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# Toxicokinetics of Amphetamines: Metabolism and Toxicokinetic Data of Designer Drugs, Amphetamine, Methamphetamine, and Their N-Alkyl Derivatives

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**Summary:** This paper reviews the toxicokinetics of amphetamines. The designer drugs MDA (methylenedioxy-amphetamine, *R,S*-1-(3′,4′-methylenedioxyphenyl)-2-propanamine), MDMA (*R,S*-methylenedioxymethamphetamine), and MDE (*R,S*-methylenedioxyethylamphetamine), as well as BDB (benzodioxolylbutanamine; *R,S*-1-(1′,3′-benzodioxol-5′-yl)-2-butanamine or *R,S*-1-(3′,4′-methylenedioxyphenyl)-2-butanamine) and MBDB (*R,S*-N-methyl-benzodioxolylbutanamine), were taken into consideration, as were the following N-alkylated amphetamine derivatives: amphetaminil, benzphetamine, clobenzorex, dimethylamphetamine, ethylamphetamine, famprofazone, fencamine, fenethylline, fenproporex, furfenorex, mefenorex, mesocarb, methamphetamine, prenylamine, and selegiline. English-language publications from 1995 to 2000 were reviewed. Papers describing identification of metabolites or cytochrome P450 isoenzyme-dependent metabolism and papers containing pharmacokinetic/toxicokinetic data were considered and summarized. The implications of toxicokinetics for toxicologic assessment or for interpretation in forensic cases are discussed. **Key Words:** Toxicokinetics—Metabolism—Cytochrome P450—Designer drugs—Amphetamine—Methamphetamine.

New therapeutic drugs are extensively studied in controlled clinical studies concerning metabolism, including cytochrome P450 (CYP) isoenzyme differentiation, and further pharmacokinetics. These data are available for interpretation of analytic data within the context of clinical or forensic toxicology and in doping control. The situation is quite different for illicit drugs or for older therapeutic agents: corresponding data are incomplete, if available at all. New illicit drugs, often designed for particular pharmacologic effects, enter the black market without any safety testing. Usually, scientific institutes perform studies for risk assessment of these drugs, but such studies are limited for ethical reasons. In many cases, corresponding metabolic and especially pharmacokinetic/toxicokinetic data for humans can be obtained only from authentic clinical or forensic cases. The aim of the present paper was to review publications describing metabolism and further pharmacokinetic/toxicokinetic data of amphetamine-type designer drugs and of amphetamine, methamphetamine, and their N-alkyl derivatives. When data were collected from case reports, often neither dosage nor time of ingestion of the corresponding drug was known; nevertheless, such data are better than no data at all.

Studies on the CYP isoenzyme-dependent metabolism, which are common parts of standard procedures in the approval process of new drugs, help in the assessment of different questions, such as variations in the formation of pharmacologically active metabolites, formation of toxic metabolites, or interactions with other

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therapeutic drugs (drug abusers often are polydrug users). All these factors have consequences for the assessment of analytic results in clinical or forensic toxicology as well as in doping control.

## AMPHETAMINE-DERIVED DESIGNER DRUGS

The designer drugs MDA (methylenedioxyamphetamine, *R,S*-1-(3′,4′-methylenedioxyphenyl)-2-propanamine, "Love Pills"), MDMA (*R,S*-methylenedioxymethamphetamine, "Adam," "Ecstasy"), and MDE (*R,S*-methylenedioxyethylamphetamine; "Eve") as well as BDB (benzodioxolylbutanamine; *R,S*-1-(1′,3′-benzodioxol-5′-yl)-2-butanamine or *R,S*-1-(3′,4′-methylenedioxyphenyl)-2-butanamine) and MBDB (*R,S*-N-methylbenzodioxolylbutanamine), have gained great popularity as "rave drugs" (1). They produce feelings of euphoria and energy and a desire to socialize. Nichols coined the term "entactogens" for this new group of drugs (2). They may lead to more or less severe intoxications (3) and impairment in driving a car (4). Therefore, in clinical and forensic toxicology, these illicit drugs are often analyzed. Commercial amphetamine immunoassays have been successfully used for screening of methylenedioxyamphetamine derivatives. For example, intake of 140 mg MDE led to immunoassay positive results up to 60 hours after ingestion (5). However, more specific procedures are necessary for confirmation and for differentiation, because nonscheduled therapeutic agents may also give positive results (6). Confirmation and quantification methods using gas chromatography–mass spectrometry, high-performance liquid chromatography, capillary electrophoresis, or thin layer chromatography were reviewed in 1998 (6) and will not be discussed in the present review.

Although the designer drugs had the reputation of being safe, several experimental studies in rats and humans, in addition to epidemiologic studies, indicated risks to humans. Recent reviews summarize the current knowledge on hepatotoxicity (7) and neurotoxicity, psychopathology, and the abuse potential of such designer drugs (1). Using positron emission tomography, Ricaurte et al (8) showed that human MDMA use resulted in brain 5-HT neurotoxicity. Because metabolites are claimed to be responsible for the hyperthermic, neurotoxic, and/or hepatotoxic effects (9,10), detailed knowledge on the CYP dependent metabolism is necessary. Also, the role of polymorphic CYP isoforms should be discussed.

In this review, we present manuscripts published from 1995 to 2000 about the metabolism of amphetamine-derived designer drugs. We also include publications on the influence of CYP isoenzymes on their metabolism. In addition, we selected and summarized toxicokinetic parameters for toxicologic interpretation of analytic results from the literature.

### Metabolism

The metabolism of amphetamine-derived designer drugs has been well studied in humans and rats (11–16). As shown in Figure 1, these designer drugs undergo predominantly two overlapping metabolic pathways: O-demethylenation to dihydroxy derivatives (catechols) followed by methylation of one of the hydroxy groups, and successive degradation of the side chain to N-dealkyl and deaminooxo metabolites. A third route via aromatic hydroxylation (not shown) has been proposed, resulting in the production of neurotoxic trihydroxyamphetamines (12,17). However, these studies were performed using rats given very high doses of MDMA. These metabolites could be detected only in brain. In addition, other studies suggest that MDMA-induced neurotoxicity may occur mainly through the production of superoxide or other radicals rather than hydroxyl free radicals such as the trihydroxy metabolites (18). The propylamines MDA, MDMA, and MDE are additionally metabolized to glycine conjugates of the corresponding 3,4-disubstituted benzoic acids, so-called hippuric acid derivatives (15). However, 3,4-dihydroxy hippuric acid and 3-methoxy-4-hydroxy hippuric acid can also be metabolites of, for instance, food preservatives (19). As expected, the hydroxy metabolites were found to be excreted in conjugated form. For example, Maurer et al (16) detected the glucuronide of the hydroxymethoxy metabolite of MDE and the sulfate of its dihydroxy metabolite in urine extracts of humans after intake of MDE.

### Influence of CYP Isoenzymes

In vitro studies in rats and humans indicated that the demethylenation of MDMA to the toxic catechols and/or their oxidation products was catalyzed by CYP2D1 in rats or by polymorphic CYP2D6 in humans (20–23). However, because CYP2D1-deficient rats could also form such catechols (24), other CYP isoenzymes or other enzymatic or nonenzymatic mechanisms should also play a role in in vivo demethylenation.

