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Population Pharmacokinetics of Caffeine and its Metabolites Theobromine, Paraxanthine and Theophylline after Inhalation in Combination with Diacetylmorphine

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Abstract: The stimulant effect of caffeine, as an additive in diacetylmorphine preparations for study purposes, may interfere with the pharmacodynamic effects of diacetylmorphine. In order to obtain insight into the pharmacology of caffeine after inhalation in heroin users, the pharmacokinetics of caffeine and its dimethylxanthine metabolites were studied. The objectives were to establish the population pharmacokinetics under these exceptional circumstances and to compare the results to published data regarding intravenous and oral administration in healthy volunteers. Diacetylmorphine preparations containing 100 mg of caffeine were used by 10 persons by inhalation. Plasma concentrations of caffeine, theobromine, paraxanthine and theophylline were measured by high performance liquid chromatography. Non-linear mixed effects modelling was used to estimate population pharmacokinetic parameters. The model was evaluated by the jack-knife procedure. Caffeine was rapidly and effectively absorbed after inhalation. Population pharmacokinetics of caffeine and its dimethylxanthine metabolites could adequately and simultaneously be described by a linear multi-compartment model. The volume of distribution for the central compartment was estimated to be 45.7 l and the apparent elimination rate constant of caffeine at 8 hr after inhalation was 0.150 hr^{-1} for a typical individual. The bioavailability was approximately 60%. The presented model adequately describes the population pharmacokinetics of caffeine and its dimethylxanthine metabolites after inhalation of the caffeine sublimate of a 100 mg tablet. Validation proved the stability of the model. Pharmacokinetics of caffeine after inhalation and intravenous administration are to a large extent similar. The bioavailability of inhaled caffeine is approximately 60% in experienced smokers.

Caffeine is widely used for a range of clinical and non-clinical purposes. It is well known for its mild central nervous system stimulant effects (Sawynok 1995).

Caffeine is extensively metabolised into xanthine and uracil derivatives. Paraxanthine is quantitatively the main demethylation product, followed by theobromine and theophylline respectively. The pharmacokinetics and metabolism of caffeine after oral and intravenous administration have been extensively studied (Bonati *et al.* 1982; Lelo *et al.* 1986; Falcao *et al.* 1997).

Caffeine pharmacokinetics after inhalation, however, have never been described. Since caffeine is a common street heroin constituent, inhalation of caffeine vapours is common in heroin users who take the drug by "chasing the dragon" (carefully heating the drug on aluminium foil and inhaling the fumes through a straw) (Huizer 1987). Due to physicochemical effects of caffeine, the sublimation temperature of diacetylmorphine base is decreased, the recovery of diacetylmorphine is slightly enhanced and its pyrolytic decomposition is reduced (Huizer 1987). In the currently described clinical trial, caffeine was used as a constituent of legal diacetylmorphine (heroin) preparations. (Van den Brink *et al.* 2003) Results from this trial on the pharmacodynamics of inhaled diacetylmorphine were previously published by Hendriks *et al.* (2001). Evidently, caffeine also has pharmacodynamic effects, besides the physicochemical effects mentioned above. In a study with 24 male healthy volunteers by Brice & Smith (2002) behavioural effects such as increased alertness (mean: +11%, $P<0.001$) and anxiety (mean: +7%, $P<0.05$) were noted on a visual analogue rating scale, after 200 mg of orally administered caffeine. When caffeine is used in diacetylmorphine preparations for study purposes (Van den Brink *et al.* 2003), its stimulant action is a drawback, as it may interfere with the establishment of the pharmacodynamic effects of diacetylmorphine. Moreover, its dimethylxanthine metabolites have a stimulant effect as well, possibly resulting from the blockade of adenosine receptors (Mattila 1984). From this perspective it is important to obtain insight into the pharmacology of caffeine as an additive of diacetylmorphine, including its metabolites.

The aim of the current study was (1) to establish the

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population pharmacokinetics of caffeine and its metabolites theobromine, paraxanthine and theophylline, after inhalation in heroin users and (2) to make a comparison to published data concerning the pharmacokinetics after intravenous and oral administration in healthy volunteers.

Materials and Methods

Volunteers and medication. In The Netherlands, a large-scale randomised trial was conducted to investigate the effectiveness of medical co-prescription of heroin and methadone to chronic, treatment-resistant heroin users (Van den Brink *et al.* 2003). To investigate the pharmacodynamics of heroin after inhalation, a preliminary study was conducted prior to this trial, at the U-Gene Research Clinical Pharmacology Unit in Utrecht. The volunteers in the preliminary study were chronic, treatment-resistant heroin users, who used substantial amounts (at least 0.5 g per day) of street heroin by inhalation. All of them were treated with methadone during 6 months prior to the study.

On 5 consecutive days, one tablet per day was used by one of two inhalation methods. The tablets contained 25 mg, 50 mg or 100 mg of diacetylmorphine base and a fixed dose of 100 mg caffeine. The two different inhalation methods were (1) inhalation after heating the tablet on a piece of aluminium foil by a lighter ("chasing the dragon"), and (2) inhalation after heating the tablet on a heating plate (for details see: Hendriks *et al.* 2001).

During the study period, starting one day prior to the first dose, the volunteers were treated with their regular dose of methadone solution once daily. They also used their prescribed medication as usual. Besides the study medication, they were not allowed to use any other addictive substances, including alcohol, however, they were allowed to take coffee and cocoa containing foodstuffs. Coffee and cocoa intake were not registered. Therefore, structural criteria were defined in the analysis plan in order to avoid potential bias.

The trial was performed in accordance with the Dutch law and with the ICH and EU Guidelines for Good Clinical Practice. The protocol was reviewed by the Central Committee on Medical Ethics of The Netherlands (KEMO).

Sampling and bioanalysis. On each of the 5 consecutive days, 14 blood samples were taken. One sample was taken prior to the inhalation of diacetylmorphine and caffeine. The samples were taken at the following time points after the end of the inhalation: 1, 2, 5, 7.5, 10, 15, 22.5, 30, 45, 60, 120, 240, 480 min.

Plasma concentrations of caffeine and its dimethylxanthine metabolites were measured by a high performance liquid chromatographic method, adopted from Tanaka (1992). In brief, the compounds of interest were isolated from plasma by organic phase extraction. To a 300 µl plasma sample, 50 µl of internal standard (N-acetaminophen 90 mg/l) and 100 µl of hydrochloric acid (1 M) were added. The mixture was extracted with 6 ml dichloromethane/isopropanol (9:1 v/v) and the extract was evaporated to dryness. The residue was then reconstituted in 200 µl acetic acid (0.05% v/v). Caffeine and its metabolites were separated on a Symmetry C18 3.5 µm column (150 mm×4.6 mm) (Waters Corporation, Milford, USA) at 30°, with a mobile phase consisting of acetic acid (0.05% v/v)/methanol (80:20 v/v). The lower limit of quantification was 200 ng/ml for caffeine and 50 ng/ml for its metabolites. The performance of the assay was validated for the range of concentrations observed in this study. Accuracy and precision were 90%–110% and <10%, respectively.

