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Mia Birkhøj Kjærstad[1*], Flemming Nielsen[1,2], Lene Nøhr-Jensen[2], Stine Zwisler[2], Kim Brøsen[2] & Helle Raun Andersen[1].

[1]*Department of Environmental Medicine, Institute of Public Health, University of Southern Denmark, JB Winsløwsvej 17, DK-5000 Odense C, Denmark*

[2]*Clinical Pharmacology, Institute of Public Health, University of Southern Denmark, JB Winsløwsvej 19, DK-5000 Odense C, Denmark*

*Corresponding author:

Mia Birkhøj Kjærstad, MSc

Tel: +45 6550 3044; fax: +45 6591 1458

E-mail address: mbirkhoej@health.sdu.dk

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Abstract

Purpose: To investigate if the ordinary use of vaginal suppository containing miconazole results in a systemic absorption that is high enough to affect the activities of CYP1A2 and CYP3A4 which are major drug and steroid metabolizing enzymes.

Methods: In 20 healthy non-pregnant women aged 18-45 years, the serum concentration of miconazole was determined following the use of a vaginal suppository containing 1200 mg miconazole. Enzyme activities of CYP1A2 and CYP3A4 were determined as metabolic ratios of caffeine (CMR=(AFMU+1MU+1MX)/17DMU) and quinidine (QMR=(3-hydroxy-quinidine/quinidine), respectively, before and 34 hours after insertion of the suppository. Miconazole was analyzed by LC-MS/MS while caffeine and metabolites were analyzed by HPLC-UV and quinidine and hydroxy-quinidine were analyzed by HPLC-fluorescence.

Results: All 20 women had measurable concentrations of miconazole in serum (mean±SD: 12.9±5.6 µg/L; range: 3.5-24.6 µg/L). Although not statistically significant, an association between the serum concentrations of miconazole and inhibition of CYP1A2 activity was indicated. No relation was observed between the CYP3A4 activity and the miconazole serum concentration.

Conclusions: Miconazole is absorbed via the vaginal mucosa to the systemic circulation in measurable concentrations. Our data indicate a concentration dependent inhibition of CYP1A2, but the effect is negligible compared to the variation in the activity of CYP1A2 and is regarded to be of no clinical significance for the women. However, further studies on the ability of miconazole to be transferred across the placenta or to interfere with the placental function are warranted to secure safe use during pregnancy.

Keywords

miconazole; antifungal; CYP1A2; CYP3A4; vaginal uptake

Introduction

Miconazole (Figure 1) is a pharmaceutical antifungal. It inhibits the fungal enzyme lanosterol 14-α-demethylase (CYP51) which regulates ergosterol synthesis essential for formation of the fungal cell membrane. Miconazole is used for the treatment of oral candidiasis in infants and vaginal mycoses in pregnant women and may reach the systemic circulation of the child and foetus if it is able to cross the mucous membrane and the placental barrier. However, the systemic bioavailability of miconazole following absorption via the vagina and the gut were reported to be low, 1.4% and 27%, respectively [1,2,3], and oral and vaginal absorbed miconazole were eliminated with half lives between 2 and 57 hours [3,4,5]. Nevertheless, drug interactions between oral gel or vaginal used miconazole and the anticoagulant warfarin have been reported indicating a systemic concentration high enough to affect biotransformation [6,7].

In a panel of *in vitro* test systems, miconazole showed potential to disrupt the endocrine system by several mechanisms [8]. Miconazole inhibited testosterone biosynthesis and the oestrogen synthesizing enzyme aromatase (CYP19) and it was a weak oestrogen receptor antagonist. In other experimental test systems, miconazole had unspecific effects on the humane cytochrome P450 (CYP) system and has been reported to inhibit CYP1A2, CYP2C9, CYP2C19, CYP2D6, CYP3A4, CYP17, CYP19, and CYP51 activities [9,10,11,12,13,14]. Though, the inhibitory potency of miconazole differed among different CYPs. CYP17 and CYP19 are involved in steroid synthesis whereas CYP1A2 and CYP3A4 are major drug metabolizing enzymes but they are also involved in oestrogen metabolism [15,16]. Disturbance of these enzymatic pathways may affect homeostasis. The developing foetal and infantile endocrine and reproductive systems are particularly

susceptible to endocrine disrupting substances [17,18,19]. Other conazoles which like miconazole, have been reported to inhibit CYP19 and testosterone synthesis *in vitro*, have been shown to disturb steroid synthesis in rat foetuses after prenatal exposure [20,21]. Consequently there is a concern that miconazole after ordinary use orally or in the vagina could result in systemic concentrations high enough to disturb the endocrine balance of the infant, the pregnant woman, or the foetus.

The primary aim of this study was therefore to investigate whether the ordinary use of vaginal suppository containing 1200 mg of miconazole resulted in a systemic absorption. The secondary aim was then to investigate if the systemic concentration of miconazole was high enough to affect enzyme activities associated with CYP1A2 and CYP3A4.

Materials and Methods

Subjects

Twenty healthy female subjects were recruited in the period of May to August 2008. The participants were mainly students at the University of Southern Denmark or staff at Odense University Hospital. Characteristics of the study subjects are shown in Table 1. After oral and written information about the study and their rights, 30 subjects gave their written consent and a written authority giving relevant personnel from the GCP-Unit and the Danish Medicines Agency access to documents and data of importance for the study. Out of these 30, four subjects regretted their enrolment and withdrew their consent. Four individuals were excluded due to use of medication, and two were excluded since their electrocardiogram (ECG) revealed irregularities. Hence, twenty subjects completed the study.

