BIOPHARMACEUTICS & DRUG DISPOSITION, VOL. 17, 81–90 (1996)

RELATIVE BIOAVAILABILITY OF FOUR CLOMIPRAMINE HYDROCHLORIDE TABLET PRODUCTS

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ABSTRACT

The relative bioavailability of clomipramine was determined in two single-blind, single-dose, randomized, crossover studies. In the first study, the relative bioavailability of the test product, 2 × 25 mg clomipramine hydrochloride tablets (Noristan Ltd.), with respect to the reference product, Anafranil® 2 × 25 mg tablets (clomipramine HCl; Ciba–Geigy (Pty) Ltd.) was determined. In the second study, the relative bioavailability of the test product, 5 × 10 mg clomipramine hydrochloride tablets (Noristan Ltd.), with respect to the reference product, Anafranil® 5 × 10 mg tablets (clomipramine HCl; Ciba–Geigy (Pty) Ltd.), was determined.

The geometric mean values for the variable C_{max} were 31·3 ng mL^{-1} for the reference and 31·6 ng mL^{-1} for the test product in study 1. The geometric mean values for the variable AUC were 736 ng h mL^{-1} and 753 ng h mL^{-1} for the reference and test, respectively. In study 2, the geometric mean C_{max} values were 25·8 ng mL^{-1} and 23·9 ng mL^{-1} for the reference and test respectively; the geometric mean AUC values were 569 ng h mL^{-1} and 547 ng h mL^{-1}.

The 90% confidence intervals for the 'test/reference' mean ratios of the plasma clomipramine pharmacokinetic variables C_{max} and $AUC_{(0-\infty)}$ (as measures of the rate and extent of absorption of clomipramine, respectively) fall within the conventional bioequivalence range of 80–125% for both studies. The test products (clomipramine HCl) are therefore bioequivalent to the reference products (Anafranil®) with respect to the rate and the extent of absorption of clomipramine in both 10 mg and 25 mg strengths.

KEY WORDS: clomipramine; bioequivalence; Anafranil®

INTRODUCTION

Clomipramine belongs to the group of tricyclic antidepressants and is a 3-chloro analogue of imipramine. The drug is recommended in depression when sedation is required, in cataplexy associated with narcolepsy and in obsessive

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CCC 0142–2782/96/010081–10

Received 24 January 1995
Accepted 7 June 1995

compulsive disorders. The daily dosage for adults may vary from 25 mg to 75 mg.[1]

Clomipramine is readily absorbed from the gastrointestinal tract and extensively demethylated by first-pass metabolism in the liver to its primary active metabolite, demethylclomipramine. Steady state concentrations of demethylclomipramine are usually 2·5 times higher than those of the parent compound.[2]

The metabolic pathways of both clomipramine and demethylclomipramine include hydroxylation and *N*-oxidation. Clomipramine is excreted in the urine, mainly in the form of its metabolites, either free or in conjugated form. Clomipramine and demethylclomipramine are widely distributed throughout the body and are extensively bound to plasma and tissue proteins.

The relative bioavailability of four clomipramine products was determined in two studies. The relative bioavailability of 10 mg clomipramine hydrochloride tablets (Noristan Ltd.) with respect to the reference product, Anafranil® 10 mg tablets (clomipramine HCl; Ciba–Geigy (Pty) Ltd.) was determined in the first study. In the second study, the relative bioavailability of 25 mg clomipramine hydrochloride tablets (Noristan Ltd.) with respect to Anafranil® 25 mg tablets (clomipramine HCl; Ciba–Geigy (Pty) Ltd.), was determined.

The pharmacokinetics of clomipramine have been less well documented with formal studies than those of other tricyclic antidepressants.[2] In this paper we report pharmacokinetic data of clomipramine from two controlled studies with 20 healthy volunteers each.

MATERIALS AND METHODS

Study population

Twenty healthy, non-smoking, female volunteers completed study 1 (age, 18–37 years) and 20 healthy, non-smoking, female volunteers completed study 2 (age, 18–24 years). No volunteer took part in both studies.

All volunteers gave written informed consent after they had received detailed instructions about the study performance, restrictions and possible adverse events that may be experienced as a result of taking the study drugs. The studies were approved by the Ethics Committee of the University of the Orange Free State, and were conducted in accordance with good clinical practice guidelines.[3] The volunteers underwent medical and clinical examinations, before and after each study.

Study design

Both were single-blind, single-dose, randomized, crossover studies with 21 day wash-out periods between profile days. Volunteers received each of two

treatment regimens once in each study, according to the randomization schedule.

Study 1. Test. 2×25 mg clomipramine hydrochloride tablets (Noristan Ltd.).

Reference. 2×25 mg Anafranil® tablets (clomipramine hydrochloride; Ciba–Geigy (Pty) Ltd.).