Pharmacokinetic data analysis. All data were fitted simultaneously with the non linear mixed effects modelling (NONMEM) program (version V, level 1.1) (GloboMax LLC, Hanover, USA) (Beal & Sheiner 1998). The first-order conditional estimation (FOCE) method with interaction between interindividual, intraindividual and residual variability was applied. The subroutine ADVAN6 was used to define the pharmacokinetic model. PDx-Pop (version 1.1j release 4) (GloboMax LLC, Hanover, USA) was used as an interface for data and output processing and for modelling management.

One cup of tea contains about 30 mg of caffeine (Khokhar & Magnusdottir 2002), which corresponds to a rise of the caffeine level of approximately 500 ng/ml. This was therefore considered a legitimate limit for data exclusion. If a measured caffeine concentration, corresponding to a time point more than 30 min. after the beginning of the inhalation, was more than 500 ng/ml higher than the previous concentration, the concerning data course was indicated as resulting from coffee or tea intake and was therefore fully (when coffee was taken in the beginning of the course) or partially (when coffee was taken at the end of the course) excluded from data analysis. Unpredictably high concentrations of theobromine during the latter part of the course were doubted as these may have been a result of cocoa intake, since theobromine is a constituent of cocoa. As cocoa is an ingredient of many different food stuffs in differing quantities, a minimal increase in theobromine levels to be indicated as resulting from cocoa intake, is difficult to assess. Therefore, if the theobromine level rose more than 50% after the first hour of the course, the inconsistent concentration and the subsequent theobromine measurements were excluded from the data analysis.

Prior to inhalation, baseline levels of each of the four relevant compounds were registered. In multiple instances, the measured baseline concentrations of the methylxanthines were too high to be accounted to the residual of the previous dose(s). This may have been due to intake of methylxanthines with coffee and cocoa during the dose intervals. To take this into account, a baseline model was developed. Between course methylxanthine intake was modelled as a bolus of each compound into the respective compartments just prior to the measurement of the baseline level. In this way, we allowed adjustment of the baseline bolus according to residuals of the previous dose(s) and introduced a residual error into the baseline level measurement.

The bioavailability of caffeine ($F_{(CAF)}$) can not be estimated from these data alone. Therefore, instead of the clearance (CL) and the volume of distribution of the central compartment (V_1), $CL/F_{(CAF)}$ and $V_1/F_{(CAF)}$ were estimated for caffeine. Formation rate constants of the metabolites were expressed as fractions ($F_{(PAR)}$, $F_{(THBR)}$, $F_{(THPH)}$ for the individual metabolites and $F_{(MET)}$ to indicate any) of the elimination rate constant of caffeine ($k_{e(CAF)}$). As no information was available to estimate the volumes of distribution of the metabolites paraxanthine, theobromine and theophylline ($V_{(PAR)}$, $V_{(THBR)}$ and $V_{(THPH)}$ respectively or $V_{(MET)}$ to indicate any), the estimates of the formation rate constants ($k_{e\ par}$, $k_{e\ thbr}$ and $k_{e\ thph}$) should be interpreted as $(F/V_{(MET)})*k_{e(CAF)}$ and the fractions of caffeine, metabolised to the respective dimethylxanthines can not be calculated from this compartmental analysis.

First, a structural model concerning the pharmacokinetics of the parent compound, caffeine, was built. Models with zero and first order absorption and with and without lag-time were tested. The inclusion of transition compartments to the absorption model was investigated. Both models with and without a peripheral compartment were applied to the data. First order elimination from the central compartment was used in all models. Thereafter, the structural model for caffeine was extended to include the metabolites theobromine, paraxanthine and theophylline.

The performance of the models was assessed by both statistical and graphical methods. Among the statistical methods, the merit of a more complex model over a less complex submodel was tested against the respective objective function values (OFV). The OFV is approximated by minus twice the maximised log-likelihood of the data. A model with one additional parameter, was considered to be significantly better than its more restricted submodel when its OFV was at least 3.84 units smaller. This difference in OFV between the fits of the two models, the log-likelihood ratio, corresponds to $P=0.05$ on the χ^2 distribution. For covariate inclusion, the critical value of the difference in OFV was set at 10.83 corresponding to $P=0.001$.

In addition, the following statistical diagnostic tools were considered: standard error and correlation matrix of parameter estimates and variance of random effects as observed by the COVARIANCE option of NONMEM. Graphical methods were facilitated by Xpose (version 3) (Division of Pharmacokinetics and Drug Therapy, Uppsala University, Sweden), a collection of applications of S-Plus (version 2000) (MathSoft Inc, Cambridge, USA), and comprised the visual assessment of various plots, such as predicted concentrations versus measured values, and weighted residuals versus predicted or measured concentrations and versus time.

The incorporation of the method of inhalation and the diacetylmorphine dose as covariates was tested to attain the intermediate model. Clinical relevance (defined as substantial impact on one or more PK parameters) and statistical significance were criteria for inclusion of covariates. The final pharmacostatistical model was selected by evaluation of random effects (interindividual variability, interoccasion variability and intraindividual residual errors) and the assessment of correlation between these random effects.

Interindividual variability for the pharmacokinetic parameters and intraindividual residual error were modelled using an exponential error model. Interoccasion variability was estimated for both the clearance and the volume of distribution of caffeine as suggested by Karlsson & Sheiner (1993). For instance, variability in $CL/F_{(CAF)}$ was estimated using $CL/F_{ij}=\theta*e^{(\eta i+\kappa j)}$ in which CL/F_{ij} represents the apparent clearance of the ith individual in the jth occasion, θ is the typical value of CL/F in the population, η_i is the interindividual random effect and κ_j is the interoccasion random effect.

Model evaluation. In order to examine if the parameter estimates were highly determined by one of the individuals in the study population, the final model was fitted to the population lacking one individual. This was repeated until every individual had been excluded from the analysis, which resulted in 10 analyses that were subject to this jack-knife method.

Comparison to caffeine pharmacokinetics after intravenous and oral administration. The model was compared to published results of three pharmacokinetic studies in healthy volunteers, who had received caffeine by intravenous injection or by oral administration of mocha coffee or caffeine capsules. Based on this comparison, the bioavailability of caffeine after inhalation was roughly estimated. Furthermore, the apparent elimination rate constant at the end of the observed time window was compared to the published results.

Results

Population pharmacokinetic model.

