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ORIGINAL ARTICLE

Comparison of repeated-dose pharmacokinetics of prolonged-release and immediate-release torasemide formulations in healthy young volunteers

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ABSTRACT

The major aim of the study was to compare the pharmacokinetic profile of repeated-dose administration of a prolonged-release (PR) formulation of torasemide with that of an immediate-release (IR) dosage. Sixteen volunteers received one daily dose, on four consecutive days, of 10 mg of torasemide-PR or torasemide-IR in a single-blind, two-treatment, two-period, repeated-dose, cross-over, sequence-randomized clinical trial. Blood samples were collected at various time points on day 1 (single-dose) and on day 4 (repeated-dose) and torasemide concentrations were analysed by LC/MS/MS. Diuretic effect and urine electrolytes were measured. Urinary urgency was subjectively assessed by visual analogue scales. Safety and tolerability were also determined. Based on logged values, bioequivalence parameters, were: on day 1, AUC_0^t ratio = 1.07 (90% CI 1.02–1.1), C_{max} ratio = 0.69 (90% CI 0.67–0.73); and on day 4, AUC_τ^{EE} ratio = 1.02 (90% CI 0.98–1.05), C_{max} ratio = 0.62 (90% CI 0.55–0.70). PR had longer t_{max} than IR and showed significantly lower fluctuations of plasma concentrations. Urine evaluations were similar with both formulations, although PR showed a lower urine volume in the first hours post-administration. Episodes of acute urinary urgency occurred later and were subjectively less intensive with PR. No significant adverse events were reported.

INTRODUCTION

Torasemide [1-isopropyl-3-([4-(3-methyl-phenylamino) pyridine]-3-sulfonyl)urea] is a diuretic agent of the pyridil sulfonylurea class that appears to have its major site of action in the ascending limb of Henle's loop [1–3]. Loop diuretics mainly inhibit the $Na^+/2Cl^-/K^+$ carrier from the luminal side of the cell [4]. Torasemide is rapidly absorbed after oral administration and has a maximal peak plasma concentration in the first hour [5,6]. Its bioavailability is about 80% and is not influenced by food [7]. It is highly protein-bound (>99%) [6–11]. The drug is metabolized to a great extent (80%) in the liver [7,9]. Renal clearance of the parent drug accounts for approximately 20% of total clearance [7–9,11]. Torasemide appears to follow a two-compartment open model [9,10] and displays linear pharmacokinetics [8,10,11]. Its plasma terminal elimination half-life is about 3.5 h [6,10,12]. Clinical trials indicate that torasemide is effective in the treatment of hypertension and edema as well as for other symptoms in patients with chronic renal failure, hepatic dysfunction or congestive heart failure [13].

Immediate-release (IR) formulations of torasemide deliver the active moiety to the systemic circulation in a short period of time, rapidly lowering plasma concentrations to subtherapeutic levels as a result of high clearance. According to the clinical prescription of the compound, this process can reduce therapeutic efficacy. These disadvantages can be minimized by administering prolonged-release (PR) formulations. A new PR formulation of torasemide has now been developed with different dose strengths, each prolonging the initial delivery rate in vitro. The formulation is a slow-release tablet containing torasemide that is manufactured by Ferrer International (SA) and it is presently authorized for use in Spain, Peru, Guatemala and Honduras.

Prolonged continuous exposure to low concentrations of diuretics appears to enhance diuretic effects and reduce the incidence of adverse reactions [14]. In a previous study in healthy volunteers (data on file), we compared the plasma pharmacokinetic profile of a single dose of a PR formulation of torasemide with that of an IR formulation. We found that the PR formulation demonstrated a significantly lower C_{max}, significantly higher t_{max} values and a similar extent of systemic exposure.

The main objective of this study, also performed in healthy volunteers, was to compare the plasma pharmacokinetic profile of a repeated-dose administration of a PR formulation of torasemide with that of an IR formulation. Further aims of the study were to evaluate the urine pharmacodynamic profile as well as the clinical safety and tolerability of the two torasemide formulations.

MATERIALS AND METHODS

Subjects

Sixteen healthy Caucasian participants (nine females and seven males) aged 20–32 years were selected from the pool of volunteers at the Drug Research Centre (Research Institute), Hospital de la Santa Creu i Sant Pau, Barcelona (Spain) and included in the study. Their body mass index was within the normal range (19–26, calculated as the ratio between body weight in kg and height in cm^2). Demographic characteristics are listed in *Table I*. All volunteers underwent a screening evaluation in the 3 weeks prior to the trial. This consisted of a medical interview, physical examination, clinical laboratory tests (hematology, chemistry and urinalysis) and a 12-lead ECG. The pre-study evaluation also included drug and alcohol testing of urine samples, serological tests (for hepatitis B and C, and HIV) and serum β-HCG (only in women). Exclusion criteria included any medication in the 15 days prior to the study, history of alcohol or drug abuse, previous allergy, and consumption of over 39 g absolute alcohol/day, 100 mg caffeine/day or 10 cigarettes/day. For women, previous vaginal childbirth was an additional exclusion criterion.

Table I Demographic characteristics of volunteers included in the study.

