Identification of the Cytochrome P450 Isoenzymes Involved in the Formation of the Main Metabolites of the Designer Drugs DOI, DOB, MDOB, and TMA-2 and Studies on Their Capability to Inhibit CYP2D6

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AIMS: 4-Iodo-2,5-dimethoxyamphetamine (DOI), 4-bromo-2,5-dimethoxyamphetamine (DOB), 4-bromo-2,5-dimethoxymethamphetamine (MDOB), and 2,4,5-trimethoxyamphetamine (TMA-2) are designer drugs which have appeared on the illicit drug market. Meanwhile, DOB and TMA-2 have been scheduled in the German Controlled Substances Act. Because of the various possibilities of interactions between different drugs especially with respect to metabolism the first aim of our study was to identify the cytochrome P450 (CYP) isoenzymes involved in the main metabolic steps of DOI, DOB, MDOB, and TMA-2. The second aim was to check whether these drugs are capable to inhibit CYP2D6, one of the major isoenzymes involved in the metabolism of xenobiotics.

METHODS: Studies on the CYP isoenzymes involved in the O-demethylation of DOI, DOB, MDOB, and TMA-2 were performed with nine individual cDNA expressed CYPs (1A2, 2A6, 2B6, 2C8, 2C9, 2C19, 2D6, 2E1, and 3A4) using microsomes of baculovirus infected insect cells as enzyme sources (details in Staack et al., Biochem. Pharmacol. 67, 235, 2004). Incubations were carried out over 30 min. They were started by adding microsomes to the incubation mixtures and terminated by protein precipitation with acetonitrile. Then, the supernatants were analyzed by LC-MS. The inhibition studies with CYP2D6 were performed by 10 min incubations and LC-MS analysis in a similar way as mentioned above. Dextromethorphan was used as CYP2D6 specific substrate (1, 2, 5, 10, 20, 50, 150, 500, 750, 1000, 1250, 1500 µM) and each of the studied drugs (250, 125, 25 µM) as well as the known CYP2D6 inhibitors fluoxetine (125 µM) and quinidine (25 nM) as inhibitors. The inhibition constants (Ki) were estimated from the Km values of dextromethorphan as obtained in presence and absence of the different inhibitors.

RESULTS: CYP2D6 was found to be the only isoenzyme involved in the O-demethylation of the studied designer drugs, but the rate of formation was low. Besides being substrates of this isoenzyme, DOI, DOB, MDOB, and TMA-2 proved to be competitive inhibitors of CYP2D6 with the Ki values of 7.1 µM, 94 µM, 13.3 µM, and 308 µM, respectively. The Ki values for quinidine and fluoxetine were 9.2 nM and 8.2 µM, respectively.

CONCLUSIONS: The exclusive metabolism of the studied designer drugs by CYP2D6 may result in considerable variations in hepatic drug elimination due to CYP2D6 poor or ultra rapid metabolism. Furthermore, different pharmacokinetics of other CYP2D6 substrates, e.g. many pharmaceuticals or designer drugs, may occur when co-administered with DOI or MDOB the Ki values of which were similar to the known potent CYP2D6 inhibitor fluoxetine. These changes of pharmacokinetics can result in higher plasma levels and even intoxications.

Keywords: Designer drugs, Cytochrome P450, Inhibition