In 2000, two papers were published clarifying the influence of CYP isoenzymes on the metabolism of designer drugs (16,25). Using rat and human liver microsomes and specific inhibitors for the main isoenzymes, Maurer et al (16) showed that N-demethylation of MDMA and MBDB was predominantly catalyzed in rats and humans by CYP1A2 and to only a minor extent by

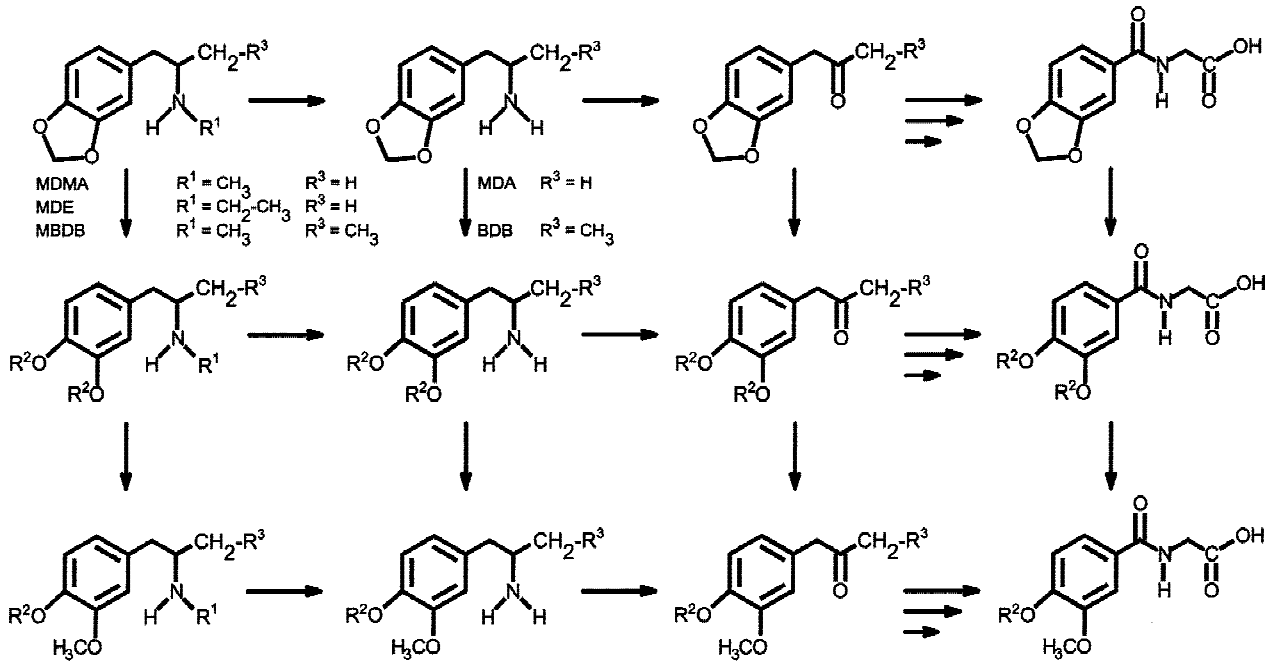


**FIG. 1.** Proposed scheme for the metabolism of the racemic MDA, MDMA, MDE, BDB, and MBDB in humans and rats. The glycine conjugates are only formed by the propylamines. The hydroxy metabolites ($R^2$ = H) are also present as glucuronic acid and/or sulfate conjugates ($R^2$ = sulfate or glucuronic acid) in urine. MDA, methylenedioxyamphetamine; MDMA, *R,S*-methylenedioxymethamphetamine; MDE, *R,S*-methylenedioxyethylamphetamine; BDB, benzodioxolylbutanamine; MBDB, *R,S*-N-methyl-benzodioxolylbutanamine. From Maurer HH [15]. Reproduced with permission.

CYP2D1 in rats or polymorphic CYP2D6 in humans. MBDB was additionally N-demethylated by CYP3A2/4. N-deethylation of MDE was predominantly catalyzed in rats and humans by CYP3A2/4, and in rats additionally to a minor extent by CYP1A2 and CYP2D1. They found that demethylenation of all tested drugs to the catechols was catalyzed in humans and rats predominantly by CYP2D1/6 and CYP3A2/4 isoenzymes. Demethylenation of MDMA and MBDB was additionally catalyzed in humans by CYP1A2. These results are summarized in Figure 2. The authors reported that demethylenation of designer drugs is also mediated by CYP-independent mechanisms. They also discussed the role of cytosolic catechol-*O*-methyl-transferase (COMT) in the toxicity of designer drugs. Massive release of catecholic neurotransmitters by designer drugs, together with catecholic designer drug metabolites, may lead to an overabundance of catechols, which might not be quantitatively detoxified by low active COMT (16,26).

Kreth et al (25) used human liver microsomes with specific inhibitors and antibodies for the different isoenzymes for their studies on demethylenation and dealkylation of MDMA, MDA, and MDE. They confirmed their results using recombinant CYP enzymes. Demethylenation of all three designer drugs followed clearly biphasic Michaelis-Menten kinetics in liver microsomes of extensive metabolizers. A low K(m) and a high K(m) demethylenase was assumed from the kinetic data. Microsomes of poor metabolizers completely lacked the low K(m) component, and the fitted kinetic parameters were in the same range as the high K(m) component of extensive metabolizer microsomes. Additional specific probes for CYP2D6 confirmed this isozyme as the exclusive low K(m) component for demethylenation. P450-selective inhibitors applied to CYP2D6-inhibited microsomes and activity measurements in a series of recombinant P450s suggested CYP1A2 as the major high K(m) component, with contributions by CYP2B6 and CYP3A4. For N-dealkylation, microsomal velocities were at least sevenfold lower than for demethylenation and were characterized by apparently monophasic kinetics. The authors found CYP2B6 to be the most important isozyme for this reaction. Inhibition data suggested that CYP1A2 and CYP3A4 might also participate in N-dealkylation of MDMA and MDE. The authors stated that the CYP2D6 poor metabolizer genotype might not be by itself a substantial risk factor for the adverse effects of MDA and MDE.

CYP2D6 polymorphism seems not to be crucial for MDMA toxicity. De la Torre et al (27) recently described the results of a double-blind, randomized, crossover and controlled clinical study (Table 1)(4,14,27–32). Eight

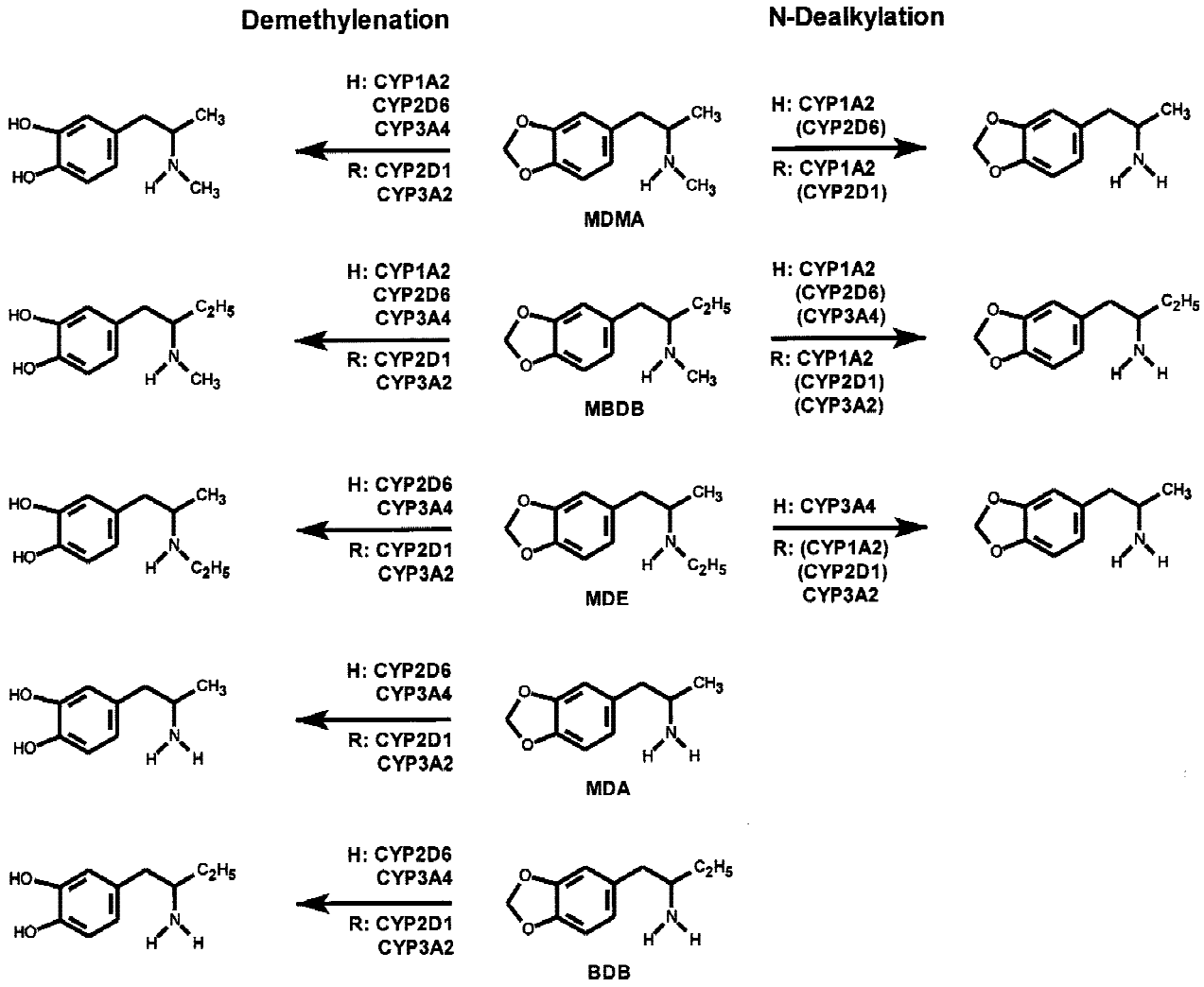


**FIG. 2.** Isoenzyme-dependent major metabolic pathways of racemic MDA, MDMA, MDE, BDB, and MBDB in humans (H) and rats (R). Metabolism by CYPs given in brackets is minor. MDA, methylenedioxyamphetamine; MDMA, *R,S*-methylenedioxymethamphetamine; MDE, *R,S*-methylenedioxyethylamphetamine; BDB, benzodioxolylbutanamine; MBDB, *R,S*-N-methyl-benzodioxolylbutanamine. From Maurer, et al. [16]. Reproduced with permission.

