The effect of ethinylestradiol-containing contraceptives on the serum concentration of olanzapine and *N*-desmethyl olanzapine

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WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT

- Olanzapine is metabolized via CYP1A2, which is inhibited by ethinylestradiol-containing contraceptives (ECC).

WHAT THIS PAPER ADDS

- A lower serum concentration of the metabolite *N*-desmethyl olanzapine during co-administration of ECC confirms a CYP1A2-inhibitory action of ethinyl estradiol.
- The CYP1A2-inhibitory action of ethinylestradiol does not provide a clinically relevant change in serum concentrations of unmetabolized olanzapine.
- Concurrent use of ECC is not expected to increase the side effect risk of olanzapine

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AIM

To investigate the potential interaction between olanzapine, a CYP1A2 substrate, and ethinylestradiol-containing contraceptives (ECC).

METHODS

The study was carried out at a routine therapeutic drug monitoring service. To identify patients who were co-administered ECC or other contraceptives, a questionnaire was sent to the physician who ordered serum monitoring of olanzapine for women aged 18–40 years during an 18 month period. The physicians were asked to provide information about contraceptive use and smoking habits. When questionnaires were returned by the physicians, the respective serum concentration data were included in the analysis. Patients were stratified into users of ECC, progestogen-based contraceptives (PBC) or no contraceptives. Dose-adjusted serum concentrations of olanzapine and the metabolite *N*-desmethyl olanzapine were compared between the subgroups.

RESULTS

A total of 149 patients were included in the study (10 ECC users and 10 PBC users). In users of ECC, we found no differences in serum concentrations of olanzapine, but significantly lower concentrations of the CYP1A2-mediated metabolite *N*-desmethyl olanzapine compared with users of PBC ($P = 0.019$) and non-contraceptive users ($P = 0.012$).

CONCLUSION

The present study confirms that ECC exhibit CYP1A2-inhibitory properties in terms of significantly lower exposure of *N*-desmethyl olanzapine. However, the inhibition does not provide clinically relevant changes in serum concentrations of olanzapine.

Introduction

Olanzapine is one of the most frequently used atypical antipsychotic agents in the treatment of schizophrenia and bipolar disorder. Olanzapine was introduced in 1996 and has been shown to be superior to most antipsychotics in terms of clinical effect [1]. On the other hand, olanzapine is more frequently associated with side effects, such as weight gain, metabolic disorders and type II diabetes mellitus, compared with other atypical antipsychotics [2]. These serious side effects, predisposing psychiatric patients to increased risk of cardiovascular events, are related to the systemic exposure of olanzapine [3, 4].

There is an extensive variability in the pharmacokinetics of olanzapine. In patients receiving a similar dose, serum concentrations of olanzapine have been shown to range more than 25-fold [5]. Individual differences in metabolism via cytochrome P450 1A2 (CYP1A2) are likely to be an important cause behind this variability, which has been associated with both genetic and non-genetic factors [6–8]. One important non-genetic factor is tobacco smoking, which induces CYP1A2 metabolism and substantially reduces serum concentrations of olanzapine [8]. On the contrary, concurrent use of CYP1A2 inhibitors, e.g. fluvoxamine and ciprofloxacin, has been reported to increase serum concentrations and the side effect risk of olanzapine [9, 10].

Estradiol-containing contraceptives (ECC) have been reported to increase systemic exposure of the CYP1A2 probe substrates tizanidine and caffeine [6, 11]. However, the potential impact of ECC on the pharmacokinetics of olanzapine has not previously been investigated. Thus, we studied the effect of ECC on serum concentrations of olanzapine and the metabolite *N*-desmethyl olanzapine, which is formed via CYP1A2.

Methods

Patients

The patients were prospectively recruited from a routine therapeutic drug monitoring (TDM) service at the Centre for Psychopharmacology, Diakonhjemmet Hospital, Oslo, Norway. All TDM files from olanzapine-treated female patients aged 18–40 years were considered for inclusion during an 18 month period (July 2007–December 2008). Patient information was extracted from the standard TDM requisition form, including entered concomitant medication, time interval between drug intake and serum sampling, and daily dosage of olanzapine. Criteria for inclusion were (i) a time interval of 10–30 h between last drug intake and serum sampling for analyte measurements, and (ii) measured analyte concentrations above the lower limit of quantification. Patients co-medicated with known inhibitors (fluvoxamine, ciprofloxacin) or inducers (carbamazepine, phenytoin and phenobarbital) of olanzapine

metabolism, as identified by browsing of TDM requisition forms, were excluded. Rofecoxib and enoxacin are also CYP1A2 inhibitors, but comedication with these agents was considered unlikely as rofecoxib was withdrawn from the global market in 2004 and enoxacin is not licensed for use in Norway.

