Environmental Health Perspectives
Vol. 98, pp. 75–80, 1992

Elucidation of Catalytic Specificities of Human Cytochrome P450 and Glutathione *S*-Transferase Enzymes and Relevance to Molecular Epidemiology

by F. Peter Guengerich,[1] Tsutomu Shimada,[1] Kevin D. Raney,[2] Chul-Ho Yun,[1] David J. Meyer,[3] Brian Ketterer,[3] Thomas M. Harris,[2] John D. Groopman,[4] and Fred F. Kadlubar[5]

A number of different approaches have been used to determine the catalytic selectivity of individual human enzymes toward procarcinogens. Studies with cytochrome P450 (P450) enzymes and glutathione *S*-transferases are summarized here, and recent work with pyrrolizidine alkaloids, aflatoxins, 4,4′-methylenebis(2-chloroaniline), and ethyl carbamate is discussed. In some cases a single enzyme can catalyze both activation and detoxication reactions of a chemical. The purification and characterization of human lung P4501A1 and the development of a noninvasive assay for human P4502E1 are also described.

Introduction

Most of the potentially genotoxic natural and synthetic chemicals to which humans are exposed are not biologically active in themselves but require transformation to reactive electrophiles (*1*). Metabolism of xenobiotic chemicals is complex, and various enzymes can activate or detoxify compounds, depending on the particular situation. Even different enzymes of a family [e.g., the cytochrome P450 (P450) group] can exert different influences by catalyzing different reactions. Levels of the various enzymes known to be involved in the metabolism of xenobiotics (*2*) often vary dramatically among individual people, owing to both genetic and environmental influences (*3,4*). There is considerable precedent from experimental animal models to hypothesize that these differences in enzyme levels may contribute to the overall risk of an individual to a particular chemical. Thus, discernment of the catalytic selectivities of families of enzymes is of interest for risk assessment. If a dominant enzyme can be implicated in a particular reaction, then it may be further considered as a potential risk factor. In people with low levels of a particular enzyme, other enzymes may participate but their enzymatic contributions will not be so important if the overall activity is low (*5*).

Several *in vitro* approaches have been used in our laboratories to identify which pro-carcinogens are oxidized by individual human P450 enzymes and conjugated by human glutathione (GSH) *S*-transferases. The rationale and some recent studies are presented here, with consideration of the further development of noninvasive assays which can be used in molecular epidemiology biomarker studies with humans at risk (Table 1).

Rationale

At least five major approaches can be used in the assigning of catalytic activities to particular P450 enzymes (*5*): *a*) Selective inhibitors may be used with microsomes. The degree to which the activity is inhibited reflects the contribution of a particular P450, if the inhibitor is truly specific. This approach may also be of use with iso-

[1]Department of Biochemistry and Center in Molecular Toxicology, Vanderbilt University School of Medicine, Nashville, TN 37232.
[2]Department of Chemistry and Center in Molecular Toxicology, Vanderbilt University School of Medicine, Nashville, TN 37235.
[3]CRC Molecular Toxicology Research Group, Middlesex School of Medicine, Cleveland Street, London, W1P 6DB, UK.
[4]Department of Environmental Health Sciences, Division of Environmental Chemistry, Johns Hopkins University School of Hygenic and Public Health, Baltimore, MD 21205.
[5]National Center for Toxicological Research, Jefferson, AR 72079.
Address reprint requests to F. P. Guengerich, Department of Biochemistry and Center in Molecular Toxicology, Vanderbilt University School of Medicine, Nashville, TN 37232-0146.

Table 1. Potential noninvasive assays for human P450s.

P450	Assay end point
1A2	Phenacetin *O*-deethylation
	Caffeine 3-demethylation
2A6	Coumarin 7-hydroxylation
2C10 (also 2C8, 2C9)	Tolbutamide hydroxylation
2D6	Debrisoquine 4-hydroxylation
	Sparteine Δ^2- and Δ^6-oxidation
	Dextromethorphan
	N-Demethylation
2E1	Chlorzoxazone 6-hydroxylation
3A4	Nifedipine oxidation
	Erythromycin *N*-demethylation
	Dapsone *N*-hydroxylation
	Lidocaine *N*-deethylation
	Cortisol 6β-hydroxylation

