Review on Interaction between Chinese Herb Metabolism and Cytochrome P450

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[Abstract] Objective: To outlines the interaction between herbs metabolism and CYP450. Methods: Refer to more than 100 pieces of literatures about CYP450 and sum up the article from conception of CYP450, study methods of CYP450, metabolism interaction and herbs metabolism. Results: study methods of CYP450 include methods in vitro and in vivo, and alteration in enzymic activity of CYP450 is reason of metabolism interaction. Conclusions: CYP450 plays an important role in drug metabolism; herbs have complicated ingredients and are reported to revoke some drug interaction often. The reason why drug interaction are caused by herbs is because these herbs affect activity or protein expression of CYP450 in organism and result in alteration of metabolism
involved CYP450 of drugs thus to cause adverse drug reaction.

Because of China joining in WTO and the further modernized research on Chinese herbal medicine, Chinese herbal medicine has been accepted and applied in more and more country, but meanwhile there’re quite some reports pointing out that combination of some herbal medicine and chemicals may cause injury. Though most people believe that herbs are of mild nature and can seldom generate side effects, clinical evidence showed that they could still result in adverse reactions, among which, interaction among metabolic medicine takes up a large proportion in drug adverse reaction. Cytochrome P450 enzyme system (CYP450) plays an important role in medicine metabolism, which is due to that Chinese herbal medicine can affect CYP450 activity or CYP450 protein expression of the body, which will result in the metabolic character changes of other medicine in which CYP 450 takes part in the metabolism, and lead in adverse reactions, etc.

On the other side, because of the complex ingredients of Chinese herbal medicine affecting the functional activity of CYP450 enzymes, and resulting in the changes of medicine disposition characters in the body, which may be one of the mechanism of different combination and different proportion of Chinese herbal medicine having different therapeutic effects, it has practical significance to further explore the
relation between Chinese herbal medicine ad CYP450 enzymes.

1. General introduction to CYP450

CYP450 belongs to hemoglobinase and is the most important oxidase family of microsomal mixed function system. It takes part in the metabolism of most endogenous and extraneous substances. CYP450 mainly exists in the liver microsomes of organism, and is also found in the gastrointestinal tract, kidney, brain, skin, lung, placenta and other tissues or organs. CYP450 is the enzyme protein formed through encoding of CYP450 gene superfamily, containing lots of isoenzymes, and the nomenclature includes three levels of family, subfamily and enzyme.

CYP doesn’t have strong selectivity for substrate. In addition to variance of race, age and gender, there’s also genetic polymorphism. It can be divided into strong, weak metabolism type according to metabolism speed, and hormone levels and day-night rhythm change also affect it, especially it can be easily affected by extraneous (mainly drugs) inducement and inhibition. The inhibition of CYP activity is the main reason that causes the drug metabolic interaction.

CYP450 having close relation with drug metabolism: 1. CYP1A2: CYP1A2 is a CYP subtype induced by multi-ring hydrocarbon, mainly expressed in the liver. The content of CYP1A2 of human is about 13% of
the total amounts of liver CYP450s; 2. CYP2A6: CYP2A6 is the main enzyme of CYP2 family, CYP2A6 is about 4% of the adult’s liver CYP450s. CYP2C: CYP2c is very complicated, containing various gene ingredients, and is a quite big family, including 4 subtypes of CYP2C8, CYP2C9, CYP2C10 and CYP2C19, CYP2C is about 20% of the total amounts of the adult’s CYP450s; 4. CYP2D6: CYP2D6 is about 2% of the total amount of CYP450s in the liver; 5. CYP2E1: CYP2E1 is about 7% of the total amount of CYP450s in the liver; 6. CYP3A4: CYP3A4 is the most ingredients of the CPY450s in the adult liver; it’s about 30%-40% of the total amount of CYP450, which is of the first.

CYP450 isoenzymes have species difference, and here’s the comparison of main CYP450 isoenzymes in the human, rat and mouse: human mainly has CYP1A1, 1A2, CYP2A6, 2A7, CYP2B6, CYP2C8, 2C9, 2C18, 2C19, CYP2D6, CYP2E1, CYP3A4, 3A5; the mouse mainly has CYP1A1, 1A2, CYP2A4, 2A5, CYP2B9, 2B10, CYP2c29, CYP2D22, CYP2E1, CYP3A11, 3A13; the rat mainly has CYP1A1, 1A2, CYP2A1, 2A2, 2A3, CYP2B1, 2B2, 2B3, CYP2C6, 2C11, CYP2D2, CYP2E1, CYP3A1, 3A2.

