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REVIEW

Cytochrome *P*450 pharmacogenetics and cancer

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The cytochromes *P*450 (CYPs) are key enzymes in cancer formation and cancer treatment. They mediate the metabolic activation of numerous precarcinogens and participate in the inactivation and activation of anticancer drugs. Since all CYPs that metabolize xenobiotics are polymorphic, much emphasis has been put on the investigation of a relationship between the distribution of specific variant CYP alleles and risk for different types of cancer, but a consistent view does not yet exist. This is to a great extent explained by the fact that the CYPs involved in activation of precarcinogens are in general not functionally polymorphic. This is in contrast to CYPs that are active in drug biotransformation where large inter-individual differences in the capacity to metabolize therapeutic drugs are seen as a consequence of polymorphic alleles with altered function. This includes also some anticancer drugs like tamoxifen and cyclophosphamide metabolized by CYP2D6, CYP2C19 and CYP2B6. Some *P*450 forms are also selectively expressed in tumours, and this could provide a mechanism for drug resistance, but also future therapies using these enzymes as drug targets can be envisioned. This review gives an up-to-date description of our current knowledge in these areas.

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Introduction to cytochrome *P*450 and cancer

Cytochrome *P*450 (CYP) enzymes (http://drnelson.utmem.edu/cytochromeP450.html) are key players in the phase I-dependent metabolism of drugs and other xenobiotics, mostly catalysing oxidations of the substrate, but occasionally also reduction reactions. As a result of the CYP-dependent metabolism, intermediates that often exert toxicity or carcinogenicity, but which also are targets for phase II enzyme dependent conjugation reactions are formed, rendering them inactive polar products suitable for excretion via the kidneys. Several exceptions are of course at hand where the phase II-dependent metabolism could produce more harmful products than the parent compounds, although this is not common. Many different cytotoxic drugs are inactivated by the action of CYP, whereas several prodrugs are activated by the action of CYP, rendering them cytotoxic and effective in cancer chemotherapy (McFadyen *et al*., 2004). Therefore, because of the important role of the CYPs in the bioactivation and inactivation of carcinogens and their participation in the activation and inactivation of anticancer drugs, they play an important role both in the aetiology of cancer diseases and as determinants of cancer therapy (Oyama *et al*., 2004; Rooseboom *et al*., 2004). These processes are mainly hepatic, but the activity of *P*450s in extrahepatic tissues might also be critical.

At present more than 57 active human *P*450 genes and 58 pseudogenes are known (Ingelman-Sundberg, 2004a; Nelson *et al*., 2004). The majority of genes are polymorphic and at the human CYP allele home page (http://www.imm.ki.se/cypalleles/) updated information is presented regarding the nomenclature and properties of the variant alleles with links to the dbSNP database (http://www.ncbi.nlm.nih.gov/projects/SNP/) and relevant literature references. At present, more than 434 different alleles of the genes encoding xenobiotic metabolizing *P*450 enzymes are presented on the page, as well as several SNPs with functional consequences, but where the corresponding allele has not yet been identified. The most polymorphic CYPs on the Web site are *CYP2B6* (48 alleles), *CYP2C9* (32), *CYP2D6* (92) and *CYP3A4* (34). Most of the functional polymorphisms are seen regarding the variability in the *CYP2A6, CYP2B6, CYP2C9, CYP2C19* and *CYP2D6* genes.

The mutations in the *CYP* genes may cause absence of enzyme, diminished enzyme expression, enzyme with altered substrate specificity or increased enzyme expression. Based on the composition of the alleles, the affected individuals might be divided into four major phenotypes: poor metabolizers (PMs), having two nonfunctional genes, intermediate metabolizers (IMs) being deficient on one allele, extensive metabolizers (EMs) having two copies of normal genes and ultrarapid metabolizers (UMs) having three or more functional active gene copies (see Ingelman-Sundberg, 2004b; Ingelman-Sundberg and Rodriguez-Antona, 2005). In the CYP gene family, the most penetrant genetic alterations are gene deletions, missense mutations and

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mutations creating splicing defects and premature stop codons. In only few examples mutations in the 5′- or 3′-untranslated regulatory regions affect the CYP phenotype. Despite this, a huge amount of literature reports association studies linking such low-penetrance polymorphisms to the incidence of severe diseases, among them various types of cancer (Agundez, 2004; Ingelman-Sundberg, 2004a).

The polymorphic xenobiotic metabolizing CYP enzymes can be mainly divided into two classes:

Class I, composed of CYP1A1, CYP1A2, CYP2E1 and CYP3A4, which are well conserved, do not have important functional polymorphisms, and are active in the metabolism of precarcinogens and drugs.

Class II, composed of CYP2B6, CYP2C9, CYP2C19 and CYP2D6, which are highly polymorphic and active in the metabolism of drugs, but not of precarcinogens.

No common polymorphic variant with a mutation in the open reading frame has been described for the Class I group of enzymes (Table 1). This is surprising in view of the lack of any important phenotype in the knockout mice for these CYP enzymes (Gonzalez, 2003). However, transgenic CYP3A4 knockout mice develop endocrine alterations (Yu *et al*., 2005) and specific functions of possible endocrine character during some phase of human development, which could explain the degree of conservation, cannot be excluded. At present only one subject with a true defective *CYP3A4* allele has been documented, where the capacity for midazolam hydroxylation was severely decreased (Westlind-Johnsson *et al*., 2006). CYP1B1 represents a special case where several rare defective alleles have been identified and their occurrence associated to glaucoma, and, in addition, many common variant haplotypes with missense mutations are distributed in the population, but their functional consequences are less pronounced. The *Cyp1b1* knockout mice develop ocular drainage structure abnormalities resembling those reported in patients having primary congenital glaucoma (Libby *et al*., 2003).

Association studies of CYP polymorphisms with cancer incidence

Owing to the important role of CYPs in the metabolic activation of precarcinogens (see Table 2), hundreds of studies aimed at finding genetic variants that could predispose to certain types of cancer have been carried out in the past. In essence, no major conclusions from these association studies can be drawn at present. This is to a great extent due to many negative studies, relatively small risk factors obtained requiring large number of cases and controls, lack of proper control of confounding factors, relatively small functional alterations between the variant alleles studied, low penetrance of the *P*450 reaction in question for the development of the cancer and strong environmental factors in the etiology

Table 1 Polymorphic cytochromes *P*450 of importance for the metabolism of drugs and carcinogens

Enzyme	*Substrates*	*Polymorphism*		
		Frequency	*Functional effects*	*Most important polymorphic variants*
CYP1A1	Carcinogens	Relatively high	Unproven	
CYP1A2	Drugs, carcinogens	High	Some	*CYP1A2*1F, CYP1A2*1K*
CYP1B1	Carcinogens, oestrogens	Rare null alleles, frequent missense mutations	At least seven haplotypes with similar activity	*CYP1B1*7*
CYP2A6	Nicotine, drugs, carcinogens	High in orientals, less frequent in Caucasians	Important for nicotine metabolism	*CYP2A6*1B, CYP2A6*4, CYP2A6*9, CYP2A6*12*
CYP2B6	Drugs	High	Reduced drug metabolism	*CYP2B6*5, CYP2B6*6 CYP2B6*16*
CYP2C8	Some drugs	High	Reduced drug metabolism	*CYP2C8*3*
CYP2C9	Drugs	Relatively low	Very significant	*CYP2C9*2, CYP2C9*3*
CYP2C19	Drugs	High	Very significant	*CYP2C19*2, CYP2C19*3, CYP2C19*17*
CYP2D6	Drugs	High	Very significant	*CYP2D6*2xn CYP2D6*4, CYP2D6*5, CYP2D6*10, CYP2D6*17*
CYP2E1	Carcinogens, solvents, few drugs	Low	No	
CYP3A4	Drugs, carcinogens	Low	No or small	*CYP3A4*1B*
CYP3A5	Drugs, carcinogens	High	Significant	*CYP3A5*3, CYP3A5*6 CYP3A5*7*
CYP3A7	Drugs, carcinogens	Low	Some	

