

Cytochrome P450

3. Clinically Significant Drug Interactions

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ABSTRACT

Recent advances in cytochrome P450 research have provided insights into the precise human cytochrome P450 complement, the role of individual P450 enzymes in drug metabolism and the various factors modulating P450 activity. Resulting from these advances is a clearer understanding of many established drug-drug and drug-food interactions, and an enhanced capacity to predict interactions likely to occur with newly introduced drugs. This final article in the three-part cytochrome P450 series describes our current understanding of drug interactions involving cytochrome P450 enzymes and highlights several clinically important examples.

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INTRODUCTION

It is well recognised that drug-drug interactions can have a significant negative impact on drug use. As evidence, we need look no further than the drug mibefradil, which was withdrawn in June 1998 solely on the basis of its propensity for inhibiting the cytochrome P450-mediated metabolism of other drugs. At the time of its withdrawal, it was reported that more than 25 drugs were 'potentially dangerous' if used in combination with mibefradil. Based on our current knowledge of the P450 superfamily and the known metabolic properties of mibefradil, most of these interactions were predictable (indeed most were listed in official monographs). Arguably, many of the harmful effects that occurred in patients who took mibefradil while taking other drugs could have been avoided if their prescribers and pharmacists had been more aware of P450-mediated drug metabolism interactions.¹ Similarly, many potentially important drug interactions with new and existing drugs can be avoided with a good working knowledge of the drugs that act as substrates, inhibitors and inducers of the relatively small number of P450 enzymes that are involved in drug metabolism. However, an understanding of basic pharmacokinetic principles is also required if this knowledge is to be applied in the clinical setting.²

As discussed in earlier articles in this series,^{3,4} there are approximately 50 individual human cytochrome P450 enzymes, yet only a small number of these are involved in the metabolism of drugs. These include the major hepatic P450, CYP3A4, the genetically variable CYP2D6 and CYP1A2 and enzymes of the CYP2C subfamily. Details regarding specific cytochrome P450 enzymes, the drugs they metabolise (i.e. substrates) and the drugs that cause inhibition and induction, are summarised in Table 1.⁵ Substrates are those drugs that significantly rely on the given enzyme for elimination from the body, while inhibitors are those compounds that are generally

capable of inhibiting the metabolism of the various substrates (therefore, the administration of the inhibitor may lead to an increased plasma concentration of the listed substrate). Inducers of the specified P450 have the capacity to increase the activity of the designated enzyme and therefore reduce the plasma concentrations of the listed substrates. It should be noted the table is not comprehensive. Information concerning the metabolic fate of many drugs is still lacking, and many drugs are metabolised by multiple P450 enzymes and are thus listed under more than one enzyme. Further, the listing of two drugs under the same P450 does not indicate a definite interaction of clinical significance. The exhaustive body of literature on cytochrome P450-mediated drug interactions has been the focus of many comprehensive reviews in recent years,⁶⁻⁹ and there is also a wealth of papers dealing with interactions for specific drug classes.

INHIBITION OF CYTOCHROME P450-MEDIATED DRUG METABOLISM

Inhibition of metabolism is probably the most common cause of clinically important pharmacokinetic drug interactions, because it can lead to a dramatic increase in the plasma concentrations of an affected drug. Inhibition generally occurs because the cytochrome P450 enzymes that are involved in drug metabolism elicit broad substrate specificity, and even though two drugs may be chemically quite distinct, they may compete with one another for binding to the catalytic site of a common enzyme. Fortunately, there are relatively few drugs that are capable of significantly inhibiting drug metabolism (Table 1). Well-characterised inhibitors include cimetidine, erythromycin, ketoconazole and quinidine, while newer drugs that are potent inhibitors include selective serotonin reuptake inhibitors and protease inhibitors. Caution should be exercised whenever one of these 'enzyme inhibitors' is added to or withdrawn from a patient's drug regimen if the patient is also taking other drugs primarily eliminated by metabolism. At the very least, to assess whether a particular drug can be safely used in combination with an enzyme inhibitor, information on the role of cytochrome P450 in the metabolism of the drug, and ideally the specific enzymes that are involved, can be extremely useful.⁶⁻⁹

Inhibition of P450-mediated drug metabolism is caused either by direct competition between two drugs for a common P450 enzyme binding site, or by mechanism-based inhibition or suicide inactivation of a P450 enzyme.