Inclusion and exclusion criteria

Inclusion criteria were healthy non-pregnant (evaluated by no detection of human chorionic gonadotrophin in a urine sample) and non-breastfeeding women aged between 18 and 45 years. They were ascertained to be in good health based on clinical examination including blood pressure, medical history, and evaluation of a standard 12-lead ECG done by the physician responsible for the study. Subjects with a daily alcohol or medical use including oral contraceptives or known allergy to miconazole, caffeine, quinidine, sodium methyl 4-hydroxybenzoate (E219), or sodium propyl 4-hydroxybenzoate (E217) were excluded. Smokers were not excluded since smoking is known to have an inducing effect on

CYP1A2, not an inhibitory effect, and since the subjects were their own controls and exclusion of smokers might hamper the recruitment of subjects.

Ethics

The study was approved by The Regional Scientific Ethical Committee for Southern Denmark (record no: S-20080022), the Danish Medicines Agency (record no: 2612-3691), and the Danish Data Protection Agency (record no: 2008-41-2011). The study was performed according to the Helsinki-II-Declaration and in accordance with guidelines of Good Clinical Practice (ICH-GCP) as monitored by the GCP-Unit, Odense University Hospital (record no: 07.035).

Study design and procedures

Initially, all participants answered a questionnaire regarding vegetable and fruit intake, smoking status, job function, and use of antifungals in the homes including personal hygiene products such as shampoo or cream.

For each subject the study lasted 11 days (Figure 2). It was a two-period study with a wash-out period of 2 days before each caffeine and quinidine administration period and of 5 days after insertion of the miconazole containing vaginal suppository.

The subjects were asked to abstain from ingesting quinine, quinidine, and methylxanthine containing food and beverages (specific items were pointed out) as well as alcohol and medication, from 48 hours before caffeine and quinidine administration and until the last blood sampling at day 8. To avoid pregnancy in the study period the subjects were asked to use condom when having intercourse from the time of pregnancy test until the insertion of

the vaginal suppository. Thereafter sexual absence was required until the end of trial at day 11.

Each subject ingested 200 mg of caffeine (batch-nr. 2151631) and 200 mg of quinidine (batch-nr. 51313011), both supplied by the Central Pharmacy, Odense University Hospital (Denmark), at 11pm on day 3. Previous studies indicate that simultaneously administrations of low doses of these two compounds do not influence the outcome of their respective metabolic ratios [22,23,24]. Morning urine was collected after a minimum of three hours. Ten ml of the urine was poured into a conical tube containing 300 µl 1 M hydrochloric acid (HCl) for the CYP1A2 activity analysis. Forty ml was transferred into each of two sterile empty test-tubes for the miconazole analysis. The urine samples were brought to the laboratory and stored at -20 ºC until analysis. At 9-10 hours after quinidine administration, blood samples (5×10 ml) were collected by vein puncture at the laboratory for the CYP3A4 activity and miconazole analysis. Blood sampling was done by either the technician or the physician involved in the study. Twenty ml was collected in heparin tubes and 30 ml in dry tubes. All blood samples were centrifuged for 10 minutes at 3500 rpm and plasma and serum were transferred to sterile cryogenic vials and kept frozen at -20 ^{o}C until analysis.

On day 6 at 11 pm the vaginal suppository was inserted and the volunteers were instructed to lie down afterwards for the night. Twenty-four hours later caffeine and quinidine were administered again for the evaluation of CYP1A2 and CYP3A4 activity after miconazole exposure. The urine and blood sampling procedures were repeated.

The time schedule was chosen based on the findings of Daneshmend (1986) [3] which showed a continuous maximally miconazole concentration in plasma from 6 to 48 hours after insertion of a 1200 mg miconazole containing suppository combined with knowledge from caffeine [25] and quinidine [23] kinetic studies.

Determination of miconazole in serum

The serum concentration of miconazole was quantified by liquid chromatography coupled to a Thermo TSQ Quantum Ultra triple quadrupole mass spectrometer (LC-HESI-MS/MS system from Thermo Scientific, Waltham, MA). Extraction procedure: A volume of 250 µl of the serum sample, 60 µl 0.2 M sodium hydroxide and 50 µl of internal standard (0.5 ng/ml tebuconazole in ethanol) was briefly vortex mixed in a 10 ml centrifugation tube. Hereafter 1 ml *tert*-butyl methylether was added and the samples were mixed horizontally for 20 minutes at 200 rpm followed by 15 minutes centrifugation at 3000 *g*. The supernatant was transferred to a new extraction tube. The extraction procedure was repeated. The two supernatants were combined and evaporated to accurate dryness at 40 °C under a gentle stream of nitrogen. The dried extract was dissolved in 125 µl acetonitrile (ACN) followed by 125 µl buffer solution (2 mM ammonium acetate and 2 mM acetic acid, pH 4.6). The sample was centrifuged for 10 minutes at 3000 *g*, transferred to a 300 µl sample vial, and injected (30 µl) onto the analytical system.

The separation was performed on a Phenomenex C12 MAX-RP column (4 µm, 150 x 2 mm) kept at 20 °C using a two solvent gradient system: (2 mM ammonium acetate and 2 mM acetic acid), pH 4.6 : ACN (95:5) (A) and (5:95) (B). The gradient profile was: 0 to 4.37 min: 40 % to 100 %B; 4.38 to 10.00 min: 100 % B; 10.01 to 11.50 min: 100 % to 40% B; 11.51 to 15.00 min: 40% B. The flow rate was 0.3 ml/min. The MS/MS-system was operated with positive polarity in SRM mode with a scan width of 0.2 m/z and a scan time of 0.250 sec. The spray voltage was 3500 V, the vaporizer temperature 350°C, the sheath gas pressure 60 AU, the aux gas pressure 45 AU and the capillary temperature 315°C.