Table 2 Precarcinogens metabolized by cytochromes *P*450

Enzyme	*Activation of carcinogens*
CYP1A1	Polycyclic aromatic hydrocarbons: benzo(*a*)pyrene, dimethylbenz[*a*]anthracene, PhIP[a]
CYP1A2	Activation of aryl and heterocyclic amines in industrial settings and food mutagens: *N*-nitrosodimethylamine, 4-aminobiphenyl, 2-acetyl-aminofluorene, *N*-nitrosodiethylamine, PhIP, IQ, aflatoxin B1
CYP1B1	Polycyclic aromatic hydrocarbons: benzo(*a*)pyrene, dimethylbenz[*a*]anthracene, benz[*a*]anthracene, 3-methylcholanthrene, DMBA, oestradiol
CYP2A6	Activation of tobacco-related *N*-nitrosamines: NNK, NNAL, NDEA, NNN, NATB, Aflatoxin B1, 1,3-butadiene, 2,6-dichlorobenzonitrile
CYP2B6	Aflatoxin B1 and 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone
CYP2E1	Low-molecular-weight toxicants and cancer suspect agents: benzene, carbon tetrachloride,chloroform, styrene, vinyl chloride, vinyl bromide, *N*-nitrosodimethylamine, NNK
CYP3A4/5/7	Diverse carcinogens: aflatoxin B1, aflatoxin G1, benzo(*a*)pyrene, naphthalene, NNN, 1-nitropyrene, 6-amino-chrysene, oestradiol, senecionine, stergmato-cystine

[a]DMBA, 7,12,-dimethylbenz[a]anthracene; IQ, 2-amino-3-methylimidazo[4,5-f]quinoline; NATB, *N*-nitrosoanatabine; NDEA, *N*-nitrosodiethylamine; NNAL, 4-(methylnitrosoamino)-1-(3-pyridyl)-1-butanol; NNK, 4-(methylnitrosoamino)-1-(3-pyridyl)-1-butanone; NNN, *N*9-nitrosonornicotine; PhIP, 2-amino-1-methyl-6-phenylimidazo-[4,5-b]pyridine.

of the type of cancer. It is beyond the aim of this review to summarize this literature.

A simple case clearly illustrating the role of a specific CYP in cancer formation is the null mice for *Cyp1b1*. Dimethylbenzanthracene (DMBA) is activated by CYP1B1, and carcinogenicity and adduct formation following DMBA injections are more frequent in mice carrying the enzyme than in the corresponding (−/−) mice (Buters *et al*., 1999, 2003). This system is simple in that: (i) only one gene product is studied, (ii) the genetic variation studied is drastic, that is, wt or null variants and (iii) one single carcinogen at high doses is studied. Furthermore, the mice are genetically identical except for the *Cyp1b1* gene variation, and exposed to the same type of environment and food, which minimizes the effect of uncontrolled factors in the study. In contrast, such clear studies relating the influence of one specific *P*450 gene on cancer risk cannot be done in humans. This is because: (i) in contrast to the clear phenotypes of PMs vs EMs frequently affecting drug metabolism, such distinct phenotypes cannot be identified for most genes encoding precarcinogen-activating *P*450s and most of the SNPs studied have only subtle functional effects (see above); (ii) the polygenic influence on genetic susceptibility to cancer is often pronounced and the effect of *P*450 variation, representing a low penetrance genetic factor, is difficult to detect; (iii) the environmental factors differ a lot between individuals and are difficult to compensate for; (iv) diet, age, hormonal status, previous diseases, etc represent confounding factors that have to be taken into account and (v) the frequencies of the polymorphisms studied are often relatively low, which would require very large well-phenotyped cohorts in order to get reliable data.

Among the CYPs studied in relation to cancer, CYP1A1 and CYP2E1 have been the most commonly investigated. Although these enzymes are involved in the activation of several different precarcinogens, the allelic variants have not shown any consistent functional effect. Thus, in 16 studies investigating *CYP1A1* polymorphisms and breast cancer, no associations have been found (Masson *et al*., 2005). In the case of CYP2E1, the c1/c2 polymorphism, an SNP at −1053 bp in the 5′-upstream regulatory region has been much studied because of one early study reporting a higher expression of the c2 allele in a gene reporter system (Hayashi *et al*., 1991). However, this result has not been reproduced by any other group and may indeed just represent an incidental finding. In our opinion this is a common tendency in the field of cancer association studies, where unfortunately sometimes little emphasis is given to the functionality of the genetic variations and association studies are performed without rigorous pre-validation. In addition, as mentioned, it is difficult to study such polymorphisms in case–control studies because of their relatively small role in the overall risk for cancer (cf. Vineis, 2002).

Cancer incidence, cancer therapy and CYP polymorphisms

CYP1A1/2

CYP1A1 is expressed extrahepatically and CYP1A2 is mainly expressed in the liver, indicating a very different basal regulation, but they share induction via the aryl hydrocarbon receptor (AhR), similarly to CYP1B1 (Hankinson, 1995). The CYP1A enzymes activate and detoxify numerous environmental polycyclic aromatic hydrocarbons (PAHs) and aromatic and heterocyclic amines present in combustion products such as cigarette smoke and charcoal-grilled foods. Thus, interindividual differences in CYP1A1/2 activity may influence individual susceptibility to cancer risk. CYP1A1 activity does not seem to be very variable, but there are large interindividual differences in CYP1A2 activity (Guengerich *et al*., 1999) and, interestingly, a study with twins phenotyped for CYP1A2 with caffeine activity showed that CYP1A2 activity was governed mainly by genetic factors, but also showed that induction by smoking is a powerful environmental factor that influences activity (Rasmussen *et al*., 2002). Common polymorphisms with important functional effects in CYP1A2 activity have not been identified, and only a couple of very rare genetic variants, *CYP1A2*7* and *CYP1A2*11*, have been described. However, two common putatively important genetic variants are *CYP1A2*1F* and *CYP1A2*1K*. The former, with a −163C > A change in intron 1, has been associated to a higher induction by smoking (Sachse *et al*., 1999) and omeprazole (Han *et al*., 2002) using caffeine as a probe drug. However, no molecular mechanism for this increased inducibility has

been provided and other studies regarding the altered inducibility of the allele have been negative. On the other hand, *CYP1A2*1F* has been associated with increased risk for colorectal cancer (Landi *et al*., 2005). The *CYP1A2*1K* allele (−739G, −729T and −163A) results in lower constitutive CYP1A2 activity and the mutation at −729 abolishes the binding of nuclear proteins, presumably of the Ets family (Aklillu *et al*., 2003). Further studies are needed in order to evaluate the functional consequences of these two variant alleles and their possible influence on carcinogen-induced cancers.