subjects (phenotyped for CYP2D6 activity) were given MDMA at doses of 75 and 125 mg, and blood levels at multiple time points were determined. With increasing doses, volunteers showed increases in MDMA concentrations that did not follow the same proportionality. The constant recovery of the hydroxy-methoxy metabolite in urine, combined with the increasing MDMA recovery, suggested a saturation or an inhibition of the demethylenation step. These observations were supported by the fact that urinary clearance was rather constant, whereas nonrenal clearance was dose-dependent. The authors concluded that the lack of linearity of MDMA pharmacokinetics (in a window of doses compatible with its recreational use) was a more general phenomenon as it concerns the whole population independent of their CYP2D6 genotype. It implies that relatively small increases in the dose of MDMA ingested lead to disproportionate rises in MDMA plasma concentrations, and hence subjects are more prone to develop acute toxicity.

There are additional data suggesting that CYP2D6 genotype may not be a risk factor for designer drug toxicity. Recently, three patients who died of MDMA intoxication were tested for their CYP2D6 genotype; all three were extensive metabolizers (33). A report on a fatal drug interaction between ritonavir, a strong inhibitor of CYP3A4 and CYP2D6, also suggests that multiple factors may influence the risk for adverse or toxic effects of amphetamine-derived designer drugs. Moreover, poor

**TABLE 1.** *Toxicokinetic data of designer drugs*

| Drugs | Dose (mg) | $C_{P\,max}$ (ng/mL) | $C_{U\,max}$ (ng/mL) | $t_{max}$ (h) | $t_{1/2}$ (h) | AUC (ng/mL*h) | Reference |
|---|---|---|---|---|---|---|---|
| MDMA | 100–120 | 331 | 28,100 | 2 | 5–10 | | 28 |
| MDA | | 15 | 2,300 | | | | |
| MDMA | 75 | 126 | | 2 | 9.5 | 995 | 29 |
| MDMA | 125 | 226 | | 2 | 9.1 | 2,235 | 29 |
| MDE | 140 | 203–465 | 24,900 | 1.5–3 | | 675–1,935 | 14,30 |
| MDA | | 7–33 | 6,700 | 3.3 | | 18–120 | |
| HME | | 67–670 | 89,690 | 1.5–4 | | 218–3,028 | |
| MBDB | 100 | | 18,577 | | | | 31 |
| BDB | | | 751 | | | | |
| MDMA | 50 | 19.8–82.8 | | 2–3 | 2.7–5.1 | 100.1–813.9 | 27 |
| MDA | | 5.1 | | 6 | 5.6 | 51.1 | |
| MDMA | 75 | 130.9 (±38.6) | | 1.8 (±0.4) | 7.7 (±3.2) | 1331.5 (±646.0) | 27 |
| MDA | | 7.8 (±2.5) | | 5.1 (±2.6) | 16.1 (±18.3) | 122.3 (±66.7) | |
| MDMA | 100 | 189.9–209.7 | | 2–3 | 5.8 | 1447.8–2256.6 | 27 |
| MDA | | 14.2–22.4 | | 4–6 | 6.3–6.4 | 61.5–345.4 | |
| MDMA | 125 | 236.4 (±58.0) | | 2.4 (±1.0) | 8.6 (±3.2) | 2623.7 (±572.9) | 27 |
| MDA | | 13.7 (±1.6) | | 7.1 (±2.8) | 27.7 (±26.0) | 215.2 (±68.5) | |
| MDMA | 150 | 441.9–486.9 | | 1.5–2 | 6.9–7.2 | 5132.8–5232.0 | 27 |
| MDA | | 31.4–34.2 | | 4–10 | 23.2–37.3 | 373.9–590.0 | |
| MDMA | — | 1–514 (median of n = 18: 76) | | | | | 4 |
| MDE | — | 1–777 (median of n = 17: 87) | | | | | 4 |
| MDA | — | 1–67 (median of n = 23: 13) | | | | | 4 |
| MDMA | | | 380–96,200 (mean of n = 34: 13400) | | | | 32 |
| MDA | | | 150–8,600 (mean of n = 34: 1600) | | | | 32 |

AUC, area under the curve (plasma concentration vs. time after ingestion; BDB, benzodioxolylbutanamine; $C_{P\,max}$, maximal plasma concentration; $C_{U\,max}$, maximal urine concentration; HME, 4-hydroxy-3-methoxyethylamphetamine; MDA, methylenedioxyamphetamine; MDMA, *R,S*-methylenedioxymethamphetamine; MDE, *R,S*-methylenedioxyethylamphetamine; $t_{max}$, time after ingestion, at which the plasma concentration is maximal; $t_{1/2}$, plasma elimination half-life.

metabolizers may be less susceptible to designer drug toxicity because fewer catechol metabolites are formed. However, because other isoforms than CYP2D6 can also catalyze demethylenation, predicting toxicity is almost impossible.

### Toxicokinetic Parameters

Determination of amphetamine-derived designer drugs and their metabolites in body fluids is often performed in clinical and forensic toxicology. Methods for this purpose can be found in different reviews (6,16,34, 35). However, much less toxicokinetic information for amphetamine-derived designer drugs has been reported. Some published data are summarized in Table 1 for the interpretation of analytic results in clinical and forensic toxicology. Some of the data were obtained from noncontrolled or single-case studies. When interpreting analytic results in correlation with these data, broad individual variations must be considered due to variations in metabolic or excretion rates or due to drug or food interactions.

## AMPHETAMINE AND ITS N-ALKYL DERIVATIVES

Amphetamine (*R,S*-1-phenyl-2-propanamine) and methamphetamine (*R,S*-N-methyl-1-phenyl-2-propanamine) are powerful stimulants of the central nervous system by acting as indirect sympathomimetic drugs. They are substrates of the monoamine transporter in the cell membrane of dopaminergic, noradrenergic, and adrenergic neurons and in the membrane of the storage vesicles. They have no affinity to the adrenoceptors or dopamine receptors. Their effects result from a nonexocytotic, carrier-mediated release of neurotransmitters. They are drugs of abuse as well as doping agents. The *S(+)* enantiomers of amphetamine and methamphetamine have five times more psychostimulant activity than the *R(−)* enantiomers. The main metabolic pathways of amphetamine and methamphetamine have been known for several years: aromatic hydroxylation, aliphatic hydroxylation, N-demethylation, oxidative deamination, N-oxidation, and conjugation of the nitrogen. The phenolic metabolites are partly excreted as conjugates. The metabolism of amphetamine and methamphetamine is not reviewed here in detail; an overview can be found

in the review by Musshoff (36). Details on the CYP isoenzyme-dependent metabolism of amphetamine and methamphetamine, which have been published in the time frame of the present review, are discussed in the corresponding section.

