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PHARMACOKINETICS AND DISPOSITION

Effects of gender and moderate smoking on the pharmacokinetics and effects of the CYP1A2 substrate tizanidine

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Abstract
Objective We studied the effects of gender and smoking on the pharmacokinetics and effects of the cytochrome P450 (CYP) 1A2 substrate tizanidine.
Methods Seventy-one healthy young volunteers (male and female nonsmokers, male smokers) ingested 4 mg tizanidine. Plasma concentrations and pharmacodynamics of tizanidine were measured, and a caffeine test was performed.
Results Among nonsmokers, the peak concentration (C_{max}) and area under concentration-time curve from 0 to infinity [AUC(0-∞)] of tizanidine did not differ significantly between females and males. However, the half-life ($t_{1/2}$) was 9% shorter in female nonsmokers than in male nonsmokers ($P<0.05$). In male smokers, the $t_{1/2}$ was 10% shorter and the weight-adjusted AUC(0-∞) 33% smaller than in male nonsmokers ($P<0.05$). The caffeine/paraxanthine ratio was 35–40% smaller ($P=0.001$) in male smokers than in nonsmoking males or females, but did not differ between males and females. Tizanidine lowered blood pressure and caused drowsiness significantly ($P<0.05$) more in females than in either male groups. The effects on blood pressure were smallest in male smokers ($P<0.05$).
Conclusions Gender by itself seems to have no clinically significant effect on the pharmacokinetics of tizanidine, whereas smoking reduces plasma concentrations and effects of tizanidine. Any possible effect of gender and smoking is largely outweighed by individual variability in CYP1A2 activity due to genetic and environmental factors and in body weight. Careful dosing of tizanidine is warranted in small females, whereas male smokers can require higher than average doses.

Keywords Gender · Smoking · Tizanidine · CYP1A2 · Caffeine test

Gender-related differences in pharmacokinetics and pharmacodynamics can be important determinants for the effectiveness and safety of drug therapy [1–4]. Gender differences have been found in the pharmacokinetics of, e.g., beta-blockers, verapamil, selective serotonin reuptake inhibitors, and caffeine [2, 3, 5]. Men have been described to have a higher activity relative to women for certain cytochrome P450 (CYP) isoenzymes, e.g., CYP1A2 and CYP2E1, whereas the activity of CYP3A4 has been slightly lower in men than in women [1, 3, 5–8]. Gender-related differences can also exist in the membrane transport of exogenous compounds [3, 4, 9, 10]. For example, a genetic polymorphism affecting the hepatic uptake transporter organic anion transporting polypeptide 1B1 has a greater effect on the pharmacokinetics of pravastatin in males than in females [10]. Other factors causing gender-related pharmacokinetic differences include the lower body weight and organ size, higher percentage of body fat, and lower glomerular filtration rate in women than in men [2–4, 7].

Exposure to the polycyclic aromatic hydrocarbons of cigarette smoke leads to induction of CYP1A2 [6, 11–20],

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and cigarette smoking can have a significant impact on the pharmacokinetics and effects of many CYP1A2-substrate drugs [6, 12, 13, 15–22]. Consequently, smoking habits should be considered in individual dosing of certain drugs metabolized by CYP1A2, such as clozapine [23, 24] and theophylline [25, 26]. Tizanidine, a centrally acting skeletal muscle relaxant, is metabolized principally by the CYP1A2 enzyme [27–32]. The oral bioavailability of tizanidine is low due to its extensive presystemic metabolism, which makes its pharmacokinetics highly susceptible to changes in CYP1A2 activity. For example, fluvoxamine, ciprofloxacin, and rofecoxib, inhibitors of CYP1A2 [32–36], strongly increase the plasma concentrations and effects of tizanidine [29–31].

Our aim was to study whether gender and moderate smoking have a clinically relevant effect on the pharmacokinetics and effects of tizanidine. In addition, we investigated whether the findings with tizanidine cosegregate with the results of the caffeine test, which is a validated index of the CYP1A2-mediated clearance of caffeine [13, 15, 17, 19, 37].

Methods

Subjects

Seventy-one healthy young volunteers (53 men and 18 women; age range 19–31 years; weight range 52–100 kg) participated in the study after giving written informed consent. As it was not possible to recruit an adequate number of female smokers, the effect of smoking was compared in men only. Of the men, 38 were nonsmokers and 15 were tobacco smokers. The smokers consumed 10–20 cigarettes daily, except for one subject, who smoked about 25 cigarettes daily, and they had smoked for about 1 year or longer, resulting in a smoking history of at least 0.5 pack-years. None of the subjects regularly consumed other kinds of tobacco products. All women were nonsmokers. Subjects' characteristics are given in Table 1.

All subjects were ascertained to be healthy by medical history, physical examination, and routine laboratory tests before entering the study. None of the subjects used oral contraceptives or other continuous medication. The women participated in the study during the days 11–26 of their ongoing menstrual cycle. For safety reasons, subjects with a systolic blood pressure lower than 110 mmHg were excluded. As the number of recruitable women fulfilling the inclusion criteria (no hormonal contraceptives, systolic blood pressure at least 110 mm Hg) and that of smoking men was limited, a larger group of nonsmoking men was included in order to increase the statistical power of the study. Of the male and female nonsmokers, 51 were included in tizanidine interaction studies also, which ran in parallel with the study reported here [29, 30, 32, 38, 39]. All plasma tizanidine concentrations for our study were determined from previously unthawed plasma samples using the same method for all samples. On the basis of the mean ± standard deviation (SD) area under concentration-time curve from 0 to infinity [AUC(0-∞)] of tizanidine (6.6±2.9 ng·h per ml) after a 4-mg oral dose [29], the total number of subjects (71) was estimated to be sufficient to detect a 35% difference in the AUC(0-∞) of tizanidine between the groups with a power of more than 80% (alpha level 5%).