CYP1B1

CYP1B1 is predominantly extra-hepatic and is frequently overexpressed in tumour tissue. Similarly to the CYP1A enzymes, CYP1B1 expression is induced by the AhR and the enzyme has an important role in the metabolism of polyaromatic carcinogens. CYP1B1 also metabolizes steroid hormones and may play a role in susceptibility to hormone-dependent cancers such as those from the breast and prostate. Five common missense mutations causing amino-acid substitutions in CYP1B1 have been identified and seven haplotypes carrying one or more of these SNPs have been characterized. For one haplotype, the corresponding enzyme CYP1B1.7 was shown to exhibit a significantly decreased capacity to metabolize estradiol (Aklillu *et al*., 2002) and benzo[*a*]-pyrene (Aklillu *et al*., 2005), which suggests a potential role in the interindividual differences in cancer risk or in hormone therapy. With respect to the metabolism of anticancer drugs, McFadyen *et al*. (2001b) showed that a cell line overexpressing CYP1B1 had a significantly decreased sensitivity towards docetaxel and Bournique and Lemarie (2002) showed that the underlying mechanism was the binding of docetaxel to CYP1B1 and an effector action of this enzyme. CYP1B1 is also involved in the 2-hydroxylation of flutamide (Rochat *et al*., 2001). In general, the CYP1B1 enzyme is not believed to play any major role for the overall clearance of drugs because of its extrahepatic localization, but it may play critical roles in the tissue-specific metabolism of certain drugs and physiological compounds.

CYP2A6

In the human CYP2A family, three genes, *CYP2A6*, *CYP2A7* and *CYP2A13*, have been reported, but *CYP2A7* is a pseudogene and *CYP2A13* mainly expressed in olfactory mucosa. The *CYP2A6* gene is highly polymorphic (Oscarson, 2001) and the variant genes of highest importance are *CYP2A6*4*, representing a gene deletion mainly present in Asian populations, *CYP2A6*9*, having a mutation in the TATA box which causes a decreased expression of the enzyme, and *CYP2A6*1B*, where a gene conversion event with *CYP2A7* creates a 3′-UTR that stabilizes the corresponding mRNA (Wang *et al*., 2006), resulting in higher metabolism *in vivo* of, for example, nicotine (Nakajima *et al*., 2001; Gambier *et al*., 2005). The most important functionally altered allele, *CYP2A6*4*, has a 7–22% allele frequency in Asians, but only 0.5–1% in Caucasians (Oscarson *et al*., 1999). Another defective allele in Caucasians is the *CYP2A6*2*, but it is very rare. Thus, *CYP2A6* does not exhibit very important polymorphism in Caucasians. As with many *CYP* genes, genotyping for the various alleles is difficult due to the risk of amplifying the homologous *CYP2A7* pseudogene, and careful controls of the primary PCR products are necessary. The higher expression of the CYP2A6 enzyme among carriers of *CYP2A6*1B* apparently affects smoking behavior (Malaiyandi *et al*., 2005), and, for example, Gambier *et al*. (2005) reported that subjects homozygous for *CYP2A6*1B* smoked more cigarettes per day as compared to subjects homozygous for *CYP2A6*1A*. In Japan, where the defective *CYP2A6*4* allele is very common, carriers of this genotype have been shown to have less risk of tobacco-induced lung cancer (Ariyoshi *et al*., 2002). This can possibly be explained by higher cigarette consumption in carriers of active *CYP2A6* alleles and/or by a higher rate of formation of carcinogenic products by the action of the active CYP2A6 enzyme.

CYP2A6 metabolizes a number of tobacco-related precarcinogens (Table 2), as well as clinically important drugs such as nicotine, coumarin, methoxyflurane, halothane, valproic acid and disulfiram. Concerning anticancer drugs, CYP2A6 catalyses the activation of tegafur to 5-fluorouracil, a drug commonly used for colorectal cancer. In one study, a patient having a poor tegafur-metabolizing phenotype was found to be heterozygous for *CYP2A6*4* and *CYP2A6*11* (Daigo *et al*., 2002). However, CYP2C8 and CYP1A2 also catalyse the activation of tegafur (Komatsu *et al*., 2000) and further investigations are needed to clarify the impact of *CYP2A6* polymorphisms on anticancer drug metabolism.

CYP2B6

The functional *CYP2B6* gene and the pseudogene *CYP2B7P* are located in the middle of the chromosome-19 cluster, which also contains the *CYP2A* and *CYP2F* subfamilies. CYP2B6 is mainly expressed in liver, where it constitutes about 3–5% of the total microsomal *P*450 pool (Gervot *et al*., 1999; Lang *et al*., 2001), but it is also detected at lower levels in extrahepatic tissues, including intestine, kidney, lung, skin and the brain (Gervot *et al*., 1999; Miksys *et al*., 2003; Yengi *et al*., 2003). CYP2B6 activity in liver microsomes varies more than 100-fold and a broad inter-individual variability of *in vivo* pharmacokinetic parameters of several CYP2B6 drug substrates suggests significant interindividual differences in the systemic exposure to a variety of drugs that are metabolized by CYP2B6 (Ekins *et al*., 1998). CYP2B6 expression is induced through proximal and distal response elements at −1.7 and −8.5 kb via constitutive androstane receptor (CAR) (Goodwin *et al*., 2001; Wang *et al*., 2003). Well-known inducers include phenobarbital and cyclophosphamide, which will cause auto-induction. *CYP2B6* is highly polymorphic and presently more than 48 different alleles have been described (http://www.imm.ki.se/CYPalleles/cyp2b6.htm); this number is increasing rapidly,

indicating that the polymorphism is even higher than previously thought. No common defective *CYP2B6* allele has been described but, instead, many variant alleles with amino-acid substitutions causing functional alterations, at least as assessed in heterologous expression systems, have been described. The role of the different *CYP2B6* alleles for the *in vivo* metabolism of drugs is still largely unknown. *CYP2B6*5* (R487C) and *CYP2B6*7* (Q172H, K262R and R487C) variants have been suggested to cause significantly reduced protein expression levels in human liver (Lang *et al*., 2001), but other studies have not confirmed this (Xie *et al*., 2003; Hesse *et al*., 2004). The *CYP2B6*6* allele (Q172H and K262R) has been associated with a decreased protein expression, but higher activity using cyclophosphamide as substrate (Xie *et al*., 2003). On the other hand, two studies showed that *CYP2B6*6* carriers have a reduced *in vivo* capacity to metabolize efavirenz (Tsuchiya *et al*., 2004; Wang *et al*., 2005) and a lower activity using bupropion as probe drug (Hesse *et al*., 2004). *CYP2B6*16* with K262R and I328T substitutions has a decreased stability that influences the *in vivo* rate of efavirenz metabolism (Wang *et al*., 2005). Additionally, there are several rare nonsynonymous SNPs, resulting in absent or nonfunctional proteins (Lang *et al*., 2004; Klein *et al*., 2005), but their role *in vivo* is not known. Further studies are thus needed in order to characterize the clinical impact of the polymorphisms identified.