N-alkylated or N,N-dialkylated amphetamine derivatives such as amphetaminil, benzphetamine, clobenzorex, dimethylamphetamine, ethylamphetamine, famprofazone, fencamine, fenethylline, fenproporex, furfenorex, mefenorex, mesocarb, methamphetamine, prenylamine, and selegiline are therapeutically used as sympathomimetics, anorectics, nonopioid analgesics, antiparkinsonian agents, or vasodilators. Figure 3 shows the chemical structures of these compounds. It is well known that these drugs are metabolically (bis)dealkylated to amphetamine or methamphetamine (6,37–43). Therefore, they are of interest in forensic toxicology and doping control, where legitimate use of these therapeutics must be differentiated from illicit amphetamine use. Immunoassays are usually used for urine screening for amphetamines to differentiate between negative and presumptively positive samples. Positive results must be confirmed by a second independent method that is at least as sensitive as the screening test and that provides the highest level of confidence in the result. Gas chromatography–mass spectrometry (GC-MS) is the most widely used method for confirmation of positive screening tests (6), because it provides high levels of specificity and sensitivity. The mandatory Guidelines for Federal Workplace Drug Testing in the United States also require GC-MS as the confirmation method. Such confirmation of positive immunoassay results for amphetamines should not be limited to detection of amphetamine and methamphetamine by GC-MS in the selected ion monitoring (SIM) mode. Those amphetamine- and methamphetamine-derived agents must be considered as part of a thorough interpretation of amphetamine-positive results. Detection of the hydroxy metabolites may help extend the time of differentiation of illicit amphetamine abuse from intake of therapeutic drugs. However, in the late phase of excretion after the intake of amphetamine-derived therapeutic agents, amphetamine and methamphetamine are often the only metabolites that can be detected in urine. In such urine samples, differentiation of illicit amphetamine or methamphetamine intake from the intake of therapeutic drugs may be impossible. In the past 2 years, two review articles have been published that discussed these problems (6,36). Detailed discussion has therefore been restricted in this review to a few recent papers.

In the following section, we will review papers on the metabolism (especially on the CYP isoenzyme-dependent metabolism) of amphetamine and those amphetamine derivatives. In addition, we selected and summarized pharmacokinetic parameters from the literature. These data may also help in differentiating (meth)-amphetamine intake from legitimate use of therapeutic drugs.

## Metabolism

The metabolism of N-alkylated amphetamine and methamphetamine derivatives was reported several years ago. However, in the time frame of this review, some interesting papers have been published describing the metabolism and discussing implications for forensic toxicology. Studies were performed on the metabolism of these therapeutics to find specific metabolites suitable for differentiation (37,44–48). Figure 4 shows metabolic pathways of N-substituted amphetamine- and methamphetamine-derived therapeutics. The main metabolic pathways are one or two fold ring hydroxylation, followed by methylation of one of the hydroxy groups; N-demethylation and/or N-dealkylation; and oxidative deamination (overlapping pathways not indicated by arrows). However, not all of these metabolites can be detected in every case. The parent compounds are usually detectable for only a few hours after ingestion and are not useful as a target compound for differentiation unless high cutoff values are chosen. Using a cutoff value as high as 500 ng/mL, fenproporex parent compound was detectable in all amphetamine-positive samples after intake of 10 mg of the drug (49). However, this method cannot be recommended for differentiation in every case, because fenproporex was not detectable in urine samples from the same study with amphetamine results of 477 ng/mL. Because only five volunteers were tested, and given the great interindividual variability, there might be a sample that contains no parent compound even if containing more than 500 ng/mL amphetamine. The corresponding hydroxy metabolites of (meth)amphetamine derivatives, which are not N-dealkylated and therefore specific for the taken drug, can be detected for a much longer time (6,50). Common procedures for confirmation of positive amphetamine immunoassay results are not suitable for the detection of such metabolites because they do not include cleavage of conjugates and the analyte is extracted at a strong alkaline pH. The (metabolic) introduction of an aromatic hydroxy group into a phenylalkylamine derivative increases the acidity of the compound and thereby changes its extractive properties. The resulting phenol bases are best extracted at their isoelectric point at pH 8 to 9. The analytic problems

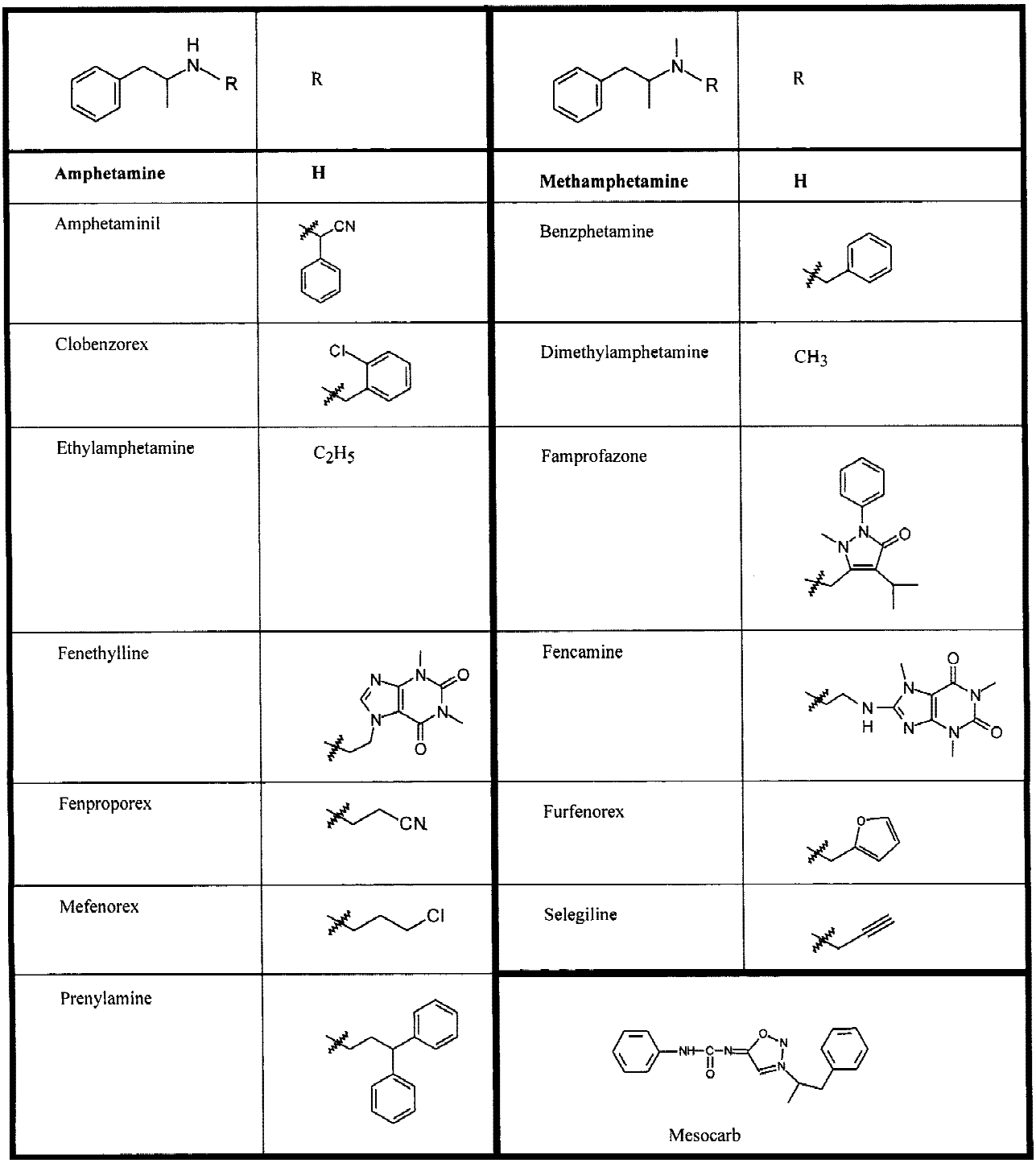


**FIG. 3.** Structures of amphetamine and methamphetamine derivatives. From Kraemer T and Maurer HH [6]. Reproduced with permission.

in differentiating amphetamine abuse from intake of legal medications were discussed in a review by Kraemer and Maurer (6).

