ORIGINAL RESEARCH ARTICLE

Clin Pharmacokinet 2011; 50 (5): 319-329
0312-5963/11/0005-0319/$49.95/0

© 2011 Adis Data Information BV. All rights reserved.

Sex Differences in 3,4-Methylenedioxymethamphetamine (MDMA; Ecstasy)-Induced Cytochrome P450 2D6 Inhibition in Humans

Samanta Yubero-Lahoz,[1,2] *Ricardo Pardo*,[1,3] *Magí Farré*,[1,3] *Brian O'Mahony*,[1,2] *Marta Torrens*,[4,5] *Cristina Mustata*,[1,3] *Clara Pérez-Mañá*,[1,3] *Marcel·lí Carbó*[1,2] and *Rafael de la Torre*[1,2]

1 Human Pharmacology and Clinical Neurosciences Research Group, Neuropsychopharmacology Program, IMIM-Hospital del Mar Research Institute, Barcelona, Spain
2 Department of Experimental and Health Sciences, Universitat Pompeu Fabra (CEXS-UPF), Barcelona, Spain
3 Department of Pharmacology, Therapeutics and Toxicology, Universitat Autònoma de Barcelona, Barcelona, Spain
4 Substances Abuse Disorders Research Group, Neuropsychopharmacology Program, IMIM-Hospital del Mar Research Institute, Institute of Neuropsychiatry and Addictions (INAD)-Hospital del Mar, Barcelona, Spain
5 Department of Psychiatry, Universitat Autònoma de Barcelona, Barcelona, Spain

Abstract

Background and Objective: 3,4-Methylenedioxymethamphetamine (MDMA; ecstasy) is a ring-substituted amphetamine widely used for recreational purposes. MDMA is predominantly *O*-demethylenated in humans by cytochrome P450 (CYP) 2D6 and is also a potent mechanism-based inhibitor of the enzyme. This study assessed the inhibition and recovery half-life of CYP2D6 and CYP3A4 activity in female subjects by administering the probe drug dextromethorphan before and repeatedly after MDMA administration. These data were compared with the data obtained from a previous study in male subjects.

Study Design: Twelve healthy female subjects who were CYP2D6 extensive metabolizers participated as outpatients in two experimental sessions. Session 1 was conducted over 2 days and session 2 over 10 days, with a minimum of 3 days between sessions. In session 1, subjects received a single oral dose of dextromethorphan 30 mg. In session 2, a 1.5 mg/kg MDMA dose was given at 0 hours, followed at 4 hours by repeated 30 mg doses of dextromethorphan over the next 10 days.

Methods: Plasma concentration-time profiles and urinary recoveries of dextromethorphan and its metabolites dextrorphan (DOR), 3-methoxymorphinan (MM) and hydroxymorphinan-3-ol (HM) were measured.

Results: MDMA given prior to dextromethorphan resulted in a 10-fold increase in the dextromethorphan maximum plasma concentration (C_{max}) and area under the plasma concentration-time curve (AUC), with corresponding decreases in DOR and HM pharmacokinetic parameters. The mean ± SD C_{max} of MDMA was 188.8 ± 16.7 ng/mL, with a time to reach C_{max} (t_{max}) of 2.0 ± 0.4 hours and an AUC from 0 to 25 hours of 2645.2 ± 170.9 mg • h/mL. The urinary recovery of the dextromethorphan dose as dextromethorphan and its main metabolites was 25.4 ± 8.9% with no MDMA pretreatment versus 6.6 ± 1.1% after 1.5 mg/kg of MDMA ($p = 0.0001$). The metabolic ratio (MR) increased almost 60-fold from 0.018 ± 0.028 to 0.998 ± 0.932 after MDMA administration, with 100% of the subjects having a value greater than the antimode of 0.3 that signified the poor-metabolizer phenotype. Data analysis of results obtained in the present study compared with those from a previous study in male subjects showed significant differences in the dextromethorphan/DOR MR in the 0- to 8-hour (session 1) and 4- to 12-hour (session 2, post MDMA) collection periods ($p = 0.032$ and $p = 0.01$, respectively). CYP2D6 activity recovered after 10 days to 90% of baseline activity, with a recovery half-life of 36.6 ± 22.9 hours. Male subjects showed a shorter recovery half-life (27.6 ± 25.1 hours). The measurement of CYP3A4 activity indicated a non-significant increase in C_{max} and AUC values of MM after drug intake, but urinary data reflected significant differences in dextromethorphan/MM MR in both

sexes, although the difference was more pronounced in women. Dextromethorphan/MM MR increased almost 3-fold from baseline.

Discussion and Conclusion: In women the pretreatment with MDMA resulted in a decrease in dextromethorphan clearance. CYP2D6 activity recovered after 10 days to 90% of baseline activity. Regarding CYP3A4 activity, there is an apparent decrease in its activity after MDMA use. In women, MDMA use has been associated with psychiatric symptoms and psychological problems that may require psychopharmacological treatment with antidepressant drugs, some of which are known CYP2D6 substrates. MDMA-induced mechanism-based inhibition of CYP2D6 is of relevance, and physicians should be advised to prescribe medications whose metabolic disposition is not regulated by CYP2D6.

Background

3,4-Methylenedioxymethamphetamine (MDMA; ecstasy) is a synthetic amphetamine derivative widely abused recreationally because of its entactogenic effects.[1,2] The serotonin syndrome (increased muscle rigidity, hyperreflexia and hyperthermia) is characteristic of acute toxicity episodes.[3] A number of case reports of severe intoxication and death after MDMA abuse have been reported.[4] A gradual loss of some cognitive functions have been observed among long-term users of MDMA, probably as a consequence of its adverse effects on the central serotonergic system.[5-9] Metabolism of MDMA in humans is rather complex and details can be found in several reports.[10-16]

O-demethylenation of MDMA is the main metabolic pathway, giving rise to 3,4-dihydroxymethamphetamine (HHMA). This reaction is mainly regulated by cytochrome P450 (CYP) 2D6, with the contribution of other CYP isoenzymes. CYP2D6 is a polymorphic enzyme responsible for the clearance of one-quarter of drugs used in therapeutics.[17-19] According to their mean effect on the CYP2D6 phenotype, genotypes can be grouped into three phenotype subgroups: poor metabolizers (PMs), extensive metabolizers (EMs), and ultrarapid metabolizers (UMs). Subjects with a PM phenotype lack two functional alleles,[20] whereas EM subjects have one or two functional alleles and UM subjects have more than two.[21-23]

The CYP2D6 phenotype is usually determined by administering a probe drug, such as dextromethorphan or debrisoquine.[20,24] *In vitro*, CYP2D6 contributes 80% to the formation of dextrorphan (DOR), and CYP3A4 contributes more than 90% to the formation of 3-methoxymorphinan (MM) from dextromethorphan as a substrate[25] and therefore is used to simultaneously assess CYP2D6 and CYP3A4 activities *in vivo* based on the determination of urine metabolic molar ratios of dextromethorphan/DOR and dextromethorphan/MM, respectively.[26-29] Both DOR and MM are partially demethylated to hydroxymorphinan (HM; hydroxymorphinan-3-ol) and all are recovered in urine, as glucuronides.[30]