Miconazole was quantified with a parent mass of 416.86 m/z fragmented to a product mass of 160.95 m/z, and was detected at a retention time of 8.66 minutes. Tebuconazole was quantified with a parent mass of 308.1 m/z to the product masses of 124.8 and 70.0 m/z and was detected at a retention time of 5.91 minutes.

Quantitation of miconazole was based on calibration curves obtained after addition of known concentrations of the compound to blank serum and water samples and then extracted and analyzed as described above. The recovery of the extraction method was > 90 %. The linearity was investigated in a range from 0.1 – 50.0 ng/ml ($R^2 > 0.98$). The interday repeatability (n=3) was < 11 %. The accuracy for the quality control samples spiked to a concentration of 5 ng/ml was -4 %. The limit of determination (LOQ) for miconazole was 0.10 ng/ml and the limit of detection (LOD) was 0.05 ng/ml.

Determination of CYP1A2 activity

Four metabolites of caffeine: 5-acetylamino-6-formylamino-3-methyluracil (AFMU), 1-methyluric acid (1MU), 1-methylxanthine (1MX), and 1,7-dimethyluric acid (17DMU) were determined in morning urine samples conserved with 1M HCl by high-performance liquid chromatography with UV detection [26]. The CYP1A2 activity was estimated by the caffeine metabolic ratio (CMR) = (AFMU+1MU+1MX)/17DMU.

Determination of CYP3A4 activity

Quinidine and 3-hydroxy-quinidine in plasma samples collected 9-10 hours after ingestion of 200 mg quinidine were analyzed using high-performance liquid chromatography as described previously [27]. The CYP3A4 activity was estimated by the quinidine metabolic ratio (QMR) = 3-hydroxy-quinidine/quinidine as suggested by [23].

Data analysis

The mean miconazole absorption was tested by a paired t-test comparing the mean miconazole concentrations in serum 34 h after and before the start of miconazole exposure. The differences in metabolic ratios are expressed as the relative differences e.g. (CMR_{after}-CMR_{before})/CMR_{before}. The mean relative differences in CMR and QMR during and before exposure to miconazole were analysed by one-sample t-tests comparing the relative differences to zero. The assumptions of normal distribution were considered fulfilled after evaluation by Q-Q plot (not shown) of the differences. The variations were roughly constant along the x-axis. Relative difference (Bland Altman) plots of CMR and QMR were used as supplements to analyse the deviation between the metabolic ratios during and before exposure to miconazole.

The association between the relative difference in CMR or QMR and the miconazole concentration in serum was examined by linear regression.

P-values <0.05 were considered statistically significant. The statistical analyses were performed using Stata/SE 9.0 for Windows (Texas, USA). Sample size calculations were based on the primary outcome, absorption of miconazole. As the background concentration of miconazole was zero and the LOQ = 0.10 µg/L we consider a concentration of at least 1 µg/L as an indication of absorption. Twenty women were considered suitable to detect miconazole in serum and would give a power >90% at a significance level of 5 %.

Results

Subjects

No subjects had adverse drug reactions. Seventeen subjects had one or more side effects. The side effects occurring were expected: head ache, trouble sleeping, diarrhea, palpitations, and vaginal itching/discomfort. All side effects were followed up and all subjects were well again after the study. Three subjects had no side effects.

Miconazole

The mean serum concentrations of absorbed miconazole in the 20 women after 34 hours use of a vaginal suppository containing 1200 mg miconazol was 12.9 ± 5.6 µg/L (mean±SD) (range: 3.5-24.6 µg/L) (Figure 3). Background serum concentrations of miconazole were below the limit of detection. The absorption of miconazole was not associated with information on menstruation during the study, age, or BMI (data not shown). The miconazole concentration in the urine samples was very high due to leakage from the vagina and therefore the urine concentration could not be used as indication of the absorbed concentration of miconazole.

CYP1A2 and CYP3A4 activity

During the chromatographic detection of caffeine and metabolites, interference from the urine sample hindered the separation of AFMU in six samples. Consequently, only 14 out of 20 subjects were included in the calculations of CYP1A2 activity. One subject had trouble swallowing the quinidine capsule and therefore 19 out of 20 subjects were included in the CYP3A4 activity determination. The urine sampling time after caffeine administration differed between the two administrations with more than one hour for five subjects, while for the rest of the subjects the difference was close to zero.

Data of CMR are presented as scatter plot showing the relative difference of CMR between the two exposure-periods against the average of CMR of the two periods (Figure 4a). The mean relative CMR difference (the horizontal full line) was shifted a bit below zero, but this was not statistically significant (Table 2). One subject had high CMRs compared to the other subjects and a relatively large change in CMR (the dot furthest to the right).

QMR data are presented in Figure 4b. The mean relative QMR difference (the horizontal full line) was shifted a bit above zero, but this was not statistically significant (Table 2). One subject (not the same having high CMRs above) had a relatively large change in quinidine metabolism.

The associations between the relative difference in CMR or QMR and serum miconazole concentrations are shown in Figure 5. A low non-statistically significant decrease ($p=0.33$) in relative CMR difference with increased concentration of miconazole in serum was indicated. There was no concentration dependent association ($p=0.89$) between QMR and miconazole.