2. Research methods of CYP450

2.1 in vitro research methods: mainly including: preparing and extracting animal or human liver microsome to study CYP450 activity
through in vitro incubation, cultivating primary generation liver cell for in vitro incubation, liver slicing for in vitro incubation, liver perfusion technique and gene recombination of liver CYP450 enzymes, etc; applying enzyme kinetics, enzyme inhibitors, specific substrate, enzyme activity involved maneuvers to analyze the influence and effect of certain CYP450 enzyme in the metabolism of studied substrate. The most commonly used CYP450 in vitro research methods now is liver microsome incubation technique, which is easy to prepare and repeat, and can be manipulated massively. It’s applied quite widespread in pre-clinic metabolism research in new drug screening, and can basically predict the metabolic characters of the drug in the human body and possible metabolic drug interaction so that decrease the risk of being eliminated because of serious drug interaction in clinic after the drug is coming into market.

2.2. in vivo research methods: in vivo CYP450 research is based on general administration to experimental animal or human body. It can directly and correctly reflect the drug metabolism of organic body in physiological state, but it can be easily affected by individual functional state and individual difference and other factors. In in vivo experiments, the metabolism of CYP450 to certain drug can be indirectly reflected according to the blood concentration of CYP450 probe drug, AUC, t1/2 and other pharmaceutical kinetics parameters, activities of CYP450
enzyme in the body can be reflected by determining the amount of metabolites of probe drug. What’s more, specific inducer or inhibitor can be used to judge the function of CYP450 isoenzymes in the studied substrate metabolism. Recently, a Cocktail probe drug was named in which several probe drugs not metabolized through enzymes are given simultaneously, its practibility and effectiveness was confirmed preliminarily and it has important applicable value for quantitative determination of drug metabolism enzyme activity in the body.

3. General introduction to metabolic drug interaction

The absorption, distribution, metabolism and discharge of drug in the body is generally termed as the endogenous process of drug, among which drug metabolism is the important segment of drug disposition in the body, it can inactivate, activate or transfer the drug into other substance and generate toxic substance. The drug metabolism in the body needs the enzymes, and CYP450 has the biggest relation with drug metabolism.

The transfer of drug can be made clear through studying drug metabolism, but the condition of drug metabolism is very complex, metabolic interaction refers to the interference during metabolism which results in the change of therapeutic effect or the toxic and side effects of the drug when drugs of two or more kinds applied simultaneously or
sequentially.

The most commonly seen reason of drug interaction is the inducement and inhibition of CYP450 enzymes. The inducement of enzyme can increase the biological transferring rate and thus lower the drug concentration, which is usually manifested as decreased drug effect; the inhibition of enzyme can increase the drug concentration, prolong the pharmacological function time and cause the increase of drug toxic reaction. Inducement or inhibition of enzyme has individual difference, race, age, disease; gene and liver function all have influence. Inducement: when a kind of drug stimulates the transfer of combined-used drug through the same or different kind of enzyme, inducement occurs, such as Rifampin inducing CYP3A4, CYP2C9, and CYP1A2. Inducer usually has specificity to specific CYP450. Inhibition, usually competitive inhibition, is drugs of two or more kinds competing for one same enzyme. The competition intensity is mainly decided by relative concentration of the drug and other specific factors, for example, Cimetidine and Ciprofloxacin are the inhibitor for CYP1A2 on Theophylline, but the inhibitory effect of Cimetidine is much stronger that that of Ciprofloxacin. The next is non-competitive inhibitory effect, because of longer functional time, this interaction has more significance. It’s found in clinic that CYP3A4 inhibitor Cyclosporin can increase the blood concentration of Cerivastatin (mainly through CYP2C8 and CYP3A4 enzyme
metabolism), while CYP3A4 inhibitor Erythromycin has no obvious influence over Cerivastatin, Cyclosporin can also increase the blood concentration of Pravastatin (non CYP3A4 enzyme substrate); Yoshihisa et al.’s research showed that Cyclosporin resulted in increased blood concentration and appearance of metabolic drug interaction mainly through inhibiting drug transport protein OATP2 to decrease the active ingestion and concentration of the liver on Cerivastatin and Pravastatin; the other was irreversible inhibitory effect, whose mechanism may be related to the formation of P450-MI complex.