Acute MDMA intoxication has been associated with high plasma concentrations[31] but not with non-functional *CYP2D6* genotypes that may explain MDMA accumulation in the body.[32] These observations suggest that acute toxicity episodes are related to MDMA exposure at high doses and/or to the fact that MDMA disposition in the body follows nonlinear pharmacokinetics.[33-35] MDMA interacts with CYP2D6 both as a substrate and as an inhibitor of its own metabolism by the formation of a complex between CYP2D6 and MDMA,[36,37] the formation of which has been shown *in vitro* in both rats and humans.[38] It is postulated that the methylenedioxy group present in the chemical structure of MDMA is responsible for the metabolic autoinhibition. Mathematical modelling of MDMA pharmacokinetic data in humans indicates that a period of 280 hours without dosing is required for the activity of CYP2D6 to return to normal.[33] In practice, inhibition of CYP2D6 activity is not >30% because of the contribution of alternative CYP isoenzymes, including CYP3A4, following its inactivation.[35] The quasi-irreversible mechanism-based inhibition (MBI) of CYP2D6 is associated with a decrease in the amount of effective enzyme such that recovery of activity depends on its *de novo* synthesis. Inhibition of CYP2D6 by MDMA and the recovery of its activity, using the dextromethorphan/DOR MR as a biomarker, in male subjects have already been reported.[39] Little is known about the pharmacology of MDMA in women and how this drug affects CYP2D6 activity. Preliminary reports suggest marked sex differences in MDMA plasma concentrations.[40]

The present study had two main objectives: to study the metabolism of dextromethorphan in women before and after MDMA intake, taking into account the activity of CYP2D6 and CYP3A4 in these two situations; and to compare the data obtained here with those previously published by Mathúna et al.[39] in males. A between-sexes comparison was made, considering three aspects: the metabolism of dextromethorphan in control conditions; its metabolism after CYP2D6 inhibition by MDMA; and the duration of CYP2D6 inactivation in both sexes and the return to baseline activity.

© 2011 Adis Data Information BV. All rights reserved.

Methods

Study Participants

Twelve healthy Caucasian females recruited by word-of-mouth were included in the study. Eligibility required self-reported recreational use of MDMA on at least ten occasions and twice in the previous year. None had a history of adverse medical or psychiatric reactions after MDMA consumption. Subjects were interviewed by a psychiatrist to exclude those with major psychiatric disorders or with a history of such disorders[41] and by a physician to exclude concomitant medical conditions. Subjects underwent a general physical examination, routine laboratory tests, urinalysis and a 12-lead ECG. The participants had a mean (SD) age of 26.9 (3.7) years, bodyweight of 56.1 (4.8) kg and height of 167 (4.8) cm. All but four were current smokers. None met criteria for drug abuse or drug dependence (except for nicotine), and all had previous experience with other psychostimulants, cannabis or hallucinogens. Subjects were informed about the study, provided written informed consent before inclusion in the study and were compensated for their participation. The study was conducted in accordance with the Declaration of Helsinki (2000), approved by the local institutional review board (the Clinical Research Ethical Committee of the Parc de Salut Mar) and authorized by the Spanish Medicines Agency (SMA; authorization no. 04-0013). All participants were phenotyped for CYP2D6 activity, using dextromethorphan; the EM phenotype was required for participation in the study.[42] Subjects were requested to refrain from consuming drugs of abuse for 2 weeks before and throughout the duration of the study. Regular ingestion of medication in the month preceding the study was an exclusion criterion, although single doses of symptomatic medication were accepted up to the week preceding the trial. Tobacco smoking was permitted 6 hours after drug administration. The women were not taking oral contraceptive steroids and all of them participated during the follicular menstrual period. At each session and before drug administration, urine samples were collected for drug testing (opioids, cocaine, amphetamines and cannabis) by a rapid test device (Instant-View®; Alpha Scientific Designs, Inc., Poway, CA, USA). A positive screening test was considered an exclusion criterion.

Study Design and Drugs

Subjects participated as outpatients in two experimental sessions. Session 1 was conducted over 2 days and session 2 over 10 days, with a minimum wash-out period of 3 days between sessions. The study design was open-label since the principal variables being measured were objective (the pharmacokinetics of dextromethorphan, its metabolites and MDMA). A pilot study was first conducted in three individuals to determine the optimal dosing intervals for dextromethorphan. Dextromethorphan tablets (Romilar®; Roche Farma, SA, Madrid, Spain) were supplied by the pharmacy at the Hospital del Mar (Barcelona, Spain). (*R,S*)-MDMA was supplied by the Spanish Ministry of Health and prepared in white, opaque, soft gelatine capsules by the pharmacy at the Hospital del Mar.

Each session began at 07:30h following an overnight fast. Drug administration commenced at 08:30h. In session 1, 30 mg of dextromethorphan was given at 0 hour. In session 2, a 1.5 mg/kg MDMA dose (minimum 75 mg; maximum 100 mg) was given at 0 hour followed by repeated 30 mg doses of dextromethorphan at 4, 24, 96, 168 and 240 hours. The dose of MDMA was chosen to be in the range reported for single recreational doses of MDMA.[43] All drugs were administered by the oral route. A light meal was provided 2 hours after initial drug administration. Blood samples were obtained through an indwelling catheter inserted into a subcutaneous vein in the forearm of the non-dominant arm. Thereafter, the subjects remained seated in a quiet room. In session 1, blood samples (8 mL) were taken at 0, 0.5, 1, 2, 4, 6, 8 and 24 hours after dextromethorphan administration; in session 2, blood samples were collected at 4, 4.5, 5, 6, 8, 10, 12 and 28 hours after MDMA administration. After centrifugation at 4°C, four 1 mL aliquots of plasma were stored at −20°C until analysis. Subjects were asked to urinate before the session commenced, to empty the bladder (pre-basal), and subsequently urine was collected for the periods of 0–8 and 8–24 hours in session 1 following dextromethorphan administration and for the periods of 0–4, 4–12, 24–32, 96–104, 168–176 and 240–248 hours in session 2 following MDMA administration. Urinary pH was measured and then acidified; aliquots were stored at −20°C until analysis. During the sessions, cardiovascular effects were recorded for safety reasons (continuous ECG, blood pressure, heart rate and temperature) but those data are not presented in this manuscript.

Genotyping

Samples of 1 mL of whole blood were taken for DNA extraction and *CYP2D6* genotyping with a DNA microarray (PHARMAchip; Progenika Biopharma SA, Vizcaya, Spain). *CYP2D6* genotypes and the corresponding dextromethorphan/DOR metabolic ratios (MRs) of subjects are shown in table I. Each genotype was classified according to the number of functional alleles. Those subjects bearing a functional allele and

© 2011 Adis Data Information BV. All rights reserved.

Table I. Cytochrome P450 (CYP) 2D6 female and male genotypes in the present study and a previous study[39]

CYP2D6 genotype	Functional alleles (n)	Subjects (n)		MR[a]
		men[39]	women	
*1/*4	1	2	1	0.0096
*9/*10	1	1	0	
*2/*4	1	1	0	
*1/*5	1	1	0	
*1/*3	1	0	1	0.0043
*4/*41	1	0	1	0.1008
*1/*10	2	1	0	
*1/*2	2	5	2	0.0121 ± 0.0012
*1/*9	2	1	0	
*2/*41	2	1	0	
*1/*1	2	1	3	0.0139 ± 0.0125
*1/*17	2	1	0	
*1/*41	2	0	1	0.0045
*2/*10	2	0	1	0.0081
*1/*35	2	0	1	0.0112
*2/*35	2	0	1	0.0051

a Mean (±SD) dextromethorphan/dextrorphan metabolic ratio (MR) in women in the present study.

a second partially functional allele were classified as having two functional alleles. Table I depicts the genotypes of female subjects analysed in the present study, and also shows the genotypes of male subjects analysed in a previous report.[39]

