, mycotoxin and synthetic prescriptions or non-prescription drugs. They may originate from mineral components, contamination and adulteration (Ernst, 2002). The determination of them is crucial to ensure the safety of the plant medicines.

Trace elements such as iron, copper, zinc and manganese are known to play important roles in biological systems, and may have relationship with specific growth location and curative efficacy of the plant, so it has been studied extensively recently. The research is often carried out combined with PCA and CA (Li et al., 2007; Ražić et al., 2003; Wesołowski et al., 2003).

In contrast, other elements, such as lead and cadmium are toxic even in trace amounts. Therefore, it has the central importance of ascertaining the content of toxic elements. Wang et al. (2010) determined trace and toxic element concentrations in *Paris polyphylla* from China by flame and graphite furnace atomic absorption spectrometry to evaluate and classify their quality. The results were analyzed with PCA and HCA which support important information for safety evaluation. Kolasani et al. (2011) determined mineral elemental concentrations of 50 Chinese medicinal herbs in acid digests with flame atomic absorption spectroscopy. The data were analyzed by PCA and HCA to understand the association between the elements and to classify the herbal samples.

PROSPECTS

The potential power of chemometrics in quality evaluation needs no further description. In the evaluation of quality of herbs, chemometrics can be used to optimize experimental procedures, pre-treat fingerprints, extract maximum useful information, and analyze results. Chemometrics is a suitable analysis method and a useful tool for estimating medicinal plants quality (Valentina Razmovski-Naumovski et al., 2010).

Meanwhile, we should keep in mind that chromatographic fingerprint is not always a perfect way to present all compounds of medicinal plants. The macromolecules (example, polysaccharides, amino acids), a major part of the bioactive compounds in medicinal plants, are often not considered in fingerprint, and should be taken into account during future development of quality evaluation. Although there are different kinds of monographs on quality evaluation of medicinal plants in some countries, sometimes we still lack an effective standard to make scientific quality estimation (Springfield and Eagles, 2005). New ways to assess the quality of medicinal plants are constantly being explored, such as combination chemical fingerprint with biological methods, biofingerprint, metabolic fingerprint (Li et al., 2008; Fan et al., 2006; Liang et al., 2009; Jiang et al., 2009b). Pharmacodynamics, quality metrology, and the export system of herbal medicine have been researched in some groups (Qi and Yu, 2010; Wang et al., 2006; He and Zeng, 2006). A perfect system of quality evaluation of quality evaluation of medicinal plants has not been achieved and will require a significant amount of work to accomplish it.

ACKNOWLEDGEMENTS

This work was supported by the Beijing Municipal Science and Technology Foundation (No. D08080203640901) granted to S.L.C. and also supported by the Open Research Fund of State Key Laboratory Breeding Base of Systematic research, Development and Utilization of Chinese Medicine Resources.

REFERENCES

Barthwal J, Smith N, Kakkar P (2008). Heavy metal accumulation in medicinal plants collected from environmentally different sites. Biomed. Environ. Sci., 21: 319-324.

Chen HS, Bhavik RB, Prem KG (2007). Toward Bayesian chemometrics- A tutorial on some recent advances. Anal. Chim. Acta, 602: 1-16.

Chen YM (2006). Traditional Chinese medicine industry development and their impact and countermeasures. Mod. Chin. Med., 8: 35-391.

Daniel KW, Mok CFT (2006). Chemical information of Chinese medicines: A challenge to chemist. Chemom. Intell. Lab. Syst., 26: 210-217.

Devi KN, Sarma HN, Kumar S (2008). Estimation of essential and trace elements in some medicinal plants by PIXE and PIGE techniques. Nuclear Instruments and Methods in Physics Research Section B: Beam Interact. Mater. Atoms, 266(8): 1605-1610.

Dirk S, Markus G (2011). Recent advances on HPLC/MS in medicinal plant analysis. J. Pharmacol., Biomed. Anal., 55: 744-757.

Ernst E (2002). Toxic heavy metals and undeclared drugs in Asian herbal medicines. Trends Pharmacol. Sci., 23: 136-139.

Fan XH, Cheng YY, Ye ZY, Lin RC, Qian ZZ (2006). Multiple chromatographic fingerprinting and its application to the quality control of herbal medicines. Anal. Chim. Acta, 555: 217-224.