A good example for differentiating amphetamine abuse from intake of a therapeutic agent by detecting the hydroxy metabolite is clobenzorex. In 1997, Maurer et al (50) showed that detection of parent compound in addition to amphetamine did not allow differentiation between a therapeutic drug and an illicit substance. Detection of hydroxyclobenzorex in addition to amphetamine

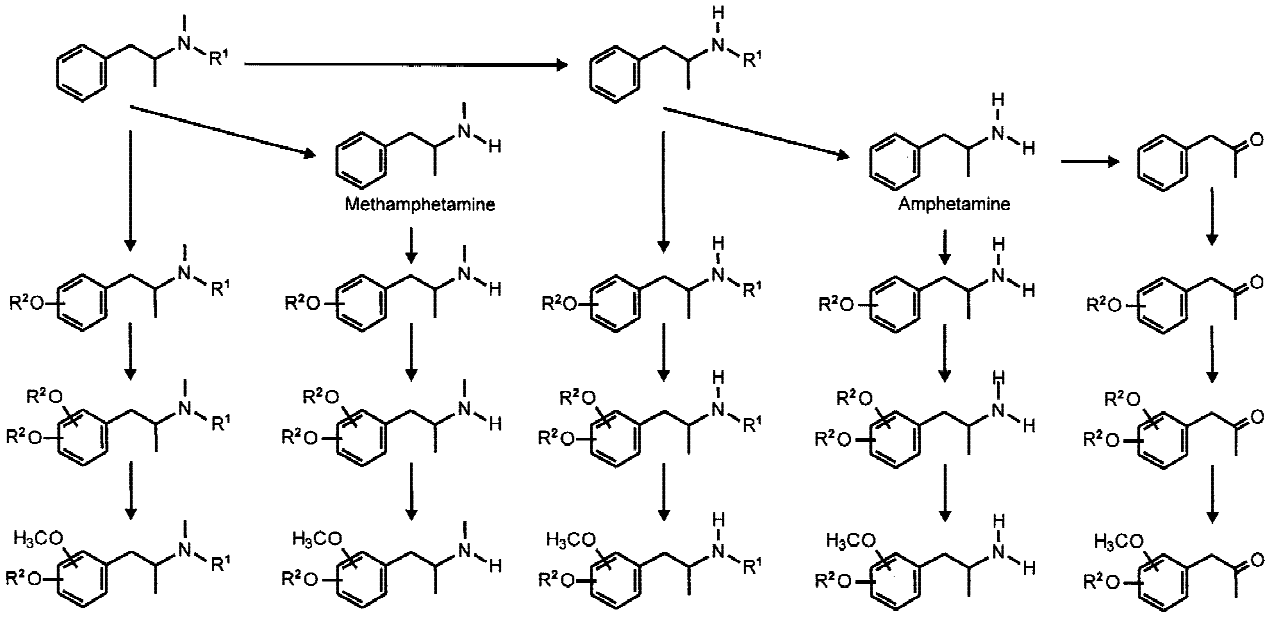


**FIG. 4.** Predominant metabolic pathways of amphetamine and methamphetamine derivatives ($R^1$ = R in Fig. 3; overlapping pathways not indicated by arrows). The hydroxy metabolites ($R^2$ = H) are also present as glucuronic acid and/or sulfate conjugates ($R^2$ = sulfate or glucuronic acid) in urine. From Kraemer T and Maurer HH [6]. Reproduced with permission.

allowed one to make this differentiation. In all urine samples that had tested positive for amphetamine, hydroxyclobenzorex was also detectable. In a series of three papers published in 1999 and 2000, these results were confirmed by other authors (44,51,52). Unfortunately, this differentiation is not possible for all of the amphetamine-derived medications. In the later phase of excretion, amphetamine and methamphetamine are often the only metabolites that can be detected in urine. In such urine samples, illicit amphetamine or methamphetamine intake cannot be differentiated from the intake of legal therapeutic drugs (37,42).

Studies on enantiomeric profiles for amphetamine or methamphetamine metabolically formed from amphetamine-derived medications have also been used to differentiate from a drug of abuse. Analytic methods for such chiral analysis were extensively reviewed by Kraemer and Maurer (6). Some of those medications are prescribed as pure enantiomers. Racemization does not occur during metabolism, so that resulting amphetamine and methamphetamine are also pure enantiomers. An analytic result of both enantiomers of amphetamine or amphetamine is therefore inconsistent with intake of a single-enantiomer therapeutic agent. This has been demonstrated for benzphetamine (46) and selegiline (53–55). If the medication contains the S(+)-enantiomer of the (meth)amphetamine derivative and only the S(+)-enantiomer of (meth)amphetamine is detected in a biosample, an abuse of amphetamine cannot be excluded in every case, because abuse of the pure S(+)-enantiomer of (meth)amphetamine is possible. In the case of selegiline, differentiation seems to be easier (53–55). Because selegiline is the pure R(−)-enantiomer, only R(−)-amphetamine and R(−)-methamphetamine are metabolically formed. Abuse of pure R(−)-enantiomers of amphetamine or methamphetamine is not known (and pharmacologically not very useful). Additional methamphetamine abuse would result in detection of both enantiomers of amphetamine and methamphetamine. As discussed above, such findings are inconsistent with selegiline intake.

It has also been proposed that changes over time in the enantiomeric ratio of amphetamine after intake of amphetamine-derived medicaments may help in differentiation. Indeed, there is a change in the enantiomeric composition of amphetamine after intake of the corresponding racemic drugs. This can be explained by stereoselectivity of the enzymes involved in metabolism of these drugs. Dopamine-beta-hydroxylase, which also catalyzes beta-hydroxylation of amphetamines to the corresponding ephedrines, reacts stereoselectively with the S(+)-enantiomer. N-dealkylation of amphetamine derivatives is also influenced by their stereochemistry. Enantioselective studies on racemic precursors have been performed, for instance on famprofazone (40,47,56,57), fenproporex (48,49,58), and prenylamine (43). However, it is questionable whether the enantiomer ratio may really help in differentiating amphetamine abuse from intake

of legal amphetamine-derived medicaments. In actual situations, it cannot be determined whether the enantiomeric ratio is caused only by metabolic dealkylation of a racemic medicament or whether racemic or pure S(+)-enantiomer of illicit amphetamine was taken alone or in combination at the same time or at different time points with the corresponding therapeutic medications.

In conclusion, a positive amphetamine or methamphetamine result in urine can be caused by intake of a legal therapeutic drug. In a later phase of excretion of such therapeutic agents, differentiation from abuse of illicit amphetamine or methamphetamine is not always possible, regardless of which method is used.

## Influence of CYP Isoenzymes

From 1995 to 2000, several papers were published that discussed the influence of CYP isoenzymes on the metabolism of amphetamine derivatives (22,23,38,59–63). Isoenzyme-dependent metabolism of anorectics, prenylamine, or famprofazone has not yet been described. However, such studies have been performed in our laboratory, and the results will be presented and published in the near future. Several publications on amphetamine or methamphetamine (22,23,38,59,60,62,64–66), additional reports on benzphetamine (59,62,67), and numerous manuscripts on selegiline (59,61,63,64,67) have described isoform-dependent metabolism of these drugs.

### *Amphetamine and Methamphetamine*

Moody et al (68) reported involvement of CYP2D in ring hydroxylation of amphetamine tested in vivo by feeding specific CYP2D inhibitors (quinidine) to rats and after application of amphetamine. Also the 4-hydroxylation of S(+)- and R(−)-methamphetamine seems to be catalyzed by CYP2D1/6. This reaction was examined in liver microsomes of Sprague-Dawley and Dark Agouti rat strains. The latter rat strain is a model of the CYP2D6 poor metabolizer phenotype. In the study, anti-P450-BTL IgG, bufuralol, and quinine, a substrate and inhibitors of CYP2D isozymes, respectively, were found to block approximately 90% of the reaction as catalyzed by microsomes from Sprague-Dawley rats. Reconstituted systems of CYP2D isozymes purified from rat liver microsomes also mediated the reaction. In microsomes from Dark Agouti rats, only minimal activity was found (60). In studies with human liver microsomes (including those from a genetically poor metabolizer with respect to CYP2D6), Lin et al (22) concluded a major involvement of CYP2D6 in the aromatic 4-hydroxylation of methamphetamine. They confirmed this in studies with recombinant CYP2D6 expressed in yeast, which was also shown to effect N-demethylation of methamphetamine. Bach et al (38) found the same results for amphetamine using microsomal preparations from cells expressing human CYP2D6. In a recent report, Law et al (66) confirmed again that CYP2D6 is selectively involved in amphetamine 4-hydroxylation. Urinary elimination of 4-hydroxy amphetamine and amphetamine was determined in male Sprague-Dawley rats pretreated with P450 inducers and inhibitors. Urinary elimination of 4-hydroxyamphetamine was significantly decreased by, and the metabolic ratio was increased by, the inhibitors 1-aminobenzotriazole, $CCl_4$, quinidine, quinine, and primaquine. Amphetamine and methamphetamine are substrates as well as competitive inhibitors of CYP2D6 (23). In conclusion, there is convincing evidence about the role of CYP2D6 in the ring hydroxylation of amphetamine and methamphetamine.

Amphetamine is oxidatively deaminated by monoamine oxidase (MAO). This deamination to phenylacetone appears to be catalyzed by CYP2C (62). The authors reported that deamination of amphetamine in rabbit was extensively inhibited only with quinidine, thought to be a specific inhibitor of CYP2D. However, deamination with purified rabbit CYP2C3 (previously identified as the major isozyme responsible for this metabolism) was extensively inhibited also with quinidine. These investigators concluded that the CYP2C isozymes played a major role in the deamination of amphetamine.