lated cells and *in vivo*. *b*) In a similar way inhibitory antibodies can be used with microsomes. Of course, this approach usually requires purification of an antigen to raise the antibody; hence this strategy has been employed in a limited number of cases. *c*) The catalytic activity can be measured in preparations (microsomes) derived from different individuals and compared to a known reaction catalyzed by a particular P450. The degree of associative attribution (the parameter r^2) should reflect the fraction of the variation that can be ascribed to a relationship between the two activities, usually interpreted as being catalyzed by the same or similarly regulated enzymes. *d*) Purified enzymes can be compared directly for catalytic activity; however, some P450s do not appear to retain their full activities expressed *in vivo* after purification (*6*) and, of course, comparison of the activities of a limited set of enzymes has implications only among the set. *e*) A cDNA can be expressed in an artificial vector system, and catalytic activities can be assessed. Some eukaryotic cell lines can be used, with transient or stable expression.

The latter two approaches are very useful for distinguishing the activities of closely related enzymes, while the first three procedures provide better estimates of the extent to which a particular enzyme contributes to the overall activity. Given the relative advantages and disadvantages of each technique, the most reliable conclusions are derived with a battery of techniques as opposed to a single one. Such knowledge is of course hypothetical in its relevance and must be considered in light of findings *in vivo*. A current list of assignments of major human P450s involved in the activation of protoxicants and procarcinogens is presented in Table 2 (*5*).

Of course, many of these same general approaches can be applied to studies with other enzymes, with some modification; however, in no other case (other than the P450s) has so extensive a menu of technical methods been applied *in vitro*, and often the noninvasive assays are more limited.

Pyrrolizidine Alkaloids

Senecionine has been studied as a prototype of the pyrrolizidine alkaloids, which can be toxic and carcinogenic in laboratory animals. Studies with rats have indicated that P450s catalyze both the formation of pyrroles and *N*-oxides, with the former considered to be an activation process and the latter a detoxication (*8*). In rats, P4503A enzymes appear to be capable of forming both pyrroles and *N*-oxides (*9*). In addition, P4503A4 appears to be the major human liver enzyme involved in both reactions (*10*). As noted elsewhere (*11*), levels of P4503A4 vary considerably among individuals. The question might there-

Table 2. Protoxicants and procarcinogens activated by human P450s (*5*).[a]

P450	Protoxicants and procarcinogens
1A1	Benzo[*a*]pyrene and other polycyclic hydrocarbons
	2-Amino-1-methyl-6-phenylimidazo[4,5-*b*] pyridine
1A2	2-Acetylaminofluorene
	2-Aminofluorene
	2-Aminoanthracene
	2-Amino-3-methylimidazo[4,5-*f*]quinoline (IQ)
	2-Amino-3,8-dimethylimidazo[4,5-*f*quinoline (MeIQ)
	2-Amino-3,8-dimethylimidazo[4,5-*f*]quinoxaline (MeIQx)
	2-Amino-3,4,8-trimethylimidazo[4,5-*f*]quinoxaline (DiMeIQx)
	2-Amino-6-methyldipyrido[1,2-*a*:3′,2′-*d*]imidazole (Glu P-1)
	3-Amino-1-methyl-5*H*-pyrido[4,3-*b*]indole (Trp P-2)
	2-Aminodipyrido[1,2-*a*:3′,2′-*d*]imidazole (Glu P-2)
	2-Naphthylamine
	4-Aminobiphenyl
	2-Amino-1-methyl-6-phenylimidazo[4,5-*b*]pyridine (PhIP)
	Acetaminophen
	4-*N*-Nitroso(methylamino)-1-(3-pyridyl)-1-butanone (NNK)
2A6[b]	*N*-Nitrosodiethylamine
	4,4′-Methylenebis(2-chloroaniline) (MOCA)
2E1	*N*-Nitrosodimethylamine
	N-Nitrosodiethylamine
	N-Nitroso-*N*-methylbenzylamine
	N-Nitrosomethylbutylamine
	Benzene
	Carbon tetrachloride
	Chloroform
	Methylene chloride
	Trichloroethylene
	Ethylene dichloride
	Ethylene dibromide
	1,2-Dichloropropane
	Styrene
	Vinyl chloride
	Vinyl bromide
	Acrylonitrile
	Ethyl carbamate
	Vinyl carbamate
	Acetaminophen
3A4	Aflatoxin B_1
	Aflatoxin G_1
	Sterigmatocystin
	Senecionine
	7,8-Dihydroxy-7,8-dihydrobenzo[*a*]pyrene
	9,10-Dihydroxy-9,10-dihydrobenzo[*b*]fluoranthene
	3,4-Dihydroxy-3,4-dihydro-7,12-dimethylbenz[*a*]anthracene
	6-Aminochrysene
	tris-(2,3-Dibromopropyl)phosphate
	1-Nitropyrene
	4,4′-Methylenebis(2-chloroaniline) (MOCA)
	17β-Estradiol