CYP2B6 participates in the metabolism of a few precarcinogens and some important therapeutic drugs such as artemisinin, ketamine, propofol, bupropion and the HIV-1 reverse transcriptase inhibitors nevirapine and efavirenz. Several potent and specific inhibitors have been described, including the anticancer agent *N*,*N*′,*N*″-triethylene thiophosphoramide (thiotepa) (Rae *et al*., 2002). With respect to the metabolism of anticancer drugs, CYP2B6 is involved in the metabolic activation of the cytotoxic prodrugs cyclophosphamide, ifosfamide, thiotepa and procarbazine (Table 3). Despite the structural similarities between cyclophosphamide and ifosfamide, they have important differences in their metabolism, toxicity and therapeutic spectrum. About 45% of a therapeutic dose of ifosfamide is typically metabolized via *N*-dechloroethylation to the toxic chloroacetaldehyde, whereas only 10% of cyclophosphamide is converted to chloroacetaldehyde (Kaijser *et al*., 1993). The activation through 4-hydroxylation is mediated mainly by CYP2B6, but also by CYP3A4, CYP2C19 and CYP2C9 for cyclophosphamide and by CYP3A4 for ifosfamide (Huang *et al*., 2000b). The 4-hydroxy-derivative is in chemical equilibrium with aldophosphamide, which can undergo chemical decom-

Table 3 Anticancer agents that are substrates for cytochromes *P*450 and their medical use

Drug	P*450 involved*	*Cancer*	*Prodrug*	P*450 involved*	*Cancer*
Docetaxel	CYP3A, (CYP1B1)	Breast, NSCLC[a], prostate	Cyclophosphamide	**CYP2B6, CYP2C19**, CYP3A4	Leukemias, lymphomas, retinoblastoma, neuroblastoma
Etoposide	CYP3A4, (CYP2E1, CYP1A2)	Testicule, SCLC	Dacarbazine	CYP1A1, CYP1A2, CYP2E1	Melanoma
Exemestane	CYP3A	Breast	Ifosfamide	CYP3A, **CYP2B6**	Cervix, soft tissue sarcoma
Flutamide	CYP1A2	Prostate	Procarbazine	CYP2B6, CYP1A	Hodgkin's disease, NHL
Fulvestrant	CYP3A	Breast	Tegafur	**CYP2A6**, CYP2C8, CYP1A2	Colon, breast, stomach
Gefitinib	CYP3A (**CYP2D6**)	NSCLC	Thiotepa	CYP3A, **CYP2B6**	Breast, bladder ovary, NHL
Idarubicin	(**CYP2D6, CYP2C9**)	AML, ANLL			
Imatinib	CYP3A	CML, GIST			
Irinotecan	CYP3A	Colon, rectum			
Letrozole	CYP3A, **CYP2A6**	Breast			
Mitoxantrone	CYP1B1, CYP3A	Breast, AML, ANLL, NHL			
Paclitaxel	CYP2C8, (CYP3A)	Ovary, breast, NSCLC, Kaposi's sarcoma			
Tamoxifen	CYP3A, **CYP2D6**, CYP1B1, **CYP2C9**, **CYP2C19**	Breast			
Teniposide	CYP3A	ALL, NHL			
Topotecan	(CYP3A)	Ovary, SCLC			
Toremifene	CYP3A, (CYP1A2)	Breast			
Vinblastine	CYP3A	Breast, testicle Hodgkin's disease, Kaposi's sarcoma			
Vincristine	CYP3A	Acute leukaemia, NHL, Hodgkin's disease, neuroblastoma, rhabdomyosarcoma			
Vindesine	CYP3A	ALL, NSCLC			
Vinorelbine	CYP3A	NSCLC, breast			

[a]NSCLC, Non-Small Cell Lung Cancer; SCLC, small Cell Lung Cancer; AML, acute myeloid leukaemia; ANLL, acute non-lymphocytic leukaemia; CML, chronic myeloid leukaemia; GIST, gastrointestinal stromal tumors; ALL, acute lymphoblastic leukaemia; NHL, non-Hodgkin's lymphoma. The contribution by the most polymorphic *P*450 forms is shown in bold.

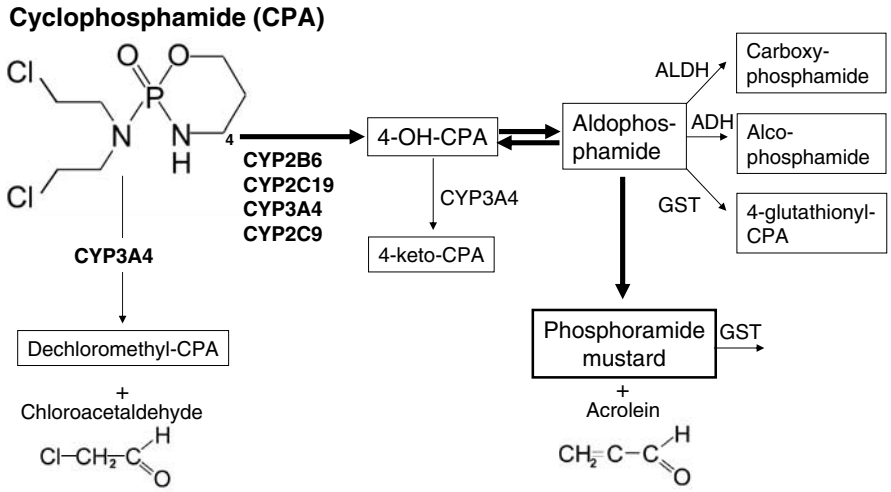

Figure 1 Chemical structure of cyclophosphamide and major biotransformation pathways. Cyclophosphamide (CPA) is activated by CYP2B6, CYP2C19, CYP2C9 and CYP3A4/5 in the liver to 4-hydroxycyclophosphamide (4-OH-CPA), which is in tautomeric equilibrium with aldophosphamide. Aldophosphamide spontaneously decompose to the alkylating metabolite phosphoramide mustard and the toxic byproduct acrolein. In a minor pathway, the dechloroacetylation of CPA mediated by CYP3A4/5 results in the toxic agent chloroacetaldehyde. In other alternative pathways the inactive 4-keto-CPA, carboxyphosphamide, alcophosphamide and 4-glutathionyl-CPA can be formed. *CYP2C19*2* allele has been shown to result in a decreased rate of CPA activation. ALDH, aldehyde dehydrogenase; ADH, alcohol dehydrogenase; GST, glutathione *S*-transferase.

position into phosphoramide mustard and acrolein (Figure 1). Phosphoramide mustard is an active DNA-alkylating metabolite and acrolein is a toxic byproduct. Owing to the important role of CYP2B6 for cyclophosphamide activation, polymorphisms of this enzyme would likely affect cyclophosphamide pharmacokinetics. In this respect, Takada *et al*. (2004) found that patients with proliferative lupus nephritis homozygous for *CYP2B6*5* and treated with pulse cyclophosphamide had a significantly higher probability of reaching end-stage renal disease with double serum creatine levels (Takada *et al*., 2004). Thiotepa undergoes oxidative desulfuration, by the action of CYP2B6 and CYP3A4, to a pharmacologically active compound with a longer plasma half-life, *N*,*N*′,*N*″-triethylene phosphoramide (TEPA) (Jacobson *et al*., 2002). CYP2B6 is also involved in the metabolism of tamoxifen; however, CYP2D6 and CYP3A4 appear to have a higher impact in its metabolism (Crewe *et al*., 2002). With respect to procarbazine, CYP2B6 and CYP1A enzymes are both involved in its activation (Goria-Gatti *et al*., 1992).