### *Benzphetamine*

Metabolism of benzphetamine (and other N,N-dialkylated amphetamines) was examined in vitro with a microsomal preparation from cells expressing human CYP2D6 (59). In contrast to selegiline, benzphetamine was not dealkylated in this test system. Ring-hydroxylated products were not found in this study.

Studies in liver microsomes from rabbits and rats and in reconstituted systems containing CYP2C subfamily isozymes showed that deamination of benzphetamine to phenylacetone had the highest activity compared with various other phenylisopropylamines (67). In a further report by the same working group, the authors showed that the CYP2C isozymes played a major role in the deamination of benzphetamine. However, N-debenzylation of benzphetamine in rat was not catalyzed by this isoenzyme. Multiple isozymes seemed to be involved in this reaction (62).

### *Selegiline*

In 1996, Sharma et al (64) reported that selegiline irreversibly inactivated the 7-ethoxy-4-trifluoromethylcoumarin O-deethylase activity of purified

CYP2B1 in a reconstituted system. Their studies on the isozyme specificity of selegiline in microsomes from rats pretreated with specific inducers showed that this inactivation was relatively specific for the major phenobarbital-inducible isozyme, CYP2B1, because significant inactivation of CYP1A1 and CYP2E1 was not observed. In their conclusions, they wondered whether CYP2D6, which they believed responsible for selegiline metabolism, might also be inactivated.

In 1994, Grace et al (69) suggested that CYP2D6 should be involved in selegiline N-dealkylation, at least in their in vitro model of recombinant yeast-expressed CYP2D6. However, in vivo studies with five poor and eight extensive metabolizers by Scheinin et al (61) showed that CYP2D6 polymorphism was not crucial for the disposition of selegiline. These authors did not find significant differences in the pharmacokinetic parameters of selegiline, desmethylselegiline, and R(−)-amphetamine between poor metabolizers and extensive metabolizers. However, the area under the serum concentration-time curve values of R(−)-methamphetamine were, on average, 46% higher in poor metabolizers than in extensive metabolizers. The inhibitory potency of selegiline, desmethylselegiline, and R(−)-methamphetamine toward dextromethorphan O-demethylase was very low. Quinidine did not inhibit the formation of desmethylselegiline or R(−)-methamphetamine from selegiline. Scheinin et al suggested involvement of CYP2D6 in a later phase of selegiline elimination, especially in the ring hydroxylation. In their study, one ultrarapid metabolizer was also tested who did not differ from the other volunteers with regard to pharmacodynamics and pharmacokinetics (with the exception of a low area under the curve of R(−)-methamphetamine).

In 2000, discussion continued on which isoenzymes may be responsible for selegiline metabolism. Bach et al (59) reported that selegiline had been dealkylated in their test system by CYP2D6 expressed in a human cell line. To clarify the interactions of selegiline with the CYP system, Valoti et al (63) treated C57BL mice with selegiline, ethanol, phenobarbital, or beta-naphthoflavone to induce different CYP isozymes. Selegiline caused a significant decrease in total CYP levels. Both phenobarbital and ethanol increased the N-depropinylation activity toward selegiline that led to the formation of methamphetamine. Ethanol alone increased the N-demethylation rate

**TABLE 2.** *Pharmacokinetic data of amphetamine derivatives*

| Drugs | Dose (mg) | $C_{P\,max}$ (ng/mL) | $C_{U\,max}$ (ng/mL) | $t_{max}$ (h) | $t_{1/2}$ (h) | AUC (ng/mL*h) | Reference |
|---|---|---|---|---|---|---|---|
| Selegiline | 10 | 2.2 ± 1.17 | | 0.9 ± 0.38 | 1.2 ± 0.56 | | 71 |
| Selegiline | 10 (poor metabolizer) | 4.9 ± 2.3 | | 0.5–1.0 | | 3.6 ± 0.9 | 61 |
| Nor-SEL | | 94.1 ± 28.9 | | 0.75–1.5 | 4.02 ± 1.15 | 234 ± 47 | |
| R(−)MA | | 82.4 ± 17.0 | | 2.5–4 | 15.4 ± 5.1 | 2,219 ± 310 | |
| R(−)AM | | 24.1 ± 4.6 | | 4–12 | 18.1 ± 5.3 | 761 ± 149 | |
| Selegiline | 10 (extensive metabolizer) | 9.3 ± 10 | | 0.5–1 | | 6.3 ± 5.5 | 61 |
| Nor-SEL | | 98.1 ± 27.1 | | 0.75–1.5 | 5.09 ± 3.04 | 228 ± 109 | |
| R(−)MA | | 72.4 ± 8.3 | | 0.75–6 | 11.6 ± 2.3 | 1,522 ± 376 | |
| R(−)AM | | 21.4 ± 3.5 | | 2.5–8 | 17.0 ± 5.8 | 640 ± 191 | |
| Benzphetamine | 50 (as hydrochloride) | | — | | | | 46 |
| MA | | | 139–965 | | | | |
| AM | | | 207–3,776 | | | | |
| Fenproporex | 10 | | 0–706 | | | | 48 |
| AM | | | 1,200–2,100 | | | | |
| Fenproporex | 10 (each for 7 days) | | 0–2,750 | | | | 49 |
| AM | | | 2,850–4,150 | | | | |
| Clobenzorex | 60 | | | | | | 45 |
| AM | | | 715–2,474 | | | | |
| Clobenzorex | 10 (each for 7 days) | | 8–47 | | | | 44 |
| AM | | | 2,900–4,700 | | | | |
| AM | 5 | | 620–3,160 | | | | 72 |
| AM | 10 | | 1,960–3,120 | | | | 72 |
| AM | 20 | | 1,510–4,570 | | | | 72 |
| Famprofazone | 50 | | | | | | 57 |
| MA | | | 10–1,967 | | | | |
| AM | | | 22–420 | | | | |
| Prenylamine | 120 | | | | | | 43 |
| AM | | | 10–1,280 | | | | |

AM, amphetamine; MA, methamphetamine; $C_{P\,max}$, maximal plasma concentration; $C_{U\,max}$, maximal urine concentration; $t_{max}$; time after ingestion, at which the plasma concentration is maximal; $t_{1/2}$, plasma elimination half-life; AUC, area under the curve (plasma concentration vs. time after ingestion).

of selegiline, resulting in the formation of nor-selegiline. Moreover, the N-dealkylation pathways of deprenyl were inhibited by 4-methylpyrazole and disulfiram, two CYP2E1 inhibitors. None of the other treatments (including specific CYP2D6 inhibition by quinine/quinidine) modified selegiline metabolism. The authors concluded that mainly CYP2E1 and to a lesser extent CYP2B isozymes should be involved in selegiline metabolism. They also suggested that by reducing CYP content, selegiline treatment might impair the metabolic disposition of other drugs given in combination regimens. Abuse of alcohol might also have implications on selegiline treatment.

In conclusion, CYP isoenzyme-dependent metabolism of selegiline is still controversial. In vivo studies and studies on liver microsomes containing the whole spectrum of CYP isoforms suggested that isoenzymes other than CYP2D6 would be responsible for selegiline dealkylation. This metabolic reaction seems to be catalyzed by CYP2D6 only in in vitro systems containing this isoform as the only one. The question is whether these systems can be representative of in vivo metabolism.

### Pharmacokinetic Parameters

The pharmacokinetic parameters of selegiline are well known from previous publications. A good overview of several parameters, including the effects of food, multiple doses, gender, age, and even other drug application routes (such as transdermal application), can be found in the publication by Mahmood (70). Therefore, not all this information has been included in Table 2(43–46,48, 49,57,61,71,72). Pharmacokinetic parameters of other amphetamine derivatives are rarely described and can also be found in Table 2. Urinary excretion of amphetamines is markedly influenced by pH in the kidney ultrafiltrate. Under acidic conditions, amphetamines are mostly ionized, and reabsorption in the kidneys hardly occurs. Under alkaline conditions, un-ionized amphetamines are readily reabsorbed in the kidneys and excretion in urine decreases. When interpreting analytic results in correlation with such data, broad individual variations must be considered due to variations in metabolic or excretion rates, due to drug or food interactions, and especially due to the fact that some data are taken from noncontrolled studies.

