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Moclobemide monotherapy *vs.* combined therapy with valproic acid or carbamazepine in depressive patients: a pharmacokinetic interaction study

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WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT

- Moclobemide (MCB) undergoes extensive both presystemic and systemic metabolism that can be affected by concomitant drugs.
- Valproic acid (VPA) and carbamazepine (CBZ) have been found to interact with psychotropic medications of all classes and many other drugs; VPA acts as a broad-spectrum inhibitor, and CBZ as a potent inducer of a variety of drug-metabolizing enzymes.
- There have been no previous studies designed to investigate a potential pharmacokinetic (PK) interaction between MCB and VPA or CBZ; however, these agents are likely to be used concomitantly for the treatment of depressive disorders.

WHAT THIS STUDY ADDS

- VPA does not significantly affect PK or metabolism of MCB at steady state.
- CBZ significantly decreases MCB exposure. This effect is time-dependent, being more pronounced after 3–5 weeks of co-administration.

AIM

To assess the impact of valproic acid (VPA) and carbamazepine (CBZ) on moclobemide (MCB) pharmacokinetics (PK) and metabolism at steady state in depressive patients.

METHODS

Twenty-one inpatients with recurrent endogenous depression received MCB (150 mg t.i.d.), either as monotherapy or in combination with VPA (500 mg b.i.d.) or CBZ (200 mg b.i.d.) in a nonrandomized manner. Steady-state plasma PK parameters of MCB and its two metabolites, Ro 12-8095 and Ro 12-5637, were derived. Clinical assessments of treatment efficacy were performed weekly using standard depression rating scales.

RESULTS

CBZ, but not VPA, was associated with decreases in the MCB AUC by 35% [from 7.794 to 5.038 mg h l^{-1}; 95% confidence interval (CI) −4.84863, −0.66194; $P = 0.01$] and C_{max} by 28% (from 1.911 to 1.383 mg l^{-1}; 95% CI −0.98197, −0.07518; $P < 0.05$), and an increase in its oral clearance by 41% (from 0.323 to 0.454 l h^{-1} kg^{-1}; 95% CI 0.00086, 0.26171; $P < 0.05$) after 4 weeks of co-administration. MCB through concentrations were also decreased, on average by 41% (from 0.950 to 0.559 mg l^{-1}; 95% CI −0.77479, −0.03301; $P < 0.05$). However, the efficacy in this group of patients was not inferior to the controls, for several possible reasons. Overall tolerability of all study medications was good.

CONCLUSIONS

VPA does not significantly affect PK or metabolism of MCB, whereas CBZ time-dependently decreases MCB exposure, probably by inducing metabolism of MCB and its major plasma metabolite. The actual clinical relevance of the observed MCB–CBZ PK interaction needs to be further evaluated in a more comprehensive study.

Introduction

Moclobemide (MCB) is a selective and reversible inhibitor of monoamine oxidase type A (MAO-A), with a broad spectrum of antidepressant activity [1]. At therapeutic doses, MCB lacks adverse anticholinergic effects and significant negative effects on psychomotor performance, cognitive function or cardiovascular system [2–4]. Therefore, it is particularly attractive in the treatment of the elderly or patients with concurrent somatic illness [1, 2, 4].

MCB is metabolized primarily via oxidative pathways (metabolized fraction >99%). Four primary and many subsequent metabolic reactions have been identified and a total of 19 metabolites have been isolated from urine [5]. The principal pathways of MCB metabolism involve *C*- and *N*-oxidation of the morpholine ring to yield its two major metabolites in plasma, Ro 12-8095 and Ro 12-5637, respectively. Ro 12-5637 retains certain MAO-A inhibitory activity, but is generally present in low concentrations, whereas the major plasma metabolite, Ro 12-8095, has no pharmacological activity [5, 6]. MCB has a short elimination half-life (1–3 h). Time and dose dependence are observed with multiple oral administrations in that clearance decreases with administration during the first week and thereafter remains constant [6–8].

Mood-stabilizing drugs, including valproic acid (VPA) and carbamazepine (CBZ), have established effects in the management of bipolar disorder, especially in mania. However, these drugs have also been shown to be effective in both bipolar and unipolar depression, including acute treatment, prevention of relapse or recurrence, and management of refractory depression (monotherapy or augmentation) [9–16].