Discussion

In this study we demonstrated that the use of a 1200 mg miconazole containing vaginal suppository resulted in a mean serum concentration of miconazole of 12.9 ± 5.6 µg/L ($n=20$) after 34 hours use. No substantial mean differences in CYP1A2 and CYP3A4 activities were observed after exposure to miconazole (exposure range: 3.5-24.6 µg/L serum). However, a weak negative and none-statistically significant association between the serum concentrations of miconazole and the relative difference in CMR was seen, indicating inhibition of CYP1A2. Due to considerable interindividual variations in the

metabolic ratios and in the absorbed miconazole concentration only marked effects on enzyme activities can be detected in this study. Post hoc calculations using the standard deviation obtained from this study show that a true relative difference of at least 0.2 could be demonstrated for both CMR (n=14) and QMR (n=19) activities at a significance level of 5 % and with a power of 80%. The association should therefore be interpreted with caution.

The large variation in serum miconazole absorption between subjects is in accordance with a previous study (mean serum concentration: 10.4±4.2 μg/L, n=11) [3]. The reason for this variation is not clarified but might be due to differences in vaginal environment and physical activity. In our study, neither menstruation during the study nor the duration of vaginal miconazole leakage (subjective evaluation of vaginal discharge) seemed to influence the systemic concentration of miconazole. The subjects were not examined for vaginal infections but a damaged mucous membrane could probably absorb more miconazole, similar to when slightly damaged skin increased the penetration of pesticides in an *in vitro* skin model [28]. Also, genetic variations in the biotransformation of miconazole could play a role [29]. Other formulations of vaginal miconazole, i.e., vaginal cream are not expected to give higher or lower systemic levels as the application is repeated every day for two weeks resulting in a similar total local dose as with the suppository.

The mean CMR (CYP1A2 activity) estimated before the exposure to miconazole was 4.3±1.5. This was very similar to the values obtained in two other studies with non-smoking Caucasian women not using oral contraceptives [26,30]. In a study with Faroese residents the overall mean (not adjusted for confounders such as gender, smoking, oral contraceptives) was 7.8±5.8 (n=307) [31] which was statistically significant different from a Danish twin study with a mean value of 5.9±3.4 (n=378) [30] indicating that either

genetic or environmental factors or both should be considered when measuring and comparing CYP1A2 activity.

None of the women in our study used oral contraceptives or other medical products. All women were Caucasian except one whose genetic background was unknown. This woman smoked occasionally and she was the only smoker included in the determination of CYP1A2 activity. No obvious deviation from the results of the other women was observed for this subject. Intraindividual variability may derive from difference in morning urine sampling time before and during miconazole exposure as the time interval differed with up to four hours. It could also be speculated that the menstrual cycle could influence the activities of the enzymes and add to the variability, though studies investigating the effect of the menstrual cycle on CYP1A2 and CYP3A4 activities did not report such an effect [32,33,34].

The mean CYP3A4 activity (based on QMR) was not affected by exposure to miconazole. The wide interindividual variability was in accordance with other studies determining CYP3A4 based on the urinary 6β-hydroxycortisol/cortisol ratio or formation clearance of 3-hydroxyquinidine [23,35]. One woman had a decreased CYP3A4 activity during the use of the miconazole vaginal suppository, but it was not the woman with the highest concentration of absorbed miconazole. The cause has not been identified, as the subjects were instructed to refrain from known CYP3A4 inhibitors. Miconazole and quinidine are both known inhibitors of P-glycoprotein and also likely substrates for this efflux transporter [36,37,38,39]. Intake of quinidine during miconazole exposure could be speculated to reduce the hepatic elimination of miconazole causing an elevation of the systemic concentration of miconazole. By inhibiting P-glycoprotein, miconazole use during pregnancy might also increase the placental transfer of other exogenous compounds that are

P-glycoprotein substrates. In placenta, P-glycoprotein is expressed both at the membrane facing maternal circulation and at the membrane facing fetal circulation [40]. If not metabolized in placenta, miconazole concentration may increase in this tissue when pregnant women are in treatment with miconazole.

Miconazole has been reported to inhibit several CYP-enzymes in human liver or lymphoblast microsomes *in vitro*, including CYP1A2 and CYP3A4, with an inhibition constant (K_i) of 3.2 and 0.028 µM [14] and a half maximal inhibitory concentration (IC_{50}) of 2.9 and 0.074 µM [41,42], respectively. This indicates, that miconazole in the mean absorbed concentration of 12.9 µg/L serum corresponding to 0.03 µM could have an effect on the enzymes *in vivo* also.

Miconazole is used for treatment of vaginal mycoses, also in pregnant women, and of oral candidiasis in infants as these treatments are regarded effective and safe in regard to a claimed negligible absorption [2,3]. The mean serum miconazole concentration of 12.9µg/L was indeed low but maintained for several days [3]. The measured biological effect in individuals using the miconazole containing suppository was negligible. However, orally administered miconazole has *in vivo* been reported to interact with the CYP2C9 substrate warfarin [43,44,7,45]. Hence, if miconazole is present in a sufficient systemic concentration, a biological effect *in vivo* of miconazole is possible. Therefore the potential adverse effects on the developing foetus and infant after miconazole exposure should be investigated further *in vivo*. Furthermore, it should be investigated if miconazole is transferred to the foetus or if miconazole concentration is increased in the placenta with the possibility to disturb development of the foetus or the placental function, respectively.

In conclusion, miconazole is absorbed via the vaginal mucosa to the systemic circulation in measurable concentrations. Our data indicate a concentration dependent inhibition of

CYP1A2, but the effect is negligible compared to the variation in the activity of CYP1A2 and regarded to be of no clinical significance for the women. However, further studies on the ability of miconazole to be transferred across the placenta or to interfere with the placental function are warranted to secure safe use during pregnancy.