## CONCLUSIONS

Identification of metabolites of amphetamine-derived designer drugs and of other amphetamine derivatives is well described. Studies in the past few years of the CYP-dependent metabolism of these drugs or medications has led to a better understanding of the toxicologic effects of designer drugs and to optimizing therapy with amphetamine derivatives. These reports also provide information to limit the risks of interactions and to understand interindividual differences in amphetamine results after intake of amphetamine-derived medicaments.

The proposition that CYP2D6 polymorphism may be responsible for the great interindividual differences in designer drug toxicity seems to be refuted. Other, nonpolymorphic isoenzymes and even CYP-independent mechanisms have been shown to be responsible for O-demethylenation of designer drugs. More studies are necessary to understand designer drug toxicity and the influence of metabolism on this toxicity. One proposal has been to study the influence of COMT, which could be important in the detoxification of catecholic designer drug metabolites.

CYP-dependent metabolism of amphetamine-derived medications has mainly been studied for selegiline. Selegiline pharmacokinetics seems to be independent from CYP2D6 polymorphism. CYP-dependent metabolism of most of the amphetamine derivatives still remains to be studied. Despite the many studies reviewed in this manuscript, data on the pharmacokinetics and toxicokinetics of amphetamines remain incomplete.

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## REFERENCES

1. Hegadoren KM, Baker GB, Bourin M. 3,4-Methylenedioxy analogues of amphetamine: defining the risks to humans. *Neurosci Biobehav Rev* 1999;23:539–53.
2. Nichols DE. Differences between the mechanism of action of MDMA, MBDB, and the classic hallucinogens. Identification of a new therapeutic class: entactogens. *J Psychoactive Drugs* 1986; 18:305–13.
3. Walubo A, Seger D. Fatal multi-organ failure after suicidal overdose with MDMA, "Ecstasy": case report and review of the literature. *Hum Exp Toxicol* 1999;18:119–25.
4. Moeller MR, Hartung M. Ecstasy and related substances—serum levels in impaired drivers. *J Anal Toxicol* 1997;21:591.
5. Ensslin HK, Kovar KA, Maurer HH. Toxicological detection of the designer drug 3,4-methylenedioxyethylamphetamine (MDE, "Eve") and its metabolites in urine by gas chromatography-mass spectrometry and fluorescence polarization immunoassay. *J Chromatogr B* 1996;683:189–97.
6. Kraemer T, Maurer HH. Determination of amphetamine, methamphetamine and amphetamine-derived designer drugs or medicaments in blood and urine. *J Chromatogr B* 1998;713:163–87.
7. Jones AL, Simpson KJ. Review article: mechanisms and management of hepatotoxicity in ecstasy (MDMA) and amphetamine intoxications. *Aliment Pharmacol Ther* 1999;13:129–33.
8. Ricaurte GA, McCann UD, Szabo Z, et al. Toxicodynamics and

long-term toxicity of the recreational drug, 3, 4-methylenedioxymethamphetamine (MDMA, "Ecstasy"). *Toxicol Lett* 2000;112: 143–146.
9. Hiramatsu M, Kumagai Y, Unger SE, et al. Metabolism of methylenedioxymethamphetamine: formation of dihydroxymethamphetamine and a quinone identified as its glutathione adduct. *J Pharmacol Exp Ther* 1990;254:521–7.
10. Carvalho F, Remiao F, Amado F, et al. d-Amphetamine interaction with glutathione in freshly isolated rat hepatocytes. *Chem Res Toxicol* 1996;9:1031–6.
11. Lim HK, Foltz RL. In vivo and in vitro metabolism of 3,4-(methylenedioxy)methamphetamine in the rat: identification of metabolites using an ion trap detector. *Chem Res Toxicol* 1988;1: 370–8.
12. Lim HK, Foltz RL. In vivo formation of aromatic hydroxylated metabolites of 3,4-(methylenedioxy)methamphetamine in the rat: identification by ion trap tandem mass spectrometric (MS/MS and MS/MS/MS) techniques. *Biol Mass Spectrom* 1991;20:677–86.
13. Maurer HH, Ensslin HK, Kovar KA. On the metabolism of designer drugs, part 1: Studies on methylenedioxyetilamfetamine (MDE) in rats. *Bull Soc Sci Med Grand-Duche Lux* 1990;127 (suppl):467–71.
14. Ensslin HK, Maurer HH, Gouzoulis E, et al. Metabolism of racemic 3,4-methylenedioxyethylamphetamine in humans. Isolation, identification, quantification, and synthesis of urinary metabolites. *Drug Metab Dispos* 1996;24:813–20.
15. Maurer HH. On the metabolism and the toxicological analysis of methylenedioxyphenylalkylamine designer drugs by gas chromatography-mass spectrometry. *Ther Drug Monit* 1996;18:465–70.
16. Maurer HH, Bickeboeller-Friedrich J, Kraemer T, et al. Toxicokinetics and analytical toxicology of amphetamine-derived designer drugs ("Ecstasy"). *Toxicol Lett* 2000;112:133–142.
17. Lim HK, Foltz RL. Ion trap tandem mass spectrometric evidence for the metabolism of 3,4-(methylenedioxy)methamphetamine to the potent neurotoxins 2,4,5-trihydroxymethamphetamine and 2,4,5-trihydroxyamphetamine. *Chem Res Toxicol* 1991;4:626–32.
18. Yeh SY. Effects of salicylate on 3,4-methylenedioxymethamphetamine (MDMA)-induced neurotoxicity in rats. *Pharmacol Biochem Behav* 1997;58:701–8.
19. Liebich HM, Forst C. Basic profiles of organic acids in urine. *J Chromatogr* 1990;525:1–14.
20. Tucker GT, Lennard MS, Ellis SW, et al. The demethylenation of methylenedioxymethamphetamine ("Ecstasy") by debrisoquine hydroxylase (CYP2D6). *Biochem Pharmacol* 1994;47:1151–6.
21. Chu T, Kumagai Y, Distefano EW, et al. Disposition of methylenedioxymethamphetamine and three metabolites in the brains of different rat strains and their possible roles in acute serotonin depletion. *Biochem Pharmacol* 1996;51:789–96.
22. Lin LY, Di Stefano EW, Schmitz DA, et al. Oxidation of methamphetamine and methylenedioxymethamphetamine by CYP2D6. *Drug Metab Dispos* 1997;25:1059–64.
23. Wu D, Otton SV, Inaba T, et al. Interactions of amphetamine analogs with human liver CYP2D6. *Biochem Pharmacol* 1997;53: 1605–12.
24. Colado MI, Williams JL, Green AR. The hyperthermic and neurotoxic effects of Ecstasy (MDMA) and 3,4 methylenedioxyamphetamine (MDA) in the Dark Agouti (DA) rat, a model of the CYP2D6 poor metabolizer phenotype. *Br J Pharmacol* 1995;115: 1281–9.
25. Kreth K, Kovar K, Schwab M, et al. Identification of the human cytochromes P450 involved in the oxidative metabolism of "Ecstasy"-related designer drugs. *Biochem Pharmacol* 2000;59: 1563–71.
26. Lavigne JA, Helzlsouer KJ, Huang HY, et al. An association between the allele coding for a low activity variant of catechol-O-methyltransferase and the risk for breast cancer. *Cancer Res* 1997; 57:5493–7.
27. De la Torre R, Farre M, Ortuno J, et al. Non-linear pharmacokinetics of MDMA ("Ecstasy") in humans. *Br J Clin Pharmacol* 2000;49:104–9.
28. Helmlin HJ, Bracher K, Bourquin D, et al. Analysis of 3,4-methylenedioxymethamphetamine (MDMA) and its metabolites in plasma and urine by HPLC-DAD and GC-MS. *J Anal Toxicol* 1996;20:432–40.
29. De la Torre R, Ortuno J, Farre M, et al. Pharmacokinetics of ecstasy (MDMA) in healthy subjects. *Ther Drug Monit* 1997;19: 585.
30. Brunnenberg M, Lindenblatt H, Gouzoulis ME, et al. Quantitation of N-ethyl-3,4-methylenedioxyamphetamine and its major metabolites in human plasma by high-performance liquid chromatography and fluorescence detection. *J Chromatogr B* 1998;719:79–85.
31. Kintz P. Excretion of MBDB and BDB in urine, saliva, and sweat following single oral administration. *J Anal Toxicol* 1997;21: 570–5.
32. Kunsman GW, Levine B, Kuhlman JJ, et al. MDA-MDMA concentrations in urine specimens. *J Anal Toxicol* 1996;20:517–21.
33. Schwab M, Seyringer E, Brauer RB, et al. Fatal MDMA intoxication [letter]. *Lancet* 1999;353:593–4.
34. Moeller MR, Steinmeyer S, Kraemer T. Determination of drugs of abuse in blood. *J Chromatogr B* 1998;713:91–109.
35. Kintz P, Samyn N. Determination of "Ecstasy" components in alternative biologic specimens. *J Chromatogr B* 1999;733:137–43.
36. Musshoff F. Illegal or legitimate use? Precursor compounds to amphetamine and methamphetamine. *Drug Metab Rev* 2000;32: 15–44.
37. Kraemer T, Theis GA, Weber AA, Maurer HH. Studies on the metabolism and toxicological detection of the amphetamine-like anorectic fenproporex in human urine by gas chromatography-mass spectrometry and fluorescence polarization immunoassay (FPIA). *J Chromatogr B* 2000;738:107–18.
38. Bach MV, Coutts RT, Baker GB. Involvement of CYP2D6 in the in vitro metabolism of amphetamine, two N- alkylamphetamines and their 4-methoxylated derivatives. *Xenobiotica* 1999;29: 719–32.
39. Szoko E, Kalasz H, Magyar K. Metabolic transformation of deprenyl enantiomers in rats. *Neurobiology Bp* 1999;7:247–54.
40. Shin HS. Stereoselective metabolism of famprofazone in humans: N-dealkylation and beta- and p-hydroxylation. *Chirality* 1997;9: 52–8.
41. Shin HS. Metabolism of selegiline in humans. Identification, excretion, and stereochemistry of urine metabolites. *Drug Metab Dispos* 1997;25:657–62.
42. Kraemer T, Vernaleken I, Maurer HH. Studies on the metabolism and toxicological detection of the amphetamine-like anorectic mefenorex in human urine by gas chromatography-mass spectrometry and fluorescence polarization immunoassay. *J Chromatogr B* 1997;702:93–102.
43. Kraemer T, Roditis SK, Peters FT, et al. Amphetamine concentrations in human urine following single-dose administration of the calcium antagonist prenylamine. In: *Proceedings of the 38th International TIAFT Meeting*, Helsinki, 2000.
44. Baden KL, Valtier S, Cody JT. Metabolic production of amphetamine following multidose administration of clobenzorex. *J Anal Toxicol* 1999;23:511–7.
45. Valtier S, Cody JT. Metabolic production of amphetamine following administration of clobenzorex. *J Forensic Sci* 1999;44:17–22.
46. Cody JT, Valtier S. Detection of amphetamine and methamphetamine following administration of benzphetamine. *J Anal Toxicol* 1998;22:299–309.
47. Musshoff F, Kraemer T. Identification of famprofazone ingestion. *Int J Legal Med* 1998;111:305–8.
48. Cody JT, Valtier S. Detection of amphetamine following administration of fenproporex. *J Anal Toxicol* 1996;20:425–31.
49. Cody JT, Valtier S, Stillman S. Amphetamine and fenproporex levels following multidose administration of fenproporex. *J Anal Toxicol* 1999;23:187–94.