[a]The purpose of this table is to summarize evidence for a major role of a single (human liver) P450 in the activation of each chemical. Of course, other P450s may contribute to some extent, particularly in individuals in whom the major enzyme is deficient. For further discussion, see Guengerich and Shimada (*5*).

[b]P4502A6 appears to make some contribution but does not appear to be the dominant enzyme in either case.

fore be raised as to whether higher levels of P450 3A4 are detrimental or protective.

Aflatoxin B_1 Oxidation

Aflatoxin B_1 (AFB_1) has several fates of relevance to its genotoxicity (Fig. 1). Our previous work led to the conclusion that, although several forms of human P450 can oxidize AFB_1 to its 8,9-oxide, P4503A4 plays a very major role in this reaction, due to both the intrinsic rate and the high levels of P4503A4 that can exist in human liver (*12,13*). In human fetal liver, the closely related P4503A7 appears to play a dominant role (*14*). The results of others have demonstrated that cDNA-expressed P4503A4 that has high activity (per nanomole P450) and that antibodies raised against P4503A enzymes are most inhibitory to activation in human liver microsomes (*7,15*). Any role for P4501A2 in bioactivation would appear to be minimal because of the stimulation seen with 7,8-benzoflavone *in vitro* (*12,13*) and the demonstrated detoxifying influence of rodent P4501A2 *in vivo* (*13,16*).

Studies in our laboratories have shown that when the activation of AFB_1 in human liver microsomes is examined by trapping the GSH conjugate (in the presence of mouse liver cytosol), the results are consistent with those in which *umu* gene response or DNA N^7-guanyl adducts are measured (*12,13*). For instance, GSH conjugate formation is well correlated with nifedipine oxidation in different microsomal preparations (Fig. 2). AFB_1 3α-hydroxylation (to form AFQ_1) was also well correlated with nifedipine oxidation ($r^2 = 0.97$) [see also Forrester et al. (*7*)] and P4503A4 levels. Such ratios of GSH conjugate and AFQ_1 formation (Fig. 2) were seen at AFB_1 concentrations as low as 2μM; it appears that the K_m for AFB_1 epoxidation is approximately 7 μM but that for AFB_1 3-hydroxylation is much higher. Even at the low AFB_1 concentration, the results of immunoinhibition, chemical inhibition, and correlation experiments are consistent with the view that both AFB_1 8,9-epoxidation and 3-hydroxylation are catalyzed primarily by P4503A4. In microsomes, however, 7,8-benzoflavone stimulated epoxidation but inhibited 3-hydro-

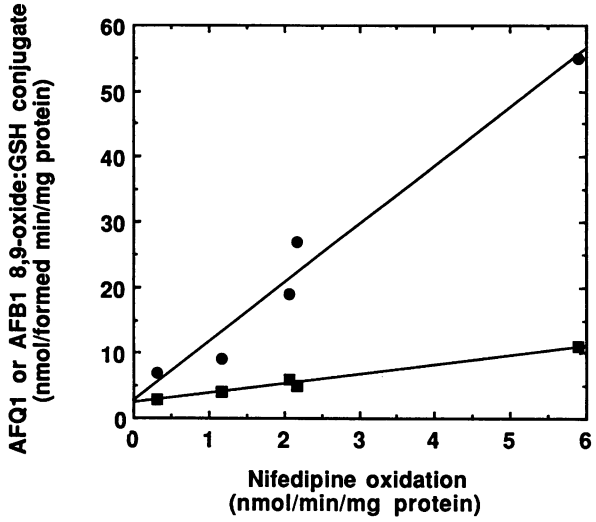

FIGURE 2. Correlation of rates of conversion of aflatoxin B_1 to aflatoxin Q_1 (●) or aflatoxin B_1 8,9-oxide trapped as its glutathione *S*-transferase conjugate (■) with rates of nifedipine oxidation in microsomal fractions prepared from different human liver samples.