Study design

The experiments comply with the current laws of Finland, and the study protocols were approved by the Ethics Committee for Studies in Healthy Subjects of the Hospital District of Helsinki and Uusimaa and the Finnish National Agency for Medicines. An open parallel group study design was used. After an overnight fast, each subject ingested a single oral dose of 4 mg of tizanidine (one Sirdalud 4 mg tablet; Novartis Pharma, Wehr, Germany) with 150 ml water. A standard meal was served 4 and 7 h after tizanidine administration. Drinking of grapefruit juice was forbidden for 1 week before each study day. Alcohol and drinks containing caffeine were not permitted on the study days, but tobacco smoking was allowed for the smokers on the day of tizanidine administration.

An oral caffeine test was performed one day before tizanidine administration [13, 18, 19, 37]. The subjects ingested 100 mg caffeine (one Cofi-Tabs 100 mg tablet; Vitabalans, Hämeenlinna, Finland) at 0900 h after having abstained from intake of caffeine for at least 18 h, and a blood sample for analysis of plasma caffeine and paraxanthine (1,7-dimethyl-xanthine) was taken from each subject 6 h after caffeine intake.

Table 1 Subjects' characteristics

	Number	Age (years)	Weight (kg)	BMI (kg/m^2)	Dose of tizanidine (mg/kg)
Male nonsmokers	38	23 (19–31)	78 (64–96)	23.4 (19.7–28.7)	0.052 (0.042–0.063)
Male smokers	15	24 (21–28)	85 (70–100)	25.6 (20.7–29.2)	0.048 (0.040–0.057)
Female nonsmokers	18	22 (19–26)	61 (52–85)	21.5 (19.3–28.7)	0.067 (0.047–0.080)

Data are given as mean (range)
BMI body mass index

The subjects were under direct medical supervision for 12 h after tizanidine administration. Fluids for intravenous infusion were available for immediate use but they were not needed.

Sampling

On the days of tizanidine administration, a forearm vein of each subject was cannulated with a plastic cannula and kept patent with an obturator. Timed blood samples were drawn before the administration of tizanidine and at 20, 40, 60, and 90 min and 2, 3, 4, 5, 7, 9, 12, and 24 h later. Blood samples (10 ml each) were taken into ethylenediaminetetraacetic-acid (EDTA)-containing tubes. Plasma was separated within 30 min.

Quantification of drug concentrations

Plasma concentrations of tizanidine were quantified using an API 2000 liquid chromatography-tandem mass spectrometry system (MDS Sciex, Toronto, ON, Canada) [38]. Chromatography was performed on an XTerra RP C18 column (3.9×100 mm; Waters Corp., Milford, MA, USA) using gradient elution. The mobile phase consisted of 10 mM ammonium acetate (pH 9.5, adjusted with 25% ammonia solution) and acetonitrile. The mass spectrometer was operated in the atmospheric pressure chemical ionization (APCI) mode with positive ion detection. The ion transitions monitored were mass-to-charge ratio (*m/z*) 254 to *m/z* 44 for tizanidine and *m/z* 230 to *m/z* 44 for internal standard, clonidine. These transitions represent the product ion of the $[M+H]^+$ ion. The limit of quantification for tizanidine was 0.05 ng/ml and the day-to-day coefficient of variation (CV) was 8.4% at 0.096 ng/ml (n=8), 6.7% at 0.96 ng/ml (n=8), and 3.8% at 9.6 ng/ml (n=8).

Plasma caffeine and paraxanthine concentrations were determined by high-performance liquid chromatography (HPLC), with β-OH-ethyltheophylline as the internal standard [40, 41]. The limit of quantification for both caffeine and paraxanthine was 0.05 mg/l, and the day-to-day CV was 6.1% at 0.95 mg/l (n=5) and 3.0% at 4.94 mg/l (n=5) for caffeine, and 1.1% at 0.995 mg/l (n=5) and 0.7% at 5.03 mg/l (n=5) for paraxanthine.

Pharmacokinetics

The pharmacokinetics of tizanidine were characterized by peak concentration in plasma (C_{max}), time to C_{max} (t_{max}), AUC(0-∞), and elimination half-life ($t_{1/2}$) using noncompartmental methods as described earlier [29]. In addition, weight-adjusted pharmacokinetic variables, $C_{max,adj}$, and $AUC(0\text{-}\infty)_{adj}$, were calculated by normalizing for 70 kg body weight.

Pharmacodynamics

Systolic and diastolic blood pressures, subjective drowsiness, subjective overall drug effect, and the Digit Symbol Substitution Test (DSST) were assessed before administration of tizanidine and immediately after each blood sampling, as described earlier [29]. For each pharmacodynamic variable, the decremental or incremental area under the effect vs. time curve from 0 to 12 h [AUC(0-12 h)] was calculated using the trapezoidal rule. In addition, the maximum responses in each pharmacodynamic variable were recorded.

Statistical analysis

Results are expressed as mean values ± SD in the tables and text. The pharmacokinetic and pharmacodynamic variables and the caffeine/paraxanthine ratio between the groups were compared by analysis of variance (ANOVA), followed by a t test for pairwise comparisons after log transformation of data when appropriate. All data were analyzed with the statistical program Systat for Windows, version 6.0.1. (SPSS Inc, Chicago, IL, USA). The differences were considered statistically significant at $P<0.05$.