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Conflict of Interest Statement

The authors declare that there are no conflicts of interest.

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Table 1 – Characteristics of the study subjects

Characteristic	
No. of subjects completing the study	20
Age, years	26 (23-43)
Weight, kg	65 (52-95)
Height, cm	169 (158-180)
BMI, kg/m^2	22.2 (18.5-31.0)
Race and nationality	16 Caucasian - Danish 3 Caucasian - Norwegian 1 unknown - Danish
Work	14 students 5 hospital workers 1 maternity leave
No. of subjects menstruating during study	9
Contraceptives	3 none 10 condom 7 intrauterine device made of cupper
Non-smokers	18

For continuous variables, data represent median (range)

Table 2 – Serum concentration of miconazole and the caffeine (CMR) and quinidine (QMR) metabolic ratio in women before and 34 h after start exposure to miconazole

	Mean±SD	Range	95% confidence interval	t-test (p-value)
Miconazole (µg/L; n=20)	12.9±5.6	3.6-24.6	10.3-15.5	<0.0001
CMR_{before} (n=14)	4.3±1.5	2.4-8.7	3.4-5.2	
CMR_{after}	3.9±0.9	2.4-6.4	3.4-4.5	
Relative $CMR_{difference}$	-0.035±0.24	-0.34-0.41	-0.17-0.11	0.60
QMR_{before} (n=19)	0.18±0.05	0.09-0.27	0.15-0.20	
QMR_{after}	0.18±0.06	0.07-0.32	0.15-0.21	
Relative $QMR_{difference}$	0.024±0.25	-0.63-0.43	-0.098-0.15	0.68

Note: Miconazole before exposure was below the limit of detection. The t-test was performed on the difference (after-before)

Figure captions

Fig. 1

Chemical structure of miconazole, a pharmaceutical imidazole fungicide

Fig. 2

Time schedule of the trial

Fig. 3

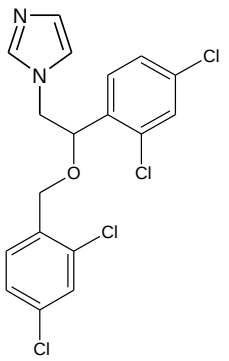
Miconazole concentration measured in serum after 34 hours use of a 1200 mg miconazole containing suppository. Each bar represents one subject. Background concentrations of serum miconazole were zero. The horizontal full line represents the mean (n=20) and the two horizontal dashed lines represent ±SD of the mean

Fig. 4

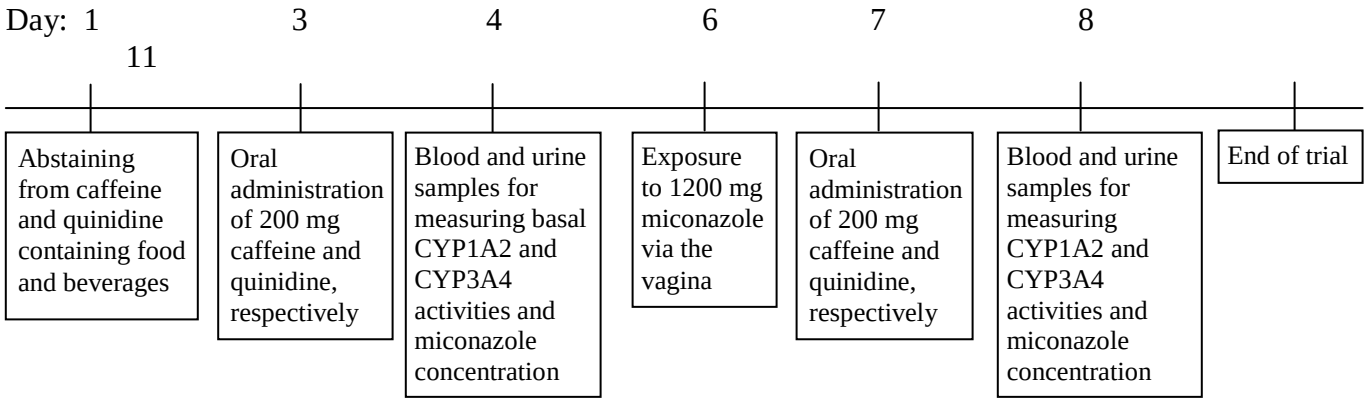
Relative difference (Bland Altman) plot of CYP1A2 activity determined by the caffeine metabolic ratio (CMR=(AFMU+1MU+1MX)/17DMU) (a) and of CYP3A4 activity determined by the quinidine metabolic ratio (QMR=3-hydroxy-quinidine/quinidine) (b). The plots help analyzing the deviation between the metabolic ratios determined 34 hours after and before exposure to miconazole started. The horizontal full line represents mean in relative CMR or QMR difference. The horizontal dashed lines represent the limits of agreement (±1.96*SD)