VPA and CBZ have been found to interact with psychotropic medications of all classes and many other drugs [17–19]. VPA acts as a broad-spectrum inhibitor of a variety of hepatic enzymes, including uridine diphosphate-glucuronyltranserases (UGTs), epoxide hydrolase, and the CYP2C enzymes (particularly CYP2C9) [20, 21]. On the other hand, CBZ is a potent inducer of several drug-metabolizing enzymes, including cytochrome P450 monooxygenases (CYPs), namely CYP3A4, CYP2C9, CYP2C19 and CYP1A2, as well as UGTs [18, 22, 23]. As a consequence of enzyme inhibition or induction, plasma concentrations of a co-administered drug may reach toxic or subtherapeutic levels, and dosage adjustment may be required to avoid adverse effects or therapeutic failure [19]. To the best of our knowledge, there have been no previous studies designed to investigate a potential pharmacokinetic (PK) interaction between MCB and VPA or CBZ, although data on combination use of MCB with mood stabilizers, including VPA and CBZ, in patients with bipolar depression exist in the literature [24]. However, as MCB undergoes extensive both presystemic and systemic metabolism [6], there is a reasonable theoretical assumption that interactions might occur at the absorption and/or elimination level. Therefore, knowledge of the potential for interactions between MCB and VPA/CBZ is an important prerequisite for ensuring optimal therapy during their concomitant use for the treatment of depressive disorders.

The purpose of the present study was to assess the impact of VPA and CBZ on MCB PK and metabolism at steady state in hospitalized patients with recurrent depressive disorder. Together with PK assessments, efficacy and safety of MCB monotherapy and combined therapy (with VPA or CBZ) were examined and compared in order to assess the clinical consequences of potential PK interactions.

Materials and methods

Patients and study design

This was a 5-week, single-centre, nonrandomized, open-label, parallel-group, steady-state, drug interaction study, which was carried out in three groups of depressive patients receiving MCB (150 mg t.i.d. at intervals of 6, 6 and 12 h), either as monotherapy (control group) or in combination with a single mood stabilizer, VPA (500 mg b.i.d.; VPA group) or CBZ (200 mg b.i.d.; CBZ group). All study medications were initiated on the same study day (day 0). The medications were administered at the end of a meal, as recommended for MCB [4]. The study was performed in accordance with the Declaration of Helsinki and its amendments and in compliance with the Guidelines of Good Clinical Practice, employing a protocol approved by the Institute of Mental Health Ethics Committee. Written, informed consent was obtained from all patients.

Hospitalized patients, 18–65 years old, who met the diagnostic criteria for F33.2, according to the ICD-10 Classification of Mental and Behavioural Disorders, were included in the study. Exclusion criteria included psychotic features, bipolar affective disorder, other primary psychiatric disorders, alcohol or drug abuse, serious somatic illness, abnormal liver or kidney function tests, pregnancy, lactation, concomitant therapy with other known nonselective cytochrome P450 inducers/inhibitors, specific inducers/inhibitors of CYP2C19, as well as drugs causing analytical interference (i.e. those interfering with determination of MCB or its metabolites in plasma). Other somatic co-medications were permitted according to clinical needs (e.g. nonsteroidal anti-inflammatory drugs, antibiotics, etc.), but substances that interfere with cytochrome P450 isoenzymes were avoided. Patients already on chronic therapy for somatic disorders, such as mild hypertension, were allowed to continue the corresponding medication at the same dose provided the co-medication was not considered to affect MCB PK. Concomitant psychotropic therapy, apart from the mood stabilizers VPA and CBZ, included benzodiazepine anxiolytics and hypnotics, as data from clinical studies revealed no relevant PK or pharmacodynamic (PD) interactions between benzodiazepines and MCB [25].

Blood sampling and analytical method

MCB PK was studied under the expected steady-state conditions (steady-state plasma levels of MCB are reached at approximately 1 week following dose adjustment [4]). Plasma samples were collected over the study period of 28 days (starting from day 7), allowing concentration profiles to be determined after the first morning dose on study days 14 and 28 (blood samples were drawn just before the first daily dose and 1, 2, 3, 4, 5 and 6 h post dosing). In addition, concentrations just before the second daily dose on study days 7, 21 and 35 were followed. Samples were stored frozen (–20°C) until analysis.

The concentrations of MCB and its two major metabolites in plasma, Ro 12-8095 (lactam metabolite) and Ro 12-5637 (*N*-oxide metabolite), were measured using a developed and validated high-performance liquid chromatographic method, as described previously [26]. The limit of quantification for all the analytes was 0.02 mg l^{-1}. The accuracy (expressed as a percentage relative error) ranged from –10.0 to 6.5%, –13.4 to 4.5%, and –10.7 to 4.1% for moclobemide, Ro 12-5637 and Ro 12-8095, respectively. Intraday precision [expressed as a coefficient of variation (CV)] was in the range 1.9–5.6, 1.5–6.1 and 2.7–7.1% for moclobemide, Ro 12-5637 and Ro 12-8095, respectively, whereas the interday CV was 3.3–13.1, 3.9–8.1 and 4.0–10.0% for moclobemide, Ro 12-5637 and Ro 12-8095, respectively. The assay was not affected by concomitant medications, since the selectivity of the method had been investigated thoroughly through the method validation studies, as well as before each patient was enrolled for concomitant drugs that had not been evaluated for interference previously.