For patients who fulfilled the inclusion criteria, a questionnaire was sent along with the TDM report to the physician who ordered serum monitoring of olanzapine. In the questionnaire, the physicians were asked to provide information about use of contraceptives and smoking habits. If contraceptives were used, information about the type of agent, including both brand- and generic name, was registered. Information about weight and height of the patients was not collected.

The study was approved by the Regional Committee for Medical and Health Research Ethics, as well as the Hospital Investigational Review Board.

Determination of olanzapine and *N*-desmethyl olanzapine

Serum concentrations of olanzapine and *N*-desmethyl olanzapine were determined by a fully validated UPLC-MS/MS method developed for routine TDM analyses. Briefly, serum samples were prepared with protein precipitation using acetonitrile. Analysis was performed on an Acquity ultraperformance liquid chromatograph with a Micromass Quattro Premier tandem MS detector (Waters, Milford, MA). Chromatographic separation was obtained by an Acquity UPLC BEH Shield RP18 column (1.7 μ m, 1.0 $\times$ 100 mm; Waters) using gradient elution at 40°C with a mix of ammonium acetate buffer (pH = 4.8) and acetonitrile (18–45%) as mobile phase. The retention times were 1.7 and 1.4 min for olanzapine and *N*-desmethyl olanzapine, respectively. Detection with multiple reaction monitoring was performed at the following transitions: m/z 313 $\rightarrow$ 256 for olanzapine and 299 $\rightarrow$ 256 for *N*-desmethyl olanzapine. Promazine was used as internal standard (transition m/z 285 $\rightarrow$ 212, retention time 2.4 min). All measured concentrations of the analytes were within the calibration curves, i.e. 10–800 nmol l⁻¹ (nM) for olanzapine and 5–300 nM for *N*-desmethyl olanzapine. Intra- and inter-assay accuracy and precision were less than 9.4% for both analytes.

Statistics

Patients were stratified into three subgroups, i.e. ECC users, progestogen-based contraceptive (PBC) users, and patients not using contraceptives. One-way ANOVA was performed to compare age, daily dose, smoking status and dose-adjusted serum concentrations (C : D ratios) between the subgroups. If the ANOVA test showed significance, subsequent analyses were performed between the subgroups by two-tailed, non-parametric Mann-Whitney tests. All statistical analyses were performed with SPSS version 17.0 (SPSS, Chicago, IL). Statistical significance was considered

Table 1

Characteristics of the study subgroups

Variable	Subgroups ECC (n = 10)	PBC (n = 10)	No contraceptives (n = 129)	P value*
Age (years)	32	27	30	0.293
Olanzapine dose (range) (mg day ⁻¹)	15 (2.5–30)	12.5 (5–20)	15 (2.5–35)	0.345
Smokers, n (%)	3 (30)	4 (40)	64 (49.5)	0.436
Time between drug intake and serum sampling (range) (h)	13 (12–15.5)	12.5 (11–24)	13 (10–27)	0.71
C : D ratio olanzapine (range)	7.83 (5.5–16.2)	7.85 (4.2–17.9)	7.67 (0.6–22.3)	0.99
C : D ratio <i>N</i> -desmethyl olanzapine (range)	1.30 (0.3–3.2)	2.20 (1.4–4.8)	1.95 (0.3–6.5)	0.033

*One-way ANOVA tests. Values are in median unless otherwise specified. ECC = ethinylestradiol-containing contraceptives. PBC = progestogen-based contraceptives. C : D ratio = dose adjusted serum concentration (nm mg⁻¹ day⁻¹).

as $P < 0.05$. GraphPad Prism version 4 was used for graphical presentations (GraphPad Software, San Diego, CA).

Results

In total, 162 of the 337 submitted questionnaires (48%) were returned, but 13 had to be excluded due to insufficient registration of information. Among the 149 remaining (included) questionnaires, 10 patients were co-administered ECC, while another 10 patients were co-administered PBC. The other 129 patients did not use contraceptives.

No significant differences in age, daily dose, smoking status, or time between drug intake and blood sampling, were found between the subgroups (Table 1). Moreover, there was no significant difference in the C : D ratio of olanzapine (Table 1). In contrast, the C : D ratio of *N*-desmethyl olanzapine was significantly different between the subgroups ($P = 0.03$ by ANOVA, Table 1). Subsequent Mann-Whitney tests showed that the C : D ratio of *N*-desmethyl olanzapine was significantly lower in the ECC subgroup (median 1.3 nm mg⁻¹ day⁻¹) compared with the PBC subgroup (median 2.2 nm mg⁻¹ day⁻¹, $P = 0.019$) and females not using contraceptives (median 1.95 nm mg⁻¹ day⁻¹, $P = 0.012$).