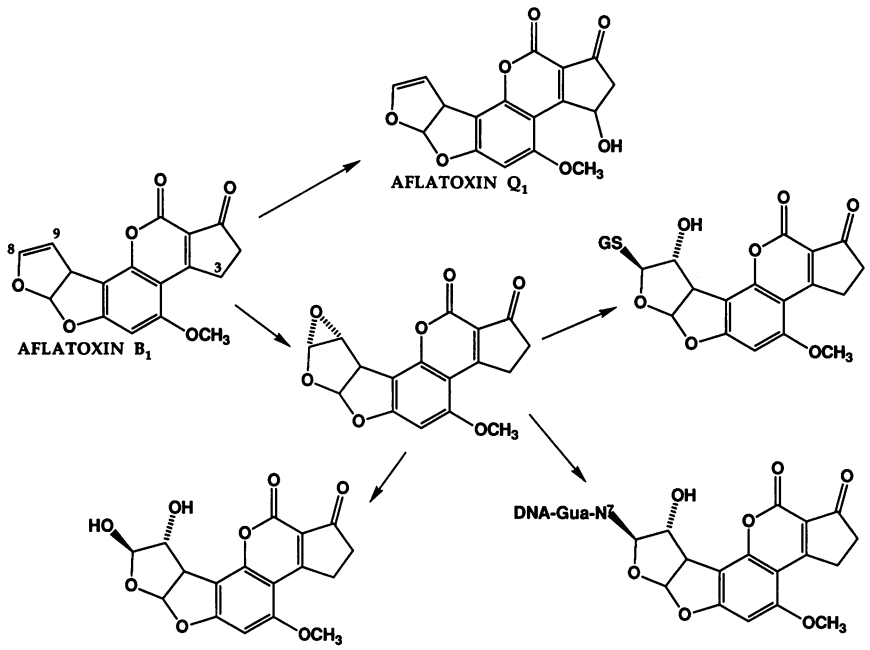

FIGURE 1. Scheme of aflatoxin B_1 metabolism.

xylation. This phenomenon was also demonstrated with purified yeast recombinant P4503A4 (Table 3) and is interpreted to reflect an allosteric effect on the enzyme which has at least two potential transition states. At low AFB_1 concentrations, below the K_m, the ratio of AFQ_1: epoxide formation should approach a constant value. Whether the presence of more P4503A4 is detrimental or beneficial may depend upon the *in vitro* assay, and the *in vivo* consequences are not yet clear; however, a correlation between urinary excretion of N^7-guanyl AFB_1 adducts and the P4503A4 marker 6β-hydroxycortisol (Table 1) has been seen in studies in the Gambia.

GSH Conjugation of AFB_1 8,9-Oxide

In rats, biliary excretion of the GSH conjugate is a major fate of AFB_1 (*17*). Mice are thought to be protected from AFB_1 by their high rates of enzymatic conjugation of AFB_1 8,9-oxide, and rats can be rendered resistant by treatment with compounds that induce GSH *S*-transferases (*18*). In work done in our laboratories, it appears that human liver cytosol has considerably less GSH *S*-transferase activity towards synthetic AFB_1 8,9-oxide than rat liver cytosol. The consequences may be of significance in the evaluation of strategies for chemoprotection.

When a group of purified rat liver GSH *S*-transferases was examined, a μ-class enzyme (4-4) (*19*) and an α class enzyme (1-1) were the most active. It is of considerable interest that what appear to be two different stereoisomeric forms of the GSH:AFB_1 8,9-oxide conjugate are formed by different enzymes in both rat and human liver, and the α and μ enzymes form different products. In rats, the conjugation activity is induced by dithiolthione, which induces α class enzymes (*20*). Of the human enzyme classes examined, the activities varied in the order of μ > α > π. The relevance of this specificity *in vivo* remains to be determined.