Analytical Methods

All chemical reagents were of the highest grade available. Dextromethorphan, DOR, MM, HM and the internal standard, levallorphan, were purchased from Sigma-Aldrich Quimica SA (Madrid, Spain). Aliquots of urine and plasma were assayed for dextromethorphan, DOR, MM and HM, using a previously reported method[44] based on solid-liquid extraction with Bond Elut Certify™ (Varian, Palo Alto, CA, USA) columns and analysis by high-performance liquid chromatography and fluorescence detection. Plasma MDMA was measured by gas chromatography-mass spectrometry.[45]

Data Analysis

The values of the maximum plasma concentrations (C_{max}), time to reach C_{max} (t_{max}), elimination half-life ($t_{½}$), elimination rate constant (k_e), area under the plasma concentration-time curve (AUC) from 0 to 8 hours (AUC_8) and from time zero to infinity (AUC_∞) were derived directly from the plasma concentration-time profiles of dextromethorphan, DOR, MM and HM. Dextromethorphan plasma clearance (CL), as the ratio between the total administered dose and AUC_∞, and volume of distribution (V_d) were also calculated. AUC values from time zero to time t (AUC_t) were determined from 0 to 8 hours (session 1 data) and from 4 to 12 hours (session 2 data), using the trapezoidal rule. AUC_8 was extrapolated to AUC_∞ by adding the last quantifiable concentration divided by k_e. k_e values were estimated by log-linear regression of terminal data points. Inhibition constant (K_i) values in both sexes were calculated taking into account MDMA plasma concentrations. All aforementioned pharmacokinetic parameters were determined using pharmacokinetic functions for Microsoft Excel (Microsoft Corporation, Redmond, CA, USA).[46]

Statistically significant differences in pharmacokinetic parameters of dextromethorphan and its metabolites during the two sessions were assessed with a paired Student's t-test (C_{max}, AUC and $t_{½}$) and the Wilcoxon signed-rank test (t_{max}). All these parameters were compared to those of a study previously reported,[39] performed in male subjects with the same experimental design. Differences were considered to be significant at $p < 0.05$. All results are expressed as mean ± SD.

The MR of dextromethorphan/DOR was determined in order to explore the recovery of the activity of CYP2D6 in male and female subjects after MDMA intake. The fold change in the MR at the mean of each urine sample period in session 2 relative to the MR value in session 1 was plotted against time after MDMA administration for each subject. An inhibitory effect sigmoid maximum effect (E_{max}) model was fitted for the individual profiles by nonlinear regression (WinNonlin version 4.1; Pharsight Corporation, Mountain View, CA, USA) with suitable weighting of the MR in the respective urine sampling interval to estimate a value of CYP2D6 recovery (equation 1):

$$E = E_{max}\left[1 - \frac{t^{\gamma}}{t_{50}^{\gamma} + t^{\gamma}}\right] \quad \text{(Eq. 1)}$$

where E is the fold change in urinary MR after MDMA administration expressed as the ratio of MR in the inhibited state to the MR in the uninhibited control state, i.e. MR_i/MR_c, at the midpoint of the interval of time; t is time (in hours), E_{max} is the value of E at $t = 0$, t_{50} is the time (in hours) required to recover 50% of the activity of CYP2D6 (or CYP2D6 recovery half-life) and γ is the steepness of the sigmoid curve. The first-order rate constant for CYP2D6 recovery (k_{CYP}) was obtained as (equation 2):

$$k_{CYP} = \frac{0.693}{t_{50}} \quad \text{(Eq. 2)}$$

© 2011 Adis Data Information BV. All rights reserved.

Urinary results were also compared to the data from the report previously mentioned.[39] Micromoles of dextromethorphan, its metabolites recovered and MR were evaluated by multiple-way ANOVA for each time period. In this way, urinary differences between women and men were studied before and after the inhibition, taking into account the recovery of CYP2D6 activity. CYP3A4 activity *in vivo* was assessed based on the determination of the urine metabolic molar ratio of dextromethorphan/MM in both sexes. K_i values were obtained following equation 3, which reflects the intensity of the inhibitory potential and offers a link between *in vitro* data obtained and *in vivo* pharmacokinetic observations:

$$K_i = \frac{I_{max,u}}{R - 1} \quad \text{(Eq. 3)}$$

The R factor is the ratio between the AUC of the substrate co-administered with the inhibitor, i.e. MDMA, and the AUC of the substrate in the uninhibited control state (AUC_i/AUC_c) of dextromethorphan, and $I_{max,u}$ refers to the maximum concentration of unbound (free) MDMA. MDMA binding to plasma protein is considered to be low[47] (less than 20%).

Because the *in vivo* exposure ([I]) can be measured, these terms are combined to yield a predictive estimate of systemic substrate clearance inhibition *in vivo*, usually designated as the AUC_i/AUC_c. In the most general form, the equation that expresses such inhibition is equation 4:

$$1 + \frac{[I]}{K_i} = \frac{AUC_i}{AUC_c} \quad \text{(Eq. 4)}$$

Results

Table II summarizes pharmacokinetic parameters for dextromethorphan, DOR, MM and HM in females. After MDMA (session 2) an increase in dextromethorphan plasma concentrations was observed, with a significant effect on C_{max} and AUC_8 (increased approximately 10-fold), compared to control (session 1). An equivalent decrease in C_{max} and AUC_8 in session 2 compared to session 1 for DOR and HM was observed. Significant differences in $t_{½}$ for both metabolites were observed between both sessions as well as significant differences in t_{max} for DOR. Plasma concentrations of MM were not significantly changed by MDMA. The mean C_{max} of MDMA was 188.8 ± 16.7 ng/mL with a t_{max} of 2.0 ± 0.4 hours and AUC from 0 to 25 hours of 2645.2 ± 170.9 mg • h/mL. The MDMA single dose of 1.5 mg/kg produced the typical effects described for this substance in an experimental laboratory setting[10] and no adverse effects were reported. Bodyweight- and dose-normalized mean plasma concentrations of dextromethorphan, DOR, MM and HM before and after a 1.5 mg/kg dose of MDMA from both sexes are shown in figure 1. Data analysis between women and men revealed that there were no statistical differences in C_{max} and $t_{½}$ for any of the metabolites during the two sessions. However, there were significant between-sexes differences in t_{max} for dextromethorphan ($p = 0.01$), DOR ($p = 0.005$) and HM ($p = 0.033$) in session 2. K_i values revealed a higher value in male compared to female subjects (185.0 ± 67.6 vs 175.0 ± 34.5 ng/mL).

Table II. Dextromethorphan (DEX) and metabolite pharmacokinetics before (session 1) and after (session 2) a 1.5 mg/kg dose of 3,4-methylenedioxymethamphetamine (MDMA; ecstasy) [n = 12][a]

Parameter	DEX		DOR		MM		HM	
	session 1	session 2	session 1	session 2	session 1	session 2	session 1	session 2
C_{max} (μg/L)	3.8 ± 4.5	31.5 ± 14.1*	514.0 ± 408.5	35.9 ± 15.2*	2.0 ± 1.7	7.2 ± 12.4	205.3 ± 93.6	26.3 ± 8.7*
t_{max} (h)[b]	2 [0.5–4]	6 [3.9–8.1]	2 [1.1–2.9]	4 [2.8–5.2]**	2 [0.5–4.2]	8 [5.4–10.6]	3 [1.7–4.3]	5 [3–7]
k_e (h^{-1})	0.2 ± 0.2	0.2 ± 0.1	0.3 ± 0.1	0.1 ± 0.1	0.2 ± 0.1	0.3 ± 0.4	0.3 ± 0.1	0.2 ± 0.3
AUC_8 (μg • h/L)	15.8 ± 26.3	145.1 ± 48.3*	1991.9 ± 1102.0	191.1 ± 89.8*	9.2 ± 8.6	26.0 ± 34.7	966.0 ± 439.8	128.0 ± 48.8*
$t_{½}$ (h)	1.7 ± 0.4	12.1 ± 12.5	2.6 ± 1.3	13.4 ± 12.6*	5.2 ± 3.9	6.3 ± 3.8	3.0 ± 1.2	16.7 ± 24.4*
CL (L/h)	3587.4 ± 3797.4	119.4 ± 66.2*	NA	NA	NA	NA	NA	NA
V_d (mL/kg)	12 967.2 ± 4964.5	1322.2 ± 1145.9*	NA	NA	NA	NA	NA	NA
AUC_∞ (μg • h/L)	29.1 ± 37.1	397.0 ± 333.1	NA	NA	NA	NA	NA	NA

a Values are expressed as mean ± SD unless specified otherwise.

b Median [range].