CYP2Cs

In humans, there are four *CYP2C* genes that have been mapped to chromosome 10q24 in the order *2C18–2C19–2C9–2C8*. The corresponding enzymes are all primarily expressed in the liver, with the exception of *CYP2C18*, which probably represents an inactive gene. The CYP2C enzymes are involved in the metabolism of about 20% of the currently used drugs. CYP2C19 catalyses the metabolism of, for example, citalopram, diazepam and omeprazole, whereas CYP2C9 is active in the metabolism of warfarin, phenytoin and nonsteroidal anti-inflammatory drugs. CYP2C8 plays an important role in about 5% of used drugs, especially antidiabetics and antimalarials. The polymorphic behaviour of CYP2C9 and CYP2C19 has been thoroughly studied and is of very high clinical significance, whereas the clinical relevance of the *CYP2C8* polymorphisms is more controversial. *CYP2C8*3* (R139K and K399R) is mainly present in Caucasians (13% frequency) and, interestingly, the allele is in linkage disequilibrium with *CYP2C9*2* (Yasar *et al*., 2002). CYP2C8.3 has been shown to exhibit a lower paclitaxel 6α-hydroxylase activity in heterologous expression systems. However, *in vivo* the data are contradictory and opposite results have been obtained using different substrates (Dai *et al*., 2001; Niemi *et al*., 2003; Martinez *et al*., 2005). Other variant alleles with missense mutations are *CYP2C8*2* and *CYP2C8*4*, but their impact on enzyme activity is unclear. *CYP2C8*5* produces a frameshift and a truncated inactive enzyme, but is very rare. The most relevant *CYP2C9* genetic variations, which result in a decreased activity, are *CYP2C9*2,* mainly present in Caucasians with a 10–15% frequency, but almost absent in Africans and Asians, and *CYP2C9*3,* which has an allele frequency of 4–10% in Caucasians, 4–7% in Asians and 1–2% in Africans (Kirchheiner and Brockmoller, 2005). Concerning *CYP2C19*, the common *CYP2C19*2* and *CYP2C19*3* alleles result in enzyme deficiency. Together, they are most common in Asians (35% allele frequency), but less common in Africans (17%) and Caucasians (15%) (Desta *et al*., 2002). One common allele (*CYP2C19*17*) causes a higher expression of the enzyme due to a mutation in the 5′-upstream region recruiting nuclear factor binding resulting in increased transcription (Sim *et al*., 2005a).

All four human CYP2C enzymes, but to a major extent CYP2C19, catalyse the activation of cyclopho-

sphamide and ifosfamide, but also CYP2B6 and other *P*450s are also implicated in these reactions (Huang *et al*., 2000b). In a study encompassing 60 cancer patients, data by Timm *et al*. (2005) indicated that those carrying the inactive *CYP2C19*2* allele had a significant decreased cyclophosphamide elimination, while no differences in elimination rates of cyclophosphamide were found between subjects of different *CYP2B6*, *CYP2C9* and *CYP3A5* genotypes (Timm *et al*., 2005). Accordingly, Takada *et al*. (2004) found that, in pulse cyclophosphamide treatment of proliferative lupus nephritis, heterozygous or homozygous *CYP2C19*2* patients had a significantly lower risk of developing premature ovarian failure, and in survival analysis patients homozygous for *CYP2C19*2* had a higher probability of a poor renal response (Takada *et al*., 2004). These studies suggest that the presence of the inactive *CYP2C19*2* causes a reduction in the metabolic activation of cyclophosphamide, thereby lowering the risk of toxicity but worsening the therapeutic response. Similarly, it could be envisioned that the rapid *CYP2C19*17* allele with an allele frequency of about 18% in Caucasians (Sim *et al*., 2005a) would cause a more efficient treatment with cyclophosphamide. It could, therefore, be suggested that predictive genotyping for *CYP2C19* would increase the success of cyclophosphamide treatment. In addition, CYP2C9 participates in the metabolism of idarubicin and both CYP2C9 and CYP2C19 are active in tamoxifen metabolism.

Paclitaxel undergoes extensive hepatic oxidative metabolism through 6α- and 3′-*p*-hydroxylations catalysed by CYP2C8 and CYP3A4, respectively. As the formation of 6α-hydroxypaclitaxel, approximately 30 times less toxic than the parent compound (Harris *et al*., 1994), has been shown to be the primary metabolic pathway of paclitaxel metabolism (Taniguchi *et al*., 2005), it would be reasonable to hypothesize that *CYP2C8* polymorphisms could influence the efficacy of paclitaxel treatment. This question has been dealt with by Nakajima *et al*. (2005), who found a 16-fold interindividual variation in the 6α-hydroxypaclitaxel area under the curve (AUC) among 23 female ovarian cancer patients, but apparently no *CYP2C8* variant alleles, due to the small study or low frequency of the alleles in Japanese. Recently, Henningsson *et al*. (2005), using 97 patients, found a 13-fold variation in paclitaxel clearance but no significant influence of the *CYP2C8*2, CYP2C8*3* and *CYP2C8*4* variant alleles. This issue, however, warrants further investigations. CYP2C8 is also involved in the metabolism of all-*trans* retinoic acid, which is given alongside chemotherapy in several cancers.

CYP2D6

There is an extensive interindividual variation in CYP2D6 activity, which to some extent is likely determined by the adaptation to the environment through metabolism of natural compounds such as alkaloids. The *CYP2D6* gene is one of the best studied human *P*450 genes and correlations between the phenotype and genotype have been extensively studied for various drugs, providing a rather well-understood molecular basis for the variation in CYP2D6 activity (Ingelman-Sundberg, 2005). The polymorphisms can result in defective or increased enzyme activity and *CYP2D6* genotypes usually exhibit large inter-ethnic differences. Increased activity results from gene duplication/amplification and individuals carrying up to 13 functional *CYP2D6* copies in one allele have been found (Johansson *et al*., 1993; Aklillu *et al*., 1996). Defective *CYP2D6* allelic variants carry gene deletions, stop codons or splicing defects, and the most common functionally altered variants are *CYP2D6*4* (15–21% in Caucasians), *CYP2D6*5* (about 3–6% in the different populations), *CYP2D6*10* (38–70% in Asians, 3–9% in Africans) and *CYP2D6*17* (20–34% in Africans). Furthermore, a large number of *CYP2D6* polymorphisms with lower frequencies, but resulting in a defective enzyme, also contribute to the extensive interindividual variation in CYP2D6 activity (http://www.imm.ki.se/CYPalleles/cyp2d6.htm).

