Cytochrome P450 comprises a superfamily of enzymes involved in oxidation of a large number of endogenous and exogenous compounds associated with pharmacological and/or toxicological actions. In human liver, P450 3A enzymes are the major P450s, followed by P450 2C and 1A2 enzymes. It has been reported that drug-induced hepatotoxicity may be caused by active intermediates formed by animal and/or human P450 enzymes from common toxicants or idiosyncratic drugs that have been withdrawn from the market. Species differences between experimental animals and humans in the roles of P450 enzymes in drug metabolism are determinant factors in evaluating drug toxicity.

Some different regioselectivity of P450 1A2 between humans and rats has been shown, presumably because of different docking of the substrate caused by up to 25% amino acid differences in P450 1A2 proteins. A pyrazolopyrimidine-derivative drug, 5-n-butyl-7-(3,4,5-trimethoxybenzoylamino)pyrazolo[1,5-a]pyrimidine, is a recent typical case exhibiting species differences in drug metabolism. Exclusively limited elevations in serum levels of aspartate- and alanine-aminotransferase were seen in a clinical study with this pyrazolopyrimidine derivative, in contrast to the findings of no toxicological potential in rats and dogs. After the first hydrolysis of the amide moiety of this drug candidate, secondary metabolism was mediated by human P450 1A2 to predominantly produce a 3-hydroxylated pyrazolopyrimidine derivative which was able to yield a quinine-imine capable of binding to cysteine or glutathione at the C6-position. Rat P450 1A2 produced a C6-hydroxylated metabolite which could not bind any SH-reagent at the C6-position. The differences in hepatotoxicity in humans and rats were consistent with the metabolic specificity of human and rat P450 1A2 toward the primary metabolite, implicating human P450 1A2 in generating species-specific reactive metabolite(s) of the drug. The hepatotoxic model compound 5-n-butyl-pyrazolo[1,5-a]pyrimidine was also activated by human liver microsomal P450 1A2 to reactive intermediate(s) in vivo in humanized chimeric mice and could relatively non-specifically bind to biomolecules such as P450 1A2 and other proteins.

Cooperativity of human liver microsomal P450 3A enzymes was recently recognized for thalidomide, which is undergoing renewed clinical interest despite its teratogenicity. Enhanced rates of midazolam or cyclosporine A clearance in the presence of thalidomide were seen in liver microsomes. Thalidomide was oxidized to 5-hydroxythalidomide and 5'-hydroxythalidomide by NADPH-fortified liver microsomes from humans and monkeys. Recombinant human P450s 3A4, 3A5, and 3A7 and monkey P450s 3A4 and 3A5 (co-expressed with NADPH-P450 reductase in bacterial membranes) also catalyzed (R)-thalidomide 5-hydroxylation. P450 3A enzymes showed sigmoidal curves for (R)-thalidomide 5-hydroxylation and also oxidized 5-hydroxythalidomide to an epoxide or dihydroxy compound. Liquid chromatography-mass spectrometry analysis revealed formation of a glutathione conjugate from (R)- and (S)-5-hydroxythalidomide catalyzed by liver microsomal P450 3A enzymes in the presence of glutathione (assigned as a conjugate of 5-hydroxythalidomide formed on the phenyl ring). These results indicate that human P450s 3A4 and 3A5 mediate thalidomide 5-hydroxylation and further oxidation leading to a glutathione conjugate, which may be of relevance in the pharmacological and toxicological actions of thalidomide via cooperativity of P450 3A enzymes.

Biography

Hiroshi Yamazaki, PhD, has been Professor of Showa Pharmaceutical Univ since 2005. He graduated from Gifu Pharmaceutical Univ Grad Sch and was a scientist at Osaka Pref. Inst Publ Hlth (-1998) and an Assoc. Prof. of Kanazawa Univ (-2001) and Hokkaido Univ (-2005). He has been the recipient of young scientist awards from the Pharmaceutical Society of Japan (2000) and the JSSX (2005) and is a JSSX Fellow (2008). He is recognized as a Highly Cited Researcher in Pharmacology by ISI/Thomson Reuters.