AUC = area under the plasma concentration-time curve; **AUC_8** = AUC from 0 to 8 hours; **AUC_∞** = AUC from time zero to infinity; **CL** = plasma clearance; **C_{max}** = maximum plasma concentration; **DOR** = dextrorphan; **HM** = hydroxymorphinan-3-ol; **k_e** = elimination rate constant; **MM** = 3-methoximorphinan; **NA** = not applicable; **$t_{½}$** = elimination half-life; **t_{max}** = time to reach C_{max}; **V_d** = volume of distribution; * $p < 0.05$ (by paired Student's t-test), ** $p = 0.008$ (by the Wilcoxon exact test for nonparametric values) vs session 1.

© 2011 Adis Data Information BV. All rights reserved.

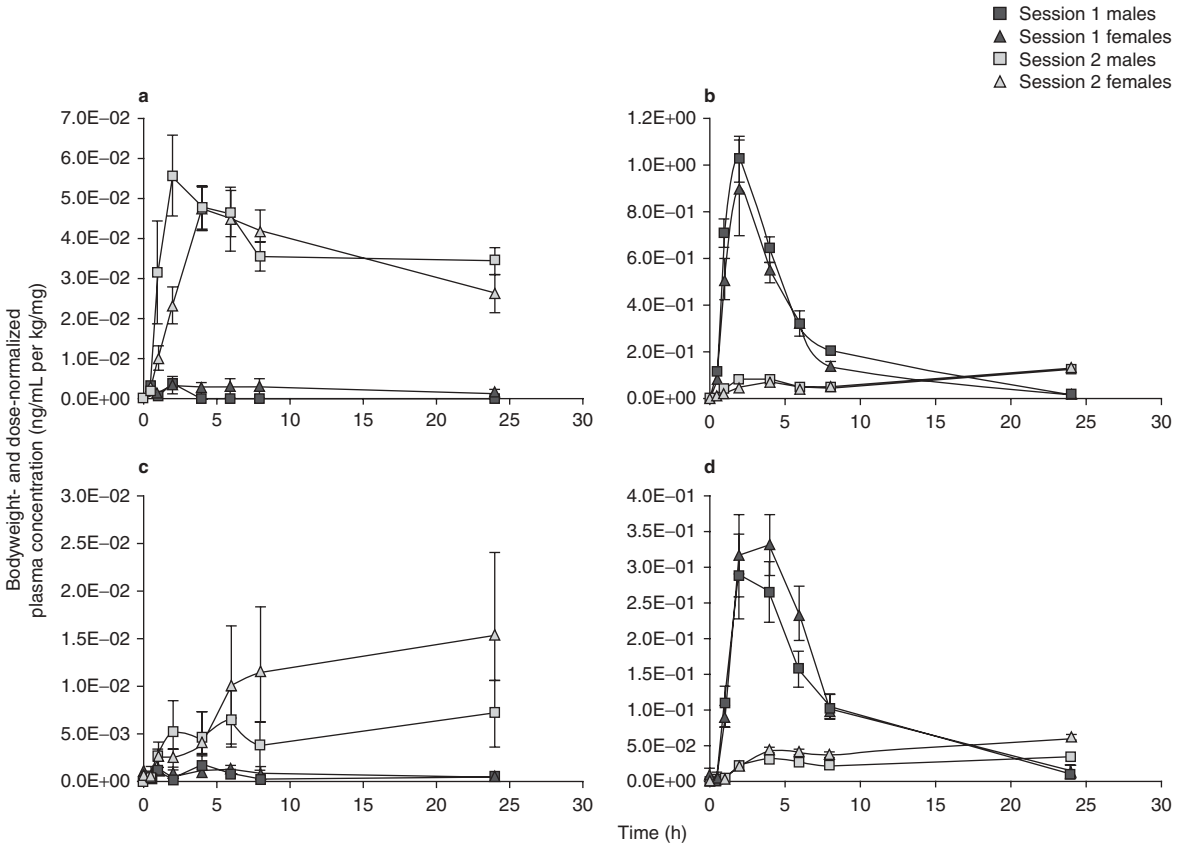

Fig. 1. Bodyweight- and dose-normalized mean (± standard error) plasma concentrations of (**a**) dextromethorphan; (**b**) dextrorphan; (**c**) 3-methoxymorphinan; and (**d**) hydroxymorphinan-3-ol in women (n = 12) and in men (n = 15)[39] before (session 1) and after (session 2) a 1.5 mg/kg dose of 3,4-methylenedioxymethamphetamine (MDMA; ecstasy).

Table III summarizes the urinary recoveries of dextromethorphan and its metabolites during the 0–8-hour collection period before and after MDMA intake in women. The urinary recovery of the dextromethorphan 30 mg dose as dextromethorphan and its main metabolites was 25.4 ± 8.9% with no MDMA pretreatment versus 6.6 ± 1.1% after MDMA 1.5 mg/kg ($p = 0.0001$). The dextromethorphan/DOR MR increased almost 60-fold from 0.018 ± 0.028 (session 1) to 0.998 ± 0.932 (session 2) in women. Figure 2 shows that after MDMA administration, 100% of the female subjects had a value greater than the antimode of 0.3 that signified the PM phenotype, while this phenocopying phenomenon was observed in only 67% of male subjects.[20,39] There were significant between-sexes differences in the rate of phenocopying after MDMA administration (χ^2 $p < 0.01$). A significant correlation was observed between MR and C_{max}(dextromethorphan)/C_{max}(DOR) [$r = 0.97$; $p = 0.0001$] in session 1 but this was marginal in session 2 [$r = 0.53$; $p = 0.073$].

The time course of CYP2D6 activity recovery in both sexes, represented by log (MR) after MDMA and the sigmoid adjustment applied, are shown in figure 3a. The excretion of dextromethorphan and DOR showed significant between-sexes differences in MR at 0–8 hours (session 1) and 4–12 hours (session 2) [$p = 0.032$ and $p = 0.01$, respectively]. Comparing all

Table III. Urinary recovery [0–8 h] of dextromethorphan (DEX) and its metabolites before (session 1) and after (session 2) a 1.5 mg/kg dose of 3,4-methylenedioxymethamphetamine (MDMA; ecstasy)[a]

Compound	Urinary excretion [0–8 h] (μmol)		Urinary recovery [0–8 h] (%)[b]	
	session 1	session 2	session 1	session 2
DEX	0.2 ± 0.2	2.4 ± 1.7*	0.2 ± 0.2	2.1 ± 1.6*
DOR	18.7 ± 8.7	2.8 ± 1.6*	16.9 ± 8.7	2.6 ± 1.6*
MM	0.1 ± 0.1	0.3 ± 0.1	0.1 ± 0.1	0.3 ± 0.1*
HM	9.0 ± 3.6	1.8 ± 1.2*	8.2 ± 3.6	1.7 ± 1.2*

a Values are expressed as mean ± SD.

b Recoveries (0–8 h) are expressed as a percentage of the administered dose.