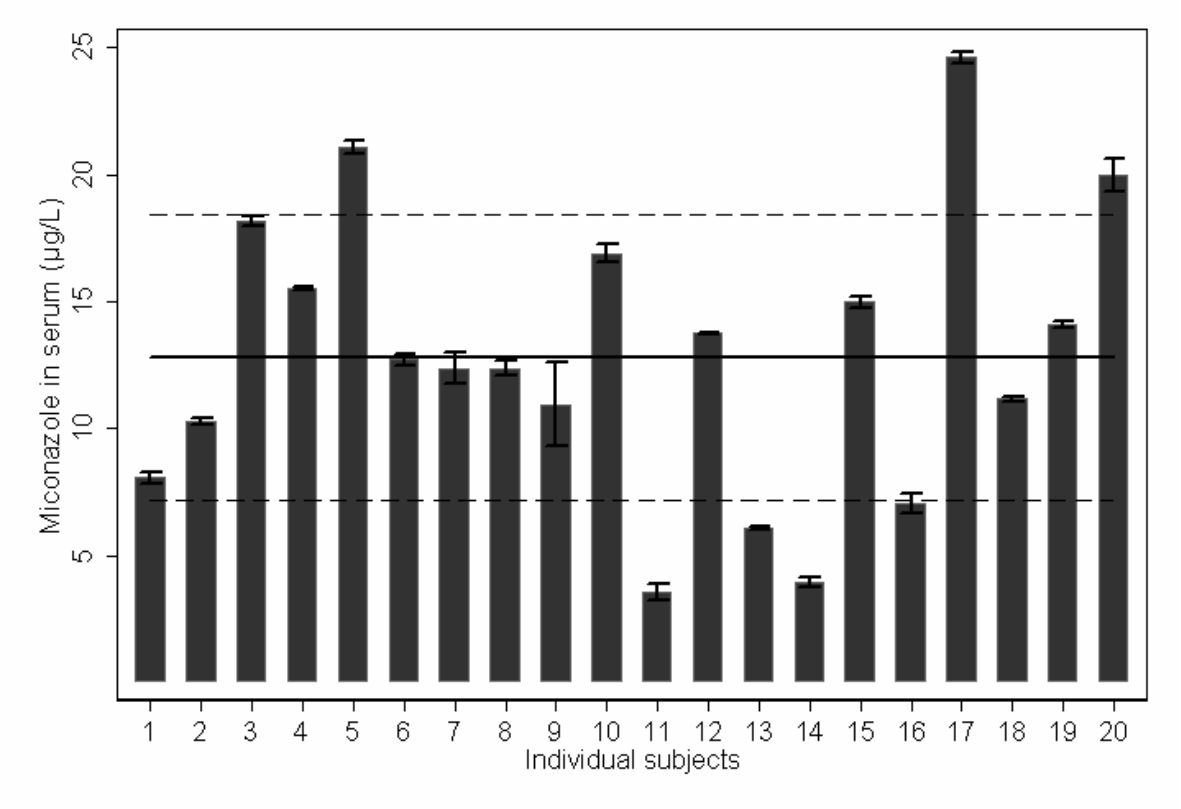
Fig. 5

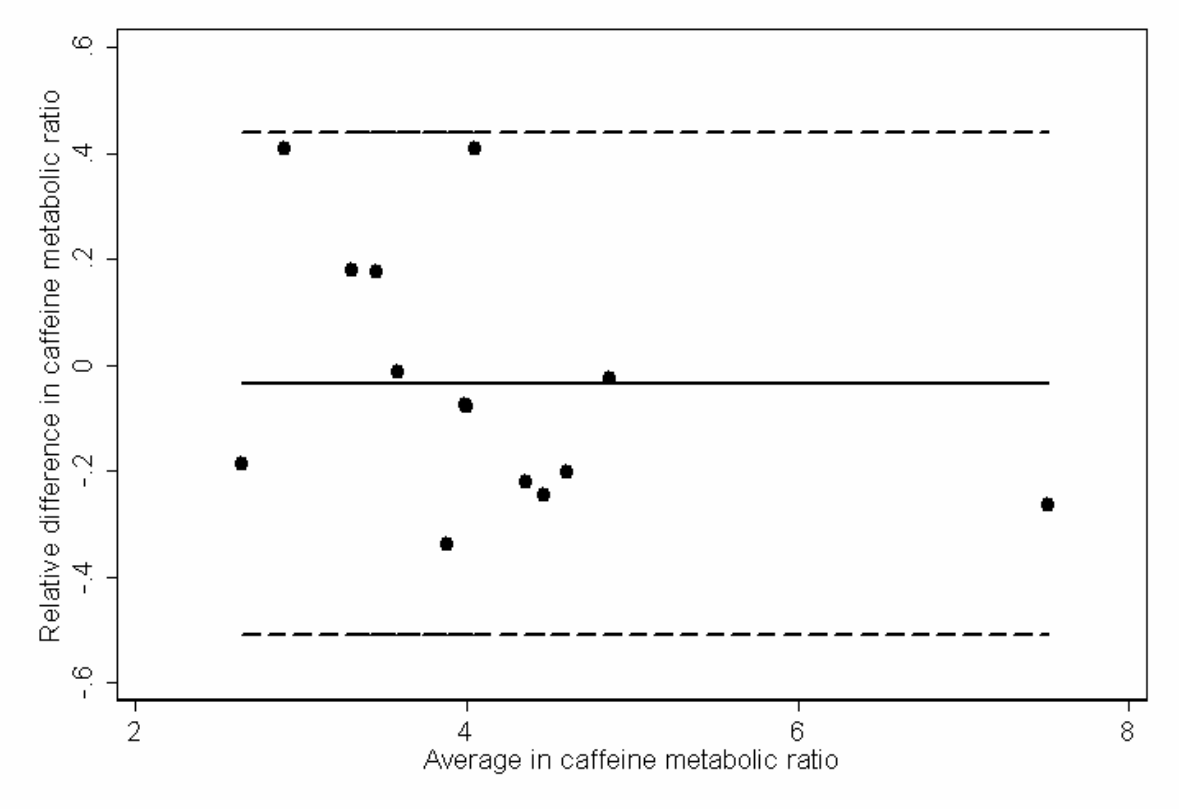
Correlation between CYP1A2 activity determined by the relative difference of caffeine metabolic ratio (a) or CYP3A4 activity determined by the relative difference of quinidine metabolic ratio (b) and miconazole concentration in serum. In plot (a) the estimated association is described by $y = - 0.0121 \times [\text{miconazole}]+0.128$ with $R^2 = 0.0738$ (P=0.33). In plot (b) no association was indicated as $y = 0.0016 \times [\text{miconazole}]+0.003$ with $R^2 = 0.0013$ (P=0.89)