Results

Effect of gender on tizanidine pharmacokinetics

The mean plasma concentrations of tizanidine were 25–30% higher in women than in nonsmoking men (Fig. 1). However, there was great interindividual variation in the pharmacokinetics of tizanidine, and a considerable overlapping was observed between groups (Figs. 1 and 2). For example, the C_{max} of tizanidine ranged 11-fold and 14-fold within the nonsmoking male and female groups.

Differences in the mean C_{max} and AUC(0-∞) of tizanidine between male and female nonsmokers were not statistically significant, although the mean values of C_{max} and AUC(0-∞) were 30% and 34% higher in women than in men (Table 2, Fig. 2), and in five of the 18 women, the C_{max} of tizanidine was higher than in any of the men (Fig. 2). However, the mean elimination $t_{1/2}$ of tizanidine was 9% shorter in the nonsmoking women than in the nonsmoking men ($P<0.05$; Table 2, Fig. 2).

Effect of smoking on tizanidine pharmacokinetics

The mean plasma concentrations of tizanidine were lower in male smokers than in nonsmokers (Fig. 1). The AUC(0-∞) of tizanidine was 38% smaller in male smokers than in male nonsmokers (Fig. 2; $P<0.05$), but the C_{max} was not

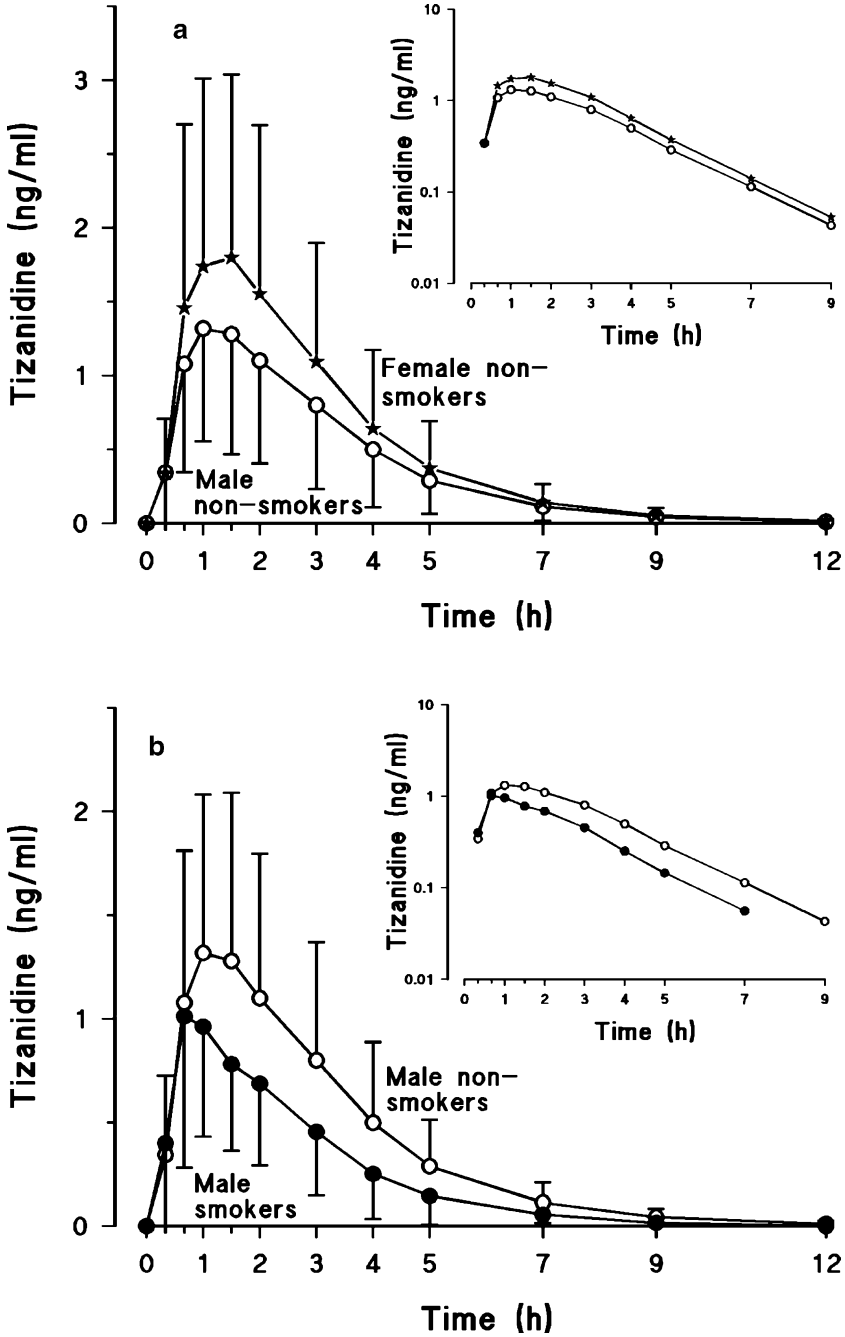


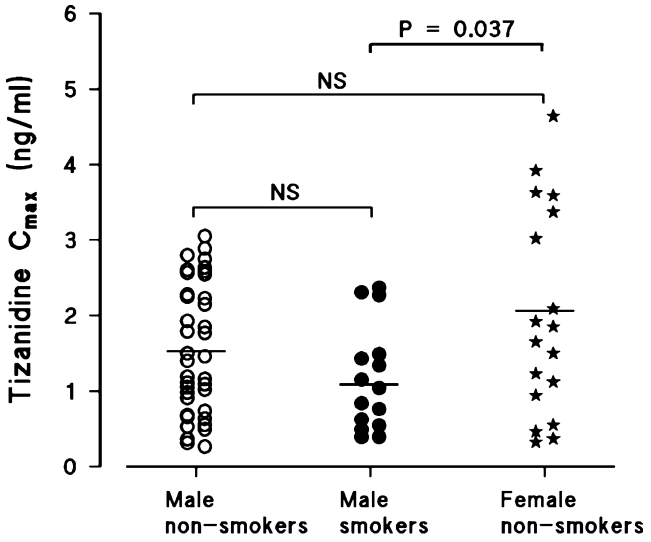
Fig. 1 Mean ± standard deviation (SD) plasma concentrations of tizanidine in 18 female nonsmokers (*stars*) and 38 male nonsmokers (*open circles*) after 4 mg tizanidine (**a**). Mean ± SD plasma concentrations of tizanidine in 15 male smokers (*solid circles*) and 38 male nonsmokers (*open circles*) after 4 mg tizanidine (**b**). *Insets* depict the same mean data on a semilogarithmic scale

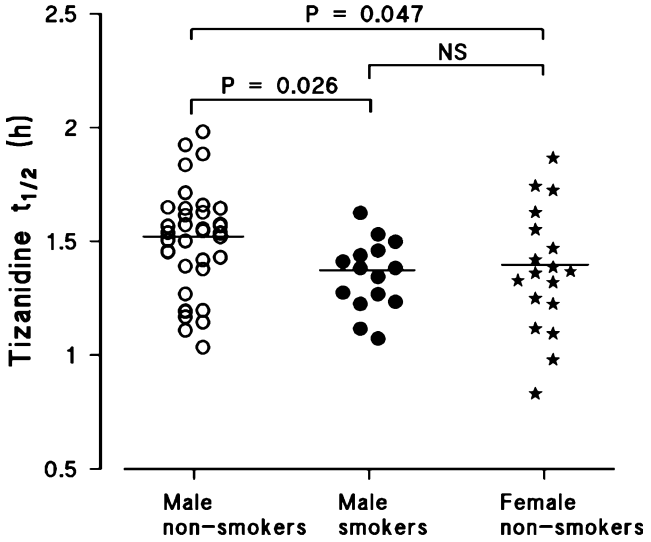