	Torasemide 10 mg (n = 16)	
Characteristic	Mean ± SD	Range
Age (years)	24.06 ± 3.3	20–32
Weight (kg)	66.04 ± 7.49	53.0–78.6
Height (cm)	171.44 ± 8.04	157–189
BMI (kg/cm^2)	22.43 ± 1.69	19.0–25.3

BMI, body mass index.

Prior to enrollment, written informed consent to participate was obtained in response to a fully written and verbal explanation of the nature of the study. The protocol was approved by the Hospital Research Ethics Committee and the Spanish Drug Agency. The study was performed in accordance with the Declaration of Helsinki and subsequent revisions as well as with the European Union Good Clinical Practice Guidelines.

Study design

The study was a single-blind, two-treatment, two-period, repeated-dose, cross-over, sequence-randomized clinical trial. The participants received once daily oral administrations of 10 mg prolonged-release torasemide (torasemide-PR) and immediate-release torasemide (torasemide-IR), both on four consecutive days. The two administrations were separated by a minimum period of 7 days wash-out.

The study was performed in four groups. The order of administration was randomized using the SPSS 14.0 program (SPSS Inc., Chicago, IL, USA), with four blocks, each of four volunteers, to achieve a balanced administration.

Sample size

Sample size was based on data from a previous study (data on file) in which an inter-subject coefficient of variation (CV) between 6 and 7% was obtained. The resulting number of necessary volunteers was 14, considering as assumptions: (i) a CV of 10% (value closest to the empirical value identified in published tables for sample-size calculations) [15]; (ii) a relative bioavailability between 0.9 and 1.1 (that is, a difference

no greater than 10% in any direction); and (iii) a power of 90%, taking into account both the use of a crossover design and log transformation data. To compensate for possible drop-outs, we enrolled a sample size of 16 subjects.

Dose selection and administration schedule

The 10 mg dose was chosen based on results from the previously mentioned bioavailability study which assessed torasemide 5 and 10 mg in both PR and IR formulations after a single oral administration. The fact that the drug has been reported to show linear pharmacokinetics within a range of 10–100 mg after single oral doses and a range of 5–80 mg after intravenous doses [2] was also taken into account.

The selected administration interval (a repeated schedule every 24 h) was based on the prescription recommendations in clinical practice [16]. The number of administrations (four) allowed to achieve steady-state plasma concentrations, taking into account an elimination half-life of around 4 h [96 h(4 days × 24 h)/4 h = $24t_{1/2}$].

Procedures

On the four experimental days, medication was administered in the early morning (08:00–09:00 hours), under fasting conditions, with 250 mL of tap water. A cannula was inserted in the cubital vein before drug-intake on day 1 and on day 4 of each study period to draw blood samples. Urine samples were collected as explained below.

Volunteers were required to stay at the center from 13 h before until 24 h after drug administration on the first and the fourth days (i.e. 4 nights and 2 days). The second dose was given in the morning before leaving the center, and on the morning of the third day of each experimental period the volunteers come to the center to receive the corresponding dose. On days 1 and 4, no food was allowed during the first 2 h after administration, and a standard breakfast, lunch and dinner were provided at 2, 6 and 12 h after drug administration. Water consumption was controlled and volunteers were required to drink 200 mL of tap water every hour in the period between +1 and +6 h (total ingestion: 250 mL + 1 L). Subjects were requested to avoid excessively salted foods on the 4 or 5 days prior to the study. Clinical tolerability and safety were assessed daily by continuous recording of adverse events (AE) and evaluation of vital signs (systolic and diastolic blood pressure, and heart rate). Laboratory tests and ECG were performed before the administration of the first dose and at +24 h post-administration of the last dose. Urinary urgency was also reported on days 1 and 4.

Blood sampling

On days 1 (single dose) and 4 (repeated dose), blood samples (6 mL) were collected in lithium heparin glass tubes immediately before and at 0.25, 0.5, 0.66, 0.83, 1, 1.16, 1.33, 1.5, 2, 3, 6, 8, 10, 12, 16 and 24 h after drug intake. Samples were centrifuged (1620 ***g***) at 4 °C for 10 min. Plasma was immediately separated into 2 aliquots and stored at −80 °C until analysis. Torasemide plasma concentrations were obtained by LC/MS/MS.

Urine collection

Urine was collected over the 12 h prior to day 1 of each experimental period and until 24 h post-medication on days 1 (single dose) and 4 (repeated dose) at the following time intervals: 0–1, 1–2, 2–4, 4–6, 6–12 and 12–24 h.

Urine obtained at each interval was collected in plastic bottles and volume was recorded. Two aliquots of 8 mL were separated and kept at −80 °C. Sodium, chloride and potassium were quantified at the collection interval prior to each experimental period and at 0 to +6 h, +6 to +12 h and from +12 to +24 h collection intervals post-medication on days 1 (single dose) and 4 (repeated dose).