4,4′-Methylenebis(2-chloroaniline)

4,4′-Methylenebis(2-chloroaniline) (MOCA) can be carcinogenic in rodents and dogs and is of concern because of its use in the curing of polyurethane. Although many of the carcinogenic arylamines are oxidized to hydroxylamines, mainly by P450 1A2 (Table 2), MOCA has been known to be an exception (*21*). Recently, we purified P4502A6 from human liver—the enzyme activates AFB_1 but does not appear to play an important role because of its low level [< 1% of total P450 (*22*)]. Purified P4502A6 catalyzed MOCA

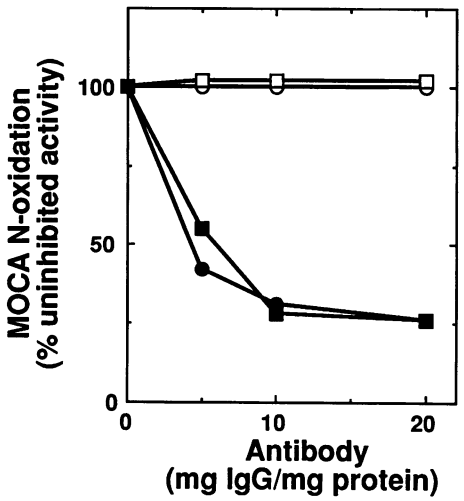

FIGURE 3. Immuno-inhibition of *N*-oxidation of 4,4′-methylenebis(2-chloroaniline) (MOCA) in two human liver samples. Pre-immune antibody (○,□); anti-human P4503A4 (●,■). Immunoglobulin G (IgG) fractions were prepared from rabbit antisera.

N-hydroxylation, but anti-P4502A6 inhibited < 20% of the activity in human liver microsomes. Anti-P4503A4, however, could inhibit up to 75% of the microsomal activity (Fig. 3), and levels of catalytic activity were well correlated with nifedipine oxidation in different liver samples (r^2 = 0.89). The P4503A4 inhibitor gestodene (*23*) could inhibit at least half of the activity in some liver samples (with inherently high activity) but was less inhibitory in samples with lower activity. Purified P4503A4 also catalyzed the reaction.

P4502E1, Ethyl Carbamate, and Chlorzoxazone

We and others have demonstrated that human P4502E1 can catalyze the oxidation of many small halogenated hydrocarbons and vinyl monomers to reactive forms (*24*). P4502E1 also oxidizes ethyl carbamate, of interest because of its use in industry and its presence in fermented beverages, to vinyl carbamate and then to the epoxide (*24,25*). The potential risk of these chemicals is still unclear, and it may be useful to have a noninvasive assay of P4502E1, since levels of the enzyme are known to vary and are induced by ethanol consumption (*26*). Human and animal P4502E1 enzymes are almost exclusively involved in

Table 3. Oxidation of aflatoxin B_1 (AFB_1) by human P4503A4.

P450	pmole Product formed min^{-1} (nmole P450)$^{-1}$[a]	
	AFQ_1	AFB_1 8,9-oxide:GSH conjugate
Yeast P450	285 ± 9	70 ± 1
Yeast P4503A4 plus 20 μM 7,8-benzoflavone	141 ± 13	260 ± 24

[a]Yeast recombinant P4503A4 (0.2 μM) was incubated with rabbit liver NADPH-P450 reductase, L-α-dilauroyl-*sn*-glyceryl-3-phosphocholine, 0.1 M potassium phosphate buffer (pH 7.7), 50 μM AFB_1, mouse liver cytosol, and an NADPH-generating system for 30 min (liver P4503A4) or 60 min (yeast P4503A4). The amounts of AFQ_1 and glutathione *S*-transferase:aflatoxin 8,9-oxide conjugate were determined by high-performance liquid chromatography.

6-hydroxylation of the muscle relaxant drug chlorzoxazone, at least as judged by studies *in vitro* (*27*). In collaborative studies with G. R. Wilkinson at Vanderbilt University, the usefulness of this reaction as a measure of P4502E1 *in vivo* is being considered. In the studies done to date, the conversion to 6-hydroxychlorzoxazone *in vivo* is inhibited by the P4502E1-selective agent disulfiram (*24*) and increased in individuals after withdrawal from consumption of large amounts of ethanol.

Lung P4501A1

Recently, what appears to be P4501A1 has been purified to electrophoretic homogeneity from human lung microsomes. The total amount of spectrally detectable P450 in human lung microsomes is very low, in the order of 10 pmole (P450)/mg protein. The properties of the enzyme are consistent with a role in the metabolism of several carcinogens. In samples with relatively high rates of oxidation of 7,8-dihydroxy-7,8-dihydrobenzo[*a*]pyrene, the role of P4501A1 is a major one, but in samples with lower activity its role appears to be less dominant. P4501A2 seems to be absent in human lung microsomes, as judged by several criteria. Another human lung microsomal P450 was partially purified, but its relevance in carcinogen metabolism is unknown.