statistically significantly lower in male smokers than in male nonsmokers (Fig. 2, Table 2). However, both the C_{max} and AUC(0-∞) of tizanidine were considerably lower in smoking men than in nonsmoking women ($P<0.05$). The $t_{1/2}$ was 10% shorter in male smokers than in male nonsmokers (Fig. 2; $P<0.05$).

Weight-adjusted pharmacokinetics of tizanidine

On average, the mean weight of the female subjects was about 20% lower than that of the nonsmoking men, and the smoking men were about 10% heavier than the nonsmoking men (Table 1). There was a negative correlation between body weight and C_{max} (Pearson $r=-0.249$, $P=0.064$) and AUC(0-∞) (Pearson $r=-0.274$, $P=0.041$) of tizanidine when all male and female nonsmokers were included. After adjusting the individual C_{max} and AUC(0-∞) values of

Fig. 2 Individual and mean values for peak plasma concentration (C_{max}), elimination half-life ($t_{1/2}$), and area under plasma concentration-time curve from time 0 to infinity (AUC(0-∞) of tizanidine in 38 male nonsmokers (*open circles*), 15 male smokers (*solid circles*), and 18 female nonsmokers (*stars*) after 4 mg tizanidine ▶





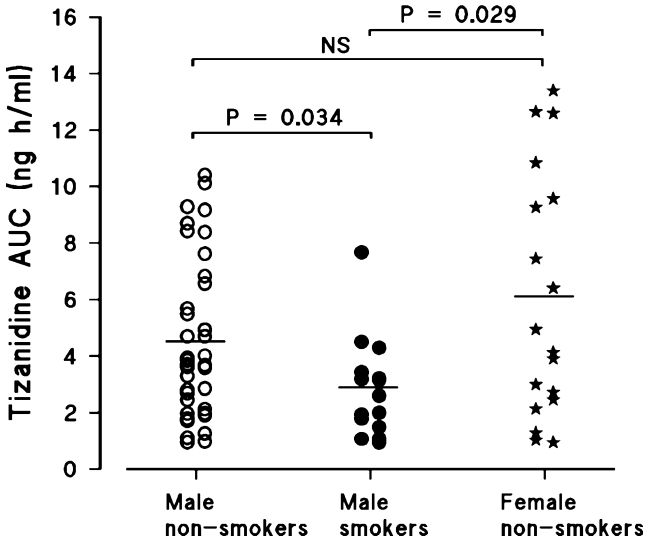


Table 2 Pharmacokinetic variables of 4 mg tizanidine in male nonsmokers, male smokers, and female nonsmokers

Variable	Male nonsmokers (n=38)	Male smokers (n=15)	Female nonsmokers (n=18)
C_{max} (ng/ml)	1.55±0.86	1.16±0.70	2.01±1.36**
$C_{max,\ adj}$ (ng/ml)	1.71±0.98	1.39±0.78	1.70±1.11
t_{max} (min)	60 (40–120)	60 (20–120)	60 (40–120)
$t_{1/2}$ (h)	1.50±0.22	1.35±0.15*	1.37±0.27*
AUC(0-∞) (ng·h/ml)	4.53±2.79	2.83±1.76*	6.04±4.34**
$AUC(0\text{-}\infty)_{adj}$ (ng·h/ml)	5.01±3.20	3.35±1.82*	5.04±3.41