Analytic methods

Torasemide plasma concentrations

Bioanalytical assays were performed at the Dr. F. Echevarne Analytical Laboratory, Barcelona, Spain. Analyses were carried out in accordance with good laboratory practices. Samples were analyzed by LC/MS/MS using a heated nebulizer interface, following a previously validated method in accordance with standard requirements [17]. Extraction was performed by protein precipitation. Two hundred microliters of plasma was deproteinized by adding 1 mL of acetonitrile. After centrifugation, 20 μL of supernatant was injected into the HPLC system. Chromatography separation was done on an analytical column Phenomenex LUNA C18 (150 × 4.6 mm) 5 μm using a mobile phase consisting of a mixture of ammonium acetate 0.05 M and acetonitrile (35 : 65 v/v) adjusted to pH 4.0. The multiple reaction monitoring was torasemide m/z 349.1 → 264.2 and sulphapiridine (internal standard) m/z 250.0 → 156.1. The calibration line ranged from 1 to 2000 ng/mL. The variation coefficients of quality

controls in the validation study were less than or equal to 8.36% for the intra-day study and less than or equal to 5.45% for the inter-day study. The relative errors of quality controls in the validation study were less than or equal to 11.67% for the intra-day study and less than or equal to 9.58% for the inter-day study.

The lowest limit of quantification was 1 ng/mL. The extraction recovery was around 90% for torasemide and the internal standard. No endogenous compounds were found to interfere with the analysis. This method met regulatory requirements for selectivity, sensitivity, goodness of fit, precision, accuracy, recovery and stability. Each sample time-point was analysed with a single determination.

Pharmacodynamics

The *diuretic effect* was monitored by measuring the volume of urine (in mL) obtained at the collection intervals as well as by computing the total volume of urine collected in the 24 h after drug intake.

Urine sodium, chloride and potassium were measured by indirect potentiometry using ion-selective electrodes (Integra 800; Roche Diagnostics SL, St. Cugat del Vallès, Barcelona, Spain). Measurements were expressed as mmol.

Urinary urgency

Urinary urgency was subjectively reported via a 100-mm-long horizontal visual analogue scale anchored by 'no urgency' and 'strong sensation of urgency'. Each time they experienced urinary urgency (event) within 0–6 h interval, the volunteers were asked to rate the intensity on the scale. Previous responses were visible. Scores were measured and expressed as mm (from 0 to 100). We also noted the number and time of urinary events for each participant in the first 6 h post-torasemide administration on days 1 and 4.

Pharmacokinetic analysis

Pharmacokinetic parameters were calculated by model-independent methods [18] using WINNONLIN 2.1 software (Pharsight, Mountain View, CA, USA).

Day 1 (single dose)

Peak plasma concentration (C_{max}) and the time to reach C_{max} (t_{max}) were obtained directly from the raw data. The terminal plasma elimination half-life ($t_{1/2}$) was calculated as follows: $t_{1/2} = 0.693/k_e$, where k_e represents the first order rate constant associated with the terminal (log-linear) portion of the curve, estimated via linear regression of time vs. log concentration. The area under the plasma concentration–time curve (AUC) from 0 to infinity (AUC_0^∞) was calculated as $AUC_0^\infty = AUC_0^{tx} + C_{tx}/k_e$, where tx is the time of the last torasemide concentration (C_{tx}) exceeding the limit of quantification. Partial AUC values with 0 and 24 h (AUC_0^{24}) as time limits were also calculated. All AUCs were calculated by applying the log-trapezoidal method. Mean residence time (MRT), a measure of drug disposition, was calculated as follows: $MRT_0^\infty = AUMC_0^\infty/AUC_0^\infty$, where $AUMC_0^\infty$ is the area under the first moment–time curve (AUMC) extrapolated to infinity.

Day 4 (repeated dose)

In addition to C_{max}, t_{max}, $t_{1/2}$, k_e and MRT, we calculated trough plasma concentration (C_{min}), AUC at steady state (AUC_τ^{EE}) and the percentage of peak-trough fluctuations (PTF). C_{min} was obtained directly from the raw data (minimum concentration between 0 and τ). AUC_τ^{EE} was calculated as the AUC between 72 and 96 h (24 h time period) by applying the log-trapezoidal method. PTF was calculated as follows: $PTF = (C_{max} - C_{min})/C_{average}$, where $C_{average} = AUC_\tau^{EE}/\tau$, in which τ is the dosing interval.

Statistical analysis

Descriptive statistics were calculated for all pharmacokinetic parameters as well as pharmacodynamic urine variables, including arithmetic mean and standard deviations.

A comparative analysis of bioavailability was separately applied to days 1 (single dose) and 4 (repeated dose) results to determine possible differences between torasemide PR and torasemide IR. An analysis of variance (ANOVA) model was used for the log-transformed AUC_0^t, AUC_0^∞ (or AUC_τ^{EE}) and C_{max} data, and the geometric means of the ratio between the two formulations with their corresponding 90% confidence interval (CI) were calculated. The sources of variation included as factors in the model were period, subject within sequence, sequence and treatment. To rule out possible experimental biases, gender and group factors were evaluated by the ANOVA model for the single and repeated-dose administrations. To accept the bioequivalence, the 90% CI had to be included within the range from 0.80 to 1.25 [19]. Comparisons for t_{max} were performed using the Wilcoxon signed-rank test. Formulations at days 1 and 4 were compared for all the remaining plasma pharmacokinetic parameters ($t_{1/2}$,

MRT, C_{min}, PTF) and for the urine pharmacodynamic variables by means of a *t*-test for repeated measures.