50. Maurer HH, Kraemer T, Ledvinka O, et al. Gas chromatography-mass spectrometry (GC-MS) and liquid chromatography-mass spectrometry (LC-MS) in toxicological analysis. Studies on the detection of clobenzorex and its metabolites within a systematic toxicological analysis procedure by GC-MS and by immunoassay and studies on the detection of alpha- and beta-amanitin in urine by atmospheric pressure ionization electrospray LC-MS. *J Chromatogr B* 1997;689:81–9.
51. Cody JT, Valtier S. A gas chromatography-mass spectrometry method for the quantitation of clobenzorex. *J Anal Toxicol* 1999; 23:603–8.
52. Valtier S, Cody JT. Differentiation of clobenzorex use from amphetamine abuse using the metabolite 4-hydroxyclobenzorex. *J Anal Toxicol* 2000;24:606–13.
53. Maurer HH, Kraemer T. Toxicological detection of selegiline and its metabolites in urine using fluorescence polarization immunoassay (FPIA) and gas chromatography-mass spectrometry (GC-MS) and differentiation by enantioselective GC-MS of the intake of selegiline from abuse of methamphetamine or amphetamine. *Arch Toxicol* 1992;66:675–8.
54. Hasegawa M, Matsubara K, Fukushima S, et al. Stereoselective analyses of selegiline metabolites: possible urinary markers for selegiline therapy. *Forensic Sci Int* 1999;101:95–106.
55. Kupiec TC, Chaturvedi AK. Stereochemical determination of selegiline metabolites in postmortem biologic specimens. *J Forensic Sci* 1999;44:222–6.
56. Neugebauer M, Khedr A, el-Rabbat N, et al. Stereoselective metabolic study of famprofazone. *Biomed Chromatogr* 1997;11: 356–61.
57. Cody JT. Enantiomeric composition of amphetamine and methamphetamine derived from the precursor compound famprofazone. *Forensic Sci Int* 1996;80:189–99.
58. Cody JT, Valtier S. Enantiomer profile for amphetamine derived from the precursor compound fenproporex. *J Anal Toxicol* 1997; 21:84.
59. Bach MV, Coutts RT, Baker GB. Metabolism of N,N-dialkylated amphetamines, including deprenyl, by CYP2D6 expressed in a human cell line. *Xenobiotica* 2000;30:297–306.
60. Lin LY, Kumagai Y, Hiratsuka A, et al. Cytochrome P4502D isozymes catalyze the 4-hydroxylation of methamphetamine enantiomers. *Drug Metab Dispos* 1995;23:610–4.
61. Scheinin H, Anttila M, Dahl ML, et al. CYP2D6 polymorphism is not crucial for the disposition of selegiline. *Clin Pharmacol Ther* 1998;64:402–11.
62. Shiiyama S, Soejima OT, Honda S, et al. Major role of the CYP2C isozymes in deamination of amphetamine and benzphetamine: evidence for the quinidine-specific inhibition of the reactions catalyzed by rabbit enzyme. *Xenobiotica* 1997;27:379–87.
63. Valoti M, Fusi F, Frosini M, et al. Cytochrome P450-dependent N-dealkylation of L-deprenyl in C57BL mouse liver microsomes: effects of in vivo pretreatment with ethanol, phenobarbital, beta-naphthoflavone and L-deprenyl. *Eur J Pharmacol* 2000;391:199–206.
64. Sharma U, Roberts ES, Hollenberg PF. Inactivation of cytochrome P4502B1 by the monoamine oxidase inhibitors R-(−)-deprenyl and clorgyline. *Drug Metab Dispos* 1996;24:669–75.
65. Gunaratna C, Kissinger PT. Investigation of stereoselective metabolism of amphetamine in rat liver microsomes by microdialysis and liquid chromatography with precolumn chiral derivatization. *J Chromatogr A* 1998;828:95–103.
66. Law MY, Slawson MH, Moody DE. Selective involvement of cytochrome P450 2D subfamily in in vivo 4-hydroxylation of amphetamine in rat. *Drug Metab Dispos* 2000;28:348–53.
67. Yamada H, Shiiyama S, Soejima Ohkuma T, et al. Deamination of amphetamines by cytochromes P450: studies on substrate specificity and regioselectivity with microsomes and purified CYP2C subfamily isozymes. *J Toxicol Sci* 1997;22:65–73.
68. Moody DE, Ruangyuttikarn W, Law MY. Quinidine inhibits in vivo metabolism of amphetamine in rats: impact upon correlation between GC/MS and immunoassay findings in rat urine. *J Anal Toxicol* 1990;14:311–7.
69. Grace JM, Kinter MT, Macdonald TL. Atypical metabolism of deprenyl and its enantiomer, (S)-(+)-N,alpha-dimethyl-N-propynylphenethylamine, by cytochrome P450 2D6. *Chem Res Toxicol* 1994;7:286–90.
70. Mahmood I. Clinical pharmacokinetics and pharmacodynamics of selegiline. An update. *Clin Pharmacokinet* 1997;33:91–102.
71. Mahmood I, Marinac JS, Willsie S, et al. Pharmacokinetics and relative bioavailability of selegiline in healthy volunteers. *Biopharm Drug Dispos* 1995;16:535–45.
72. Poklis A, Still J, Slattum PW, et al. Urinary excretion of d-amphetamine following oral doses in humans: implications for urine drug testing. *J Anal Toxicol* 1998;22:481–6.