REVIEW

Study of pharmacodynamic material basis of single-component Chinese medicine based on metabolomics

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Metabolomics is the holistic qualification and quantification of metabolites in vivo. Metabolomics explores the relative relationship between metabolites and physiological or pathological changes. Its characteristics of integrity, high throughput and the 'end of the end' has a prominent advantage in the study of the Chinese medicine overall efficacy and the mechanisms of multitarget effects. The present article briefly summarizes research regarding single-component Chinese medicine efficacy and functional mechanisms based on the science and techology of metabolomics.

Key Words: *Chinese medicine; Functional mechanism; Metabolomics*

Metabolomics is a relatively recent subject originated by Nicholson et al (1), and was developed after genomics and proteomic chemistry. Metabolomics is a quantitative and qualitative analysis of all the low-molecular-weight metabolites present in an organism or cell in a particular physiological period using modern technology of analysis and detection (2). The metabolite map holistically inspects, under different exogenous factors (eg, drug intervention, environmental change, disease interference, etc), the types and quantity of change, and regularity and relativity, of endogenous metabolites (3-6). Metabolomics studies the organism as a whole, and systematically studies high throughput, which coincides with the overall concept, diagnosis and treatment priciples of Chinese medicine (7-9). It plays an important role in many respects such as exploring the biological essences of syndromes of traditional Chinese medicine (TCM) (10), early diagnosis of disease (11,12), the study of pathogenesis, the quality of TCM (13-15), toxicity (16-19) and the regular pattern of prescription (20,21) as well as the study of medicinal effectiveness and action mechanism. The present article reviews the general analysis of single-component medicine in metabolomics to provide new methods for low toxicity and multiple target research of the the pharmacodynamic material basis of TCM .

RESEARCH CONTENT AND PROCESS OF METABOLOMICS

The main research objects of metabolomics are urine, blood plasma or serum, saliva, and cell and tissue extracts, with a relative molecular mass <1000 Da. Metabolomics' whole research process is divided into four stages: sample collecting and pretreatment; detection and recognition by instrument; data collating and analyzing; and interpretation of biologically significant markers (Figure 1).The main detection techniques include nuclear magnetic resonance (NMR) (22), liquid chromatography-mass spectrometry (LC-MS) (23), gas chromatography-MS (GC-MS) (24), electrophoresis-MS and plasma MS (25). Massive amounts of raw data are obtained using these methods, and must be processed using statistics and modern stoichiometry to extract potential information and interpret the biological significance. Data analysis and pattern recognition is often used: unsupervised classification methods such as principal component analysis, hierarchical cluster analysis and nonlinear mapping; and supervised classification methods such as partial least squares method discriminant analysis (PLS-DA), orthogonal partial least squares, and K recent neighbor neural network (26).

METABONOMIC STUDY OF THE PHARMACODYNAMIC MATERIAL BASIS OF SINGLE HERBS

TCM has the characteristics of multiple components and multiple targets, and metabonomics provides the possibility to establish the related "composition-activity relationship" by identifying the effective components of Chinese medicine on the whole .

Ding (27) established the fingerprint of the volatile oil from curcuma by using GC-MS, studied the relationship between various volatile oils constituents and antitumor activity, and built the optimal "composition-activity relationship" model. Wu et al (28) analyzed the relationship between *Salvia miltiorrhiza* components and antioxidant abilities by using the research strategy of the metabonomics, and realized the activity prediction of composition. Chen (29) also used GC-MS technique to analyze chemical constituents of volatile oil of *Ligusticum chuanxiong* Hort (LCH), data mining the volatile oil chemical composition and relevance of vascular active data, and discriminated the main component of LCH that has a vasodilator effect. Li et al (30) based on the least angle regression algorithm, research the relationship between LCH constituents combination and vascular activity and realized the activity optimization of constituents combination. Li et al (31) described the pharmacometabolomics study investigating the therapeutic basis and metabolic effects of *Epimedium brevicornum* Maxim on hydrocortisone-induced rat using ultra-performance (UP) LC-MS. The results showed that icariin and epimedin C may be the main pharmacodynamic material basis of *Epimedium brevicornum* Maxim. Zhang (32) used the composition-activity relationship to identify antitumour compounds from turmeric based on orthogonal partial least squares and compatibility research, and finally obtained seven optimum combinations. Yu et al (33) explored the antibacterial mode of *Hemsleya pengxianensis* against *Staphylococcus aureus*. The results revealed the antibacterial mechanism to be similar to vancomycin by inhibiting cell wall synthesis, and found the main antibacterial component is cucurbitacins. In addition, using the same method, they found that magnoflorine is the main antibacterial component of *Aquilegia oxysepala* Trautv (34), and learned that the antibacterial effect of *Radix tinosporae* is due to jatrorrhizine and martin (35). Wang et al (36) studied the pharmacodynamic material basis of honey-stir-baked and liquorice-stir-boiled *Polygalae* radix (PR) by using plant metabonomic technology based on ^{1}H-NMR and UPLC. Result shows that the processing may change the medicinal properties so as to lead the various pharmacological activities. Che et al (37) and Wang (38)