Linear pharmacokinetics of torasemide after repeated administration of the PR and the IR formulations were separately evaluated by assessing whether the 95% CI of the ratio of AUC_0^∞ (single dose) to AUC_τ^{EE} (repeated dose) geometric means were within the range of 0.80–1.25.

The level of significance in all contrast hypotheses was 5% with a bilateral approach. All statistical analysis was performed with SPSS 14.0.

RESULTS

All 16 participants completed the trial and were compliant with the study protocol. Intention-to-treat data and per protocol analyses were thus coincident. Absence of bias was evidenced when ANOVA was applied to parameters corresponding to plasma concentrations obtained after first administration of both formulations (gender: $P = 0.177$, 0.559, 0.589 for C_{max}, AUC_0^t, AUC_0^∞, respectively; group: $P = 0.710$, 0.493, 0.485 for C_{max}, AUC_0^t, AUC_0^∞, respectively) and after repeated administration (gender: $P = 0.608$, 0.328, for C_{max}, AUC_τ^{EE} respectively; group: $P = 0.959$, 0.507 for C_{max}, AUC_τ^{EE}, respectively).

Pharmacokinetic analysis

Day 1 (single dose)

Figure 1a depicts the mean plasma concentration time profile for both formulations (PR and IR) after a single dose of 10 mg. Individual plasma concentrations ranged from 1.03 to 1504.61 ng/mL for PR and 1.92 to 2003.00 ng/mL for IR. Concentrations were detected at 15 min post-dose in 15 of 16 subjects after PR administration and in all volunteers after IR administration. They remained detectable for at least 24 h in all volunteers after both administrations.

Table II shows the results of the comparative analysis of bioavailability. The PR formulation exhibited a significantly lower C_{max} compared with the IR formulation (90% CI 0.67–0.73). However, total systemic exposure to the drug was similar for both formulations for both AUC_0^t and AUC_0^∞. The ratio between the two areas $[(AUC_0^t/AUC_0^\infty) \times 100]$ was ≥80% in all the volunteers after both administrations, assuring that the number of scheduled experimental samples was sufficient to adequately characterize the pharmacokinetic plasma profile. No significant period ($P = 0.802$, 0.901, 0.886 for C_{max}, AUC_0^t, AUC_0^∞, respectively) or sequence ($P = 0.509$, 0.532, 0.526 for C_{max}, AUC_0^t, AUC_0^∞, respectively) factors were evidenced when the ANOVA model was applied to the bioavailability parameters.

Additional plasma pharmacokinetic parameters are summarized in *Table III*. No significant differences were observed in plasma (k_e, $t_{1/2}$) pharmacokinetic parameters between the two formulations except for t_{max} and MRT which were significantly higher after PR administration in comparison with IR administration (1.45 vs. 0.79 h; $P = 0.001$ and 4.19 vs. 3.48 h; $P = 0.001$ respectively).

Day 4 (repeated dose)

Figure 1b depicts the mean plasma concentration time profile for both formulations (PR and IR) after repeated administration of 10 mg. Individual plasma concentrations ranged from 2.66 to 1317.18 ng/mL after PR administration and from 1.64 to 2439.30 ng/mL after IR administration. Concentrations were detected at 15 min post-dose in 15 of 16 subjects after PR administration and in all volunteers after IR, and they remained detectable for at least 24 h in all volunteers after both administrations. Mean plasma concentration values obtained at the different troughs during torasemide administration (+24 h after the first, third and fourth doses) were 5.70 ± 3.57, 6.01 ± 3.52 and 5.53 ± 2.77 ng/mL after PR, and 4.69 ± 2.89, 5.06 ± 4.58 and 4.68 ± 3.34 ng/mL after IR. No differences over time were observed after either administration (ANOVA $P = 0.398$ and 0.678 for PR and IR, respectively).

Table II shows the results of the comparative analysis of bioavailability. The PR formulation exhibited a significantly lower C_{max} than the IR formulation (90% CI 0.55–0.70). C_{max} values were out of the lower limit of bioequivalence 90% acceptance criteria (0.80–1.25).

However, total systemic exposure to the drug was similar for both formulations for AUC_τ^{EE}. No significant period ($P = 0.911$, 0.910 for C_{max}, AUC_τ^{EE}, respectively) or sequence ($P = 0.159$, 0.255 for C_{max}, AUC_τ^{EE}, respectively) factors was evidenced when the ANOVA model was applied to the bioavailability parameters.

Additional plasma pharmacokinetic parameters are summarized in *Table III*. No significant differences were found between the two formulations in pharmacokinetic parameters k_e, $t_{1/2}$, and C_{min}. However, t_{max} and MRT were significantly higher after PR administration than after IR administration (1.80 vs. 0.90 h, $P = 0.003$ and 4.31 vs. 3.46 h, $P = 0.001$, respectively). Fluctuations of plasma concentrations, represented by PTF values were significantly lower after PR administration (669.04% vs. 1114.02%, $P = 0.001$).