The time that is taken for the full impact of an inhibitory interaction to be elicited generally depends on the pharmacokinetic properties of the inhibitor and the affected drug—as a general rule, it may take about 4 half-lives of either the inhibitor or the affected substrate—whichever is longer. If both the inhibitor and the affected drug have short half-lives (e.g. cimetidine as an inhibitor of theophylline metabolism), the interaction would probably be maximal within 2 to 4 days and clearance of

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Table 1. Cytochrome P450 (CYP450) enzymes involved in drug metabolism⁵

CYP450	Substrates	Inhibitors	Inducers
CYP1A2	Amitriptyline, caffeine, clomipramine, clozapine, imipramine, mexilitine, oestradiol, olanzapine, paracetamol, propranolol, tacrine, theophylline, R-warfarin	Cimetidine, fluvoxamine, grapefruit juice, quinolone antibiotics (ciprofloxacin, enoxacin, norfloxacin)	Cigarette smoking, omeprazole, phenobarbitone, phenytoin, polycyclic aromatic hydrocarbons (e.g. charbroiled meat)
CYP2C9	Diclofenac, indomethacin, losartan, naproxen, phenytoin, piroxicam, tenoxicam, tetrahydrocannabinol, tolbutamide, S-warfarin	Amiodarone, chloramphenicol, cimetidine, fluconazole, fluoxetine, isoniazid, omeprazole, sertraline, sulfaphenazole, sulfipyrazone	Rifampicin
CYP2C19	Citalopram, clomipramine, clozapine, diazepam, imipramine, lansoprazole, methylphenobarbitone, omeprazole, pentamidine, phenytoin, propranolol, topiramate	Fluoxetine, fluvastatin, fluvoxamine, isoniazid, omeprazole, sertraline, ticlopidine, tranlycypromine	Rifampicin
CYP2D6	Alprenolol, amitriptyline, chlorpromazine, citalopram, clomipramine, clozapine, codeine, desipramine, dextromethorphan, diphenhydramine, doxepin, encainide, fenfluramine, flecainide, fluoxetine, fluphenazine, haloperidol, hydrocodone, imipramine, labetalol, methadone, metoprolol, mexiletine, mianserin, nortriptyline, ondansetron, oxprenolol, oxycodone, paroxetine, perhexiline, perphenazine, pethidine, procainamide, promethazine, propafenone, propranolol, risperidone, thioridazine, timolol, trimipramine, venlafaxine	Amiodarone, cimetidine, fenfluramine, haloperidol, mibefradil, quinidine, propafenone, ritonavir, selective serotonin reuptake inhibitors (all SSRIs inhibit 2D6 with fluoxetine and paroxetine the most potent), thioridazine, yohimbine	
CYP2E1	Caffeine, dapson, ethanol, halothane, paracetamol, theophylline	Cimetidine, disulfiram	Ethanol, isoniazid
CYP3A4	Alprazolam, amiodarone, amitriptyline, astemizole, atorvastatin, budesonide, buprenorphine, busulphan, carbamazepine, cisapride, clarithromycin, clomipramine, clonazepam, clozapine, cocaine, cortisol, cyclophosphamide, cyclosporin, dapson, dexamethasone, dextromethorphan, digitoxin, diltiazem, diazepam, doxorubicin, erythromycin, ethinyloestradiol, ethosuximide, etoposide, felodipine, fentanyl, fexofenadine, flutamide, ifosfamide, imipramine, indinavir, ketoconazole, loratadine, losartan, lovastatin, miconazole, midazolam, nifedipine, nelfinavir, oestradiol, omeprazole, ondansetron, paclitaxel, propafenone, quinidine, ritonavir, saquinavir, sertraline, simvastatin, tacrolimus, tamoxifen, teniposide, tetrahydrocannabinol, theophylline, trazadone, troleandomycin, verapamil, vinblastine, vincristine, warfarin	Amiodarone, cannabinoids, cimetidine, clarithromycin, clotrimazole, delavirdine, diltiazem, erythromycin, fluoxetine (due to norfluoxetine metabolite), fluvoxamine, grapefruit juice, itraconazole, ketoconazole, metronidazole, miconazole, nefazodone, paroxetine, protease inhibitors (all inhibit 3A4 with ritonavir the most potent), troleandomycin	Carbamazepine, ethosuximide, glucocorticoids, phenobarbitone, phenytoin, rifampicin, sulfadimidine, nevirapine, sulfipyrazone