Fang F, Li SJ, Guan Y (2004). PCA on medicinal materials of three species of Isodon based on their HPLC. Bull. Botan. Res., 24: 351-355.

Feng XS, Dong HY (2002). Data mining in establishing fingerprint spectrum of Chinese traditional medicines. Prog. Pharm. Sci., 26: 198-201.

Gan F, Ye RY (2006). New approach on similarity analysis of chromatographic fingerprint of herbal medicine. J. Chromatogr. A, 1104: 100-105.

Ganzera M, Zhao J, Khan IA (2003). *Hypericum perforatum* – Chemical profiling and quantitative results of St. John's Wort products by an improved high-performance liquid chromatography method. J. Pharmacol. Sci., 91: 623-630.

Gong F, Liang YZ, Xie PS, Chau FT (2003). Information theory applied to chromatographic fingerprint of herbal medicine for quality control. J. Chromatogr. A, 1002: 25-40.

Gong F, Liang YZ, Fung YS (2004). Correction of retention time shifts for chromatographic fingerprints of herbal medicines. J. Chromatogr. A, 1029: 173-183.

Gong F, Wang BT, Liang YZ, Chau FT, Fung YS (2006). Variable selection for discriminating herbal medicines with chromatographic fingerprints. Anal. Chim. Acta, 572: 265-271.

Guo S, Duan JA, Tang YP, Su SL, Shang EX, Ni SM, Qian DW (2009). High performance liquid chromatography - Two wave length detection of triterpenoid acids from the fruits of *Ziziphus jujube* containing various cultivars in different regions and classification using chemometric analysis. J. Pharm. Biomed. Anal., 49: 1296-1302.

Guo Q, Xia LV, Li Tan, Yu BY (2009). Simultaneous determination of 26 pesticide residues in 5 Chinese medicinal materials using solid-phase extraction and GC-ECD Method. Chin. J. Nat. Med.,