As illustrated in Figure 1, there was large inter-individual variability in the C : D ratios of both olanzapine and *N*-desmethyl olanzapine across the whole study population. For olanzapine and the metabolite, C : D ratios ranged 37-fold (0.6–22.3 nm mg⁻¹ day⁻¹) and 24-fold (0.28–6.48 nm mg⁻¹ day⁻¹), respectively.

Discussion

In this study, dose-adjusted serum concentrations of olanzapine and the metabolite *N*-desmethyl olanzapine were compared in women aged 18–40 years with respect to co-administration of ECC or not. For olanzapine, no difference in serum concentration was found between the ECC

Figure 1

Scatter plots showing individual dose-adjusted serum concentrations of olanzapine (A) and *N*-desmethyl olanzapine (B) in patients receiving ethinylestradiol-containing contraceptives (ECC), progestogen-based contraceptives (PBC) or no contraceptives. Values are in nm mg⁻¹ day⁻¹ and lines indicate median values

subgroup and patients using PBC or those not using contraceptives at all. In contrast, a significantly lower serum concentration of the metabolite *N*-desmethyl olanzapine was observed in patients using ECC contraceptives.

The 35–40% lower serum concentration of *N*-desmethyl olanzapine in ECC users confirms the CYP1A2-inhibitory properties of ethinylestradiol. ECC have previously been shown to provide a four-fold increase in systemic exposure of the CYP1A2 probe substrate tizanidine [11]. However, this degree of interaction is moderate compared with the 10- and 33-fold increase in tizanidine exposure reported during co-administration of the potent CYP1A2 inhibitors ciprofloxacin and fluvoxamine, respectively [12, 13]. The moderate CYP1A2-inhibitory potential of ethinylestradiol may explain the lack of effect of ECC on serum concentrations of olanzapine.

Another possible explanation for the lack of effect on the parent drug might be that ECC, in parallel with the inhibition of CYP1A2, induce alternative metabolic pathways of olanzapine. The phase II metabolic enzyme uridine diphosphate glucuronosyltransferase 1A4 (UGT1A4) is involved to a similar extent as CYP1A2 in the metabolism of olanzapine [14, 15]. ECC have been shown to induce UGT1A4-mediated metabolism of the antiepileptic drug lamotrigine [16]. The inducing effect of ethinylestradiol on UGT1A4 might therefore have counterbalanced the inhibition of CYP1A2 in the present study, and hence reduced formation and serum concentrations of the CYP1A2-mediated metabolite without affecting the parent drug.

Both environmental and genetic factors could influence the pharmacokinetics of olanzapine. Besides concurrent use of enzyme inhibitors or inducers, age and sex have been associated as determinants of the individual variability in olanzapine exposure [5, 17, 18]. Polymorphisms in genes encoding CYP1A2 and UGT1A4 have been linked to variability in olanzapine exposure as well. In homozygous carriers of the variant allele *CYP1A2*1F*, 20–30% lower serum concentrations of olanzapine have been reported [19], while a recent study found that the *UGT1A4 142 T>G* polymorphism was of equal importance as sex and cigarette smoking for variability in olanzapine exposure [20].

The potential pharmacological importance of *N*-desmethyl olanzapine is not fully elucidated. Despite that animal studies have indicated no or limited pharmacological activity of olanzapine metabolites in terms of affinities towards serotonergic and dopaminergic receptors [21]. A study in psychiatric patients found that c-peptide and insulin concentrations were inversely correlated with the serum concentration of *N*-desmethyl olanzapine [22]. This latter observation might actually indicate a 'side effect-balancing' ability of the metabolite. However, as the level of the metabolite is only about one fifth that of the parent drug, we consider it unlikely that the reduction in *N*-desmethyl olanzapine exposure during concurrent use of ECC is of clinical relevance.

Due to the naturalistic setting of this study, non-adherence to the prescribed drug treatment is a potential methodological limitation. Moreover, we do not have full control over concurrent use of other drugs or herbal agents, which may affect serum concentrations of olanzapine and the metabolite. Patient height and weight are also potential factors of pharmacokinetic importance, but this kind of information was not for consideration in the questionnaire. In fact, sex hormones may affect body weight and composition, and from a theoretical point of view, this aspect might be a confounding factor with respect to the hypothesized impact of ethinylestradiol on CYP1A2-mediated metabolism of olanzapine. Finally, the questionnaire to the physicians did not include information about the number of cigarettes smoked per day. However, in a previous study, we showed that in patients smoking more than six cigarettes per day, there was no significant difference in the induction of olanzapine metabolism [23].

In summary, use of ECC had no effect on serum concentrations of olanzapine, but significantly reduced serum concentrations of the CYP1A2-mediated metabolite *N*-desmethyl olanzapine. We conclude that co-medication with ECC is unlikely to affect the clinical response of olanzapine.

Competing Interests

There are no competing interests to declare.

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