Conclusions

A number of approaches can be used to characterize the selectivity of individual human enzymes *in vitro*, and our current views regarding catalytic selectivity are summarized here. *In vitro* approaches and observations on the metabolism of drugs have led to the development of noninvasive assays which can be used to phenotype individuals for enzymes hypothesized to contribute to the metabolism of procarcinogens. In some cases, a single enzyme can both activate and detoxify a single chemical, and work *in vivo* will be required to evaluate the significance of variation in enzyme levels.

This manuscript was presented at the Conference on Biomonitoring and Susceptibility Markers in Human Cancer: Applications in Molecular Epidemiology and Risk Assessment that was held in Kailua-Kona, Hawaii, 26 October–1 November 1991.

Supported in part by U.S. Public Health Service grant nos. CA 44353, ES 00267, and ES 07028. J.D.G. is a recipient of a Research Career Development Award K04 CA 01517.

REFERENCES

1. Miller, E. C., and Miller, J. A. Searches for ultimate chemical carcinogens and their reactions with cellular macromolecules. Cancer 47: 2327-2345 (1981).
2. Jakoby, W. B. Detoxication enzymes. In: Enzymatic Basis of Detoxication, Vol. 1 (W. B. Jakoby, Ed.), Academic Press, New York, 1980, pp. 1-6.
3. Conney, A. H., and Kappas, A. Interindividual differences in the metabolism of xenobiotics. Carcinogenesis 6: 147-165 (1985).
4. Guengerich, F. P., and Turvy, C. G. Comparison of levels of several human microsomal cytochrome P450 enzymes and epoxide hydrolase in normal and disease states using immunochemical analysis of surgical liver samples. J. Pharmacol. Exp. Ther. 256: 1189-1194 (1991).
5. Guengerich, F. P., and Shimada, T. Oxidation of toxic and carcinogenic chemicals by human cytochrome P450 enzymes. Chem. Res. Toxicol. 4: 391–407 (1991).
6. Guengerich, F. P., Martin, M. V., Beaune, P. H., Kremers, P., Wolff, T., and Waxman, D. J. Characterization of rat and human liver microsomal cytochrome P450 forms involved in nifedipine oxidation, a prototype for genetic polymorphism in oxidative drug metabolism. J. Biol. Chem. 261: 5051-5060 (1986).
7. Forrester, L. M., Neal, G. E., Judah, D. J., Glancey, M. J., and Wolf, C. R. Evidence for involvement of multiple forms of cytochrome P450 in aflatoxin B_1 metabolism in human liver. Proc. Natl. Acad. Sci. USA 87: 8306-8310 (1990).
8. Guengerich, F. P., and Liebler, D. C. Enzymatic activation of chemicals to toxic metabolites. CRC Crit. Rev. Toxicol. 14: 259-307 (1985).
9. Williams, D. E., Reed, R. L., Kedzierski, B., Guengerich, F. P., and Buhler, D. C. Bioactivation and detoxication of the pyrrolizidine alkaloid senecionine by cytochrome P450 isozymes in rat liver. Drug Metab. Dispos. 17: 387-392 (1989).
10. Miranda, C. L., Reed, R. L., Guengerich, F. P., and Buhler, D. R. Role of cytochrome P450IIIA4 in the metabolism of the pyrrolizidine alkaloid senecionine in human liver. Carcinogenesis 12: 515-519 (1991).
11. Guengerich, F. P. Oxidation of 17α-ethynylestradiol by human liver cytochrome P450. Mol. Pharmacol. 33: 500-508 (1988).
12. Shimada, T., and Guengerich, F. P. Evidence for cytochrome $P450_{NF}$, the nifedipine oxidase, being the principal enzyme involved in the bioactivation of aflatoxins in human liver. Proc. Natl. Acad. Sci. USA 86: 462-465 (1989).