1992, there’s a report in Britain on the event that Hismanal and Terfenadine may induce complications of heart diseases in the patients. After that, CSM (Committee of Safety in Medicine) repeatedly warn clinical patients that do not use Hismanal at a dosage bigger than recommended dosage, and do not use it together with Erythromycin and ketoconazole. Non-sedative anti-histamine Terfenadine can close the potassium ion channel of cardiac muscle and prolong its action potential so that the QT period in ECG is prolonged and ventricular arrhythmia is resulted in. Actually, while using under therapeutic dosage, because of the first pass effect of the liver; Terfenadine has been transferred into Fexofenadine, which is non-toxic to the heart. Only after the pathway of transferring to Fexofenadine is blocked, can Terfenadine result in obvious ventricular arrhythmia, so Terfenadine is prohibited to be combined with
CYP450 inhibitors. Since the appearance of Fexofenadine, most countries gradually prohibited the application of Terfenadine. Jan. 1992-Sep. 1996, English pharmacologist de Ahaja FJ et al. carried an experiment research on whether non-sedative anti-histamine will result in ventricular arrhythmia, the relations between 5 non-sedative anti-histamine of Avastin, Astemizole, Loratadine, Terfenadine and Cetirizine and ventricular arrhythmia were studied, and the influence of age, gender, dosage, administration time as well as their combination with P450 inhibitor over the results were studied. The results showed that Hismanal had the relatively highest risk rate of inducing arrhythmia, and its metabolite still had influence over the heart. 1999, Janssen Company voluntarily retreated Hismanal globally. 1993, the event of interaction between 5-Fu and Sorivudine happened in Japan, which resulted in 15 patients with cancer complicated by herpes zoster died of 5-Fu poisoning.

The above are simple examples of serious consequences resulted from metabolic interactions of western medicines in clinic, the following are commonly seen inhibitory of some CYP450 isoenzymes: CYP1A2: ciprofloxacin, enoxacin, levofloxacin, lomefloxacin, norfloxacin, ofloxacin, isoniazid, selegline; CYP2C9: amiodarone, benzbromarone, fluconazol, ketoconazole, miconazole, fluvastatin, sulfinpyrazone; CYP2C19: isoniazid, desmethylinertraline, fluvoxamine, imipramine, mephenytoin, fluconazol, ritonavir, diazepam, nicardipine, warfarin,
cimetidine, omeprazole; CYP2D6: amiodarone, mexiletine, quinindium, propafenone, amitriptyline, chlorpromazine, fluoxetine, sertraline, fluvoxamine, verapamil, diphenhydramine, cimetidine, omeprazole, ritonavir; CYP3A4: clarithromycin, erythromycin, roxithromycin, cimetidine, ciclosporin, ketoconazole, fluconazole, itraconazole, verapamil, felodipine, nifedipine, fluoxetine, fluvoxamine, ritonavir.

4. Chinese herbal medicine-medicine interaction and CYP450

4.1 Current research conditions in China Domestic scholars’ report of research on the relations between Chinese herbal medicine and CYP450 mainly focused on exploring the influence and effect of certain herb (formula) or herb ingredient on certain CYP450 isoenzymes in in vitro experiments. In recent 10 years, main herbal compound (formula) having influence over CYP450 found at home are: Formula: Yi Kun Ning, Xue Sai Tong, Er Xian Tang, Ju Li San Jie Wan, Xiao Zhi Hu Gan Jiao Nang, Qing Gan Tiao Zhi Yin, etc.; Single Chinese herbal medicine: dang gui, dan shen, ku shen, ren shen, li lu, gua lou, bai ji, ban xia, bei mu, wu tou, huang qin, gan cao, jiang huang, wu wei zi, yin chen, yin xing ye, qing hao, gout eng, mu tong, du zhong, chai hu, bai zhu, da qing ye, ze xie, chen pi, tu si zi, chuang xiong qin, guan ye lian qiao, wu bei zi, e zhu, cang zhu, bai zhu, fu ling, niu bang zi, ju luo, ying xing; Herbal ingredients: aristolochic acid, puerarin, flavonoid ingredient, pilose antler
polysaccharides, furocoumarin, chlorogenic acid, baicalin, phillyrin, kaempfetol, isorhamnetin and quercetin, etc. Among all above Chinese herbs (formulas) or herbal ingredients, those who having quite some different research on effects over CYP450 and confirmed by repeated experiment are mainly aristolochic acid, flavonoid-containing herbs, dang gui, dan shen, ren shen, li lu, gua lou, bai ji, ban jia, bei mu, wu tou, huang qin, gan cao, jiang huang, wu wei zi, yin xing ye, furocoumarin chemicals, guan ye lian qiao, baicalin and phillyrin.