the affected drug would be expected to return to normal within a similar timeframe after cessation of the inhibitor.¹⁰ In contrast, the half-life of warfarin is 2 days and that of amiodarone, which inhibits the metabolism of warfarin via CYP3A4 and CYP2C9, is much longer. In this case, since amiodarone has the longer half-life, it might take one month or longer for the full impact of the interaction to be observed. In this clinical scenario, prolonged monitoring of blood clotting is warranted.¹¹

Even though a drug may be a substrate for a particular cytochrome P450 enzyme, it does not necessarily follow that it will be capable of inhibiting the metabolism of other drugs that are also substrates for the same enzyme. To appreciate this, it is necessary to define the term K_i , which is the concentration of the ‘inhibitor’ that will cause a two-fold reduction in the rate of metabolism of a second drug at a given concentration. In most cases, K_i values are obtained *in vitro* by studying the effects of the ‘inhibitor’ on the metabolism of a particular substrate. Human liver microsomes are the most appro-

prate model for these *in vitro* studies. If, in the clinical setting, the unbound concentration of the ‘inhibitor’ in the vicinity of the enzyme is much less than its K_i value, then inhibition will not be discernible.

In human liver microsomes, the reported K_i value for ketoconazole inhibition of CYP3A4 was found to be just 0.015 μM ,¹² while antifungal effects occur at plasma concentrations of about 10 μM . Even allowing for the extensive plasma protein binding of ketoconazole *in vivo* (99%), the unbound concentration in plasma, and presumably within the cell cytosol is approximately 0.1 μM —much greater than its *in vitro* K_i value. Therefore, it is entirely predictable that ketoconazole would be a strong inhibitor of CYP3A4-mediated drug metabolism *in vivo*.

As a general rule, drugs that are affected by cytochrome P450-mediated drug interactions are those that rely extensively on metabolism for elimination from the body. For such drugs, only a relatively small fraction of a dose is excreted unchanged in urine or bile. Moreover, a drug whose metabolism is carried out predominantly

by one individual P450 enzyme is particularly susceptible since inhibition of that one enzyme can cause a substantial reduction in the metabolic clearance. The conversion of terfenadine (now withdrawn because of cytochrome P450-mediated drug interactions) to its carboxylic acid metabolite fexofenadine is catalysed almost exclusively by CYP3A4, and inhibition of this enzyme by drugs such as azole antifungals caused significant elevations in the blood levels of the parent compound, which was cardiotoxic.¹³

Although inhibition of cytochrome P450-mediated metabolism is a considerable problem in clinical practice, there are circumstances where the phenomenon can be exploited. Rightly or wrongly, the use of diltiazem, an inhibitor of CYP3A4, to reduce the dosage requirements and the cost of cyclosporin (a CYP3A4 substrate) is well documented.¹⁴⁻¹⁶ A more recent example includes the combined use of the protease inhibitors, saquinavir and ritonavir, to substantially increase the bioavailability of oral saquinavir.¹⁷ A more acceptable strategy could be the development of pharmacologically inactive substrates that would inhibit metabolism, and therefore reduce dose and/or dose frequency, without adding to the risk of toxicity.