DOR = dextrorphan; **HM** = hydroxymorphinan-3-ol; **MM** = 3-methoxymorphinan; * $p < 0.05$ vs session 1.

© 2011 Adis Data Information BV. All rights reserved.

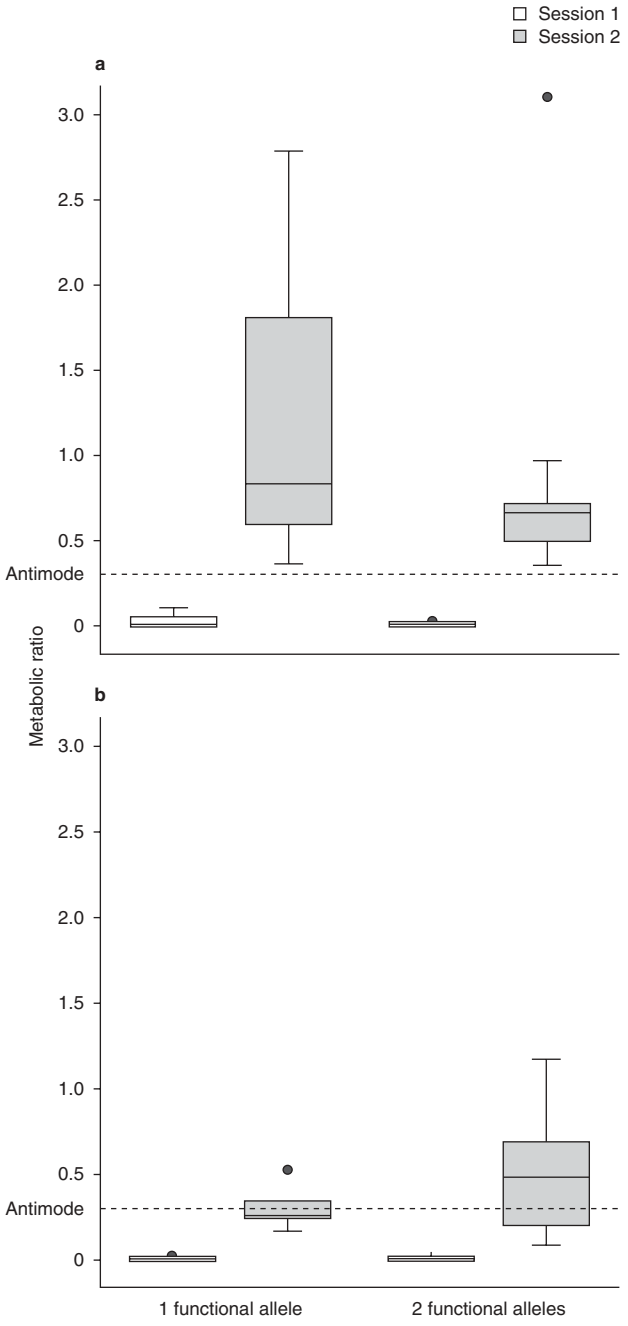

Fig. 2. Urinary dextromethorphan/dextrorphan metabolic ratio changes (**a**) in women and (**b**) in men[39] before (session 1) and after (session 2) a single recreational dose of 3,4-methylenedioxymethamphetamine (MDMA; ecstasy), taking the antimode value (0.3) as a reference. A comparison was made between women (n = 12) and men (n = 15)[39] to further study this change depending on the genotype of the subjects. The box represents the median and the 25th and 75th percentiles of the data, the whiskers represent the range and the black circles represent the outliers.

metabolites separately between females and males, dextromethorphan excretion in session 1 is not statistically significant (p = 0.526). However, DOR, MM and HM concentrations in these time periods are close to statistical significance (p = 0.08, p = 0.08 and p = 0.05, respectively) but reach significance (p = 0.039, p = 0.013 and p = 0.044, respectively) when CYP2D6 activity (240–248 hours) recovered almost to baseline values.

The predicted values generated by the sigmoid fitting correlate well (r = 0.91 ± 0.14) with the observed values and the plot of residuals against time showed a randomly scattered distribution. CYP2D6 activity recovered after 10 days, with a half-life of 36.6 ± 22.9 hours. k_{CYP} was 0.0293 ± 0.0198 h^{-1}. Men showed a shorter recovery half-life (27.6 ± 25.1 hours) and therefore higher k_{CYP} values (0.0627 ± 0.0754 h^{-1}). E_{max} values were 151.4 ± 188.1 ng/mL and 198.3 ± 198.1 ng/mL in females and males, respectively.

Significant higher values of the steepness of sigmoid curve were observed in women versus men (2.61 ± 1.00 vs 1.86 ± 0.68,

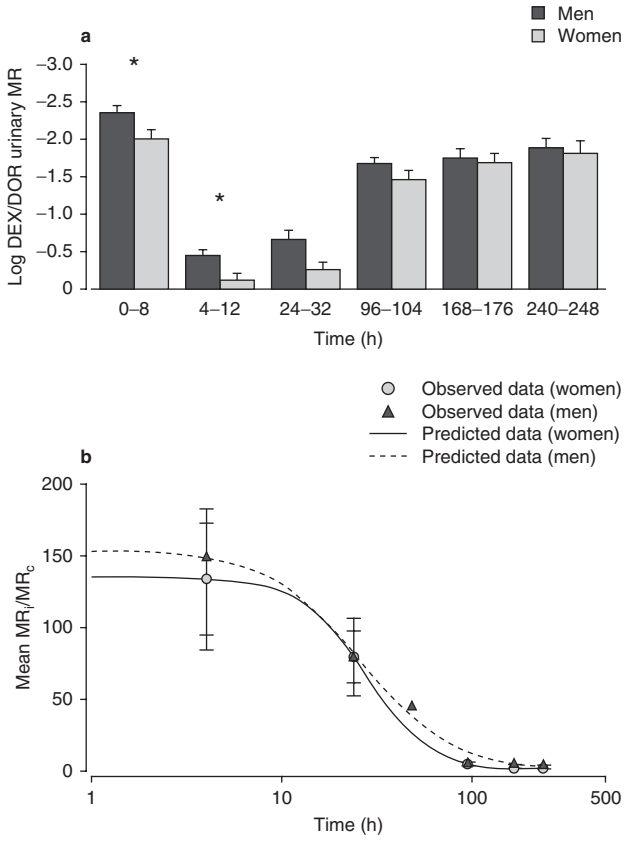

Fig. 3. (**a**) Recovery of cytochrome P450 2D6 activity over time following a 1.5 mg/kg oral dose of 3,4-methylenedioxymethamphetamine (MDMA; ecstasy) as measured by the log-transformed dextromethorphan (DEX)/dextrorphan (DOR) urinary metabolic ratio (MR) before (0–8 h) and after MDMA intake (all others periods of time) in women (n = 12) and in men (n = 15).[39] * p < 0.05 between females and males at the corresponding time periods. The error bars represent the standard error. (**b**) Data for women and men, fitting to the sigmoid model. **MR_c** = MR in the uninhibited (control) state; **MR_i** = MR in the inhibited state.

© 2011 Adis Data Information BV. All rights reserved.

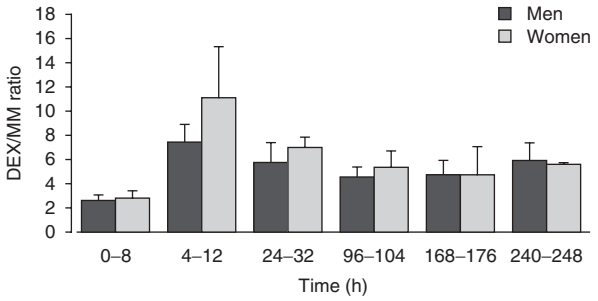

Fig. 4. Cytochrome P450 3A4 activity measured by the relationship between urinary excretion of dextromethorphan (DEX) and methoxymorphinan (MM) in women (n = 12) and in men (n = 15).[39] The basal period of time (0–8 h) was compared to all other periods after the inhibition. The error bars represent the standard error.

respectively; $p < 0.05$). Figure 3b shows the adjustment between observed and predicted data points in the sigmoid model.