CYP2D6 is involved in the metabolism of 20–25% of all drugs in clinical use, and it has a special impact on the treatment of psychiatric and cardiovascular diseases. By contrast, the role of CYP2D6 in the metabolism of precarcinogens is minor and the polymorphism of the enzyme is apparently without importance for interindividual differences in susceptibility for cancer. CYP2D6 has been shown to play a crucial role in the metabolism of tamoxifen, which is an estrogen receptor modulator widely used for the endocrine treatment of all stages of hormone receptor-positive breast cancer. Tamoxifen is activated by the CYP system to antioestrogenic metabolites that are more potent than the parent compound (Jin *et al*., 2005). *In vitro* studies implicated many CYP isoforms such as CYP3A, CYP2D6, CYP2C9, CYP2C19, CYP2B6 and CYP1A2, in the biotransformation of tamoxifen. However, the key metabolites of tamoxifen seem to be 4-hydroxytamoxifen and endoxifen, formed primarily by CYP2D6, and *N*-desmethyltamoxifen, formed primarily by CYP3A4 (Figure 2). In patients receiving tamoxifen, the most abundant compounds in plasma are *N*-desmethyltamoxifen and endoxifen, and it has been shown that endoxifen has approximately 100 times greater affinity for the oestrogen receptor than tamoxifen and *N*-desmethyltamoxifen (Jordan *et al*., 1977; Clarke *et al*., 2003; Jin *et al*., 2005). As endoxifen is mainly formed by the action of CYP2D6, patients with defective *CYP2D6* alleles would obtain less benefit from tamoxifen therapy than those carrying functional copies of *CYP2D6*. Thus, in a study of 80 women with breast cancer starting tamoxifen adjuvant therapy, the plasma concentrations of endoxifen after 4 months of therapy were significantly lower in patients being homozygous or heterozygous for defective *CYP2D6* genes as compared to those with two functional alleles (Jin *et al*., 2005). Additionally, those subjects using CYP2D6 inhibitors had 58% reduction in the plasma concentration of endoxifen. The *CYP2D6* genotype is also relevant for cancer patients with respect

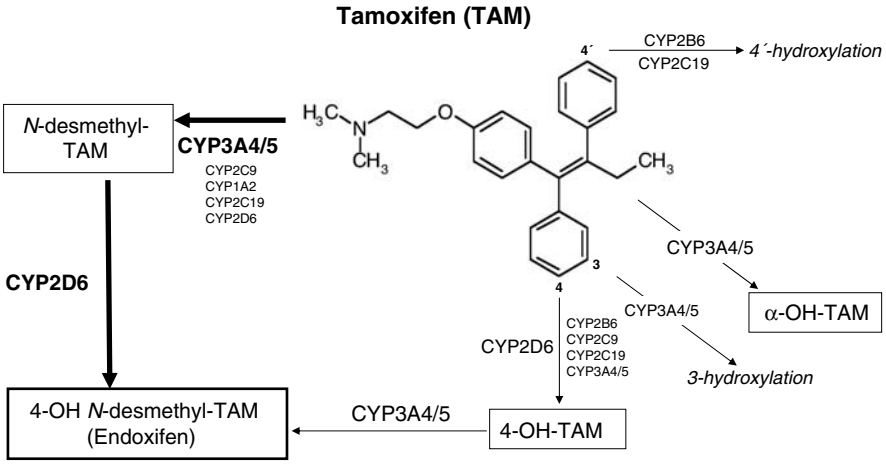

Figure 2 Chemical structure of tamoxifen and major biotransformation pathways. CYP3A4/5 are the more efficient enzymes responsible for the *N*-demethylation of tamoxifen (TAM), whereas the generations of endoxifen and 4-hydroxytamoxifen (4-OH-TAM) are predominantly catalysed by CYP2D6. Other CYP isoforms, including CYP2C19, CYP2C9, CYP2B6 and CYP1A2, have also been shown to participate in the metabolism of tamoxifen. The most abundant compounds in plasma are *N*-desmethyltamoxifen and endoxifen, and endoxifen has approximately 100 times greater affinity for the oestrogen receptor than tamoxifen and *N*-desmethyltamoxifen. *CYP2D6* polymorphisms have been shown to affect the plasma concentrations of endofixen.

to the action of the antiemetic drugs tropisetron and ondasetron. Lower plasma levels and higher frequency and intensity of vomiting were found in subjects carrying a higher number of active *CYP2D6* gene copies (Kaiser *et al*., 2002).

CYP2E1

CYP2E1 is responsible for the metabolism and activation of a large number of low-molecular-weight chemicals, solvents, cancer suspect agents and a few drugs (Table 2). Thus, CYP2E1 might be an important determinant of human susceptibility to toxicity and carcinogenicity of industrial and environmental chemicals. However, polymorphisms affecting CYP2E1 expression or activity have not been found, probably because of high conservation due to a critical role of the enzyme in gluconeogenesis during conditions of starvation. By contrast, induction of the enzyme by, for example, alcohol might provide a more important factor for interindividual susceptibility to cancer in reactions mediated by CYP2E1.

CYP3A4/5

The human *CYP3A* locus carries four genes, but only *CYP3A4, CYP3A5* and *CYP3A7* encode active enzymes relevant for the metabolism of a wide range of structurally different xenobiotics. The expression of these enzymes is regulated in a tissue-specific manner, the *P*450s being predominant in the liver and gastrointestinal tract. During fetal stages CYP3A4 is absent, while CYP3A7 expression is maximum. In adult life the predominant hepatic *P*450 is CYP3A4, with some individuals also exhibiting a significant expression of the 'fetal' CYP3A7 (Lacroix *et al*., 1997; Sim *et al*., 2005b). CYP3A5 is mainly absent from Caucasian livers but contributes to CYP3A activity in Africans (Kuehl *et al*., 2001). The substrate specificities of the CYP3A enzymes are overlapping, but CYP3A4 usually exhibits a higher specific activity towards many CYP3A substrates when compared to CYP3A5 and CYP3A7 (Williams *et al*., 2002). The CYP3A enzymes are involved in the metabolism of about 50% of all drugs currently on the market (Li *et al*., 1995) and they participate in the metabolic activation and metabolism of several carcinogens such as aflatoxin B and also of anticancer drugs (see Tables 2 and 3). Interindividual variation in CYP3A activity, thus, has a major impact on pharmacokinetics and metabolism of a majority of different drugs. Generally, a five-fold interindividual variability in clearance of CYP3A substrates *in vivo* has been found with some scarce 'outliers'. This variation can be caused by environmental factors or drugs that inhibit or induce CYP3A enzymes but, additionally, it has been shown that the variation is determined to a high extent by genetic factors (Ozdemir *et al*., 2000). Important genetic polymorphisms that severely decrease the expression of CYP3A5 protein have been described, that is, *CYP3A5*3*, *CYP3A5*6* and *CYP3A5*7* (see Kuehl *et al*., 2001; Lee *et al*., 2003). However, this is not true for CYP3A4 since, despite the analysis of thousands of subjects, no major functionally variant allele has been found at an allele frequency higher than 0.1%. The only allele that appears to influence the CYP3A4 expression is *CYP3A4*1B*, common in Africans and present at 5% frequency in Caucasians, through alteration of nuclear proteins binding to the polymorphic element (Rodriguez-Antona *et al*., 2005). The distribution of the *CYP3A4*1B* allele has been associated to prostate and lung cancer, although the data are generally conflicting (Rebbeck *et al*., 1998; Paris *et al*., 1999; Spurdle *et al*., 2002; Tayeb *et al*., 2002, 2003; Dally *et al*., 2003). The basis for any genetic background for the interindividual variation in CYP3A4 expression remains a challenge.