4.2 Research on the relations between combination of Chinese herbal medicine and CYP450.

CYP450 enzymes may be one of the internal factors that cause different functions according to combination and proportion of Chinese herbal medicine. Domestic scholars had certain discovery on the relations between combination of Chinese herbal medicine and Cyp450 enzymes, among them, there’re relatively more research on the relations between medicines in “eighteen incompatible medicine” and CYP450.

Yu Donghua et al. studied the liver toxicity before and after huang yao zi in combination with dang gui, it showed that huang yao zi may induce the mRNA expression of CYP1A3 and CYP2E1, so that its own pre-toxic substance turned into hepatic toxic substance and resulted in hepatotoxicity, when dang gui was in combination with huang yao zi, the inhibition of expression level of mRNA was one of its mechanism of
antagonizing the toxicity of huang yao zi. Ye Xuan et al. studied the influence of ren shen over CYP450 enzymes and the changes of related CYP450 subtype enzyme activity after combining with li lu at general animal level and molecular level, the research showed that single application of ren shen and combination with li lu lowered total content of CYP450 enzymes in the microsome of the liver in rats, increased the activity of CPY1A and inhibited the activity of CYP3A, held that the changes of CYP450 expression and activity affected the metabolism clearance and pharmacological and toxicological functions, which would cause the interaction between ren shen and li ru and thus generate incompatibility of combination. Xiao Chenrong et al studied the influence of ban xia, bei mu, gua lou, bai lian, bai ji in combination with wu tou which were in “eighteen incompatible medicine” over the contents of CYP450 enzymes in rat liver, using spectrophotometer to determine the contents of CYP450 and cytochrome b5 in liver microsome of rats. The result showed that comparing with corresponding single herb groups, combination groups could significantly lower the contents of CYP450 enzymes and b5. Dai Fangguo et al studied the relationship between combination of gan sui and gan cao and CYP450 in liver microsome of rats, found that combination had great influence over the activity of liver CYP1A2 and CYP3A4. Xi Lijun et al used probe drug method to evaluate the influence of combination of sheng jiang and ban xia over
CYP450 enzymes. The experiment showed that ban xia, sheng jiang, and combination of ban xia and sheng jiang could all induce CYP450 enzymes, meanwhile, compared with single application, combination of ban xia and sheng jiang had more inducing effect on CYP enzymes and could counteract the inhibitory effect of single application of ban xia on CYP enzymes. Wang Yuguang et al. explored the mechanism of “eighteen incompatible medicine” in Chinese herbal medicine through hepatic drug enzymes, the results showed that Chinese herbal medicine dan shen, ku shen, ren shen and their combination with li lu all inhibited the contents of CYP450 enzymes and the enzyme activity of main drug metabolism enzymes CYP3A and CYP2E1 to different degrees, which indicated that it could just be due to the inhibitory effect of drug metabolism enzymes caused by combination of the three herbs and li lu that caused the slow-down of the metabolism of certain substances in hyper-toxic herb li lu and thus resulted in the increase of toxicity and unexpected interactions of Chinese herbal medicines. Combination of gua lou, bai ji, ban xia, bei mu and wu tou generates inhibitory effect to different degrees on CYP3A and CYP1A2 at genetic, protein and enzyme activity levels according to their functional intensity. gan cao and its combination with hai zao, da ji, gan sui, yuan hua all had including effect on mRNA expression and enzyme activity of CYP3A, especially the combination of hai zao, da ji, yuan hua and gan cao. Hu Xiqin et al
studied the influence of he shou wu and prepared he shou wu in combination with fu ling over liver microsome CYP450 in rats. The research showed that he shou wu, he shou wu combining large dosage of fu ling could increase the content of CYP450 in liver microsome of rats, but none administration groups of prepared he shou wu showed significant influence on it. Jin ketao et al. illustrated the mechanism of interaction in the combination of wu tou and bai ji based on drug metabolism enzyme, found that the combination of wu tou and bai ji could inhibit the enzyme activity of CYP2A1/2, there’s CYP3A1/2-based drug interaction in the combination of wu tou and bai ji.