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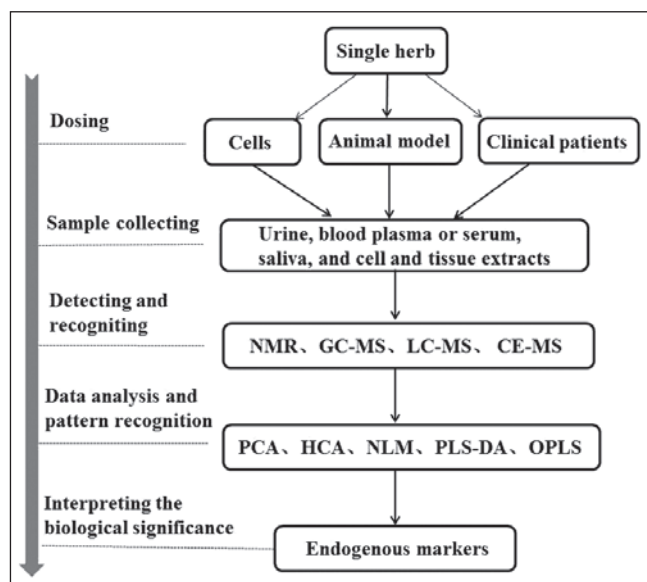*Telephone 18826411201, e-mail jida2468@163.com*

Figure 1) *Research content and process of the metabolomics. CE-MS Capillary electrophoresis–mass spectrometry; GC-MS Gas chromatography–mass spectrometry; HCA High content analysis; LC-MS Liqid chromatography–mass spectrometry; NMR Nuclear magnetic resonance; OPLS Orthogonal partial least squares; PCA Principal component analysis; PLS-DA Partial least squares discriminant analysis*

have studied the main metabolites in urine and blood of humans and rats that had been dosed with baicalin. They found that baicalein is the main intermediate of drug metabolites, and constitutes the pharmacodynamic material basis of baicalin.

METABONOMIC RESEARCH ON MECHANISMS OF SINGLE CHINESE MEDICINE

Metabolomics study not only reveals the metabolic processes of TCM, itself with complex characteristics among multicomponents which have interaction, but also speculates the drug-induced change of endogenous metabolites and related pathways to clarify the action of mechanism of a drug.

Lu et al (39) explored the effect of ginsenosides on the metabolism of spontaneously hypertensive rats using GC-MS. Results showed that after the intervention of ginsenosides, the levels of the specific biomarkers – palmitic acid, galactose pyranthrone, linoleic acid and tyrosine – in the blood plasma of spontaneously hypertensive rats had a downward trend and the metabolism returned to normal. Also, the action of mechanism of berberine was confirmed to be associated with the decreased levels of free fatty acid in blood plasma of rats with type I diabetes (40). Meng et al (41), using the metabolomics approach, found eight kinds of potential biomarkers in cold condensation syndrome rats, and the mechanism of action of chuangxiong, a heat-natured medicine, appeared to inhibit the conversion of phosphatidylcholine to phospholipase A2, which produces arachidonic acid ,causing decline in the level of thromboxane synthesis and affecting the metabolic pathways of phosphatidylcholine, thus easin g the symptoms of blood stasis. Yang et al (42), using UPLC-MS, made a comparison of the effects of red peony root and white peony root on toe swelling of rats caused by diagonal carageen glue. Results indicated that endogenous biological markers such as F2α, prostaglandin E3, leukotriene A4, prostaglandin E2 and glutathione play an important role in the anti-inflammatory action of red peony root and white peony root, and lays the foundation for further research on the different mechanisms of action between red peony root and white peony root. Likewise, Li et al (43) used rapid resolution LC-MS, principal component analysis, PLS-DA and other chemometrics methods to select four characteristic endogenous metabolites deriving from chronic renal failure, and found the possibility that rhubarb inhibits