7(3): 210-216.
He QF, Zeng QJ (2006). The Research on the Quality Evaluated of Chinese Traditional Medicine Based on ANN-ES. J. Jinzhu Univ. Guiyang, 1: 27-31.
Huang BK, Huang LQ, Qin LP (2008). Cluster analysis of NIR fingerprint of four species plants in *Valeriana officinalis* L. J. Chin. Med. Mater., 31: 1494-1496.
Jiang Y, Li SP, Wang YT, Chen JX, Tu PF (2009a). Differentiation of Herba Cistanches by fingerprint with high-performance liquid chromatography–diode array detection–mass spectrometry. J. Chromatogr. A, 1216: 2156-2162.
Jiang Y, Bruno D, Tu PF, Tu P, Barbin Y (2009b). Recent analytical approaches in quality control of traditional Chinese medicines-A review. Anal. Chim. Acta., 657: 9-18.
Jurgen WE (2004). Chemometrics in analytical chemistry. Anal. Bioanal. Chem., 380: 368-369.
Kateman G (1983). Meeting reports: International conference on chemometrics in analytical chemistry. Trends Anal. Chem., 2(3): 11-12.
Kong WJ, Zhao YL, Xiao XH, Cheng Jin, Li ZL (2009). Quantitative and chemical fingerprint analysis for quality control of Rhizoma Coptidis chinensis based on UPLC-PAD combined with chemometrics methods. Phytomedicine, 16: 950-959.
Kolasani A, Xu H, Millikan M (2011). Evaluation of mineral content of Chinese medicinal herbs used to improve kidney function with chemometrics. Food Chem., 127: 1465-1471.
Lai YH, Ni YN, Serge K (2010). Authentication of Cassia seeds on the basis of two-wavelength HPLC fingerprinting with the use of chemometrics. Chin. Chem. Lett., 21: 213-216.
Li P, Qi LW, Liu EH, Zhou JL, Wen XD (2008). Analysis of Chinese herbal medicines with holistic approaches and integrated evaluation models. Trends Anal. Chem., 27: 66-77.
Li YJ, Zhu L (2005). Initial Study on the Way of Chemometrics into our Country. J. Xinchang Agric. Coll., 19: 87-89.
Li JF, Gao JH, Zhu BL (2007). Analysis of the Contents of Trace Elements in Five Rutaceae Family Plants. J. Qingdao Univ. Sci. Technol., 28: 400-402.
Liang XM, Jin Y, Wang YP, Jin GW, Fu Q, Xiao YS (2009). Qualitative and quantitative analysis in quality control of traditional Chinese medicines. J. Chromatogr. A. 1216: 2033-2044.
Liang YZ, Xie PS, Kelvin C (2004). Quality control of herbal medicines. J. Chromatogr. B. 812: 53-70.
Liang YZ, Yu RQ (1999). The development of chemometrics in China. Chemistry, 10: 14-18.
Lin P, Wang YZ, Zhu FB, Chen QM (2011). Fingerprint profile of active components for *Artemisia selengensis* Turcz by HPLC-PAD combined with chemometrics. Food Chem., 125: 1064-1071.
Liu YS, Meng QH, Chen R, Wang JS, Jiang SM, HU YZ (2004). Improvement of similarity measure: pears on product moment correlation coefficient. J. Chin. Pharm. Sci., 13: 180 - 186.
Marek W, Paweł K (2003). Thermoanalytical, chemical and principal component analysis of plant drugs. Int. J. Pharm., 262: 29-37.
Marston A (2007). Role of advances in chromatographic techniques in phytochemistry. Phytochemistry, 68: 2786–2798.
Naumovski VR, Wannit TK, Kimble B (2010). Multiple chromatographic and chemometric methods for quality standardisation of Chinese herbal medicines. Mode. Tradit. Chin. Med. Mater. Med., 12(1): 99-106.
Ni YN, Lai YH, Sarina B, Serge K (2009). Multi-wavelength HPLC fingerprints from complex substances: An exploratory chemometrics study of the Cassia seed example. Anal. Chim. Acta, 647: 149-158.
Philip KH (2003). The evolution of chemometrics. Anal. Chim. Acta., 500: 365-377.
Qi J, Yu BY (2010). A new methodology for the quality evaluation of traditional Chinese medicine-integrated spectrum-effect fingerprint research. Chin. J. Nat. Med., 8: 171-175.
Ražić S, Onjia A, Potkonjak B (2003). Trace elements analysis of *Echinacea purpurea*-herbal medicinal plant. J. Pharm. Biomed. Anal., 33: 845-850.
Song JZ, Li SL, Zhou Y (2010). A novel approach to rapidly explore analytical markers for quality control of Radix *Salviae Miltiorrhizae* extract granules by robust principal component Analysis with ultra-highperformance liquid chromatography–ultraviolet quadrupole time-of-flight mass spectrometry. J. Pharm. Biomed. Anal., 53: 279-286.
Springfield EP, Eagles SG (2005). Quality assessment of South African herbal medicines by means of HPLC fingerprinting. J. Ethnopharmacol., 101: 75-83.
Wold S (1991). Chemometrics, why, what and where to next?. J. Pharm. Biomed. Anal., 9: 589-596.
Wold S, Sjöström M (1998). Chemometrics present and future success. Chemom. Intell. Lab. Syst., 44: 3-14.
Tian RT, Xie PS, Liu HP (2009). Evaluation of traditional Chinese herbal medicine: Chaihu (*Bupleuri Radix*) by both high-performance liquid chromatographic and high-performance thin-layer chromatographic fingerprint and chemometric analysis. J. Chromatogr. A, 1216: 2150-2155.
Wang ZM, Gao HM, Fu XT, Wang WH (2006). Multi-components quantitation by one marker new method for quality evaluation of Chinese herbal medicine. Chin. J. Chin. Mater. Med., 31: 1925-1928.
Wang WQ, Peng J, Wan J (2007). Chemical pattern recognition of quality assessment of *Folium Isatidis* from various habitats. Chin. Trad. Herbal Drugs, 38: 921-925.
Wang HW, Liu YQ (2010). Evaluation of trace and toxic element concentrations in *Paris polyphylla* from China with empirical and chemometrics approaches. Food Chem., 121: 887-892.
Wiberg K, Andersson M, Hagman A (2004). Peak purity determination with principal component analysis of high-performance liquid chromatography-diode array detection data. J. Chromatogr. A, 1029: 13-20.
Wong SK, Tsui SK, Kwan SY (2002). Analysis of proprietary Chinese medicines for the presence of toxic ingredients by LC/ MS/MS. J. Pharmacol. Biomed. Anal., 30: 161-170.
Wu HL, Liang YZ, Yu RQ (1999). Analytical Chemometrics. Anal. Lab., 18: 94-100.
Xie PS, Chen SB, Liang YZ, Wang XH, Tian RT, Roy U (2006). Chromatographic fingerprint analysis - A rational approach for quality assessment of traditional Chinese herbal medicine. J. Chromatogr. A, 1112: 171-180.
Xie PS, Albert YL (2009). Understanding the traditional aspect of Chinese medicine in order to achieve meaningful quality control of Chinese materia medica. J. Chromatogr. A, 1216: 1933-1940.
Xu CJ, Liang YZ, Chau FT, Foo-Tim C, Yvan VH (2006). Pretreatments of chromatographic fingerprints for quality control of herbal medicines. J. Chromatogr. A, 1134: 253-259.
Xu YQ, Huang H, Zhou Q, Zhou HT, Hu SL, Sun SQ (2003). The application of fingerprint infrared spectra and clustering analysis in the discrimination of geographical origin of *Paeonia lactiflora* Pall. Chin. J. Anal. Chem., 31: 5-9.
Xu ZS, Zhang X, Li ZY, Qi J (2007). Research of the HPLC fingerprint of gastrodia elata blume produced in jinzhai county, anhui province. Anhui Med. Pharm. J., 11: 200-201.
Xu SJ, Yang L, Tian RT, Wang Z, Liu Z, Xie P, Feng Q (2009). Species differentiation and quality assessment of *Radix Paeoniae Rubra* (Chi-shao) by means of high-performance liquid chromatographic fingerprint. J. Chromatogr. A, 1216: 2163-2168.
Yang SL, Gao HQ (1999). Good agriculture practice. World Sci. Technol., 1: 54 - 55.
Yang J, Chen LH, Zhang Q, Lai MX, Wang Q (2007). Quality assessment of Cortex Cinnamomi by HPLC chemical fingerprint, principle component analysis and cluster analysis. J. Separ. Sci., 30: 1276–1283.
Yi LC, Wu H, Liang YZ (2008). Chromatographic fingerprint and quality control of traditional Chinese medicines. Chin. J. Chromatogr., 26: 166-171.
Yu CH, Li QT, Wang J (2002). The research of evaluating quality of Liu Wei Di Huang Wan by cluster analysis. J. ShanDong Coll. Trad. Chin. Med., 26: 375-378.
Yu JM, Peng YL, Liang YZ (2010). Analysis of essential oil components in Kaempferia galanga L. by GC-MS and chemometric resolution method. J. Hunan Univ. Arts Sci., 22: 24- 27.
Yu RQ (1991). Introduction of chemometrics. Hunan educ. press.
Yu RQ (1992). Chemometrics in China. Chemom. Intell. Lab. Syst., 14: 23-39.