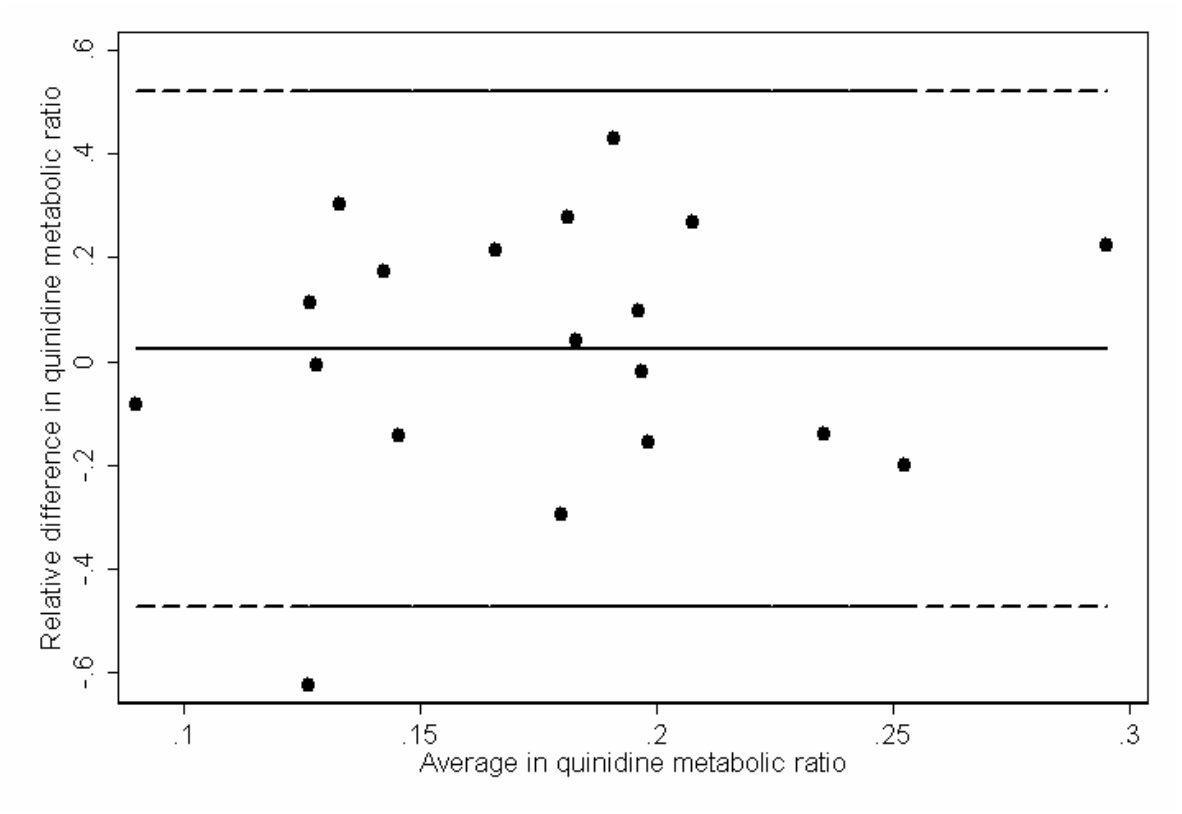


Miconazole


Day: 1
11
3
4
6
7
8
Abstaining from caffeine and quinidine containing food and beverages
Oral administration of 200 mg caffeine and quinidine, respectively
Blood and urine samples for measuring basal CYP1A2 and CYP3A4 activities and miconazole concentration
Exposure to 1200 mg miconazole via the vagina
Oral administration of 200 mg caffeine and quinidine, respectively
Blood and urine samples for measuring CYP1A2 and CYP3A4 activities and miconazole concentration
End of trial


Miconazole in serum (µg/L)
5
10
15
20
25
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20
Individual subjects


Relative difference in caffeine metabolic ratio
-.6
-.4
-.2
0
.2
.4
.6
2
4
6
8
Average in caffeine metabolic ratio