For some drugs, cytochrome P450-mediated metabolism represents pharmacological or toxicological activation. In the case of codeine, for example, conversion to the active moiety morphine relies on CYP2D6-mediated O-demethylation. Inhibition of this process by drugs such as quinidine can potentially reduce the clinical benefits of codeine. A recent study compared the effect of quinidine (an inhibitor of CYP2D6) on the pharmacokinetics and pharmacodynamics of codeine in Caucasian and Chinese subjects.¹⁸ Codeine O-demethylation was significantly reduced after quinidine in both ethnic groups. Interestingly, the extent of inhibition was greater in the Caucasian population, and in Caucasians there was a marked reduction in codeine's pharmacodynamic effects. These authors concluded that the quinidine inhibition of codeine O-demethylation is influenced by ethnicity with the effect being greater in Caucasians. This is an excellent example of how the clinical impact of a drug interaction can be modulated by genetic factors.¹⁸ As our understanding of cytochrome P450 expression develops, it may become possible to identify subgroups of patients that are most susceptible to a particular drug-drug interaction.

INDUCTION OF CYTOCHROME P450-MEDIATED DRUG METABOLISM

Induction of metabolism arises when one drug induces or stimulates the synthesis (or alternatively reduces the natural degradation) of enzymes involved in the metabolism of another. Rifampicin is perhaps the most widely recognised cytochrome P450 enzyme inducer—this drug increases hepatic levels of CYP3A4 and CYP2C enzymes resulting in an increase in the intrinsic metabolic clearance of a wide range of drugs. These include, predictably, those drugs that are highly reliant on the affected enzymes (e.g. cyclosporin, metoprolol, quinidine, theophylline and tolbutamide).¹⁹ Other notable drugs that induce cytochrome P450-mediated drug metabolism include phenytoin, phenobarbitone and carbamazepine. In addition to drugs, cytochrome P450-mediated drug metabolism can be induced by a diverse range of non-

drug chemicals including dietary-derived substances and environmental contaminants. Cigarette smoke is a potent inducer of CYP1A2 and can reduce the plasma levels of drugs that are substrates for this enzyme, including caffeine, theophylline and warfarin.

Because the introduction of an enzyme inducer leads to a reduction in the plasma concentrations of a drug that is a substrate for the inducible cytochrome P450, it is most likely to lead to a lack of drug effect. However, when an inducing agent is withdrawn from the regimen of a patient receiving multiple-drug therapy, there is a possibility that the plasma concentrations of other drugs may increase. The impact of such a scenario is likely to be felt in coffee drinkers who decide to quit smoking. As stated earlier, cigarette smoke is known to accelerate the metabolism of a range of drugs including caffeine by inducing CYP1A2.²⁰ Therefore, cessation of smoking can be associated with a reduction in hepatic CYP1A2 and a resultant increase in the plasma concentration of caffeine. This may contribute to the perceived symptoms of the tobacco withdrawal syndrome, including headache and agitation.

ASSESSMENT OF CYTOCHROME P450 EFFECTS DURING DRUG DEVELOPMENT

The various drug regulatory authorities have recognised the clinical importance of drug interactions arising from inhibition and induction of cytochrome P450. As a result, a guidance document has been issued aimed at ensuring that the propensity for a new drug to become involved in such interactions is understood at an early stage of drug development.²¹ Regulatory authorities further recognise that much of the required information for predicting such interactions in humans can be obtained from *in vitro* experiments. This has become possible because of the substantial progress that has been made over the last 20 years in our understanding of the cytochrome P450 superfamily and of the factors that govern and regulate *in vivo* drug metabolism.⁷ More recently, mathematical models have been developed that assist in deciding whether an *in vitro* interaction will be of concern in clinical practice.²²

As new drugs become available on the market, specific details regarding the cytochrome P450 enzymes that are responsible for their metabolism will be included in the official monographs. It should therefore be possible to anticipate the wisdom of using the drug in patients taking known enzyme inducers or inhibitors. If, for example, it is reported that a new drug relies extensively on CYP3A4-mediated metabolism, then one would anticipate that inhibitors of this enzyme, such as ketoconazole, would interact with the new drug. However, history has shown that warnings in product information are not sufficient for preventing the coadministration of potentially dangerous drug combinations.