Ten volunteers were included in the study and, from 5 occasions per person, a total of 2545 plasma concentrations (total of caffeine, theobromine, paraxanthine and theophylline) was available for pharmacokinetic analysis. Due to potential caffeine intake, 9 courses were completely excluded from the analysis and 2 other courses were re-

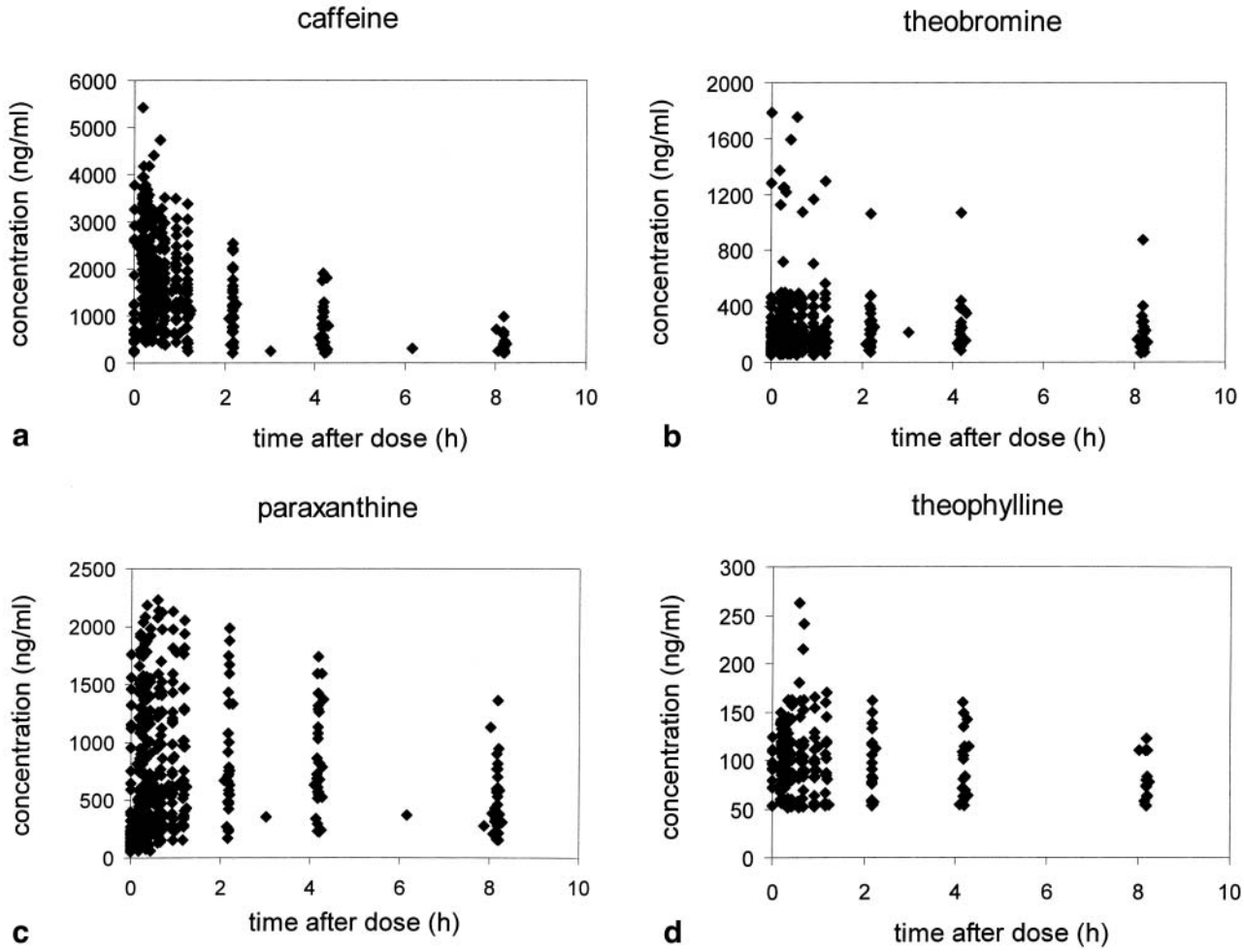

Fig. 1. Concentration (ng/ml) versus time after dose (hr) plots of caffeine (a) and its dimethylxanthine metabolites (theobromine (b), paraxanthine (c) and theophylline (d)).

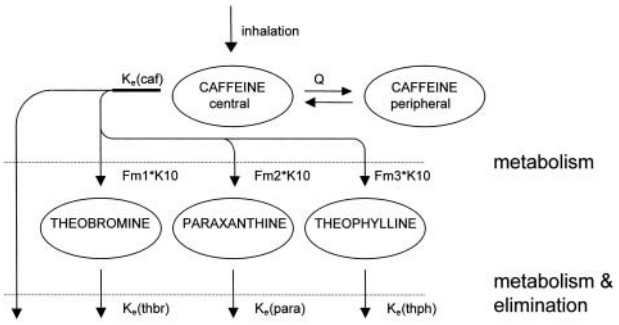

Fig. 2. Structural model of the pharmacokinetics of caffeine and three metabolites. Compartment 1=[caffeine central], compartment 2 =[theobromine], compartment 3=[paraxanthine], compartment 4=[theophylline], compartment 5=[caffeine peripheral].

stricted to the first 6 hr. In 13 cases, one or more theobromine concentrations that were possibly raised due to cocoa intake, were excluded. Careful examination of the remaining individual plasma concentration versus time profiles showed that all abnormalities had indeed been eliminated by application of the restrictions, which were predefined in the analysis plan. Thus, the criteria were well suitable to correct for xanthine intake with foodstuffs. Raw data are depicted in fig. 1.

Caffeine plasma concentrations were best described by a two compartmental model and its metabolites by single compartment models. Fig. 2 depicts the structural model of the pharmacokinetics of caffeine and its dimethylxanthine metabolites.

Table 1 shows the final estimates of the pharmacokinetic parameters, the inter- and intraindividual variabilities and the relative correlation coefficients of the final model. $CL/F_{(CAF)}$ was estimated to be 8.37 l/hr on an average. The population values of the central compartment $V_1/F_{(CAF)}$ and the peripheral compartment $V_2/F_{(CAF)}$ were estimated to be 76.2 l and 214 l respectively. The interindividual and interoccasion variability of $CL/F_{(CAF)}$ and $V_1/F_{(CAF)}$, prob-

Table 1.

Parameter estimates of the final population pharmacokinetic model and the stability of the parameters using a jack-knife evaluation procedure.