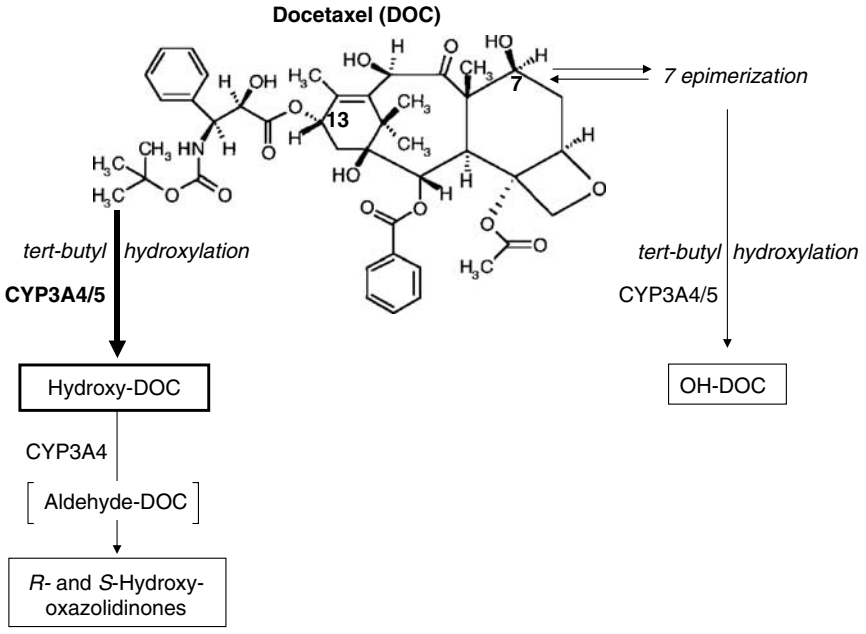

Figure 3 Chemical structure of docetaxel and major biotransformation pathways. Docetaxel (DOC) is inactivated in the liver by CYP3A4/5 through successive oxidations of the tert-butyl ester group of the C13-side chain, mainly the direct hydroxylation to an alcohol (hydroxyl-DOC) and a subsequent oxidation to an unstable aldehyde, which cycles to two stereomeric hydroxyoxazolidinones. DOC can also 7-epimerize to a diasteromer, which undergoes metabolic pathways similar to DOC. CYP3A4 phenotype has been shown to influence DOC pharmacokinetics and therapeutic outcome.

With respect to the action of anticancer drugs, the variability of CYP3A4 is expected to influence the outcome of several different treatments. Docetaxel is metabolized by CYP3A4 to inactive hydroxylated derivatives (Figure 3) and, therefore, a high CYP3A4 activity would result in a poor therapeutic outcome of the drug. Accordingly, in cancer patients treated with docetaxel in combination with the potent CYP3A4 inhibitor ketoconazole, a 49% decrease in docetaxel clearance was found (Engels *et al*., 2004). Similarly, hepatic CYP3A4 activity measured by the erythromycin breath test and midazolam clearance predicted docetaxel clearance, finding the greatest toxicity in patients with the lowest CYP3A4 activity (Hirth *et al*., 2000; Goh *et al*., 2002). Furthermore, Yamamoto *et al*. (2005) phenotyped CYP3A4 in patients with advanced non-small-cell lung cancer by measuring urinary 6-beta-OHF after cortisol administration and found that an individualized dosing method, based on CYP3A4 phenotyping, decreased the pharmacokinetic variability of docetaxel when compared to body-surface area-based dosing (Yamamoto *et al*., 2005). In addition, CYP3A4 expression in breast tumour tissue has been shown to predict therapeutic response to docetaxel (Miyoshi *et al*., 2002, 2005). Similarly to docetaxel, irinotecan is inactivated by CYP3A4 and induction of CYP3A4 in patients receiving irinotecan results in a significant decrease in the formation of the toxic metabolite of this drug (Friedman *et al*., 1999; Mathijssen *et al*., 2002). Additionally, Mathijssen *et al*. (2004) showed that CYP3A4 phenotype, as assessed by midazolam clearance, is statistically significantly associated with irinotecan pharmacokinetics. With respect to the already described *CYP3A* genotypes, a combination of *CYP3A4, CYP3A5, GSTM1* and *GSTT1* genotypes was shown to influence the probability of treatment failure after high-dose adjuvant chemotherapy for node-positive breast cancer (DeMichele *et al*., 2005). Table 3 also shows other important anticancer agents metabolized by CYP3A4, including taxanes, vinca-alkaloids and new drugs such as imatinib and gefitinib.

*P*450 expression in tumours

In addition to an interindividual variability in the pharmacokinetics of anticancer drugs caused by hepatic CYPs, an altered CYP activity in the tumour cells could result in an altered drug efficacy. Cancer cells by means of genetic or epigenetic mechanisms, due to their higher DNA instability and more frequent alterations in chromatin structure than nontumour cells, could alter *P*450 transcription. The capacity of the tumours to metabolize drugs is a potential means to achieve optimal therapy by activation of prodrugs in the cancer cells; however, it is also a potential mechanism of resistance to therapy by an increased inactivation of anticancer drugs caused by an overexpression of *P*450s. Many studies have reported the presence of drug-metabolizing enzymes in tumours (Dhaini *et al*., 2003; Gharavi and El-Kadi, 2004; Oyama *et al*., 2004; Downie *et al*., 2005; Kumarakulasingham *et al*., 2005). However, differences in the quantification and sampling techniques and heterogeneous patient populations have resulted, in

some cases, in conflicting data, making it difficult to conclude about any impact on deactivation of anticancer agents or activation of prodrugs.

With respect to the impact of tumour *P*450s on drug therapy outcome, Tanaka *et al*. (2004), using 19 human cancer cell lines and eight common anticancer drugs, measured the cytotoxic activity of the drugs and performed cDNA microarray analysis to identify associations between specific gene expression and effect of the drug in question (Tanaka *et al*., 2004). In all, 12 genes with proven functional significance to drug sensitivity, which included *CYP2C8* and *CYP3A4*, were selected and prediction models to accurately predict the *in vitro* efficacy of the drugs were developed. The *in vivo* relevance of the model was tested for 5-fluorouracil treatment in gastric cancer patients. The model of predictive value in terms of survival, time to treatment failure and tumour growth showed that the tumour phenotype was indeed related to the therapeutic response to 5-fluorouracil (Tanaka *et al*., 2004). Miyoshi *et al*. (2002, 2005) showed that a low CYP3A4 expression in breast tumours, as determined at mRNA and protein level, resulted in a better response to docetaxel, which is inactivated by CYP3A4. Similarly, Dhaini *et al*. (2003) showed that a high CYP3A expression in osteosarcoma tumours from 18 patients predicted metastasis and poor prognosis. CYP3A4 is involved in the oxidation of compounds that are usually used as chemotherapeutic agents for the treatment of osteosarcomas such as etoposide, ifosfamide, cyclophosphamide and doxorubicin, suggesting that the response to these drugs could be worse in tumours with high CYP3A expression, increasing the risk of metastasis. Therefore, the main hepatic drug metabolizing *P*450 enzymes if expressed in the tumour cells could influence the success of drug therapy.