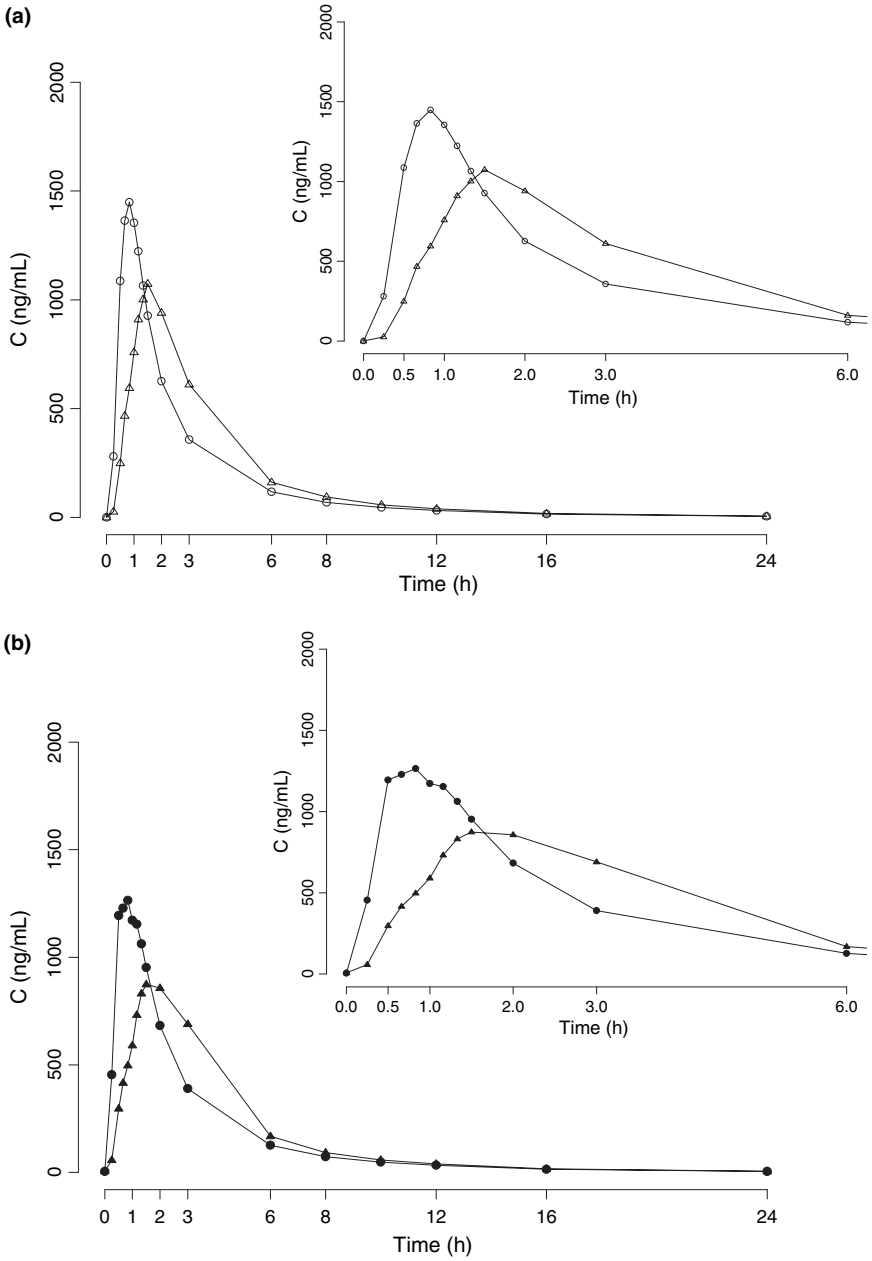

Figure 1 Mean plasma concentration–time profiles of 10 mg oral torasemide (a) on day 1 (single dose, in zoom from 0 to 6 h) and (b) on day 4 (repeated once daily dose, in zoom from 0 to 6 h) in 16 healthy volunteers of both sexes. (Δ, ▲) PR formulation and (○, ●) IR formulation.

Table II Pharmacokinetic parameters for plasma torasemide 10 mg: geometric mean ± standard deviation (n = 16). Plasma log-transformed data.

Parameter	Torasemide-PR Mean ± SD	Torasemide-IR Mean ± SD	F (PR/IR)	90% CI
Day 1 (single dose)				
AUC_0^t (ng h/mL)	3685.59 ± 660.03	3448.56 ± 565.98	1.07	1.02–1.11
AUC_0^∞ (ng h/mL)	3718.04 ± 680.93	3476.50 ± 582.36	1.07	1.02–1.11
C_{max} (ng/mL)	1127.08 ± 170.74	1610.38 ± 229.30	**0.69**	0.67–0.73
Day 4 (repeated dose)				
AUC_τ^{EE} (ng h/mL)	3604.72 ± 685.15	3550.49 ± 658.82	1.02	0.98–1.05
C_{max} (ng/mL)	1000.50 ± 152.28	1605.48 ± 357.05	**0.62**	0.55–0.70

Bold values indicate that the PR formulation exhibited a significantly lower C_{max} compared with the IR formulation.