Clinical assessments

The Newcastle diagnostic scale [27] was used for differential diagnosis of endogenous and neurotic depression; the assessment was performed before the initiation of treatment (day 0). Symptom severity and clinical efficacy were assessed weekly (on days 7, 14, 21, 28 and 35) using the 17-item Hamilton Depression Rating Scale (HAMD) [28] and two Clinical Global Impression (CGI) scales [29], the CGI–Severity of Illness (CGI–S) and the CGI–Global Improvement scales (CGI–I). The CGI–S assessed the clinician's impression of the current state of the patient's illness on a seven-point scale (1 = 'normal/not at all ill' to 7 = 'among the most extremely ill patients'). The CGI–I assessed the patient's improvement or deterioration since the beginning of the study using the scores, 1 ('very much improved') to 7 ('very much worse'). Ratings were carried out by the same investigator at each assessment.

Safety was monitored throughout the study by assessing adverse events (AE) reports, laboratory tests (haematological and biochemical) and vital signs (blood pressure and heart rate). The overall tolerability of the treatment was assessed using a four-point scale (very good, good, moderate, and poor) [24].

Pharmacokinetic and statistical analysis

To achieve the validity of the PK comparisons, the dose of MCB was the same in all patients because MCB exhibits nonlinear, dose-dependent PK [6]. PK parameters were calculated for a 6-h morning dosing interval. All PK analyses were performed using noncompartmental methods with WinNonlin®, version 4.1 (Pharsight Corp., Mountain View, CA, USA). The maximum plasma concentration (C_{max}) and the time to its occurrence (t_{max}), as well as trough levels [concentrations obtained just prior to the second daily dose ($C_{min(6)}$)] were read directly from the concentration–time data. The terminal disposition rate constants of MCB and the two metabolites (λ_z) were obtained by log-linear regression analysis of the terminal phase of the plasma concentration–time curves. The terminal disposition half-lives ($t_{1/2}$) were calculated as ln $2/\lambda_z$. The areas under the concentration–time curves within the 6-h dosing interval at steady state (AUC) for MCB and the two metabolites were estimated using the trapezoidal rule. The linear rule was used for the ascending part, and the log trapezoidal rule for the descending part of the curve, up to the last measured concentration. The oral clearance of MCB (CL/*F*) was calculated as the dose/AUC, and the apparent volume of distribution based on the terminal phase (V_z/F) as CL/λ_z. The average concentration (C_{av}) of MCB during the 6-h dosing interval was calculated from the expression AUC/6. For each metabolite, the metabolic ratios (defined as MCB/metabolite AUC ratios) were estimated.

A previously developed physiological PK–PD model [30] was used for estimation of the relationship between MCB plasma concentration and MAO-A activity. The PD model parameters were used to simulate the MAO-A inhibition time course from measured plasma levels of MCB.

PK and clinical data were analysed using both parametric and nonparametric statistical tests. As comparable differences were found, only results using parametric tests were presented.

After testing for homogeneity of variances using the Levene test, plasma PK parameters of MCB and its two metabolites were compared among the three patient groups by analysis of variance (ANOVA), followed by a Dunnett's test to compare the VPA or the CBZ group with the control group. Statistical analysis was performed on PK profile 1 (day 14) and PK profile 2 (day 28) of MCB, and the two metabolites to test for variability and evaluate a time dependency of potential PK drug–drug interactions. For each patient group, pairwise comparison (paired *t*-test) of the two PK profiles was also performed. For t_{max}, the nonparametric tests (Kruskal–Wallis or Wilcoxon's signed rank test) were used. ANOVA or χ^2 test was used to analyse efficacy parameters (the χ^2 test was performed for proportions). All tests were two-tailed. A value of $P < 0.05$ was considered to be statistically significant. Unless otherwise

stated, data are presented as the means ± SD, except for t_{max}, which is presented as the median and range.

Results

Characteristics of the study population

A total of 21 inpatients (seven in each group) of both genders were enrolled, and all patients completed the study. According to the Newcastle scale, all patients had the diagnosis of endogenous depression (a score ≥6 [27]). There were no statistically significant differences in the mean total scores among the three groups of patients (7.2 ± 0.8, 6.8 ± 0.6 and 7.6 ± 1.2 in the control, VPA and CBZ groups, respectively). The complete blood count, and renal (plasma creatinine) and liver function tests were normal for all patients.

All patients were White. There were no statistically significant age, weight or gender differences among the groups. The use of additional medications was in compliance with the predefined criteria (i.e. inclusion/exclusion criteria). Benzodiazepines, followed by β-adrenergic antagonists and other antihypertensives, were the most frequently used co-medications in all treatment groups (Table 1). The proportion of patients on concomitant therapy with any of these drugs did not significantly differ among the groups. Other co-medications (cefalexin, ranitidine and famotidine) were taken by one patient each. Patients had already been on the stable doses of all co-medications prior to study enrolment, and the dosages were unchanged throughout the study, with the exception of the antibiotic, which was taken for 9 days. Well-known PK and PD properties of the concomitant drugs and MCB did not give any reason to suspect that relevant interactions could occur.