Data are given as mean ± standard deviation (SD), except for t_{max} data, which are given as median and range
Cmax peak concentration in plasma, *adj* weight-adjusted, *tmax* time to reach peak concentration in plasma, *t1/2* half-life, *AUC(0-∞)* area under plasma concentration-time curve from time 0 to infinity
* $P<0.05$ vs. male nonsmokers
** $P<0.05$ vs. male smokers

tizanidine for body weight, the only significant difference between the groups was that the $AUC(0\text{-}\infty)_{adj}$ of tizanidine was 33% (P=0.023) smaller in male smokers than in male nonsmokers (Table 2).

Pharmacodynamic effects of tizanidine

Tizanidine decreased systolic blood pressure and increased subjective drowsiness and drug effect significantly more in female nonsmokers than in male nonsmokers or smokers (Table 3). In addition, the effects of tizanidine on blood pressure were weaker in smoking than in nonsmoking men.

Caffeine test

The caffeine/paraxanthine ratio ranged about 4-fold within the groups of male and female nonsmokers, but the mean ratios were close to each other in both of these groups (Fig. 3). However, the caffeine/paraxanthine ratio was 35–40% smaller in male smokers than in male or female nonsmokers (Fig. 3; $P<0.001$). There was a linear relationship between the AUC(0-∞) of tizanidine and the caffeine/paraxanthine ratio in the entire population (Pearson r=0.696, $P<0.001$).

Discussion

The results of this study show that smoking can affect the pharmacokinetics of tizanidine, a CYP1A2 substrate with an extensive presystemic metabolism; the mean AUC of tizanidine was about 30–40% smaller in male smokers than

Table 3 Pharmacodynamic variables: maximum changes from baseline values (minimum or maximum) and incremental/decremental area under effect vs. time curve from time 0 to 5 h [AUC(0–5)] for blood pressures, psychomotor tests [subjective drowsiness, subjective drug effect, and Digit Symbol Substitution Test (DSST)] after 4 mg tizanidine in male nonsmokers, male smokers, and female nonsmokers

Variable	Male nonsmokers (n=38)	Male smokers (n=15)	Female nonsmokers (n=18)
Systolic blood pressure			
Maximum change (mmHg)	−14±8	−10±6	−17±9[a]
Decremental AUC(0–5) (mmHg·h)	−21±34	−4±24*	−45±37*[c]
Diastolic blood pressure			
Maximum change (mmHg)	−11±6	−8±4	−14±5[b]
Decremental AUC(0–5) (mmHg·h)	−23±25	−11±16*	−36±26[b]
Drowsiness (VAS)			
Maximum change (mm)	15±13	16±13	39±20***[c]
Incremental AUC(0–5) (mm·h)	−22±64	−20±45	47±94**[a]
Drug effect (VAS)			
Maximum change (mm)	10±12	12±16	28±25**[a]
Incremental AUC(0–5) (mm·h)	20±33	16±23	53±54*[a]
DSST			
Maximum change (symbols/2 min)	−9±6	−8±5	−10±6
Decremental AUC(0–5) (symbols/2 min·h)	−7±25	−6±20	−15±19

Data are given as mean ± standard deviation (SD)
VAS visual analog scale
* P<0.05, ** P<0.01, *** P<0.001 vs. male nonsmokers
[a] P<0.05, [b] P<0.01, [c] P<0.001 vs. male smokers

in male nonsmokers. On the other hand, the gender-related differences in tizanidine pharmacokinetics were minor. Yet, the highest individual plasma tizanidine concentrations were observed in nonsmoking women. It should be noted that there were greater than 10-fold interindividual differences independent of gender and smoking habits in the C_{max} and AUC(0-∞) of tizanidine within the study groups. This variability, which is unavoidable in this kind of parallel group study, reduced the statistical significance of the effects of gender and smoking. For obvious reasons, a crossover study design, although statistically more sensitive to demonstrate differences, was not possible.

Fluvoxamine, ciprofloxacin, and rofecoxib, inhibitors of CYP1A2 [32–36], have increased the AUC of tizanidine 33-fold, 10-fold, and 14-fold, respectively [29–31]. These findings, together with in vitro results, suggest that the contribution of CYP1A2 to the total elimination of tizanidine is at least 90%. In contrast to tizanidine, caffeine lacks presystemic metabolism [42]. Accordingly, the caffeine/paraxanthine ratio mainly reflects the CYP1A2-mediated systemic elimination of caffeine [13, 15, 17, 19, 37]. The effects of CYP1A2 inhibitors on the AUC and C_{max} of tizanidine were shown to be greater than their effects on the caffeine test [29–31, 39], suggesting that a CYP1A2 substrate with extensive presystemic metabolism can be particularly susceptible to factors affecting CYP1A2 activity.