Table III Pharmacokinetic parameters for plasma torasemide 10 mg after single-dose (day 1) and repeated-dose (day 4) administration: arithmetic mean ± standard deviation (n = 16, each dose).

	Torasemide-PR Mean ± SD		Torasemide-IR Mean ± SD		Paired t-test PR–IR	
Parameter	Single dose	Repeated dose	Single dose	Repeated dose	Single dose	Repeated dose
t_{max} (h)[a]	1.50 ± (1.00–2.00)	1.50 ± (0.66–3.00)	0.75 ± (0.50–1.33)	0.66 ± (0.5–2.00)	**0.001**	**0.003**
k_e (h^{-1})	0.171 ± 0.02	0.169 ± 0.01	0.168 ± 0.02	0.171 ± 0.01	0.465	0.458
$t_{1/2}$ (h)	4.08 ± 0.42	4.12 ± 0.31	4.18 ± 0.52	4.07 ± 0.33	0.345	0.467
MRT (h)	4.19 ± 0.58	4.31 ± 0.58	3.48 ± 0.58	3.46 ± 0.72	**0.001**	**0.001**
C_{min} (ng/mL)	NA	5.31 ± 2.81	NA	4.29 ± 3.20	NA	0.053
PTF (%)	NA	669.03 ± 97.70	NA	1114.01 ± 251.16	NA	**0.001**

[a]Median ± (minimum–maximum) and Wilcoxon signed-rank test.

NA, not appropriate.

Bold values indicate that t_{max} and MRT are significantly higher after the PR administration in comparison with IR administration. PTF is significantly lower after PR administration in comparison with IR administration.

Linear pharmacokinetics after repeated dosage were confirmed for both formulations, as systemic exposure to the drug was similar after single (AUC_0^∞) and repeated (AUC_τ^{EE}) schedules. For PR administration, the ratio was 96.95 and the confidence interval (95% CI) was 92.44–101.67. for IR administration, the ratio was 102.12 and the confidence interval (95% CI) was 96.75–107.80.

Urine pharmacodynamic analysis

Day 1 (single dose)

Diuretic effect: There were no differences in the urine volume between PR and IR torasemide administrations in the pre-administration collection interval (from −12 h to basal), in the total interval after drug administration (from basal to +24 h), or in the individual post-administration collection intervals (basal to +1 h, +1 to +2 h, +2 to +4 h, +4 to +6 h, +6 to +12 h and +12 to +24 h). However, a tendency to produce a lower volume of urine after PR administration was observed at the 0 to +1 h collection interval (PR: 393.20 mL, IR: 573.00 mL; P = 0.060) (*Figure 2*, upper panel).

Electrolytic effect: There were no differences in urine sodium, chloride or potassium amounts between PR and IR administration in the pre-administration collection interval (from −12 h to basal), in the total interval after drug administration (from basal to +24 h), or in the individual post-administration collection intervals.

Day 4 (repeated dose)

Diuretic effect: There were no differences in the urine volume between PR and IR administrations in the basal to +24 h interval after drug administration or in the majority of the individual post-administration collection intervals (+1 to +2 h, +2 to +4 h, +4 to +6 h, +6 to +12 h and +12 to +24 h). However, the urine volume was significantly lower after PR administration in the basal to +1 h (PR: 455.0 mL; IR: 578.27 mL, P = 0.049) collection interval (*Figure 2*, lower panel).

Electrolytic effect: There were no differences in sodium, chloride and potassium urine amounts between PR and IR administrations in the basal to +24 h interval after drug administration or in the individual post-administration collection intervals.

Urinary urgency

Day 1 (single dose)

After PR administration, all 16 volunteers had from one to three successive episodes of urinary urgency within the 0–6 h interval, at +1.16, +2.01 and +3.28 h mean time-points, and with a mean subjective evaluation intensity of 79.81, 84.44 and 69.06.

After IR administration, all 16 volunteers had from one to three successive episodes of urinary urgency within the 0–6 h interval at +0.55, +1.39 and +1.44 h mean time-points and with a mean subjective evaluation intensity of 85.50, 84.69 and 73.56.

Day 4 (repeated dose)

After PR administration, all 16 volunteers had one or two episodes of urinary urgency within the 0–6 h interval at +1.37 and +3.03 h mean time-points. The mean subjective evaluation of intensity was 81.25 and 74.06, respectively.