		Full data set		Jack-knife data	
		Estimate	(CV%)	Median estimate	(range)
Pharmacokinetic parameters					
$V_1/F_{(CAF)}$	(L)	76.2	(17.8)	77.9	(68.3–82.8)
$CL/F_{(CAF)}$	(L/h)	8.37	(18.5)	8.38	(7.48–9.09)
$V_2/F_{(CAF)}$	(L)	214	(17.1)	214	(197–242)
$Q/F_{(CAF)}$	(L/h)	9.74	(16.5)	9.82	(8.60–10.70)
$F_{(THBR)}/V_{(THBR)}$	(L^{-1})	0.00537	(36.8)	0.00574	(0.00446–0.00693)
$F_{(PARA)}/V_{(PARA)}$	(L^{-1})	0.0288	(27.5)	0.0283	(0.0260–0.0340)
$F_{(THPH)}/V_{(THPH)}$	(L^{-1})	0.00342	(31.9)	0.00342	(0.00265–0.00426)
$k_{e\ (THBR)}$	(h^{-1})	0.117	(19.1)	0.118	(0.100–0.130)
$k_{e\ (PARA)}$	(h^{-1})	0.308	(16.4)	0.309	(0.277–0.339)
$k_{e\ (THPH)}$	(h^{-1})	0.313	(26.9)	0.316	(0.220–0.351)
Interindividual variability					
$\eta\ V_1/F_{(CAF)}$	(%)	51.9	(12.5)	52.0	(47.0–54.7)
$\eta\ CL/F_{(CAF)}$	(%)	56.3	(27.8)	58.8	(34.5–64.6)
$\eta\ F/V_{(THBR)}$	(%)	124	(20.3)	129.2	(104.9–136.0)
$\eta\ F/V_{(PARA)}$	(%)	65.5	(24.1)	64.7	(47.5–75.3)
$\eta\ F/V_{(THPH)}$	(%)	64.4	(27.5)	65.8	(43.5–73.2)
$\rho\ (\eta V_1/F_{(CAF)}, \eta CL/F_{(CAF)})$		0.42	(107.4)	0.44	(0.10–0.63)
$\rho\ (\eta F/V_{(THBR)}, \eta\ F/V_{(PARA)})$		0.55	(95.9)	0.56	(0.23–0.82)
$\rho\ (\eta F/V_{(PARA)}, \eta\ F/V_{(THPH)})$		0.74	(80.8)	0.74	(0.57–0.98)
$\rho\ (\eta F/V_{(THBR)}, \eta\ F/V_{(THPH)})$		0.83	(84.4)	0.82	(0.47–0.98)
Interoccasion variability					
$\kappa\ V_1/F_{(CAF)}$	(%)	29.8	(16.2)	30.4	(25.2–31.5)
$\kappa\ CL/F_{(CAF)}$	(%)	17.9	(17.4)	18.4	(12.5–19.0)
$\rho\ (\kappa V_1/F_{(CAF)}, \kappa CL/F_{(CAF)})$		0.67	(34.8)	0.65	(0.53–0.77)
Residual variability					
$\varepsilon_{(CAF)}$	(%)	26.2	(11.5)	26.5	(23.3–27.4)
$\varepsilon_{(THBR)}$	(%)	22.6	(14.9)	23.1	(20.0–23.8)
$\varepsilon_{(PARA)}$	(%)	21.3	(11.1)	21.5	(19.4–22.2)
$\varepsilon_{(THPH)}$	(%)	22.2	(16.5)	22.5	(19.4–23.7)

Abbreviations used in table 1.

V: volume of distribution (V_1: central compartment, V_2: peripheral compartment); F: bioavailability (caffeine) or fraction caffeine converted to the metabolite (paraxanthine, theobromine, theophylline); CL: clearance; Q: intercompartmental clearance between central and peripheral compartment; k_e: elimination rate constant; caf: caffeine; thbr: theobromine; para: paraxanthine; thph: theophylline; η: interindividual variability; ρ: correlation coefficient; κ: interoccasion variability; ε: residual variability.

ably partly due to variation in $F_{(CAF)}$, were moderate. On the condition that the volumes of distribution of the metabolites were approximately equal, paraxanthine was the main dimethylxanthine metabolite ($F_{(PARA)}/V_{(PARA)}$= 0.0288 l^{-1}), followed by theobromine ($F_{(THBR)}/V_{(THBR)}$= 0.00537 l^{-1}) and theophylline ($F_{(THPH)}/V_{(THPH)}$=0.00342 l^{-1}) respectively. Formation rate constants of the metabolites were subject to large interindividual variability (64.4%–124%). Residual errors were small and of comparable size for all compounds (21.3%–26.2%). From the results in table 1 it can be concluded that all parameters could be estimated with an acceptable precision.

Goodness of fit plots for the final model are depicted in fig. 3 and fig. 4. As can be seen from these plots, the developed model enabled adequate description of the pharmacokinetics of caffeine and the dimethylxanthines.

After start of inhalation, caffeine very rapidly appeared in the circulation indicating a rapid absorption process in the lungs. The duration of inhalation was 10 min. on average (range 7–14 min.). However, no plasma concentration data were available during the inhalation. Therefore, the absorption process was difficult to assess. The intake rate of caffeine was not expected to be constant, but probably decreased in time as the tablet gradually got smaller. It was supposed that the absorption rate was maximal shortly after start of inhalation. Initially, the data could adequately be described as if it was a bolus administration into the central compartment. After inclusion of metabolite data, the smoking process was further investigated. The data indeed best supported a bolus administration to describe the inhalation of caffeine.

None of the tested covariates (the method of inhalation and the diacetylmorphine dose) showed a relation with the individual estimates for $CL/F_{(CAF)}$ or $V_1/F_{(CAF)}$ and were, therefore, not included in the model.

Through dependence of both the clearance ($CL/F_{(CAF)}$) and the volume of distribution ($V_1/F_{(CAF)}$) on $F_{(CAF)}$, the individual parameter estimates were expected to be correlated. Indeed the correlation coefficient was 0.42. Correlation between the individual values of formation rate constants of the metabolites was observed as well. Covariance between interindividual variability of $CL/F_{(CAF)}$ and $V_1/F_{(CAF)}$ and between the formation rate constants of the metabolites were included in the final model.