Figure 4 displays CYP3A4 activity before (0–8 hours) and after (all other collection periods) the administration of MDMA in both sexes. The data show an increase in C_{max} and AUC values of MM after drug intake that are not significantly different from baseline levels. The urinary data reflect significant differences in dextromethorphan/MM ratios after MDMA intake compared to the control situation in both sexes, although it was more pronounced in women. Dextromethorphan/MM ratios increased almost 3-fold from 0.311 ± 0.310 to 0.860 ± 0.382 after MDMA administration in women and there was a 2.5-fold-increase in men (from 0.325 ± 0.283 to 0.751 ± 0.331). There were no between-sexes differences in CYP3A4 activity before or after MDMA administration. The measurement of CYP3A4 activity indicated that when CYP2D6 is impaired, CYP3A4 activity is apparently decreased in both sexes.

Discussion

In the present study we report, for the first time, MDMA-induced CYP2D6 inhibition as well as enzyme activity recovery over time in women, using dextromethorphan as probe drug to monitor CYP2D6 activity.

A single recreational dose of MDMA caused a dramatic increase in the systemic exposure to dextromethorphan with the simultaneous decrease in plasma concentrations of its primary metabolite, DOR. Since no statistically significant differences in dextromethorphan t_{max} were observed, differences in dextromethorphan pharmacokinetics were likely due to metabolic changes and not to absorption and distribution processes. This was also reflected in a marked decrease in dextromethorphan metabolism seen in the urinary recovery of dextromethorphan, but also in dextromethorphan/DOR MR, which had not fully recovered to its basal level after 10 days of MDMA intake. These data, compatible with a potent MBI of CYP2D6, agree with studies previously published.[36,37,39] CYP2D6 inhibition is translated in a rapid phenocopying to apparent PM status, meaning a poor enzyme capability to metabolize dextromethorphan to dextrorphan after a single dose of MDMA, explaining the inability to relate acute toxicity to the *CYP2D6* genotype.[48]

Regarding DOR, HM and MM, several pharmacokinetic parameters were altered after dextromethorphan administration following MDMA pretreatment in women. Changes in DOR plasma concentrations are easily explained because the *O*-demethylation pathway from dextromethorphan to DOR is regulated by CYP2D6. Consequently, HM plasma concentrations were altered as its formation is mainly dependent on DOR formation. Plasma concentrations of MM were not significantly changed by pretreatment with MDMA because its formation is regulated by CYP isoenzymes other than CYP2D6.

There are significant differences between both sexes in CYP2D6 activity *in vivo* using dextromethorphan as probe drug at baseline and these sex-related differences persisted after CYP2D6 inhibition. At baseline level, urinary MRs differ between sexes because of a higher recovery of dextromethorphan in women. Figure 2 shows that once CYP2D6 is inhibited, MR in women displays values that are still higher than those of men. Moreover, women show a larger variability in MR values after MDMA administration compared to men, in particular in subjects displaying one functional allele. However, a higher-fold increase in MR in men than in women after MDMA intake is observed, and therefore, a stronger inhibition is observed. Calculated K_i values also corroborate this phenomenon. All subjects included in this study were confirmed to be EMs on the basis of a dextromethorphan phenotyping test. Beyond this phenotype, certain heterogeneity in the genotype associated with it is expected (table I). However, between-sexes differences observed in dextromethorphan disposition are not attributable to a dissimilar distribution of *CYP2D6* genotypes as their distribution among subjects was quite the same (one-third of the subjects had one functional allele and two-thirds had two functional alleles in both sexes). The sigmoid inhibitory model used to analyse the data provided a good fit for the fold increase at $t = 0$ when CYP2D6 is inhibited by MDMA, showing the potent inhibitory effect on the metabolism of dextromethorphan (figure 3b).

The time course of CYP2D6 recovery shows no significant between-sexes differences in terms of the t_{50}. Values estimated in men (27.6 ± 25.1 hours) are similar to a corrected mean recovery half-life value (37.4 ± 11.6 hours) previously reported[39]

© 2011 Adis Data Information BV. All rights reserved.

after modelling a monoexponential decay of the MR over time. The sigmoid model applied in the present report shows that despite the absence of between-sexes differences in the E_{max} and t_{50} values, a significant effect on the sigmoidicity of the fitting is observed. This sigmoidicity factor (γ) greater than 1 indicates a steeper recovery of CYP2D6 that is significantly more pronounced in women than in men. Between-sexes differences in *de novo* CYP2D6 protein synthesis after MBI of the isoenzyme could be the cause of these differences observed in the steepness of the sigmoid curves, but further studies and different models are required to explain the more pronounced shape of functional recovery of CYP2D6 in women.

Observing the data in figure 3 and assuming that CYP2D6 recovers 90% of full activity within 3–4 half-lives, this value seems to underestimate the recovery because CYP2D6 activity has not recovered fully even after 10 days.

The clinical implication of MDMA-induced MBI of CYP2D6 activity is that within 2 hours, subjects display the PM phenotype after drug intake irrespective of their original genotype. Therefore, recreational MDMA users are exposed to a higher probability of relative overdose and, therefore, an increased risk of suffering adverse effects from CYP2D6 substrates including MDMA (repeated doses in the same session). CYP2D6-impaired metabolic capacity would be a contributory factor to drug side effects and CYP2D6 EMs are not protected against adverse events.[49] CYP2D6 substrates include tricyclic antidepressants (amitriptyline, clomipramine, desipramine), serotonin reuptake inhibitor antidepressants (citalopram, fluoxetine, paroxetine), amphetamine-like compounds (methamphetamine), antipsychotics (haloperidol, perphenazine), antiarrhythmics (aprindine), antiemetics (dolasetron, ondansetron), β-adrenoceptor antagonists (alprenolol, metropolol) and opioids (codeine, tramadol).[20,50] MDMA users display a higher prevalence of psychopathology particularly of mood disorders when compared to non-drug users.[51,52] It is in this context that the MDMA-induced MBI of CYP2D6 is of relevance, and physicians should be advised to prescribe medications whose metabolic disposition is not regulated by CYP2D6.

The dextromethorphan/DOR urinary MR is sensitive to changes in the renal clearance of dextromethorphan, for example, due to urine pH-dependent excretion.[53,54] However, we did not detect any influence of urinary pH or renal function. The homogeneity of the population involved in the study, in terms of their dietary and life-style habits, may explain this observation.

Plasma concentration profiles, pharmacokinetic parameters and urinary recovery of dextromethorphan and metabolites in women in the absence of pretreatment with MDMA are in agreement with those described in previous studies.[54,55] Phenotyping of 15 men and 12 women with dextromethorphan in the current study suggests that CYP2D6 activity might be greater in men. These results are consistent with some previous studies.[54-57] In contrast, other groups found either no difference[57,58] or decreased CYP2D6 activity in men compared to women.[48,59]

CYP3A4 is partially involved in the *O*-demethylenation of MDMA and MDA to HHMA and HHA, respectively. Even considering the minor contribution of CYP3A4 to metabolic disposition of MDMA, its activity was monitored because other enzymes may become more important once CYP2D6 is inhibited. Segura et al.[60] showed there was a conversion from MDMA to HHMA *in vivo*, despite the CYP2D6 inhibition by paroxetine suggesting alternative metabolic pathways. *In vitro* assays demonstrated that other enzymes such as CYP1A2 and, to a lower extent, CYP2B6 and CYP3A4, may participate in the conversion from MDMA to HHMA.[18] CYP3A4 is also involved in the metabolism of dextromethorphan and its activity can be monitored with the MR of dextromethorphan/MM. However, CYP2D6 and CYP3A4 metabolic activities are not strictly independent from each other.[49] Comparing CYP2D6 and CYP3A4 metabolic activity, it has been shown that while dextromethorphan levels increase almost 10-fold once MDMA is administered, DOR metabolite levels decrease dramatically 10-fold, reflecting a potent metabolic inhibition not shown in the CYP3A4-regulated pathway.