4.3 Current research conditions abroad

4.3.1 guan ye lian qiao  It’s found in clinic that when combined with other medicine, guan ye lian qiao could lower the steady state blood concentration of various medicine, including amitriptyline, ciclosporin, digoxin, fexofenadine, indinavir, simvastatin and warfarin, etc, and the molecular mechanism was that various active substance of guan ye lian qiao could activate pregnane X receptor and thus induce the activity of human CYP450 isoenzymes CYP3A4, P-glycoprotein and other enzymes.

4.3.2 ren shen  The activation of transcription of human CYP1A1 gene is mediated by aromatic hydrocarbon receptor, ginsenoside Rg1 and Rb1 could significantly increase the expression of CYP1A1 mRNA in
human Hep G2 cells because ginsenoside Rg1 and Rb1 increased the ability of aromatic hydrocarbon receptor integrating oligonucleotide.

4.3.3 yin xing Some research showed that yin xing in combination with aspirin, acetaminophen, warfarin and ibuprofen etc. could lead to spontaneous bleeding of patients; in combination with anti-epilepsy drugs, it could reduce the therapeutic effects of the drugs. Etheridge AS et al’s research showed that yin xing extract could inhibit the activity of CYP2C8 in the liver microsome of human in vitro.

4.3.4 Milk thistle: There’s a research showed that combination of milk thistle and indinavir could lower the blood concentration of indinavir in vivo in healthy volunteers, and the mechanism lied in that milkthistle could include the activity of CYP2D6. This had dual effects in clinic: one hand it can have the side effect of lowering therapeutic effects of drugs, on the other hand, it can reduce the injury of some toxin such as phenyltoin and ethanol.

4.3.5. ling zhi Reishi polysaccharides, the main active substance of ling zhi had inhibitory effect on the enzyme activity of CYP2E1, CYP1A2 and CYP3A.

5. Conclusion

Chinese herbal medicine is being used more and more widely because of its pure nature property and other advantages. The ingredients
of Chinese herbal medicine are very complex, so more emphasis should be placed on the interactions between Chinese herbal medicine and medicine. More and more clinical and pharmacological experiments proved that many Chinese herbal medicine had influence over CYP450, so the combination of Chinese herbal medicine-Chinese herbal medicine or Chinese herbal medicine-western medicine or compound preparations could increase therapeutic effect, lower toxic and side effects and exert the effects that single medicine can’t have, which showed the advantages of reasonable combination of medicine; on the contrary, unreasonable combination of medicine could lower the therapeutic effect of medicine or increase side or toxic effects.

Currently, the research on the relations between Chinese herbal medicine and CYP450 enzymes at home are not systematic, it’s also limited by the experimental conditions, such as possible false positive results in in vitro research in animals to predict the effect of Chinese herbal medicine on CYP450 enzyme, and result deviation due to polygenic differences. Further research on transcription and translation of single gene and enzyme activity are also absent. Researches on relations between Chinese herbal medicine and CYP450 enzymes made scholars abroad are also limited in some commonly used botanic medicine, which are usually food additives or adjuvant medicine.

The mechanism of Chinese herbal medicine is quite complex,
pharmacokinetic rules of most Chinese herbal medicine are not clear yet. The future task is to further explore the influence of Chinese herbal medicine over CYP450 enzymes and the mechanism of CYP450 enzymes on various kind of Chinese herbal medicine, reveal comprehensively the rules of metabolism, medicine effect and side/toxic effects when Chinese herbal medicine are used in combination, predict the medicine interaction caused by combination of medicine in clinic, prevent side effects of medicine combination and make sure of the reasonability of clinical administration and popularization of Chinese herbal medicine.