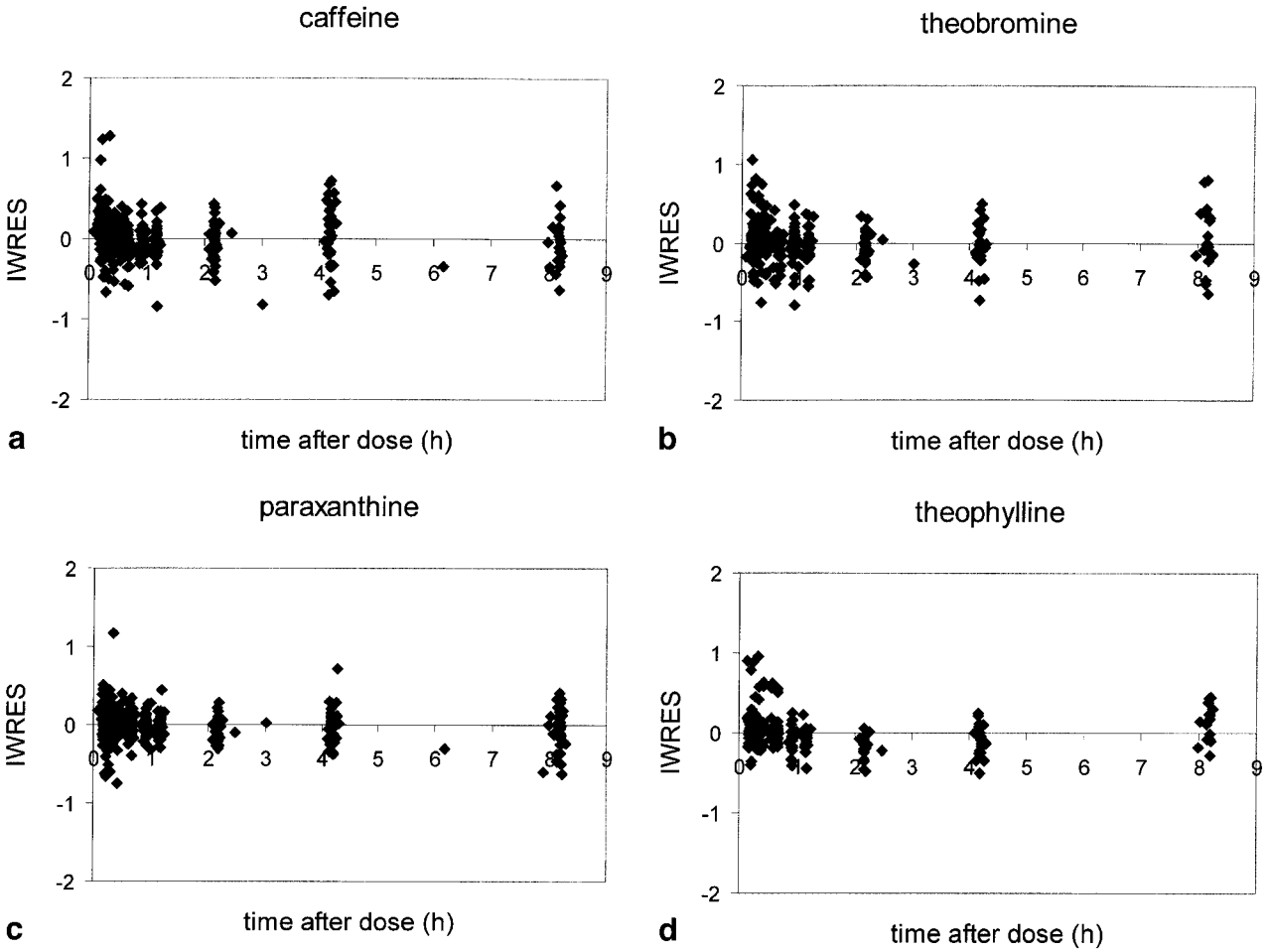

Fig. 3. Individual weighted residuals (IWRES) versus time after dose (h) for caffeine (a), theobromine (b), paraxanthine (c) and theophylline (d).

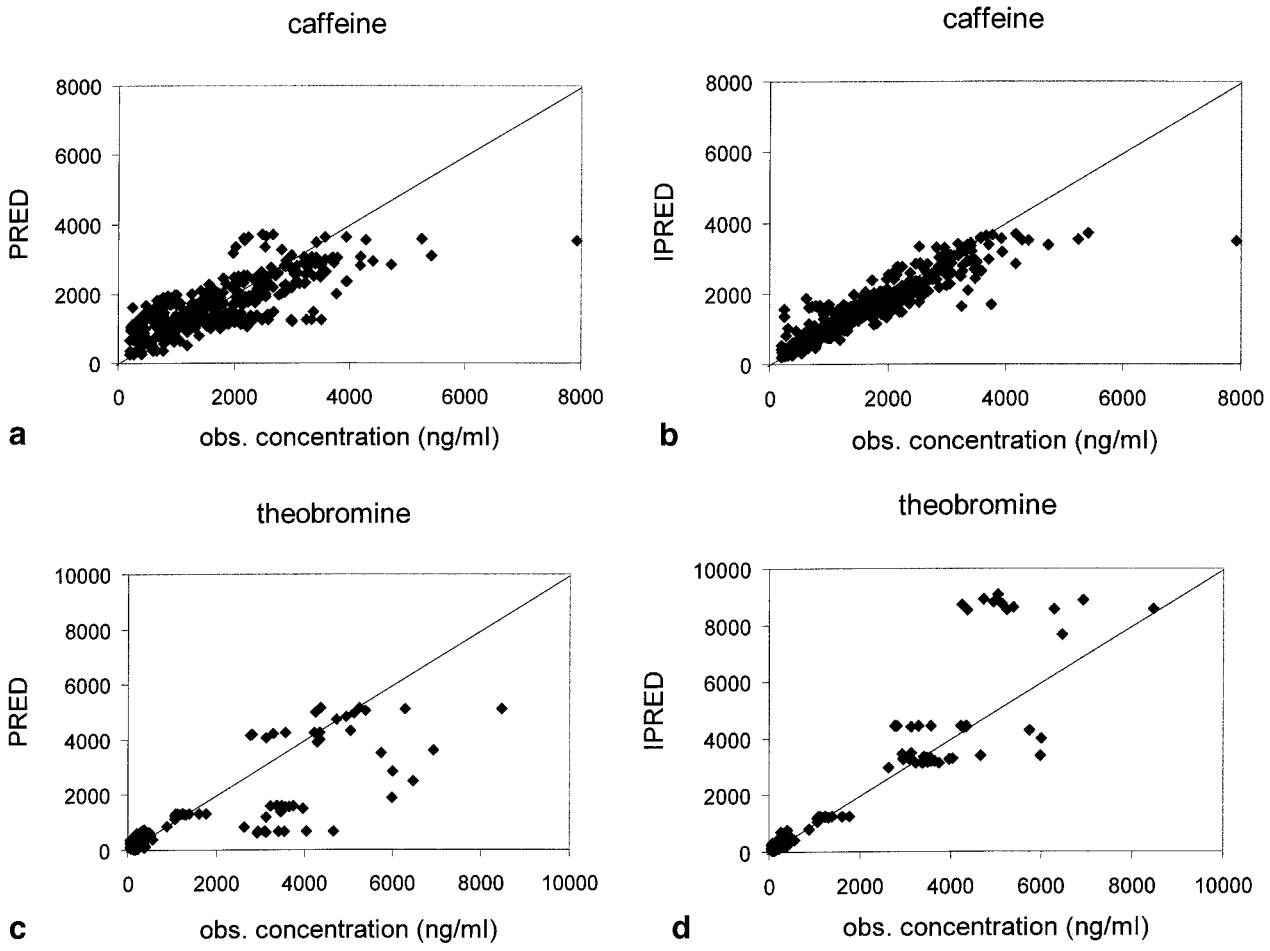

Fig. 4c and 4d show that most predicted and measured theobromine concentrations are lower than 2000 ng/ml. However, much higher concentrations up to 8000 ng/ml are observed. All measured and predicted concentrations higher than 2000 ng/ml are related to the theobromine levels of a single exceptional individual.