In our study, the average plasma concentrations of tizanidine were about 30% higher in nonsmoking women than in nonsmoking men. This statistically nonsignificant difference can be explained by the smaller body size of women compared with men. Accordingly, gender had virtually no effect on the weight-adjusted C_{max} and AUC (0-∞) of tizanidine or on the caffeine test. In some previous studies, CYP1A2 activity, measured using caffeine-based indexes, was slightly lower in women than in men, even when oral contraceptive users were excluded from the analysis [1, 6, 15, 17]. On the other hand, some studies found no gender differences in CYP1A2 activity or in the pharmacokinetics of CYP1A2 substrates [20, 43, 44]. Nevertheless, the effect of gender (per se) on CYP1A2 activity is largely outweighed by the wide interdividual variability in CYP1A2 activity due to other factors. In our study, the half-life of tizanidine was slightly shorter in women than in men. Possible explanations for this finding could be differences in hepatic blood flow or volume of distribution of tizanidine between men and women.

Smoking can increase the clearance and reduce the plasma concentrations of many CYP1A2 substrates, e.g., caffeine, theophylline, melatonin, clozapine, riluzole, lidocaine, and ropivacaine [11, 13, 15–18, 21, 22, 24–26, 45]. In general, the AUC of CYP1A2 substrates without

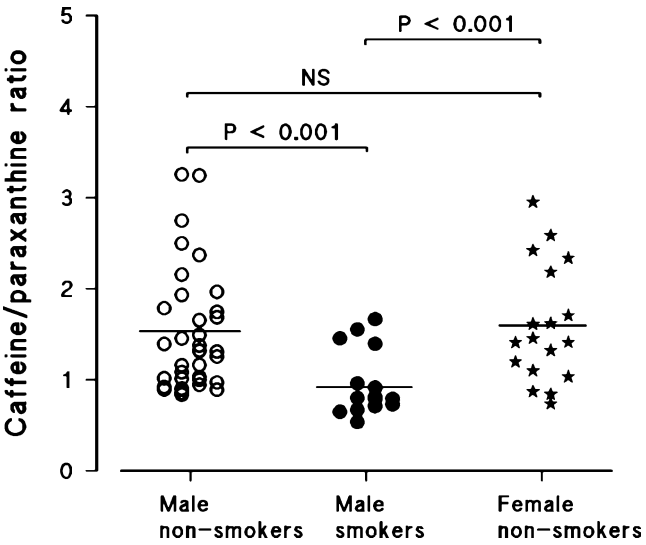


Fig. 3 Individual and mean values for the caffeine/paraxanthine ratio in 38 male nonsmokers (*open circles*), 15 male smokers (*solid circles*), and 18 female nonsmokers (*stars*)

significant presystemic metabolism, such as caffeine and theophylline, is about 30–50% smaller in smokers than in nonsmokers [11, 16, 25, 26]. In our study, the AUC of tizanidine (even after weight adjustment) and the caffeine/paraxanthine ratio were more than 30% smaller, and the half-life of tizanidine was about 10% shorter in smoking men consuming 10–20 cigarettes per day than in nonsmoking men. Thus, our findings suggest that the effect of smoking on CYP1A2 substrates with extensive presystemic metabolism is not markedly different from the effect on substrates without presystemic metabolism. On the other hand, the AUC of melatonin in smokers is more than 50% smaller before smoking abstinence than after a 1-week abstinence [22]. Hepatic CYP1A2 activity decreases to a noninduced level within about 1 week after smoking cessation [18]. However, induction of CYP1A2 by smoking is dependent on the number of cigarettes smoked daily [13, 17, 46]. Accordingly, the effect of heavy smoking on the pharmacokinetics of tizanidine could be greater than what was observed in our study with moderate smokers.

Rifampicin reduces the AUC of tizanidine by about 50%, mainly by inducing its first-pass metabolism (no effect on $t_{1/2}$), and has only a minor effect (23%) on the caffeine test [38]. Thus, the effect of rifampicin seems to differ from that of smoking (30–40% reduction in the caffeine/paraxanthine ratio and a small reduction in the half-life of tizanidine). A possible explanation for this slight difference is that rifampicin could have induced some minor CYP1A2-independent pathway in the elimination of tizanidine.

The hemodynamic and psychomotor effects of tizanidine correlate well with its plasma concentrations [29]. Accordingly, the effects of tizanidine on blood pressure and subjective drug effects were stronger in nonsmoking women than in nonsmoking or smoking men, and its effects on blood pressure were weaker in smoking than in nonsmoking men (Table 3), in line with the differences in mean tizanidine concentrations (Fig. 1). These findings may be of clinical significance. For example, cessation of heavy smoking could increase the AUC of tizanidine by more than 50% and enhance its blood-pressure-lowering and central nervous system effects in a patient whose tizanidine dose has been titrated to its maximum before changes in smoking habits take place. Vice versa, beginning tobacco smoking might decrease the efficacy of tizanidine.

The present findings suggest that some women may be sensitive to the effects of tizanidine due to their smaller body size compared with men. In our study, five of the 18 women had a higher peak plasma tizanidine concentration than any of the men, and the AUC of tizanidine correlated with body weight. An additional factor that can predispose women to the adverse effects of tizanidine is the use of oral contraceptives or postmenopausal hormone replacement therapy, which are established inhibitors of CYP1A2 [6, 14, 15, 17, 20, 25, 47, 48]. Contraceptives containing ethinylestradiol and gestodene have increased the mean C_{max} and AUC(0-∞) of tizanidine about 3- to 4-fold, leading to increased pharmacodynamic effects [39]. In our study, none of the women used hormonal contraceptives because we wanted to study the effect of gender per se.