Demographic and medication details for the patients included in the study are summarized in Table 1.

Pharmacokinetics

Patient compliance was dictated by the fact that medication was provided by medical personnel. Nevertheless, noncompliance was observed in one patient in the control group who missed the morning dose on day 14, as detected by visual inspection of the plasma concentration–time curve (no absorption phase). These data were not taken into account for the calculation of mean PK parameters, except for the $t_{1/2}$.

The mean plasma concentration–time profiles of MCB and the two metabolites on days 14 and 28 for each group are presented in Figure 1.

On day 14, no statistically significant difference was observed in any of the calculated PK parameters among the three groups, except for the t_{max} of MCB and the $t_{1/2}$ of the major plasma metabolite, Ro 12-8095, which were significantly lower in CBZ-treated patients compared with the controls (Table 2). Statistical analysis of PK parameters of MCB and its metabolites obtained on day 28 again revealed no difference between the control and VPA groups; however, C_{max}, C_{av}, the AUC of MCB, as well as the AUC and $t_{1/2}$ of Ro 12-8095, were significantly decreased (by 28, 35, 35, 46 and 60%, respectively) in the CBZ-treated patients in comparison with the controls. CL/*F* of MCB was increased by 41% in the CBZ group ($P < 0.05$) (Table 2). The differences in the PK parameters of Ro 12-5637 were all statistically nonsignificant.

To assess the effect of CBZ on MCB disposition more fully, a statistical comparison of PK parameters between days 14 and 28 was performed for each patient group. There were no statistically significant differences in any PK parameter for the control and VPA groups, demonstrating the achievement of steady state. For the CBZ group, a significantly decreased C_{max} (14.3%), C_{av} (18.4%) and AUC (18.4%) of MCB as well as an increased CL/*F* (18.4%) and V_z/F (21.4%) were found on day 28 (Table 2), suggesting that effects of CBZ on MCB PK were time dependent.

Compared with the controls, the mean steady-state trough levels ($C_{min(6)}$) of MCB were 34–52% lower in CBZ-treated patients; these differences reached statistical significance on days 21–35 ($P < 0.05$; Table 3). The mean plasma trough levels of the two metabolites were also reduced, by 47–65 and 51–68% for Ro 12-5637 and Ro 12-8095, respectively, in comparison with the controls; the

Table 1

Demographic and medication details for the patients included in the study

	Control group ($n = 7$)	VPA group ($n = 7$)	CBZ group ($n = 7$)
Gender distribution	6 F/1 M	5 F/2 M	5 F/2 M
Age (years)*	52 ± 4	48 ± 10	53 ± 8
Body weight (kg)*	64 ± 13	72 ± 13	68 ± 9
Concomitant psychotropic medication (mg day^{-1}†)	Diazepam (15), bromazepam (4.5–9)	Bromazepam (3–12), lorazepam (3), nitrazepam (5)	Diazepam (10–15), alprazolam (0.5–1), lorazepam (2–3), nitrazepam (5)
Concomitant nonpsychotropic medication (mg day^{-1}†)	Atenolol (50), cefalexin (2000), enalapril (5), nifedipine (40)	Metoprolol (50), enalapril (5–10), ranitidine (300)	Metoprolol (50), propranolol (20), famotidine (20), enalapril (5), amlodipine (5)

*Expressed as mean ± SD. †Expressed as a dose range.