Data in the right columns of table 1 resulted from the jack-knife procedure. The validation proved that the results were not highly based on a single individual. Exclusion of the exceptional individual with high theobromine levels did not significantly change the parameter estimates. Furthermore, very little variability in the estimates of the all parameters was observed in the jack-knife runs. This is an indication for proper stability of the model presented.

Comparison to caffeine pharmacokinetics after intravenous and oral administration.

Table 2 shows the results of three previously published pharmacokinetic studies. Healthy volunteers received caffeine (1) by an intravenous bolus injection (Renner *et al.* 1984), (2) by oral intake of mocha coffee (Bonati *et al.* 1982) or (3) by oral intake of caffeine capsules (Lelo *et al.* 1986). The absolute bioavailability of caffeine after intravenous or oral administration was shown to be complete by Blanch-

Table 2.

Mean pharmacokinetic parameters of caffeine, from previous study results.

Route of administration	Formulation	k_a	V (l)	CL (L/hr)	k_e (h^{-1})	Number of subjects	Literature reference
Intravenous	Injection		44.8[1]	8.48[1]	0.189	10	(Renner *et al.* 1984)
Oral	Coffee	5.6	53.5	7.14	0.133	4	(Bonati *et al.* 1982)
Oral	Capsule	1.3–2.4[2]	51.5[1]	8.70[1]	0.169	6	(Lelo *et al.* 1986)

[1] Result was normalised to a parameter value for 70 kg subjects.
[2] No absorption rate constant was reported by Lelo *et al.* (1986). Therefore, the reported value for k_a was adopted from Kamimori *et al.* (2002).

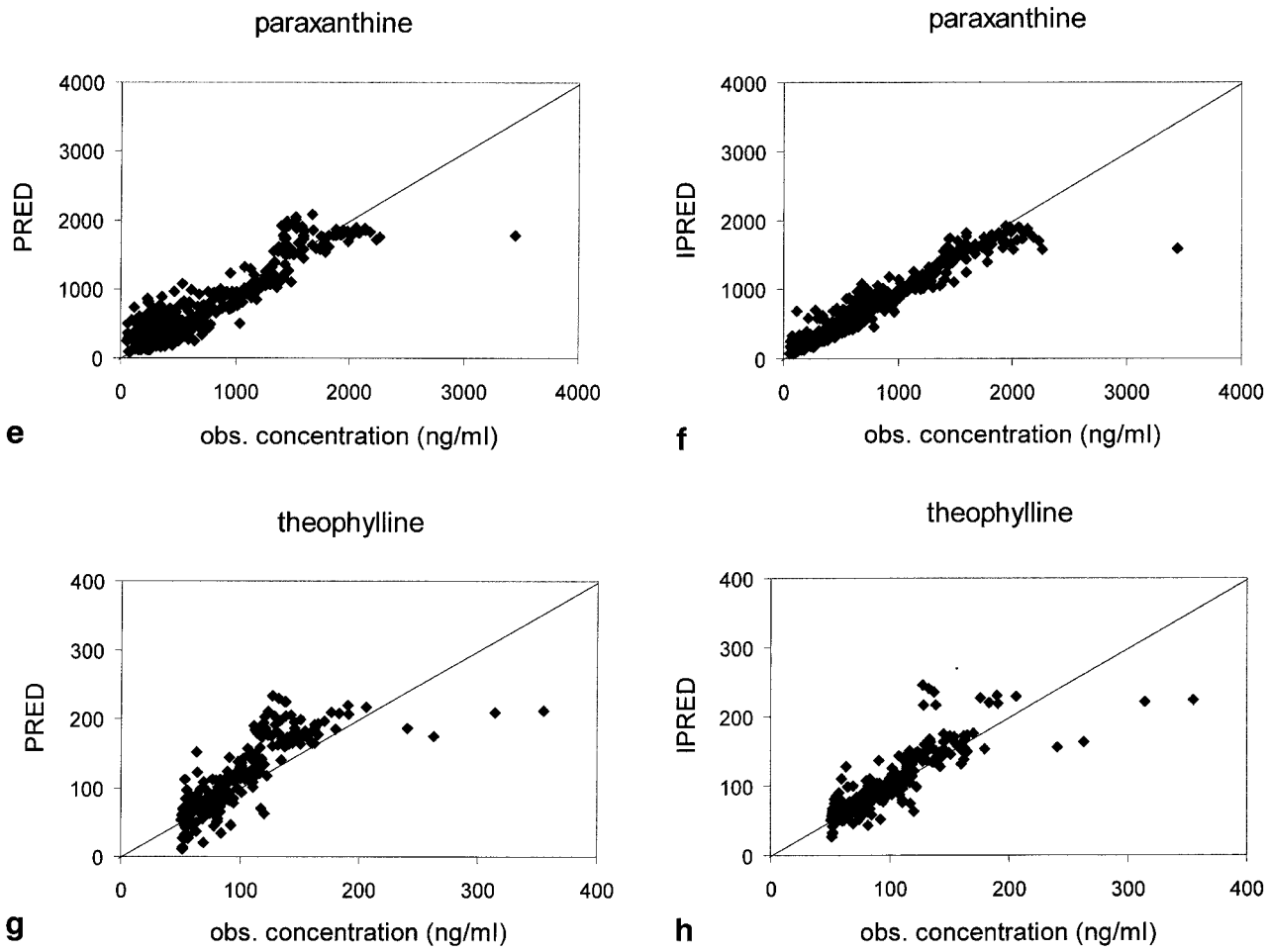

Fig. 4. Model predicted (PRED) and individual Bayesian predicted concentrations (IPRED) (ng/ml) versus observed concentrations (ng/ml) for caffeine (a and b), theobromine (c and d), paraxanthine (e and f) and theophylline (g and h).