Study 2. Test. 5×10 mg clomipramine hydrochloride tablets (Noristan Ltd.).

Reference. 5×10 mg Anafranil® tablets (clomipramine hydrochloride; Ciba–Geigy (Pty) Ltd.)

Study performance

Volunteers were not allowed to take any medication, with the exception of oral contraceptives and the study drugs, for 14 d prior to and during the studies. Strenuous physical activities or ingestion of alcohol or caffeine containing food or beverages were not allowed for 24 h before and after administration of the study drugs.

On the morning of profile days for both studies, the volunteers reported to the FARMOVS clinic after an overnight fast of at least 10 h. After insertion of an indwelling venous cannula, and after predose blood samples had been drawn, the volunteers ingested the appropriate study drugs, according to the randomization schedule, with 100 mL tap water. The volunteers remained recumbent for 8 h after drug administration, except for bladder voiding after 3 and 6 h. A standardized meal was served 5 h after drug administration. The volunteers received 200 mL tap water at 2, 4, and 7 h after drug administration and 200 mL orange juice with their meal. Eight hours after drug administration, the volunteers were allowed to leave the FARMOVS clinic with the proviso that they would return for the subsequent sample collections. Food and fluid intake was allowed *ad libitum* after the volunteers had left the FARMOVS clinic.

Blood sampling

Blood samples were collected, handled and assayed under similar conditions for both studies 1 and 2.

Blood samples (10 mL) were taken *via* an indwelling venous cannula according to the following time schedule: before drug administration (0 h), 1, 2, 3, 4, 5, 6, 7, and 8 h and thereafter by venipuncture at 10, 12, 24, 36, 48, 60, 72, 84, 96, 120, and 144 h after drug administration.

Within 10 min of collection, the blood samples were centrifuged at 950–1240 *g* for 9–11 min and from each sample two aliquots of plasma were

transferred to labelled tubes. All samples were handled at room temperature before storage at −20 °C pending clomipramine assays.

Assay performance

To 0·5 mL plasma in a 5 mL glass culture tube was added 0·5 mL water containing 20 ng imipramine as internal standard and the following solid phase extraction (SPE) procedure performed. The sample was loaded onto an activated Bond-Elut, C8, 100 mg (Varian) solid phase extraction column. After washing with 1 mL water followed by 1·5 mL of acetonitrile:water (1:1) and 0·5 mL acetonitrile, the analyte was eluted with 1 mL methanol and the eluate evaporated to dryness under nitrogen. The residue was reconstituted in 50 μL methanol of which 20 μL was injected onto the HPLC column (Waters Nova-Pak C18, 4 μm, 150 mm × 3.9 mm ID). Detection was by an electrochemical detector (Coulochem model 5100A using a model 5011 analytical cell with the screen electrode at +0.4 V and the sample electrode at +0.7 V).

Between-day coefficients of variation, determined from quality control samples processed together with each batch of samples run, were between 4·5% and 10% for concentrations ranging between 4·7 ng mL^{-1} and 47 ng mL^{-1} and the accuracy was 95–102%. The limit of quantification was set at 2 ng mL^{-1}.

Pharmacokinetic variables

To compare the rate and extent of absorption of clomipramine in both studies, the following pharmacokinetic variables were calculated for each volunteer and product using the actual blood sampling times:

(i) the maximum concentration (C_{max});
(ii) the time to maximum concentration (t_{max});
(iii) the apparent terminal half-life ($t_{1/2\,z}$);
(iv) the area under the plasma concentration–time data pairs ($AUC_{(0-t\ last)}$);
(v) the area under the plasma concentration–time data pairs, with extrapolation to infinity ($AUC_{(0-\infty)}$);
(vi) the ratio of C_{max} and $AUC_{(0-\infty)}$($C_{max}/AUC_{(0-\infty)}$); and
(vii) the total mean time in the system (MT_{vsys}).

In addition, the relative total clearance (CL_{tot}/f) was calculated. CL_{tot}/f was also normalized for body mass.

Statistical analysis

The test and reference treatments of each study were compared with respect to relevant pharmacokinetic variables using an analysis of variance with volunteer, product, and period effects after a logarithmic transformation of the

Table 1. Summary of pharmacokinetic data for clomipramine (dose 2 × 25 mg clomipramine HCl tablets; $n = 20$)