In addition, some extrahepatic *P*450s, many of which have major roles in the metabolism of endogenous substrates and are not involved in xenobiotic biotransformation, have been found to be overexpressed in tumour tissue. These include CYP1B1, CYP2J2, CYP2W1 and CYP4Z1. The AhR binds several carcinogens that are metabolized by the CYP1 enzymes and regulates the expression of *CYP1B1* (Nebert *et al*., 2004), which has been found to be overexpressed in a large number of tumours, including cancers of the prostate, kidney, ovarian, breast and colon tumours (Murray *et al*., 1997; McFadyen *et al*., 1999, 2001a; Gibson *et al*., 2003; Tokizane *et al*., 2005). CYP2J2 is able to metabolize arachidonic acid to epoxyeicosatrienoic acids, which have been suggested to play a role in angiogenesis and to exert antiapoptotic effects (Chen *et al*., 2001; Jiang *et al*., 2005; Pozzi *et al*., 2005). CYP2J2 was much overexpressed relative to adjacent normal tissue in the majority of tumours examined, which included esophageal squamous cell carcinoma, esophageal adenocarcinoma, pulmonary squamous cell carcinoma, pulmonary adenocarcinoma, small-cell pulmonary carcinoma, breast carcinoma, stomach carcinoma, liver carcinoma and colon adenocarcinoma (Jiang *et al*., 2005). CYP2W1 has been shown to be almost exclusively expressed during embryogenesis and in adult humans it is mainly detected in tumour tissue samples, more frequently from colon and adrenal gland (Karlgren *et al*., 2005, submitted). With respect to CYP4Z1, it is regulated by the glucocorticoid and progesterone receptors and has been shown to be overexpressed preferentially in breast carcinoma tissue and mammary gland (Rieger *et al*., 2004; Savas *et al*., 2005).

*P*450 as a drug target in cancer therapy

A major objective of cancer research is the development of therapeutic agents specifically targeted to tumour cells. *P*450s expressed at higher levels in the tumour cells than in the surrounding normal tissue offer therapeutic options by the activation of prodrugs specifically in the cancer cells and avoiding undesirable systemic effects (see Riddick *et al*., 2005). In this respect, there are therapeutic options and opportunities arising from both the enhanced endogenous expression of CYP in tumours and CYP-mediated gene therapy. Concerning endogenous overexpression of individual forms of *P*450 enzymes in tumour cells, CYP1B1 is the best studied example, because although several CYP1As, CYP2Cs and CYP3As exhibit enhanced expression in some tumour cells, these enzymes display considerable expression in normal tissue, mainly in the liver. On the other hand, CYP1B1 mRNA and protein expression has been found in a wide range of malignant tumours and in metastatic disease (McFadyen *et al*., 2001a), but the CYP1B1 protein is generally not detected in normal tissue at important levels (Gibson *et al*., 2003). Taking advantage of this, several agents activated by CYP1B1 are currently in preclinical evaluation, such as resveratrol and phortress (Potter *et al*., 2002; Leong *et al*., 2003); in addition, there is a CYP1B1 vaccine (Zyc300) in phase I/II trials, aimed to destroy cancer cells through induction of T-cell response (Gribben *et al*., 2005). Similar strategies could be initiated with other *P*450s mainly identified in tumour cells, such as CYP2W1, CYP2J2 and CYP4Z1, after identification of an appropriate prodrug. The polymorphism of these genes in relation to the success of *P*450-based cancer therapy remains to be elucidated.

Gene therapy offers another approach to get a differential *P*450 expression between tumour/normal tissue, where an exogenous *P450* gene and a prodrug activated by that *P*450 are delivered to the tumour. The enzyme expression can be genetically controlled or its delivery targeted to ensure tumour selectivity. The gene-directed enzyme prodrug therapy systems with CYP have been mainly based on cyclophosphamides, which needs to be activated mainly by CYP2B6. Expression of CYP enzymes has been shown to sensitize cells to both cyclophosphamide in a range of cell lines *in vitro* and the bystander effect is mediated through the soluble derivative 4-hydroxycyclophosphamide. Hepatic *P*450 enzymes like CYP2B6 or CYP2B1 as well as *P*450 reductase have been inserted into 9L gliosarcoma cells by viral transfection in order to facilitate tumour growth suppression in cultured cells and in xenogaft models

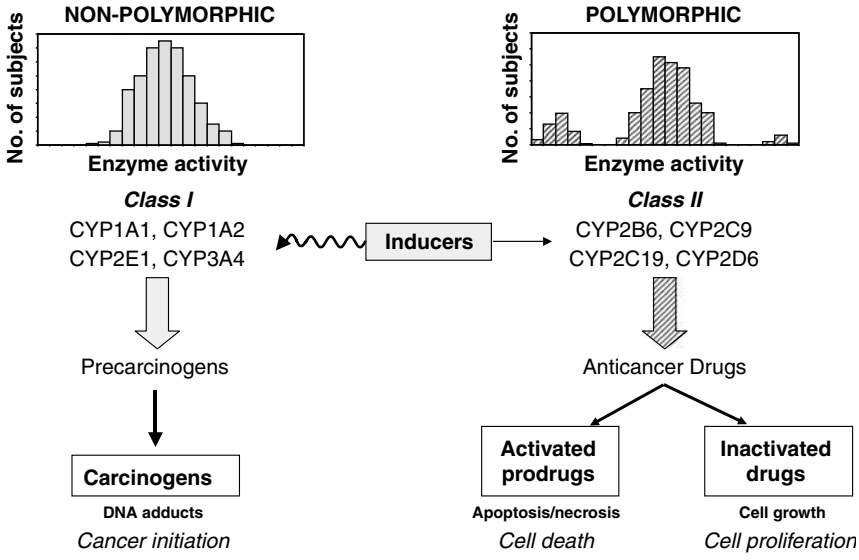

Figure 4 Xenobiotic metabolizing CYP enzymes and cancer. The CYP enzymes can be divided into two different groups: Class I composed of CYP1A1, CYP1A2, CYP2E1 and CYP3A4 and Class II composed of CYP2B6, CYP2C9, CYP2C19 and CYP2D6. Class I enzymes are in general well conserved and active in the metabolism of precarcinogens and drugs, while Class II enzymes have important functional polymorphisms and are active in the metabolism of drugs, but not of precarcinogens. Inducers play an important role in regulating the expression of Class I enzymes, with the exception of CYP2E1, and to a minor extent of Class II enzymes. Class I *P*450s are important for the aetiology of cancer diseases, while Class II *P*450s play an important role in cancer therapy.

upon treatment with anticancer agents (Huang *et al*., 2000a; Roy and Waxman, 2005). In addition, a combinatory treatment of cyclophosphamide and another drug impairing the hepatic expression of *P*450 reductase and hence minimizing the hepatic activation of cyclophosphamide can be used (Huang *et al*., 2000a). *In vitro* and animal models showed promising effects of this approach (McFadyen *et al*., 2004; Dachs *et al*., 2005) and, for example, in a trial of 14 patients with inoperable pancreatic cancer, the median survival was doubled in the treatment group compared to historical controls and 1-year survival improved three-fold (Lohr *et al*., 2001; Salmons *et al*., 2003).

Conclusions

CYPs have important roles in activation and inactivation of both precarcinogens and of anticancer drugs (see Figure 4). Interindividual differences in the *P*450-mediated actions are caused both by environmental and genetic factors. Due to the relatively high extent of conservation of genes encoding CYPs participating in the activation of precarcinogens, the genetic factors are less important determinants of individual susceptibility, whereas inducers of *P*450s like smoking, ethanol, etc., appear to be more relevant factors for such variability. An exception might be CYP2A6 in Asia, where the functional polymorphism is pronounced. The metabolism of several anticancer drugs is catalysed by specific polymorphic forms of CYP, like CYP2B6, CYP2C19 and CYP2D6. Here the knowledge about the different CYP alleles distributed in the populations and their functional consequences is relatively well known, whereas the impact of the polymorphism for *in vivo* treatment with anticancer drugs remains largely to be elucidated. The recent achievements in using the polymorphic *P*450 as drug targets in cancer therapy are promising and could provide a novel and effective alternative of future cancer therapy.

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