13. Shimada, T., Iwasaki, M., Martin, M. V., and Guengerich, F. P. Human liver microsomal cytochrome P450 enzymes involved in the bioactivation of procarcinogens detected by *umu* gene response in *Salmonella typhimurium* TA1535/pSK1002. Cancer Res. 49: 3218-3228 (1989).
14. Kitada, M., Taneda, M., Ohta, K., Nagashima, K., Itahashi, K., and Kamataki, T. Metabolic activation of aflatoxin B_1 and 2-amino-3-methylimidazo[4,5-*f*]-quinoline by human adult and fetal livers. Cancer Res. 50: 2641-2645 (1990).
15. Aoyama, T., Yamano, S., Guzelian, P. S., Gelboin, H. V., and Gonzalez, F. J. 5 of 12 forms of vaccinia virus-expressed human hepatic cytochrome-P450 metabolically activate aflatoxin B_1. Proc. Natl. Acad. Sci. USA 87: 4790-4793 (1990).
16. Butler, M. A., Iwasaki, M., Guengerich, F. P., and Kadlubar, F. F. Human cytochrome $P450_{PA}$ (P450IA2), the phenacetin *O*-deethylase, is primarily responsible for the hepatic 3-demethylation of caffeine and *N*-oxidation of carcinogenic arylamines. Proc. Natl. Acad. Sci. USA 86: 7696-7700 (1989).
17. Holeski, C. J., Eaton, D. L., Monroe, D. H., and Bellamy, G. M. Effects of phenobarbital on the biliary excretion of aflatoxin P_1-glucuronide and aflatoxin B_1-S-glutathione. Xenobiotica 17: 139-153 (1987).
18. Kensler, T. W., Egner, P. A., Dolan, P. M., Groopman, J. D., and Roebuck, B. D. Mechanism of protection against aflatoxin tumorigenicity in rats fed 5-(2-pyrazinyl)-4-methyl-1,2-dithiol-3-thione (Oltipraz) and related 1,2-dithiol-3-thiones and 1,2-dithiol-3-ones. Cancer Res. 47: 4271-4277 (1987).
19. Ketterer, B., Meyer, D. J., and Clark, A. G. Soluble glutathione transferase isozymes. In: Glutathione Conjugation: Mechanisms and Biological Significance (H. Sies and B. Ketterer, Eds.), Academic Press, London, 1988, pp. 74-135.
20. Meyer, D. J., Coles, B., Harris, J., Gilmore, K. S., Raney, K., Harris, T. M., Guengerich, F. P., Kensler, T. W., and Ketterer, B. Induction of rat liver GSH transferases by 1,2-dithiol-3-thione illustrates both anticarcinogenic and tumor-promoting properties. In: Anticarcinogenesis and Antimutagenesis Mechanisms III (G. Bronzetti and D. L. Shankel, Eds.), Plenum Press, New York, in press.
21. Butler, M. A., Guengerich, F. P., and Kadlubar, F. F. Metabolic oxidation of the carcinogens 4-aminobiphenyl and 4,4′-methylene-bis-(2-chloroaniline) by human hepatic microsomes and by purified rat hepatic cytochrome P450 monooxygenases. Cancer Res. 49: 25-31 (1989).
22. Yun, C-H., Shimada, T., and Guengerich, F. P. Purification and characterization of human liver microsomal cytochrome P450 2A6. Mol. Pharmacol. 40: 679–685 (1991).
23. Guengerich, F. P. Mechanism-based inactivation of human liver cytochrome P450 IIIA4 by gestodene. Chem. Res. Toxicol. 3: 363-371 (1990).

24. Guengerich, F. P., Kim, D-H., and Iwasaki, M. Role of human cytochrome P450 IIE1 in the oxidation of many low molecular weight cancer suspects. Chem. Res. Toxicol. 4: 168-179 (1991).
25. Guengerich, F. P., and Kim, D-H. Enzymatic oxidation of ethyl carbamate to vinyl carbamate and its role as an intermediate in the formation of 1,N^6-ethenoadenosine. Chem. Res. Toxicol. 4: 413–421 (1991).
26. Perrot, N., Nalpas, B., Yang, C. S., and Beaune, P. H. Modulation of cytochrome P450 isozymes in human liver, by ethanol and drug intake. Eur. J. Clin. Invest. 19: 549-555 (1989).
27. Peter, R., Böcker, R. G., Beaune, P. H., Iwasaki, M., Guengerich, F. P., and Yang, C.-S. Hydroxylation of chlorzoxazone as a specific probe for human liver cytochrome P450 IIE1. Chem. Res. Toxicol. 3: 566-573 (1990).