ard & Sawers (1983). Fig. 5a shows the typical concentration versus time curves for caffeine after a 100 mg dose for the previously published and the current studies, given that the bioavailability of caffeine is 100% in all four occasions. Caffeine plasma concentrations are typically lower after inhalation than after oral or intravenous administration of a 100 mg dose. From fig. 5a, it can therefore be concluded that the bioavailability of caffeine after inhalation is lower than 100%. As shown in fig. 5b, the concentration versus time profile after inhalation of a 100 mg dose corresponds well to the typical profiles after oral or intravenous administration of a 60 mg dose. As a result, the absolute bioavailability of caffeine administered by inhalation is estimated to be approximately 60%. Consequently, the volume of distribution of the central compartment $V_{1(CAF)}$ for the current study is estimated to be 45.7 l which corresponds well to previously reported volumes of distribution (44.8 l (Renner *et al.* 1984), 53.5 l (Bonati *et al.* 1982) and 51.5 l (Lelo *et al.* 1986)). The apparent elimination rate constant $k_{e\,(CAF)}$ at 8 hr after inhalation was 0.150 hr^{-1} for the current study. This also corresponds well to previously reported elimination rate constants (0.189 hr^{-1} (Renner *et al.* 1984), 0.133 hr^{-1} (Bonati *et al.* 1982) and 0.169 hr^{-1} (Lelo *et al.* 1986)).

Discussion

The bioavailability of caffeine was estimated to be approximately 60% after inhalation. For the co-administered diacetylmorphine a bioavailability of 45% was reported (Hendriks *et al.* 2001). The incomplete bioavailability of heroin is due to decomposition during the heating process and incomplete inhalation of the fumes. Huizer (1987) reported that the recovery rate of heroin base during intermittent heating of a 1:1 mixture with caffeine was 76%. By combining these results, it can be concluded that approximately 60% (=0.45/0.76) of the fumes is inhaled by experienced smokers using the "chasing the dragon" inhalation method. As caffeine is rather heat-stable (Brenneisen & Hasler 2002), its bioavailability is indeed expected to be close to the inhaled fraction. The method of inhalation and the diacetylmorphine dose were tested as covariates related to $V_1/F_{(CAF)}$ and $CL/F_{(CAF)}$. Both parameters were likely to have influenced the bioavailability of caffeine. This was expected

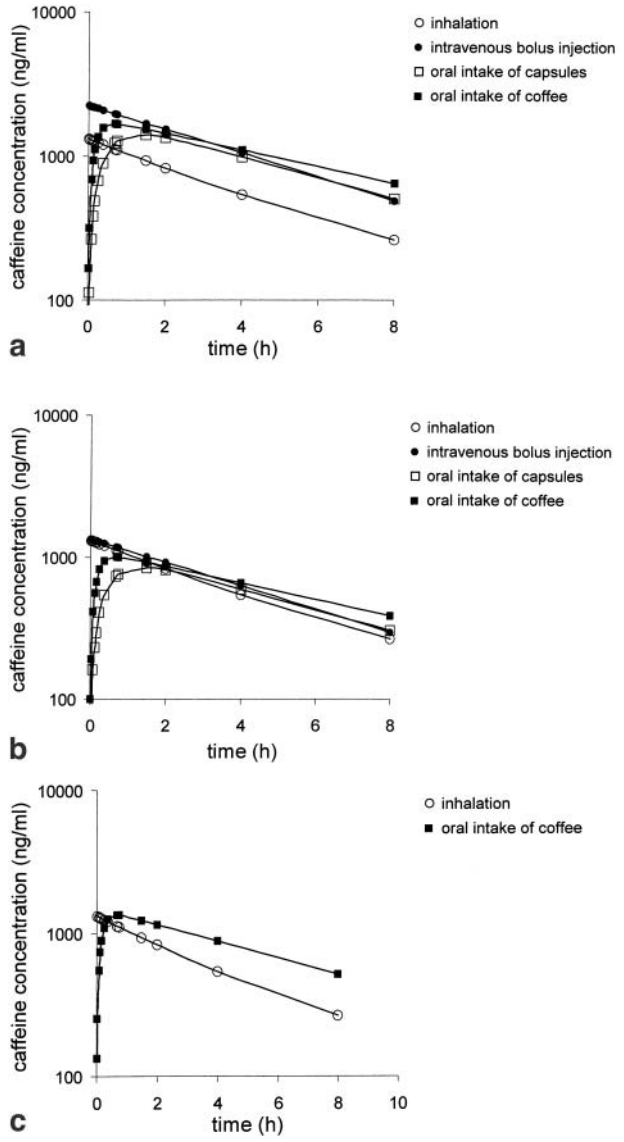

Fig. 5. Typical concentration versus time profiles of caffeine (a) after administration of 100 mg and assuming that the bioavailability is complete for all routes of administration (b) after administration of 60 mg for the oral and intravenous routes of administration and 100 mg for inhalation (c) after intake of 1 cup of coffee and after inhalation of 100 mg dose.

because of their potential to affect the yield of the sublimation process, by causing variations in the attained temperature and in the ratio of caffeine and diacetylmorphine in the tablets. However, no influence was observed.

Caffeine is metabolised by CYP enzymes (demethylation), xanthine oxidase (formation of uric acid metabolites) and/or N-acetyltransferase (acetylation). As far as the metabolites observed in this study are concerned, only CYP mediated metabolism is relevant. All demethylation reactions are predominantly catalysed by CYP1A2. This CYP isoform is exclusively responsible for the N3-demethylation of caffeine (formation of paraxanthine), but the N1- and N7-demethylation (formation of theobromine and theophylline) are also catalysed by CYP2E1 (Bechtel *et al.* 1993; Yuan *et al.* 2002). As all dimethylxanthines are predominantly formed through CYP1A2 catalysed reactions, the formation rate constants are expected to be correlated. This was indeed observed in the pharmacokinetic analysis (correlation coefficients were 0.55, 0.74 and 0.83). Polymorphism of the CYP1A2 isoenzyme is likely to partially explain interindividual variability of the formation rate constants and their correlation. In the current study, the patient population (n=10) was too small to conduct a genotyping analysis and to determine the relationship between CYP1A2 genotype and caffeine metabolism phenotype.

According to Bonati *et al.* (1982), the metabolic rate constant ratio for the dimethylxanthines after oral administration of caffeine was 10:78:12 (theobromine: paraxanthine: theophylline). In the present study, the estimates for $F/V_{(MET)}$ were in the proportion of 14:77:9. On the condition that the volumes of distribution of the metabolites are about equal, the present results after caffeine inhalation correspond well with previous data.

The model was corrected for unregistered caffeine and cocoa intake in the best possible way. Especially one particular subject showed extremely high theobromine concentrations, whereas the levels of paraxanthine and theophylline were in the normal range. The estimates for the formation and elimination rate constants for theobromine for this subject were not exceptional, but the baseline level was particularly high. A possible explanation could be a high intake of cocoa products, but it remains remarkable that the theobromine concentration is high throughout the total follow up time of 5 days.