Conclusion

In this study carried out in 12 female subjects, pretreatment with MDMA resulted in an alteration of dextromethorphan metabolism, which was characterized by an increase in dextromethorphan plasma concentrations and a decrease in dextromethorphan clearance. These results were compared with a previous study in male subjects. Between-sexes differences in CYP2D6 activity in basal conditions (dextromethorphan MR) were observed; after MDMA intake and CYP2D6 inhibition, these differences persisted for some hours but afterwards CYP2D6 recovery followed a similar time pattern and reached baseline activity 10 days after MDMA exposure. These results contribute to a better understanding of CYP2D6 activity in both sexes as well as the clinical implications associated with these pharmacokinetic changes.

Acknowledgements

Authorship credit should be equally distributed among the authors, independently of the order in which they are listed.

© 2011 Adis Data Information BV. All rights reserved.

This study was supported by the US National Institute on Drug Abuse (NIDA; grant no. 5R01BA017987-01), the Spanish Ministry of Education and Science (grant no. MICINN FI09/00355), the Spanish Network on Addiction Disorders (FIS-RTA; grant no. RD06/0001/0026) and the Spanish Agency for Administration of University and Research Grants (AGAUR; grant no. 2009 SGR 718). All authors have no conflicts of interests that are directly relevant to the content of this study.

References

1. Parrott AC. Human psychopharmacology of ecstasy (MDMA): a review of 15 years of empirical research. Hum Psychopharmacol 2001; 16: 557-77
2. De la Torre R, Farré M, Roset PN, et al. Human pharmacology of MDMA. Ther Drug Monit 2004 Apr; 26 (2) 137-44
3. Mas M, Farré M, de la Torre R, et al. Cardiovascular and neuroendocrine effects and pharmacokinetics of 3,4-methylenedioxymethamphetamine in humans. J Pharmacol Exp Ther 1999 Jul; 290 (1): 136-45
4. Camí J, Farré M. Drug addiction. New Engl J Med 2003; 349: 975-86
5. Henry JA, Jeffreys KJ, Dawling S. Toxicity and deaths from 3,4-methylenedioxymethamphetamine ('ecstasy'). Lancet 1992 Aug; 340 (8816): 384-7
6. Capela JP, Carmo H, Remião F, et al. Molecular and cellular mechanisms of ecstasy-induced neurotoxicity: an overview. Mol Neurobiol 2009; 39: 210-71
7. Murphy PN, Wareing M, Fisk JE, et al. Executive working memory deficits in abstinent ecstasy/MDMA users: a critical review. Neuropsychobiology 2009 Nov; 60 (3-4): 159-75
8. Gudelsky GA, Yamamoto BK. Actions of 3,4-methylenedioxymethamphetamine (MDMA) on cerebral dopaminergic, serotonergic and cholinergic neurons. Pharmacol Biochem Behav 2008 Aug; 90 (2): 198-207
9. Baumann MH, Rothman RB. Neural and cardiac toxicities associated with 3,4-methylenedioxymethamphetamine (MDMA). Int Rev Neurobiol 2009; 88: 257-96
10. de la Torre R, Farré M, Roset PN, et al. Pharmacology of MDMA in humans. Ann N Y Acad Sci 2000; 914: 225-37
11. Segura M, Ortuño J, Farré M, et al. 3,4-Dihydroxymethamphetamine (HHMA): a major in vivo MDMA metabolite in humans. Chem Res Toxicol 2001; 14: 1203-8
12. Easton N, Fry J, O'Shea E, et al. Synthesis, in vitro formation, and behavioural effects of glutathione regioisomers of alpha-methyldopamine with relevance to MDA and MDMA (ecstasy). Brain Res 2003; 987 (2): 144-54
13. 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 Aug; 254: 521-7
14. Perfetti X, O'Mathúna B, Pizarro N, et al. Neurotoxic thioether adducts of 3,4-methylenedioxymethamphetamine identified in human urine after ecstasy ingestion. Drug Metab Dispos 2009 Jul; 37 (7): 1448-55
15. Capela JP, Macedo C, Branco PS, et al. Neurotoxicity mechanisms of thioether ecstasy metabolites. Neuroscience 2007 Jun; 146 (4): 1743-57
16. Monks TJ, Jones DC, Bai F, et al. The role of metabolism in 3,4-(+)-methylenedioxyamphetamine and 3,4-(+)-methylenedioxymethamphetamine (ecstasy) toxicity. Ther Drug Monit 2004 Apr; 26 (2): 132-6
17. Tucker GT, Lennard MS, Ellis SW, et al. The demethylenation of the methylenedioxymethamphetamine ('ecstasy') by cytochrome P4502D6 by human debrisoquine hydroxylase (CYP2D6). Biochem Pharmacol 1994; 47 (7): 1151-6
18. 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 Jun; 59 (12): 1563-71
19. Meyer MR, Peters FT, Maurer HH. The role of human hepatic cytochrome P450 isozymes in the metabolism of racemic 3,4-methylenedioxy-methamphetamine and its enantiomers. Drug Metab Dispos 2008 Nov; 36 (11): 2345-54
20. Zanger UM, Raimundo S, Eichelbaum M. Cytochrome P450 2D6: overview and update on pharmacology, genetics, biochemistry. Naunyn Schmiedebergs Arch Pharmacol 2004; 369: 23-37
21. Bogni A, Monshouwer M, Moscone A, et al. Sibstrate specific metabolism by polymorphinc cythochrome P450 2D6 alleles. Toxicol In Vitro 2005; 19: 621-9
22. Zhou SF. Polymorphism of human cytochrome P450 2D6 and its clinical significance: part I. Clin Pharmacokinet 2009; 48 (11): 689-723
23. McElroy S, Sachse C, Brockmoller J, et al. CYP2D6 genotyping as an alternative to phenotyping for determination of metabolic status in a clinical trial setting. AAPS PharmSci 2000; 2 (4): E33
24. Woktczak A, Rychlik-Sych M, Krochmalska-Ulacha E, et al. CYP2D6 phenotyping with dextormethorphan. Pharmacol Rep 2007; 59: 734-8
25. Yu A, Haining RL. Comparative contribution to dextromethorphan metabolism by cytochrome P450 isoforms in vitro: can dextromethorphan be used as a dual probe for both CTP2D6 and CYP3A activities? Drug Metab Dispos 2001; 29: 1514-20
26. Frank D, Jaehde U, Fuhr U. Evaluation of probe drugs and pharmacokinetic metrics for CYP2D6 phenotyping. Eur J Clin Pharmacol 2007; 63: 321-33
27. Spatzenegger M, Jaeger W. Clinical importance of hepatic cytochrome P450 in drug metabolism. Drug Metabol Rev 1995; 27 (3): 397-417
28. Ducharme J, Abdullah S, Wainer IW. Dextromethorphan as an in vivo probe for the simultaneous determination of CYP2D6 and CYP3A activity. J Chromatogr B Biomed Appl 1996 Mar; 678 (1): 113-28
29. Jacqz-Aigrain E, Funck-Brentano C, Cresteil T. CYP2D6- and CYP3A-dependent metabolism of dextromethorphan in humans. Pharmacogenetics 1993 Aug; 3 (4): 197-204
30. Chladek J, Zimova G, Beranek M, et al. In-vivo indices of CYP2D6 activity: comparison of dextromethorphan metabolic ratios in 4-h urine and 3-h plasma. Eur J Clin Pharmacol 2000; 56: 651-7
31. Greene SL, Dargan PI, O'Connor N, et al. Multiple toxicity from 3,4-methylenedioxymethamphetamine ('ecstasy'). Am J Emerg Med 2003 Mar; 21 (2): 121-4
32. Gilhooly TC, Daly AK. CYP2D6 deficiency, a factor in ecstasy related deaths? Br J Clin Pharmacol 2002; 54: 69-70
33. de la Torre R, Farré M, Ortuño J, et al. Non-linear pharmacokinetics of MDMA ('ecstasy') in humans. Br J Clin Pharmacol 2000 Feb; 49 (2): 104-9
34. Farré M, de la Torre R, Mathúna BO, et al. Repeated doses administration of MDMA in humans: pharmacological effects and pharmacokinetics. Psychopharmacology 2004; 173: 364-75
35. Baumann MH, Zolkowska D, Kim I, et al. Effects of dose and route of administration on pharmacokinetics of (±)-3,4-methylenedioxymethamphetamine in the rat. Drug Metab Dispos 2009 Nov; 37 (11): 2163-70
36. Yang J, Jamei M, Heydari A, et al. Implications of mechanism-based inhibition of CYP2D6 for the pharmacokinetics and toxicity of MDMA. J Psychopharmacol 2006; 20: 842-9
37. de la Torre R, Farré M. Neurotoxicity of MDMA (ecstasy): the limitations of scaling from animals to humans. Trends Pharmacol Sci 2004 Oct; 25 (10): 505-8
38. Delaforge M, Jaouen M, Bouielle G. Inhibitory metabolite complex formation of methylenedioxymethamphetamine with rat and human cytochrome P450: particular involvement of CYP 2D. Environ Toxicol Pharmacol 1999; 7: 153-8
39. Mathúna OB, Farré M, Rostami-Hodjegan A, et al. The consequences of 3,4-methylenedioxymethamphetamine induced CYP2D6 inhibition in humans. J Clin Psychopharmacol 2008; 28: 525-31
40. Kolbrich EA, Goodwin RS, Gorelick DA, et al. Plasma pharmacokinetics of 3,4-methylenedioxymethamphetamine after controlled oral administration to young adults. Ther Drug Monit 2008 Jun; 30 (3): 320-32
41. Torrens M, Serrano D, Astals M, et al. Diagnosing comorbid psychiatric disorders in substance abusers: validity of the Spanish versions of the Psychiatric Research Interview for Substance and Mental Disorders and the Structured Clinical interview for DSM-IV. Am J Psychiatry 2004; 161: 1231-7