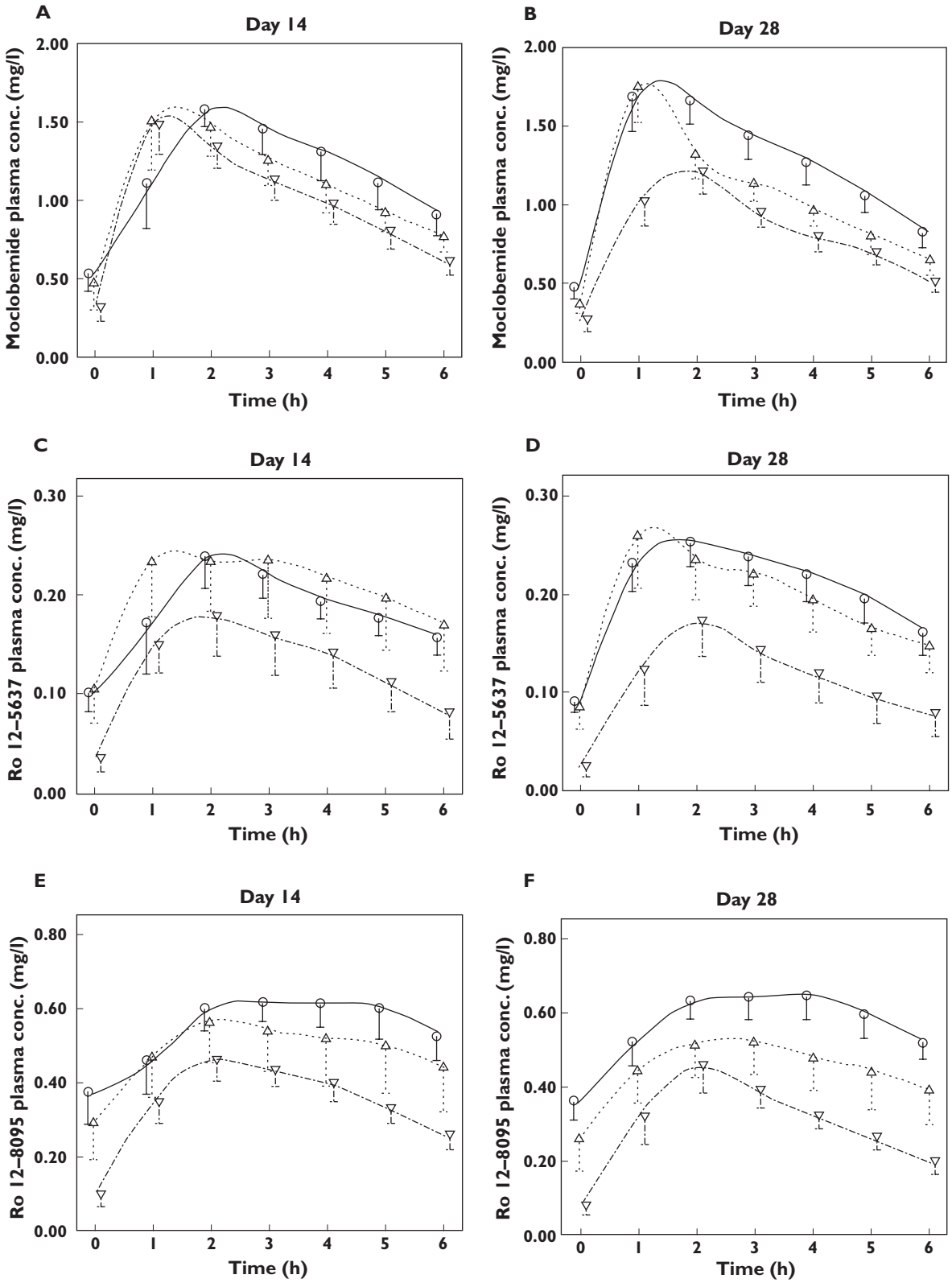

Figure 1

Mean (± SEM) steady-state plasma concentrations of moclobemide (A,B) and the two metabolites, Ro 12-5637 (C,D) and Ro 12-8095 (E,F), in patients on monotherapy (Control group) and combined therapy with valproic acid (VPA group) or carbamazepine (CBZ group). Control (–○–); VPA (·△·); CBZ (·▽·)

Table 2

Steady-state plasma PK parameters of moclobemide (MCB; 150 mg t.i.d.) and its two major metabolites, Ro 12-5637 and Ro 12-8095, in patients on monotherapy (control group) and combined therapy with valproic acid (500 mg b.i.d.; VPA group) or carbamazepine (200 mg b.i.d.; CBZ group)

Parameter	Control group Day 14 ($n = 6$)	Day 28 ($n = 7$)	VPA group Day 14 ($n = 7$)	Day 28 ($n = 7$)	CBZ group Day 14 ($n = 7$)	Day 28 ($n = 7$)
Moclobemide						
t_{max} (h)	2 (1–4)	1 (1–2)	2 (1–3)	1 (1–3)	1 (1–2)*	2 (1–3)
C_{max} (mg l^{-1})	1.82 ± 0.39	1.91 ± 0.34	1.78 ± 0.62	1.86 ± 0.42	1.61 ± 0.25	1.38 ± 0.28*†
C_{av} (mg l^{-1})	1.23 ± 0.30	1.30 ± 0.34	1.15 ± 0.46	1.09 ± 0.29	1.03 ± 0.29	0.84 ± 0.16**†
AUC (mg h^{-1} l^{-1})	7.36 ± 1.83	7.79 ± 2.02	6.91 ± 2.74	6.52 ± 1.72	6.18 ± 1.75	5.04 ± 0.97**†
CL/F (l h^{-1} kg^{-1})	0.35 ± 0.07	0.32 ± 0.06	0.34 ± 0.14	0.35 ± 0.13	0.38 ± 0.12	0.45 ± 0.10*†
$t_{1/2}$ (h)	3.47 ± 0.56‡	3.77 ± 1.13	3.85 ± 0.91	3.66 ± 1.13	3.13 ± 0.55	3.17 ± 0.50
V_z/F (l kg^{-1})	1.70 ± 0.29	1.73 ± 0.49	1.77 ± 0.31	1.75 ± 0.42	1.68 ± 0.36	2.04 ± 0.37†
Ro 12-5637						
C_{max} (mg l^{-1})	0.27 ± 0.08	0.28 ± 0.13	0.27 ± 0.15	0.27 ± 0.13	0.19 ± 0.10	0.18 ± 0.09
AUC (mg h^{-1} l^{-1})	1.13 ± 0.33	1.26 ± 0.38	1.25 ± 0.85	1.19 ± 0.57	0.79 ± 0.48	0.70 ± 0.43
Metabolic ratio§	6.74 ± 1.73	6.41 ± 1.72	6.46 ± 2.03	6.16 ± 1.99	11.09 ± 7.12	9.29 ± 4.54
$t_{1/2}$ (h)	4.58 ± 0.82‡	4.79 ± 1.18	4.68 ± 1.67	4.54 ± 1.30	3.20 ± 0.71	3.31 ± 1.12
Ro 12-8095						
C_{max}(mg l^{-1})	0.67 ± 0.17	0.67 ± 0.15	0.60 ± 0.32	0.55 ± 0.26	0.48 ± 0.14	0.47 ± 0.17
AUC (mg h^{-1} l^{-1})	3.41 ± 1.06	3.46 ± 0.92	2.96 ± 1.80	2.72 ± 1.48	2.14 ± 0.61	1.88 ± 0.60*
Metabolic ratio§	2.36 ± 1.04	2.50 ± 1.24	2.68 ± 1.05	2.71 ± 0.97	3.09 ± 1.18	3.12 ± 1.63
$t_{1/2}$ (h)	7.62 ± 4.43‡	7.87 ± 3.38	7.07 ± 3.87	6.31 ± 2.67	3.21 ± 0.61*	3.12 ± 1.23**