The maximal model predicted plasma concentrations of caffeine in a typical individual are 1312 ng/ml after inhalation of a 100 mg caffeine tablet and 1335 ng/ml after consumption of one cup of coffee, containing about 80 mg of caffeine (fig. 5c). The pharmacodynamic effect of inhalation of 100 mg caffeine is therefore expected to be approximately equal to the effect of intake of one cup of coffee.

The behavioural effects of 200 mg caffeine in healthy volunteers reported by Brice & Smith (2002) were mild. It should however be noted that sensitivity to caffeine is known to be highly variable and especially patients with particular psychiatric disorders can be abnormally sensitive to caffeine (Lee *et al.* 1988; Bruce *et al.* 1992). As this may apply to particular subjects from the study population, inhalation of the sublimate of a 100 mg tablet of caffeine may occasionally induce anxiety and may therefore interfere with the establishment of the pharmacodynamic effects of diacetylmorphine in clinical trials. In consecutive study programmes in The Netherlands for the investigation of medical prescription of pharmaceutically prepared heroin to heroin addicts, caffeine dosages were decreased to $^1/_3$ of the dosage of diacetylmorphine.

In conclusion, when diacetylmorphine is administered for study purposes, after inhalation of the sublimate of 100 mg caffeine together with diacetylmorphine base, the pharma-

codynamic effects of caffeine may interfere with the pharmacodynamic effects of diacetylmorphine. The current study focused on the pharmacokinetics of caffeine after inhalation in heroin users. The model presented in this paper adequately describes the population pharmacokinetics of caffeine and its metabolites theobromine, paraxanthine and theophylline after inhalation of the caffeine sublimate of a 100 mg tablet. Validation by the jack-knife procedure proved the stability of the model. The pharmacokinetics of caffeine administered by inhalation are to a large extent similar to the pharmacokinetics of intravenously administered caffeine. The bioavailability of inhaled caffeine is approximately 60%.

References

Beal, S. L. & L. B. Sheiner: NONMEM User's Guide. 1998.

Bechtel, Y. C., C. Bonaiti-Pellie, N. Poisson, J. Magnette & P. R. Bechtel: A population and family study of N-acetyltransferase using caffeine urinary metabolites. *Clin. Pharmacol. Ther.* 1993, **54**, 134–141.

Blanchard, J. & S. J. Sawers: The absolute bioavailability of caffeine in man. *Eur. J. Clin. Pharmacol.* 1983, **24**, 93–98.

Bonati, M., R. Latini, F. Galletti, J. F. Young, G. Tognoni & S. Garattini: Caffeine disposition after oral doses. *Clin. Pharmacol. Therap.* 1982, **32**, 98–106.

Brenneisen, R. & F. Hasler: GC/MS determination of pyrolysis products from diacetylmorphine and adulterants of street heroin samples. *J. Forensic Sci.* 2002, **47**, 885–888.

Brice, C. F. & A. P. Smith: Effects of caffeine on mood and performance: a study of realistic consumption. *Psychopharmacology (Berl.)* 2002, **164**, 188–192.

Bruce, M., N. Scott, P. Shine & M. Lader: Anxiogenic effects of caffeine in patients with anxiety disorders. *Arch. Gen. Psychiatry* 1992, **49**, 867–869.

Falcao, A. C., M. M. Fernandez de Gatta, M. F. Delgado Iribarnegaray, B. D. Santos, M. J. Garcia, A. Dominguez-Gil & J. M. Lanao: Population pharmacokinetics of caffeine in premature neonates. *Eur. J. Clin. Pharmacol.* 1997, **52**, 211–217.

Hendriks, V. M., W. van den Brink, P. Blanken, I. J. Bosman & J. M. van Ree: Heroin self-administration by means of 'chasing the dragon': pharmacodynamics and bioavailability of inhaled heroin. *Eur. Neuropsychopharmacol.* 2001, **11**, 241–252.

Huizer, H.: Analytical studies on illicit heroin. V. Efficacy of volatilization during heroin smoking. *Pharm. Weekbl. Sci.* 1987, **9**, 203–211.

Kamimori, G. H., C. S. Karyekar, R. Otterstetter, D. S. Cox, T. J. Balkin, G. L. Belenky & N. D. Eddington: The rate of absorption and relative bioavailability of caffeine administered in chewing gum versus capsules to normal healthy volunteers. *Int. J. Pharm.* 2002, **234**, 159–167.

Karlsson, M. O. & L. B. Sheiner: The importance of modeling interoccasion variability in population pharmacokinetic analyses. *J. Pharmacokinet. Biopharm.* 1993, **21**, 735–750.

Khokhar, S. & S. G. Magnusdottir: Total phenol, catechin, and caffeine contents of teas commonly consumed in the United kingdom. *J. Agric. Food Chem.* 2002, **50**, 565–570.

Lee, M. A., P. Flegel, J. F. Greden & O. G. Cameron: Anxiogenic effects of caffeine on panic and depressed patients. *Amer. J. Psychiatry* 1988, **145**, 632–635.

Lelo, A., D. J. Birkett, R. A. Robson & J. O. Miners: Comparative pharmacokinetics of caffeine and its primary demethylated metabolites paraxanthine, theobromine and theophylline in man. *Brit. J. Clin. Pharmacol.* 1986, **22**, 177–182.

Mattila, M. J.: Interactions of benzodiazepines on psychomotor skills. *Brit. J. Clin. Pharmacol.* 1984, **18**, 21S–26S.

Renner, E., H. Wietholtz, P. Huguenin, M. J. Arnaud & R. Preisig: Caffeine: a model compound for measuring liver function. *Hepatology* 1984, **4**, 38–46.

Sawynok, J.: Pharmacological rationale for the clinical use of caffeine. *Drugs* 1995, **49**, 37–50.

Tanaka, E.: Simultaneous determination of caffeine and its primary demethylated metabolites in human plasma by high-performance liquid chromatography. *J. Chromatogr.* 1992, **575**, 311–314.

Van den Brink, W., V. M. Hendriks, P. Blanken, M. W. Koeter, B. J. van Zwieten & J. M. van Ree: Medical prescription of heroin to treatment resistant heroin addicts: two randomised controlled trials. *Brit. Med. J.* 2003, **327**, 310.

Yuan, R., S. Madani, X. X. Wei, K. Reynolds & S. M. Huang: Evaluation of cytochrome P450 probe substrates commonly used by the pharmaceutical industry to study in vitro drug interactions. *Drug Metab. Dispos.* 2002, **30**, 1311–1319.