the catecholamines generating in the blood of the chronic renal failure rats, the decomposition of phosphate esters and the production of inflammatory mediators, causing the recovery of D-glutamic acid metabolism, D-glutamine metabolism and methionine cycle. Wang et al (44), by using NMR in combination with PLS-DA, analyzed the changes in endogenous metabolites in urine after intraperitoneal injection of allitridum in rats and found that allitridum has an impact on Kreb's cycle and the level of acetone bodies, suggesting that studying this metabonomic interaction contributes to understand the liver-protective mechanism of garlic. By analyzing the change of serum metabolomics in kidney yang deficiency rats induced by hydrocortisone, *Morinda officinalis,* with an effect of kidney-replenishing, was confirmed to enhance the methyl transfer reaction of the organism's lack of vital energy, improve energy metabolism and reverse amino acid metabolic disorders; this same effect was not seen on intestinal flora (45). Drynaria rhizome, which may associate with the regulation of energy metabolism, metabolism of intestinal bacteria, and amino acid metabolism and antioxidative damage (46), will not only regulate the metabolic disorders in glucocorticoid-induced osteoporosis rats, but also has an effect of kidney-replenishing (47). Using GC-MS metabonomics technology, Wang et al (48) initially demonstrated that *Bupleurum chinense* DC, with its multiple targets, may participate in enzyme inhibition, neurotransmitters, sugar-lipid metabolism, amino acids metabolism, energy metabolism and more aspects to collaboratively exert the antipyretic effect. Gu et al (49) studied the treatment of total ginsenosides to diabetic cardiomyopathy by investigating urine metabolomics and found three kinds of biomarkers, whose mechanism may be that total ginsenosides regulate the citric acid cycle, fatty acid metabolism and oxidative stress. *Poria cocos* skin, which is possibly involved in the metabolism of phospholipids, energy and amino acids, was found to be effective in regulating six biomarkers, including creatinine, in a study on serum metabonomics of model rats with kidney disease (50). Detoxification with licorice was showed to be associated with the increased free bile acid and bile acid conjugated with glycine in the plasma metabolome of rats (51), whereas the anti-inflammatory action of violet magnolia was speculated to be associated with the metabolism of fatty acids and cholesterol in in the plasma metabolome of rats with inflammation (52). Followed by the metabonomics with LC-MS, Cao (53) analyzed metabolite changes in blood plasma and urine after the interference of total glucosides of peony in acute liver injury rats model induced by CCl4, and detected the retracement of alanine, proline and glycine, suggesting that total glucosides of peony have a preventive effect on CCl4-induced acute liver injury rats model, and it's action mechanism may relate to the Krebs cycle and the synthesis of glutathione eliminating oxygen free radicals.

EXISTING PROBLEMS AND PROSPECTS IN RESEARCH OF METABOLOMICS IN TCM

Metabolomics shares multitarget, global and systematic principles with with TCM, which validate the unique advantages of the study of efficacy and mechanisms of TCM. Metabonomics has developed rapidly in recent years; however, it is not perfect and there are still some problems and challenges in the modern research of TCM. First, most studies of metabolism are only qualitative analyses rather than quantitative analyses. Second, due to the complexity of metabolites, current technology cannot comprehensively analyze all metabolites in an organism, and differences in concentrations among the metabolites is also a significant challenge for detection sensitivity. Nevertheless, although a variety of data analysis methods have been developed, most are only suitable for linear data and there are some challenges to processing nonlinear data. Finally, the support of complete databases is required by metabonomics (54-56). Metabonomics, with the increasing development and improvement, will fit in the process of modernization and internationalization of TCM in a better way, providing new ideas, new methods and new platform to reveal the mysteries of TCM.

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