In conclusion, gender by itself seems to have no clinically significant effect on the pharmacokinetics of tizanidine, whereas smoking reduces tizanidine plasma concentrations and effects. However, any possible effects of gender and moderate smoking are largely outweighed by individual variability in CYP1A2 activity, due to other genetic and environmental factors, and by differences in body weight. Higher than average doses of tizanidine may be required in male smokers, whereas careful dosing of tizanidine may be warranted in nonsmoking individuals with a low body weight, such as many females, and during smoking cessation.

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References

1. Relling MV, Lin JS, Ayers GD, Evans WE (1992) Racial and gender differences in N-acetyltransferase, xanthine oxidase, and CYP1A2 activities. Clin Pharmacol Ther 52:643–658
2. Fletcher CV, Acosta EP, Strykowski JM (1994) Gender differences in human pharmacokinetics and pharmacodynamics. J Adolesc Health 15:619–629
3. Meibohm B, Beierle I, Derendorf H (2002) How important are gender differences in pharmacokinetics? Clin Pharmacokinet 41:329–342
4. Gandhi M, Aweeka F, Greenblatt RM, Blaschke TF (2004) Sex differences in pharmacokinetics and pharmacodynamics. Annu Rev Pharmacol Toxicol 44:499–523
5. Bies RR, Bigos KL, Pollock BG (2003) Gender differences in the pharmacokinetics and pharmacodynamics of antidepressants. J Gend Specif Med 6:12–20
6. Rasmussen BB, Brix TH, Kyvik KO, Brøsen K (2002) The interindividual differences in the 3-demethylation of caffeine alias CYP1A2 is determined by both genetic and environmental factors. Pharmacogenetics 12:473–478
7. Schwartz JB (2003) The influence of sex on pharmacokinetics. Clin Pharmacokinet 42:107–121
8. Cotreau MM, von Moltke LL, Greenblatt DJ (2005) The influence of age and sex on the clearance of cytochrome P450 3A substrates. Clin Pharmacokinet 44:33–60
9. Morris ME, Lee HJ, Predko LM (2003) Gender differences in the membrane transport of endogenous and exogenous compounds. Pharmacol Rev 55:229–240
10. Niemi M, Pasanen MK, Neuvonen PJ (2006) SLCO1B1 polymorphism and sex affect the pharmacokinetics of pravastatin but not fluvastatin. Clin Pharmacol Ther 80:356–366