*$P < 0.05$; **$P < 0.01$ *vs.* the control group of the same day; †$P < 0.05$ *vs.* day 14 (intragroup comparison). ‡$n = 7$; §$AUC_{MCB}/AUC_{metabolite}$. Values are expressed as mean ± SD with the exception of t_{max}, for which the median (range) is reported.

Table 3

Steady-state trough plasma concentrations ($C_{min(6)}$) of moclobemide (MCB;150 mg t.i.d.) and its two metabolites, Ro 125637 and Ro 12-8095, in patients on monotherapy (control group, $n = 7$) and combined therapy with valproic acid (500 mg b.i.d.; VPA group, $n = 7$) or carbamazepine (200 mg b.i.d.; CBZ group, $n = 7$)

Concentration (mg l^{-1}) Compound	Group	Day 7	Day 14	Day 21	Day 28	Day 35
MCB	Control	1.03 ± 0.29	0.92 ± 0.33†	1.00 ± 0.26	0.84 ± 0.27	0.97 ± 0.31
	VPA	1.03 ± 0.51	0.78 ± 0.39	0.77 ± 0.36	0.66 ± 0.26	0.79 ± 0.39
	CBZ	0.60 ± 0.27	0.61 ± 0.23	0.60 ± 0.24*	0.51 ± 0.15*	0.47 ± 0.16*
Ro 12-5637	Control	0.19 ± 0.07	0.16 ± 0.04†	0.17 ± 0.03	0.16 ± 0.06	0.20 ± 0.07
	VPA	0.20 ± 0.14	0.18 ± 0.13	0.17 ± 0.12	0.15 ± 0.07	0.16 ± 0.06
	CBZ	0.08 ± 0.04	0.08 ± 0.07	0.09 ± 0.08	0.08 ± 0.06*	0.07 ± 0.05**
Ro 12-8095	Control	0.59 ± 0.14	0.53 ± 0.17†	0.65 ± 0.20	0.52 ± 0.12	0.63 ± 0.19
	VPA	0.47 ± 0.28	0.45 ± 0.32	0.48 ± 0.32	0.42 ± 0.27	0.40 ± 0.22
	CBZ	0.26 ± 0.11**	0.26 ± 0.09	0.26 ± 0.05**	0.20 ± 0.09**	0.20 ± 0.07***

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$ *vs.* control group. †$n = 6$. Values are expressed as mean ± SD.

differences were statistically significant on days 28 and 35 for Ro 12-5637 and days 7, 21, 28 and 35 for Ro 12-8095. There were no statistically significant differences in trough plasma concentrations of MCB and the two metabolites in VPA-treated patients compared with the controls.

The average MAO-A inhibition estimated from the mean steady-state MCB plasma concentrations over a 6-h dosage interval was 80.7, 79.3 and 76.1% on day 14 for the control, CBZ and VPA groups, respectively, and 80.8, 77.3 and 72.9% on day 28 for the control, CBZ and VPA groups, respectively.