FOOD-DRUG INTERACTIONS

Dietary changes can alter the expression and activity of hepatic cytochrome P450 enzymes and although this can lead to alterations in drug metabolism, the magnitude of the change in systemic clearance is generally small.²³ Classic examples include the ability of an increased intake of cruciferous vegetables or charcoal-broiled beef to induce the oxidative metabolism of drugs such as theophylline. The 1990s has seen much publicity given to

what has become known as the 'grapefruit-juice interaction'. Interest in this seemingly unlikely interaction stems from a study in 1991, in which it was demonstrated that a single glass of grapefruit juice caused a 2-3 fold increase in the plasma levels of felodipine—a similar amount of orange juice did not affect the pharmacokinetics of the drug.²⁴ It is believed that a component of grapefruit juice—possibly a flavonoid or furanocoumarin—inhibits the intestinal (but not hepatic) CYP3A4-mediated metabolism of felodipine and at least 25 other drugs that are metabolised in this way. The growing list of drugs affected by grapefruit juice include a range of calcium channel blockers, cyclosporin, midazolam, terfenadine, ethinyl oestradiol, simvastatin, lovastatin and buspirone.^{25,26}

The discovery of the grapefruit interaction has created an increased awareness of the potential for dietary substances to alter drug metabolism, and as we learn more about how food-drug metabolism interactions contribute to variability in drug response, we may be in a position to advise patients taking particular drugs on what specific foods should be avoided. Alternatively, it is feasible that foods such as grapefruit juice could be used to reduce dosage requirements.

Inhibition of Intestinal CYP3A4

In humans, CYP3A4 is expressed not only in the liver, but in various extra-hepatic tissues, most notably the intestine. For some drugs, including cyclosporin, saquinavir and midazolam, a significant fraction of an oral dose can actually be metabolised prior to reaching the mesenteric blood. The potential exists for this intestinal metabolism to be reduced significantly by inhibitors of CYP3A4, and when one considers that concentrations of these inhibitors within the intestine during the absorption process will be orders of magnitude higher than the corresponding levels in plasma, it is apparent that intestinal inhibition may in some circumstances take on great importance. The oral antiviral drug saquinavir has a very low bioavailability due to extensive presystemic intestinal and hepatic metabolism via CYP3A4—indeed its oral bioavailability is only about 4%. Coadministration of the related compound ritonavir, which exerts potent CYP3A4 inhibitory properties, caused a 50-fold increase in the area under the plasma concentration-time curve of a single oral dose of saquinavir.¹⁷ It was concluded that the large effect of ritonavir on the pharmacokinetics of saquinavir is due to an inhibition of CYP3A4-mediated metabolism of the latter drug in the intestine during absorption, in the liver during the hepatic first-pass, and in the liver during the post-absorptive period. In addition to increasing the bioavailability of saquinavir, ritonavir also reduced inter-subject variability in its pharmacokinetics—it essentially converted saquinavir from a drug with highly variable pharmacokinetics to one in which intersubject variability was relatively small.

SUMMARY

As our understanding of cytochrome P450 has increased, so too has our ability to predict potential drug interactions, both *in vitro* and *in vivo*. Despite this increased sophistication in our overall knowledge of P450s and the drug metabolism process, there is no place for complacency in the delivery of this information to the clinic. In a recent review of the mibefradil withdrawal,¹ Krayen-

bühl and colleagues from the Swiss Intercantonal Office for the Control of Medicines concluded, firstly, that 'when interpreting the results of interaction studies, it is important to consider not only the mean of the interaction effect but also the observed and the theoretically conceivable extreme effects in individual subjects' and, secondly, that 'a drug with a high interaction potential may represent a high risk even if an adequate warning is included in the product information'.

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