After IR administration, 15 volunteers presented one or two episodes of urinary urgency at +1.12 and +3.03 h mean time-points. The mean subjective

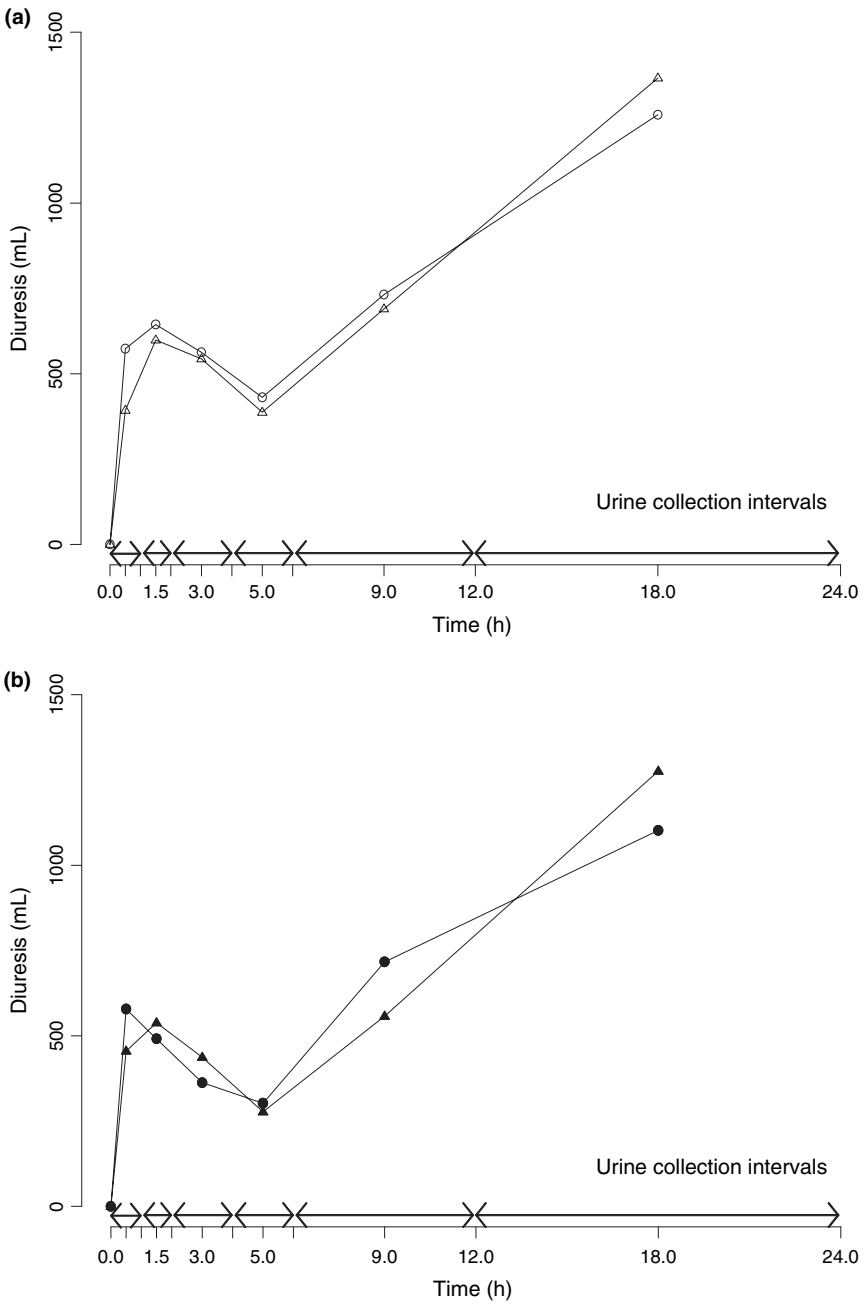

Figure 2 Mean urine volume excreted after 10 mg oral torasemide both on day 1 (single dose: upper panel) and day 4 (repeated once daily dose: lower panel) in 16 healthy volunteers of both sexes. (Δ, ▲) PR formulation and (○, ●) IR formulation. Data are plotted against the midpoint of the collection interval.

evaluation of intensity was 83.87 and 77.20, respectively. One volunteer presented only one episode.

Safety and tolerability

A total of 26 adverse events (AE) were reported during the study (16 when receiving PR and 10 when receiving IR). When PR was administered, 10 AE were qualified as 'not drug-related' (three decreases in hemoglobin, two dysmenorrheas, two viral gastroenteritis, one tonsillitis, one vomiting, one general discomfort) and only six as 'possibly drug-related' (two tachycardia, two nervousness, one diarrhea, one general discomfort). When IR was administered three AE were qualified as 'not drug related' (two decreases in hemoglobin, one urinary retention and seven as 'possibly drug-related' (three headaches, two nervousness, one dizziness, one diarrhea). No serious AE were reported and no subject withdrew because of drug intolerance.

No clinically significant abnormal trends or values were observed during the study either in vital signs

(systolic and diastolic blood pressure, heart rate) or in ECG parameters. Although some laboratory findings were outside the normal range, these deviations were generally minor and were not considered clinically relevant.

DISCUSSION

The main objective of this study was to compare the plasma pharmacokinetic profile of a PR formulation of torasemide with that of an IR formulation after repeated-dose administration in healthy volunteers. Our findings show that after single and repeated administrations of 10 mg oral doses of torasemide-PR and torasemide-IR, the former showed a typical plasma pharmacokinetic profile of a PR form. When the PR and IR formulations were compared we observed a lower peak of plasma levels in the former, represented by reduced and delayed C_{max} values. Nevertheless, a similar extent of systemic exposure, represented by AUC values, was seen. Additionally, after repeated administration, the PR formulation showed significantly lower fluctuations of plasma concentrations.