Efficacy

All three groups of patients had a similar significant improvement in HAMD and CGI–S scores; over a 4-week evaluation period (days 7–35), the mean reduction of HAMD total score was 38.1 [from 24.0 (±3.1) to 14.9 (±2.6)], 37.5 [from 22.9 (±1.6) to 14.3 (±2.7)] and 39.3% [from 24.0 (±2.2) to 14.6 (±2.63)], whereas the mean decline in CGI–S was 39.4 [from 4.7 (±0.5) to 2.9 (±0.7)], 36.7 [from 4.3 (±0.5) to 2.7 (±0.8)] and 41.2% [from 4.9 (±0.4) to 2.9 (±0.7)] for the control, VPA and CBZ groups, respectively. However, there were no statistically signifi-

cant differences in the mean improvement per week among the three groups at any time ($P > 0.05$). The following CGI–I mean scores were observed at the final evaluation: 2.1 (±0.7), 2.4 (±0.5) and 2.3 (±0.5) for the control, VPA and CBZ groups, respectively, with the difference being statistically nonsignificant ($P > 0.05$). Although the difference among the groups at treatment end-point was nonsignificant, the proportion of patients with CGI–I score of 1 or 2 ('very much improved' or 'much improved') differed at study week 3: three patients (42.8%) in the CBZ group and no patients in either the control or VPA groups ($P < 0.05$). At week 4, the response rates pointed in the same direction [four patients (57.1%) in the CBZ group *vs.* two patients (28.6%) in both the control and VPA groups], but this difference failed to reach statistical significance.

Safety

A total of three AEs were reported, one in each patient group: agitation (control group), sexual dysfunction (VPA group), and drowsiness (CBZ group). All AEs were mild and expected, and lasted <3 weeks. No serious AEs were reported. Furthermore, there were no clinically significant changes in blood pressure, pulse rate or clinical laboratory variables throughout the study. Overall tolerability of the study medications in all patient groups was rated as 'very good' or 'good'.

Discussion

The obtained steady-state PK parameters of MCB in patients on monotherapy were in the range of corresponding values reported previously for different multiple oral dose schedules [6]. As reported previously [5, 6], the *C*-oxidized metabolite of MCB (Ro 12-8095) was dominant in human plasma compared with the *N*-oxidized metabolite (Ro 12-5637). Since morpholine *C*-oxidation is catalysed by CYP2C19 [31], its production is affected by CYP2C19 inducers/inhibitors. Relevant interactions involving this mechanism have already been described in the literature (e.g. with cimetidine [32] and omeprazole [33]). We hypothesized that this metabolic pathway might also have been influenced by VPA and/or CBZ. Morpholine *N*-oxidation is predominantly catalysed by flavin-containing monooxygenase (FMO) [34], which is not easily induced or readily inhibited [35], therefore potential drug–drug interactions including this metabolic pathway are less likely to occur.

The present study, however, showed no statistically significant effect of VPA on PK and two major metabolic pathways of MCB, although, contrary to our initial expectations, there was a trend for lower plasma concentrations of MCB in patients on combined therapy with VPA compared with the group on monotherapy (Figure 1A,B; Table 3). Concentrations of the major plasma metabolite, Ro 12-8095, were also nonsignificantly decreased in the VPA-treated patients (Figure 1E,F; Table 3), secondary to the decrease in plasma concentrations of the parent drug. Although VPA is a well-known inhibitor of certain metabolic enzymes, an increase in oral clearance of topiramate (by 13%) [36] and clonazepam (by 14%) [37] were reported during VPA concomitant therapy.

Except for the t_{max}, no statistically significant differences in the PK parameters of MCB were found on day 14 between CBZ-treated patients and the controls. The difference in t_{max} was the consequence of a surprisingly high value (4 h) observed in one patient in the control group, probably caused by food (it has been reported that food ingestion results in no difference in C_{max}, CL/*F*, or relative bioavailability, but has a significant effect on the rate of MCB absorption, reflected in changes in t_{max} [6]). This difference, however, was considered clinically nonsignificant. In contrast, a significant decrease in C_{max}, C_{av} and AUC of MCB was observed in CBZ-treated patients in comparison with the controls on day 28, indicating reduced bioavailability of MCB in patients on the combination therapy. The $t_{1/2}$ of MCB was nonsignificantly decreased (by 16%), in spite of a significant increase in CL/*F* of MCB. This may indicate that the influence of CBZ is greater on the absorption phase of MCB than on its elimination phase. MCB undergoes a substantial first-pass metabolism, which becomes saturated following multiple doses (e.g. bioavailability increases from 56% following a single 100-mg dose to 90% following dosing with 100 mg t.i.d. for 15 days) [6]. It is possible that CBZ might induce first-pass metabolism of MCB, thus diminishing its oral bioavailability. The results of the present study (for both between-group comparisons as well as statistical comparison of PK parameters between days 14 and 28, performed for each group separately) suggested that 3–4 weeks were necessary until the effect of CBZ on MCB PK became statistically significant. Therefore, the potential effect of CBZ on CYP2C19 activity *in vivo* might be a function of time, and this could be a possible explanation for the conflicting data on this matter contained in the literature [17, 18, 22, 23, 38, 39].