Relative difference in quinidine metabolic ratio
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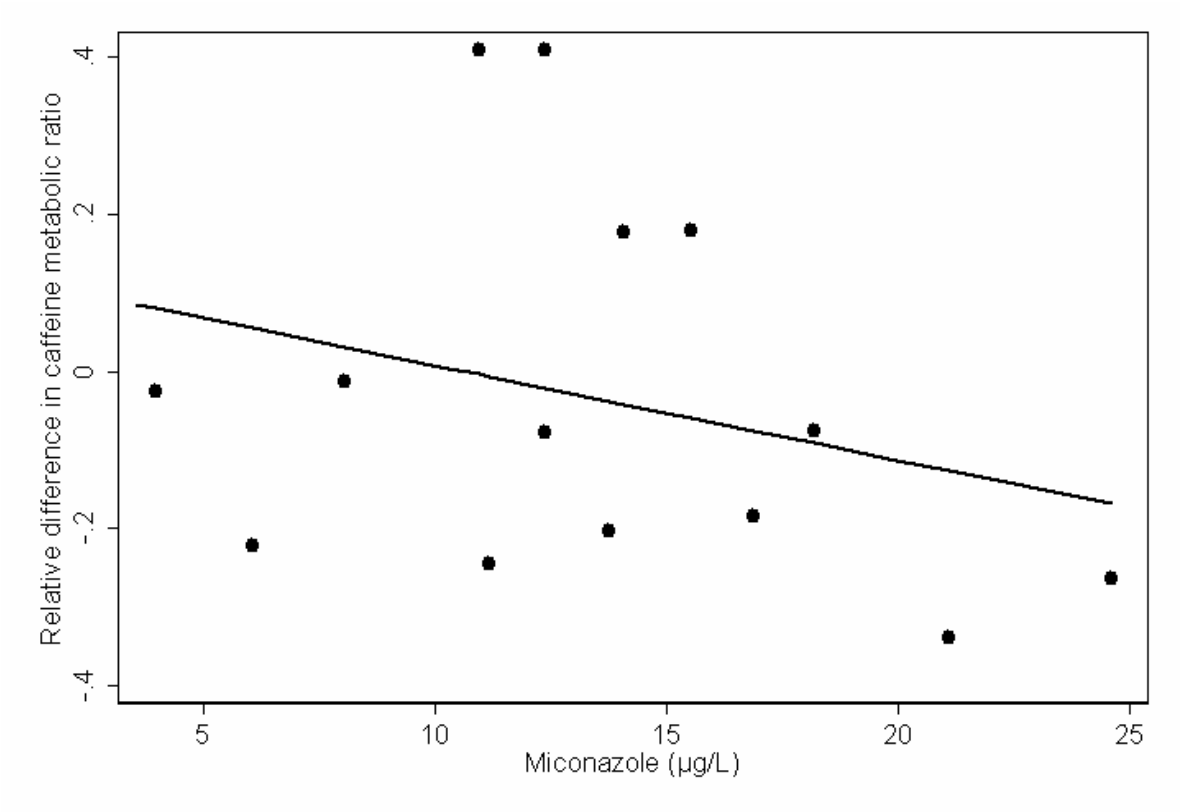
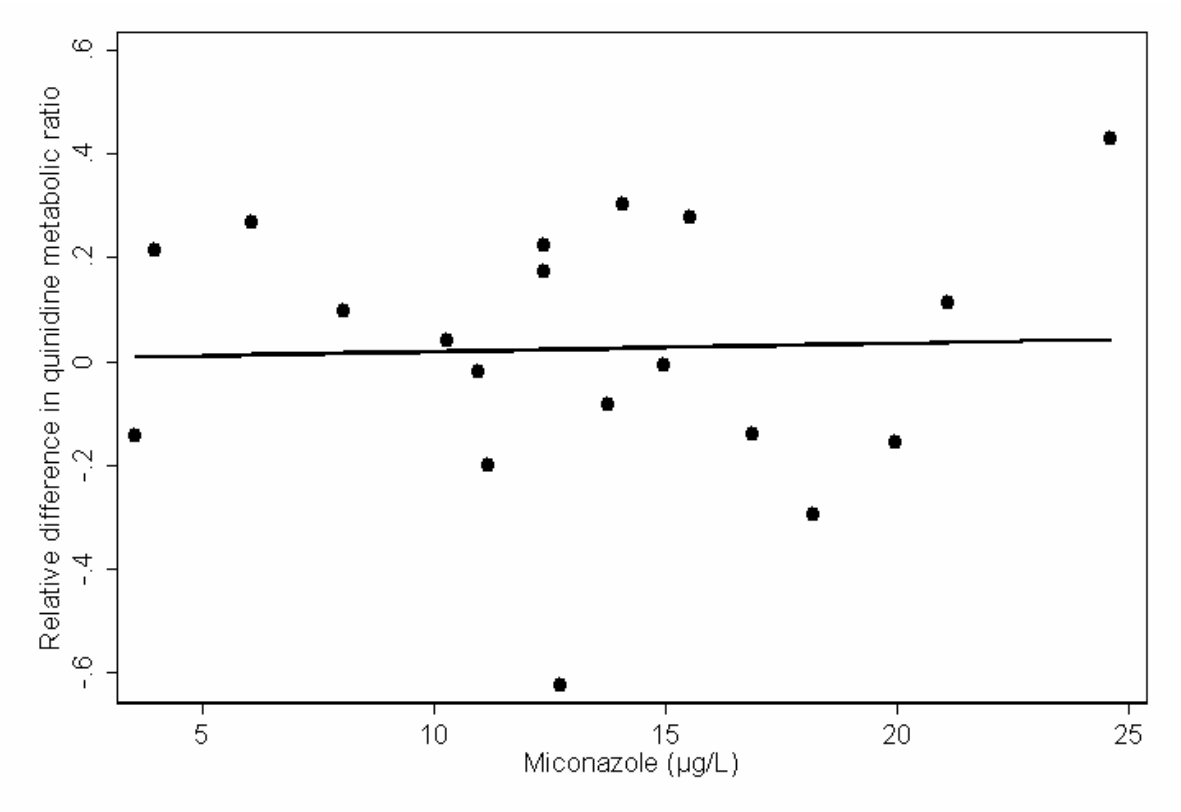
-.4
-.2
0
.2
.4
.6
.1
.15
.2
.25
.3
Average in quinidine metabolic ratio


Relative difference in caffeine metabolic ratio
-.4
-.2
0
.2
.4
5
10
15
20
25
Miconazole (µg/L)


Relative difference in quinidine metabolic ratio
-.6
-.4
-.2
0
.2
.4
.6
5
10
15
20
25
Miconazole (µg/L)