Evaluation of the rate of absorption (C_{max} and t_{max}) after a repeated administration schedule clearly indicated that after 4 days of a once daily 10 mg oral dose, the PR formulation showed lower torasemide concentration peaks and more delayed concentration–time profiles (significantly longer t_{max}) than the IR formulation. This demonstrated that the PR formulation has a slower absorption rate. The extent of absorption of the two formulations was comparable with the 90% confidence intervals for the ratio of AUC_{τ}^{EE}, and was within the accepted equivalence range of 0.8–1.25 [19]. This indicates that almost the same proportion of torasemide reached the systemic circulation with both formulations. However, mean residence time was significantly higher with the PR formulation.

The PR formulation showed a significantly lower fluctuation index than IR, in keeping with the more sustained plasma levels of torasemide-PR. This was not only the consequence of the lower C_{max} achieved after each administration but also the result of a higher C_{min}. However, these higher C_{min} did not lead to a progressive accumulation of the compound as there were no differences between mean plasma concentration values obtained at the different troughs assayed (+24 h after the first, third and fourth doses). This finding demonstrates that a steady-state plasma concentration–time profile has been achieved [20].

Results concerning the rate and extent of absorption after repeated administration (day 4) were in total accordance with those obtained at the first single administration (day 1) (C_{max}, AUC_0^t, AUC_0^{∞}, t_{max}) and with those found using the same torasemide formulation but after single oral administration of doses of 5 and 10 mg (data on file). This shows the linearity of torasemide plasma pharmacokinetics after repeated administration, as the 95% confidence interval of the ratio of the AUC_0^{∞} (single dose) to AUC_{τ}^{EE} (repeated dose) geometric means were within the accepted equivalence range of 0.80–1.25, after both PR and IR formulations [21]. This pharmacokinetic linearity after repeated administration has been reported previously after dosage increases [1,8].

The values of the pharmacokinetic parameters calculated for the PR and IR formulations, (AUC_{0-t}, $AUC_{0-\infty}$, C_{max}, t_{max}, k_e, $t_{1/2}$) were similar to those obtained in our previous study in healthy volunteers. The parameters for the IR formulation were also similar to previously reported data [1,2,5,6,12,22].

The pharmacodynamic profile of the two formulations was also evaluated in this study. Urine volume as well as total amount of sodium, chloride and potassium after drug administration (from basal to +24 h period) did not differ between PR and IR. When analysing shorter time periods, differences between PR and IR were found. Urine volume was lower after PR than after IR at the basal to +1 h collection interval after single administration and even more so after repeated administration. Thus, changes observed in plasma pharmacokinetic profile between both formulations had a limited effect on pharmacodynamics, showing the PR formulation lower effects in the first hour post-administration, while effects remained similar throughout the drug administration period. This was in accordance with results obtained in our previous study administering only a single oral dose of 5 and 10 mg torasemide.

To evaluate clinical safety and tolerability of both torasemide formulations, urinary urgency was evaluated by subjective reports using visual analog scales. The incidence of subjective urgency was similar with both formulations, after both single and repeated administration. However, with the PR formulation, these events occurred later than with the IR formulation, generally at double the time, especially after the single dose, and they were quantified with an approximate average of 4% lower intensity. These differences, although slight, reflect the lower urine volume obtained with the PR formulation during the first hour post-administration and suggest a more physiologic diuresis in comparison with

the IR formulation. The total number of possible drug-related side-effects was similar for both torasemide formulations. No serious AE were observed. There were no changes in vital signs or ECG parameters throughout the study. Furthermore, laboratory assessments prior to and following treatment did not show any changes that could be attributed to the study medications.

Concerning the loop of Henle diuretics, a relatively slow drug input to the site of action has been shown to reduce the disadvantages associated with rapid changes in plasma levels, thereby leading to increased efficiency. Wakelkamp et al. [23] observed this behavior in controlled release formulations of furosemide. Slow input at the site of action might also help to prevent the compensatory and antagonistic renal sodium retention that may occur during diuretic activity and after the drug effect has subsided [24,25]. Additionally, and especially in outpatients, it could be important to avoid an acute increase in diuresis as this could interfere with normal daily activity [26]. It would therefore be desirable to use PR formulations that provide therapeutic plasma levels with lower fluctuations between C_{max} and C_{min} once the steady-state is achieved, reducing the probability to attain subtherapeutic or toxic levels.

These findings indicate that the PR formulation of torasemide has a lower C_{max} than the IR formulation. A similar extent of systemic exposure is maintained after repeated daily administrations, leading to lower fluctuations in plasma concentrations during the dosing interval. This plasma pharmacokinetic profile is associated with urinary urgency occurring later in time and subjectively quantified as slightly less intensive.

CONCLUSION

After single and repeated administration of torasemide-PR and torasemide-IR, the extent of systemic exposure (AUC) was similar for both formulations. However, torasemide-PR had a slower rate of absorption (C_{max}) and thus presented a lower fluctuation of plasma concentrations. Urine evaluations were similar with both formulations, and episodes of acute urinary urgency occurred later and were subjectively less intensive with PR. The PR-formulation was well tolerated and showed a good safety profile.

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