© 2011 Adis Data Information BV. All rights reserved.

42. Schmid B, Bircher J, Preisig R, et al. Polymorphic dextromethorphan metabolism: co-segregation of oxidative O-demethylation with debrisoquin hydroxylation. Clin Pharmacol Ther 1985; 38: 618-24

43. Parrott AC. Is ecstasy MDMA? A review of the proportion of ecstasy tablets containing MDMA, their dosage levels, and the changing perceptions of purity. Psychopharmacology 2004; 173: 234-41

44. de la Torre R, Farré M, Mathúna BO, et al. MDMA (ecstasy) pharmacokinetics in a CYP2D6 poor metaboliser and nine CYP2D6 extensive metabolisers. Eur J Clin Pharmacol 2005; 61: 551-4

45. Pizarro N, Ortuno J, Farré M, et al. Determination of MDMA and its metabolites in blood and urine by gas chromatography-mass spectrometry and analysis of enantiomers by capillary electrophoresis. J Anal Toxicol 2002; 26: 157-65

46. Usansky JI, Desai A, Tang-Liu D. Pharmacokinetic and pharmacokinetic resources: pharmacokinetic software. PK functions for Microsoft Excel [online]. Available from URL: http://www.pharmpk.com/soft.html [Accessed 2011 Feb 7]

47. de la Torre R, Farré M, Navarro M, et al. Clinical pharmacokinetics of amfetamine and related substances: monitoring in conventional and non-conventional matrices. Clin Pharmacokinet 2004; 43 (3): 157-85

48. Labbe L, Sirois C, Pilote S, et al. Effect of gender, sex hormones, time variables and physiological urinary pH on apparent CYP2D6 activity as assessed by metabolic ratios of marker substrates. Pharmacogenetics 2000; 10: 425-38

49. Funck-Brentano C, Boëlle PY, Verstuyft C, et al. Measurement of CYP2D6 and CYP3A4 activity in vivo with dextromethorphan: sources of variability and predictors of adverse effects in 419 healthy subjects. Eur J Clin Pharmacol 2005; 61: 821-9

50. Kirchheiner J, Seeringer A. Clinical implications of pharmacogenetics of cytochrome P450 drug metabolizing enzymes. Biochim Biophys Acta 2007; 1770: 489-94

51. Soar K, Turner JJ, Parrott AC. Problematic versus non-problematic ecstasy/MDMA use: the influence of drug usage patterns and pre-existing psychiatric factors. J Psychopharmacol 2006; 20: 417-24

52. Martín-Santos R, Torrens M, Poudevida S, et al. 5-HTTLPR polymorphism, mood disorders and MDMA use in a 3-year follow-up study. Addict Biol 2010 Jan; 15 (1): 15-22

53. Ozdemir M, Crewe KH, Tucker GT, et al. Assessment of in vivo CYP2D6 activity: differential sensitivity of commonly used probes to urine pH. J Clin Pharmacol 2004; 44: 1398-404

54. McCune JS, Lindley C, Decker JL, et al. Lack of gender differences and large intrasubject variability in cytochrome P450 activity measured by phenotyping with dextromethorphan. J Clin Pharmacol 2001; 41: 723-31

55. Kashuba AD, Nafziger AN, Kearns GL, et al. Quantification of intraindividual variability and the influence of menstrual cycle phase on CYP2D6 activity as measured by dextromethorphan phenotyping. Pharmacogenetics 1998 Oct; 8 (5): 403-10

56. Franconi F, Brunelleschi S, Steardo L, et al. Gender differences in drug responses. Pharmacol Res 2007 Feb; 55 (2): 81-95

57. Schwartz JB. The influence of sex on pharmacokinetics. Clin Pharmacokinet 2003; 42 (2): 107-21

58. Anderson GD. Gender differences in pharmacological response. Int Rev Neurobiol 2008; 83: 1-10

59. Hägg S, Spigset O, Dahlqvist R. Influence of gender and oral contraceptives on CYP2D6 and CYP2C18 activity in healthy volunteers. Br J Clin Pharmacol 2001 Feb; 51 (2): 169-73

60. Segura M, Farré M, Pichini S, et al. Contribution of cytochrome P450 2D6 to 3,4-methylenedioxymethamphetamine disposition in humans. Clin Pharmacokinet 2005; 44 (6): 649-60

Correspondence: Dr *Rafael de la Torre*, Human Pharmacology and Clinical Neurosciences Research Group, Neuropsychopharmacology Program, IMIM-Hospital del Mar Research Institute, Parc de Recerca Biomèdica de Barcelona, Doctor Aiguader, 88, 08003 Barcelona, Spain.
E-mail: rtorre@imim.es

© 2011 Adis Data Information BV. All rights reserved.