11. Parsons WD, Neims AH (1978) Effect of smoking on caffeine clearance. Clin Pharmacol Ther 24:40–45
12. Sesardic D, Boobis AR, Edwards RJ, Davies DS (1988) A form of cytochrome P450 in man, orthologous to form d in the rat, catalyses the O-deethylation of phenacetin and is inducible by cigarette smoking. Br J Clin Pharmacol 26:363–372
13. Kalow W, Tang BK (1991) Caffeine as a metabolic probe: exploration of the enzyme-inducing effect of cigarette smoking. Clin Pharmacol Ther 49:44–48
14. Vistisen K, Loft S, Poulsen HE (1991) Cytochrome P450 IA2 activity in man measured by caffeine metabolism: effect of smoking, broccoli and exercise. Adv Exp Med Biol 283:407–411
15. Rasmussen BB, Brøsen K (1996) Determination of urinary metabolites of caffeine for the assessment of cytochrome P4501A2, xanthine oxidase, and N-acetyltransferase activity in humans. Ther Drug Monit 18:254–262
16. Zevin S, Benowitz NL (1999) Drug interactions with tobacco smoking. An update. Clin Pharmacokinet 36:425–438
17. Tantcheva-Poor I, Zaigler M, Rietbrock S, Fuhr U (1999) Estimation of cytochrome P-450 CYP1A2 activity in 863 healthy Caucasians using a saliva-based caffeine test. Pharmacogenetics 9:131–144
18. Faber MS, Fuhr U (2004) Time response of cytochrome P450 1A2 activity on cessation of heavy smoking. Clin Pharmacol Ther 76:178–184
19. Faber MS, Jetter A, Fuhr U (2005) Assessment of CYP1A2 activity in clinical practice: why, how, and when? Basic Clin Pharmacol Toxicol 97:125–134
20. Petersen MS, Halling J, Damkier P, Nielsen F, Grandjean P, Weihe P, Brøsen K (2006) Caffeine N3-demethylation (CYP1A2) in a population with an increased exposure to polychlorinated biphenyls. Eur J Clin Pharmacol 62:1041–1048
21. Jokinen MJ, Olkkola KT, Ahonen J, Neuvonen PJ (2001) Effect of rifampin and tobacco smoking on the pharmacokinetics of ropivacaine. Clin Pharmacol Ther 70:344–350
22. Ursing C, von Bahr C, Brismar K, Röjdmark S (2005) Influence of cigarette smoking on melatonin levels in man. Eur J Clin Pharmacol 61:197–201
23. Rostami-Hodjegan A, Amin AM, Spencer EP, Lennard MS, Tucker GT, Flanagan RJ (2004) Influence of dose, cigarette smoking, age, sex, and metabolic activity on plasma clozapine concentrations: a predictive model and nomograms to aid clozapine dose adjustment and to assess compliance in individual patients. J Clin Psychopharmacol 24:70–78
24. Seppälä NH, Leinonen EV, Lehtonen ML, Kivistö KT (1999) Clozapine serum concentrations are lower in smoking than in nonsmoking schizophrenic patients. Pharmacol Toxicol 85:244–246
25. Gardner MJ, Tornatore KM, Jusko WJ, Kanarkowski R (1983) Effects of tobacco smoking and oral contraceptive use on theophylline disposition. Br J Clin Pharmacol 16:271–280
26. Grygiel JJ, Birkett DJ (1981) Cigarette smoking and theophylline clearance and metabolism. Clin Pharmacol Ther 30:491–496
27. Koch P, Hirst DR, von Wartburg BR (1989) Biological fate of sirdalud in animals and man. Xenobiotica 19:1255–1265
28. Granfors MT, Backman JT, Laitila J, Neuvonen PJ (2004) Tizanidine is mainly metabolized by cytochrome p450 1A2 in vitro. Br J Clin Pharmacol 57:349–353
29. Granfors MT, Backman JT, Neuvonen M, Ahonen J, Neuvonen PJ (2004) Fluvoxamine drastically increases concentrations and effects of tizanidine: a potentially hazardous interaction. Clin Pharmacol Ther 75:331–341
30. Granfors MT, Backman JT, Neuvonen M, Neuvonen PJ (2004) Ciprofloxacin greatly increases concentrations and hypotensive effect of tizanidine by inhibiting its cytochrome P450 1A2-mediated presystemic metabolism. Clin Pharmacol Ther 76:598–606
31. Backman JT, Karjalainen MJ, Neuvonen M, Laitila J, Neuvonen PJ (2006) Rofecoxib is a potent inhibitor of cytochrome P450 1A2: studies with tizanidine and caffeine in healthy subjects. Br J Clin Pharmacol 62:345–357
32. Karjalainen MJ, Neuvonen PJ, Backman JT (2006) Rofecoxib is a potent, metabolism-dependent inhibitor of CYP1A2: implications for in vitro prediction of drug interactions. Drug Metab Dispos 34:2091–2096
33. Harder S, Fuhr U, Staib AH, Wolff T (1989) Ciprofloxacin-caffeine: a drug interaction established using in vivo and in vitro investigations. Am J Med 87:89S–91S
34. Fuhr U, Wolff T, Harder S, Schymanski P, Staib AH (1990) Quinolone inhibition of cytochrome P-450-dependent caffeine metabolism in human liver microsomes. Drug Metab Dispos 18:1005–1010
35. Brøsen K, Skjelbo E, Rasmussen BB, Poulsen HE, Loft S (1993) Fluvoxamine is a potent inhibitor of cytochrome P4501A2. Biochem Pharmacol 45:1211–1214
36. Wang JS, Backman JT, Wen X, Taavitsainen P, Neuvonen PJ, Kivistö KT (1999) Fluvoxamine is a more potent inhibitor of lidocaine metabolism than ketoconazole and erythromycin in vitro. Pharmacol Toxicol 85:201–205
37. Kalow W, Tang BK (1991) Use of caffeine metabolite ratios to explore CYP1A2 and xanthine oxidase activities. Clin Pharmacol Ther 50:508–519
38. Backman JT, Granfors MT, Neuvonen PJ (2006) Rifampicin is only a weak inducer of CYP1A2-mediated presystemic and systemic metabolism: studies with tizanidine and caffeine. Eur J Clin Pharmacol 62:451–461
39. Granfors MT, Backman JT, Laitila J, Neuvonen PJ (2005) Oral contraceptives containing ethinyl estradiol and gestodene markedly increase plasma concentrations and effects of tizanidine by inhibiting cytochrome P450 1A2. Clin Pharmacol Ther 78:400–411
40. Holland DT, Godfredsen KA, Page T, Connor JD (1998) Simple high-performance liquid chromatography method for the simultaneous determination of serum caffeine and paraxanthine following rapid sample preparation. J Chromatogr B Biomed Sci Appl 707:105–110
41. Pickard CE, Stewart AD, Hartley R, Lucock MD (1986) A rapid HPLC method for monitoring plasma levels of caffeine and theophylline using solid phase extraction columns. Ann Clin Biochem 23(Pt 4):440–446
42. Dollery C (1999) Caffeine. In: Dollery C (ed) Therapeutic drugs. Churchill Livingstone, Edinburgh, pp C4–C6
43. Bebia Z, Buch SC, Wilson JW, Frye RF, Romkes M, Cecchetti A, Chaves-Gnecco D, Branch RA (2004) Bioequivalence revisited: influence of age and sex on CYP enzymes. Clin Pharmacol Ther 76:618–627
44. Oztekin S, Mavioglu O, Elar Z, Guven H, Kalkan S, Gurpinar T (2005) The effects of gender and menopause on serum lidocaine levels in smokers. Eur J Drug Metab Pharmacokinet 30:231–234
45. Bruno R, Vivier N, Montay G, Le Liboux A, Powe LK, Delumeau JC, Rhodes GR (1997) Population pharmacokinetics of riluzole in patients with amyotrophic lateral sclerosis. Clin Pharmacol Ther 62:518–526
46. Schrenk D, Brockmeier D, Mörike K, Bock KW, Eichelbaum M (1998) A distribution study of CYP1A2 phenotypes among smokers and non-smokers in a cohort of healthy Caucasian volunteers. Eur J Clin Pharmacol 53:361–367
47. Bock KW, Schrenk D, Forster A, Griese EU, Mörike K, Brockmeier D, Eichelbaum M (1994) The influence of environmental and genetic factors on CYP2D6, CYP1A2 and UDP-glucuronosyltransferases in man using sparteine, caffeine, and paracetamol as probes. Pharmacogenetics 4:209–218
48. Laine K, Palovaara S, Tapanainen P, Manninen P (1999) Plasma tacrine concentrations are significantly increased by concomitant hormone replacement therapy. Clin Pharmacol Ther 66:602–608