The finding of reduced plasma concentrations of the metabolites in patients on combination therapy with CBZ (Figure 1E,F; Table 3) may be at least partly regarded as a subsequent result from decreased concentrations of the parent compound. The plasma disposition kinetics of the metabolites following multiple administration of therapeutic doses of MCB have not been thoroughly determined previously, as a result of low concentrations at the usual MCB dosage and/or sampling schedule, which did not allow a complete description [6]. However, it has been reported that the plasma concentration of Ro 12-8095 in extensive metabolizers of CYP2C19 declines with longer $t_{1/2}$ than the parent compound, indicating elimination-limited kinetics of the metabolite, whereas the elimination of the other metabolite, Ro 12-5637, is formation limited [31]. A similar situation was observed in the present study in both patients on monotherapy and combination therapy with VPA (Figure 1). In the CBZ-treated patients,

however, plasma concentrations of all three compounds declined in parallel (Figure 1) with similar $t_{1/2}$ (Table 2). Finding of a significantly decreased $t_{1/2}$ of the major plasma metabolite in this group of patients compared with the controls suggested that the metabolism of Ro 12-8095 was also induced by CBZ, although the 6-h sampling time might have been too short for accurate determination of the Ro 12-8095 $t_{1/2}$ in patients on monotherapy and combination therapy with VPA. It has been reported that the lactam derivative, Ro 12-8095, undergoes further degradation to mainly acids with <1% of the oral dose of MCB being recovered in urine as this metabolite [5, 6]. On day 28, the AUC of Ro 12-8095 was significantly decreased in CBZ-treated patients compared with the controls, whereas the metabolic ratio ($AUC_{MCB}/AUC_{Ro12\text{-}8095}$) was nonsignificantly increased, suggesting that the AUC of the parent compound and the AUC of its major metabolite decreased in nearly the same proportion. Therefore, if CBZ stimulated the clearance (CL) of formation of Ro 12-8095, the increase in its elimination CL would be proportional. It has already been reported that CBZ, as a broad-spectrum inducer, can decrease plasma concentrations of both drug and its major metabolite. Such an effect of CBZ has been described, for example, for risperidone [40] and simvastatin [41].

In general, higher interindividual variability was observed in the PK parameters of the two metabolites compared with the MCB ones, particularly in patients on combination therapy with VPA or CBZ (Table 2). This is associated with the variable contribution of different metabolic pathways and pharmacogenetic behaviour of the two metabolic enzymes, CYP2C19 and FMO [6, 35], involved in the principal pathways of MCB metabolism. Therefore, for a more comprehensive delineation of the effect of CBZ on MCB metabolism and disposition of the metabolites, a larger study enrolling previously genotyped subjects would be required. However, irrespective of the magnitude of interaction, it is highly unlikely that the metabolites contribute to any significant extent to the clinical effects of MCB (see Introduction); thus, further investigation in this field would probably be of limited clinical relevance.

The addition of the mood stabilizers, VPA or CBZ, did not seem to impair safety. No clear differences between the treatment groups could be shown with respect to improvement on the clinical rating scales for depression, although the interpretation of results of clinical assessments is limited by small sample size, study design (open-label, nonrandomized study, without placebo group) and short study duration (the duration of the study was insufficient to draw a definite conclusion on the number of responders). Despite significantly decreased moclobemide exposure in patients on combination therapy with CBZ, the measured efficacy of the treatment in this group of patients was not inferior to the controls. Moreover, some indicators of a possible earlier onset of antidepressive action in patients on combination therapy with MCB and CBZ were noticed (i.e. higher proportion of patients with CGI–I score of 1 or 2 found in the CBZ group at earlier evaluations), although the small number of patients included precludes a more reliable conclusion. These preliminary observations might have been partly attributable to the beneficial effects of CBZ on depressive symptoms. It has been reported that CBZ itself has some acute antidepressant efficacy [42] and also potentiates the actions of antidepressants in patients with depression [14, 15]. However, the estimated difference of 8% in average MAO-A inhibition between the CBZ group and controls was much lower than the actual difference in average MCB plasma concentrations, which might have been another explanation for the lack of clinical manifestations of the decreased MCB exposure. Therefore, the actual clinical relevance of the observed PK interaction between MCB and CBZ needs to be further evaluated in a more comprehensive study of an adequate design, using higher doses of MCB in combination with CBZ in order to investigate the possibility of higher therapeutic efficacy, particularly in patients with more severe symptomatology (600 mg b.i.d. MCB has been reported to be the maximum tolerated dose regimen in healthy subjects [43]).

In conclusion, the statistical analysis of PK parameters indicates the absence of significant effect of VPA on MCB PK and metabolism at steady state. CBZ co-administration, on the other hand, significantly decreases bioavailability of MCB, and reduces steady-state plasma concentrations of its major, but pharmacologically inactive, plasma metabolite. Decreased absorption of MCB and enhanced elimination of both MCB and the metabolite are possible explanations for the mechanism underlying the observed PK interaction between MCB and CBZ.

Competing interests

None to declare.

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