Zeng ZD, Liang YZ, Wang YL, Li BY (2006). Alternative moving window fact or analysis for comparis on analysis between complex chromatographic data. J. Chromatogr. A, 1107: 273-285.

Zeng ZD, Xu CJ, Liang YZ, Li XR, Liang LM, Xu QS, Zhao CX, Li BY, Foo-Tim C (2003). Sectional moving window factor analysis for diagnosing elution chromatographic patterns. Chemom. Intell. Lab. Syst., 69: 89-101.

Zhai HL, Hu FD, Huang XY, Chen JH (2010). The application of digital image recognition to the analysis of two-dimensional fingerprints. Anal. Chim. Acta., 657: 131-135

Zhang GF, Chen CJ, Chen ZP (2008). Analysis of principle component and chemistry type of essential oil from *Cinnamomum camphora* leaf in Fujian Province. J. Plant Resour. Environ., 17: 24-27.

Zhang FL (1994). Cluster analysis and quality research of traditional Chinese medicine. The People's Medical Publishing House.

Zhao CX, Li XN, Liang YZ, Fang HZ, Huang LF, Guo FQ (2006). Comparative analysis of chemical components of essential oils from different samples of *Rhododendron* with the help of chemometrics methods. Chemom. Intell. Lab. Syst., 82: 218-228.

Zhu HB, Wang YZ, Liang H, Chen QM, Zhao P, Tao J (2010). Identification of *Portulaca oleracea L.* from different sources using GC–MS and FT-IR spectroscopy. Talanta, 81: 129-135.