Variable	Unit	Anafranil® (reference) Geometric mean	SD	Range	Clomipramine HCl (test) Geometric mean	SD	Range	Mean ratio (%)[a]	90% CI (%)[b]
C_{max}	ng mL^{-1}	31·3	1·48	14·2–66·4	31·6	1·43	10·7–50·3	101	93–109
t_{max}[c]	h	4·0		2·0–6·0	4·0		2·0–7·0	0·5	0–1·5
$AUC_{(0-t\ last)}$	ng h mL^{-1}	616	1·98	154–1796	619	2·05	75·6–1597	101	91–112
$AUC_{(0-\infty)}$	ng h mL^{-1}	736	1·97	177–2173	753	1·95	131–1797	102	92–113
C_{max}/AUC	h^{-1}	0·043	1·43	0·023–0·084	0·042	1·45	0·026–0·085	98	92–106
$t_{1/2z}$	h	23·5	1·97	6·96–55·2	23·8	1·89	6·47–65·2	101	86–120
MT_{vsys}	h	36·0	1·76	13·1–80·7	37·5	1·72	13·6–89·5		
CL_{tot}/f	mL min^{-1}	1015	1·97	344–4211	991	1·95	416–5695		
CL_{tot}/f[d]	mL min^{-1} kg^{-1}	17·3	1·92	6·48–71·4	16·9	1·92	7·84–96·5		

[a]Geometric mean of individual 'test/reference' ratios.
[b]90% CI for the 'test/reference' mean ratio after logarithmic transformation of the data.
[c]Medians, non-parametric estimate of the 'test − reference' median difference and corresponding 90% CI.
[d]Normalized for body mass.

Table 2. Summary of pharmacokinetic data for clomipramine (dose 5 × 10 mg clomipramine HCl tablets; $n = 20$)

		Anafranil® (reference)			Clomipramine HCl (test)				
Variable	Unit	mean	Geometric SD	Range	mean	Geometric SD	Range	Mean ratio (%)[a]	90% CI (%)[b]
C_{max}	ng mL^{-1}	25·8	1·48	8·81–66·3	23·9	1·47	10·9–39·1	92·5	84·8–101
t_{max}[c]	h	4·00		3·00–8·00	4·50		3·00–7·00	0·50	0·00–1·00
$AUC_{(0-t\ last)}$	ng h mL^{-1}	467	1·98	77·8–2037	429	2·20	69·0–1168	91·9	81·7–103
$AUC_{(0-\infty)}$	ng h mL^{-1}	569	1·92	138–2518	547	2·18	104–1775	96·2	85·1–109
C_{max}/AUC	h^{-1}	0·045	1·40	0·026–0·084	0·044	1·62	0·018–0·110	96·0	86·4–107
$t_{1/2\,z}$	h	24·1	2·23	6·39–87·6	27·1	2·43	5·12–107	112·5	89·4–142
MT_{vsys}	h	34·7	1·82	13·4–92·3	38·2	2·06	9·44–136		
CL_{tot}/f	mL min^{-1}	1313	1·92	297–5422	1364	2·18	421–7213		
CL_{tot}/f[d]	mL min^{-1} kg^{-1}	21·1	1·94	5·20–82·2	21·9	2·19	7·38–125		

[a]Geometric mean of individual 'test/reference' ratios.
[b]90% CI for the 'test/reference' mean ratio after logarithmic transformation of the data.
[c]Medians, non-parametric estimate of the 'test − reference' median difference and corresponding 90% CI.
[d]Normalized for body mass.

data. Point estimates and 90% confidence intervals (CI) for the 'test/reference' mean ratios of these variables were calculated.[4] Bioequivalence of the test and reference product was assessed on the basis of these CIs, in relation to the conventional bioequivalence range of 80–125%. In addition, a non-parametric point estimate and 90% CI for the 'test − reference' median difference of t_{max} was calculated.[5]

RESULTS

Safety results

There were no clinically significant adverse events or changes in vital signs, clinical chemistry, and haematological variables during either study.

The following adverse events were reported in both studies: dizziness, nausea, and headache. In addition, some volunteers reported jaw stiffness, abdominal pain, diarrhoea, and vomiting in study 2.

Pharmacokinetic results

The pharmacokinetic variables for studies 1 and 2 are summarized in Tables 1 and 2, respectively. Table 3 gives the intersubject and intrasubject coefficients of variation (CV) for the variables C_{max}, AUC, C_{max}/AUC, and $t_{1/2z}$.[6] The geometric mean plasma clomipramine concentrations are represented in Figures 1 and 2, respectively. In study 1, the geometric mean values for the variable C_{max}were 31·3 ng mL^{-1} for the reference and 31·6 ng mL^{-1} for the test product. The geometric mean values for the variable AUC were 736 ng h mL^{-1}

Table 3. Intrasubject, intersubject, and pooled CVs of plasma clomipramine pharmacokinetic variables calculated from log-transformed data

Variable	Intrasubject CV_ε (%)	Intersubject CV_s (%)	Pooled CV_p (%)	Pooled (reference) CV_R (%)	Pooled (test) CV_T (%)
Study 1 (dose 2 × 25 mg clomipramine HCl tablets; $n = 20$)					
C_{max}	14·5	35·3	38·5	39·0	38·0
$AUC_{(0-\infty)}$	18·8	73·4	77·0	76·6	77·4
C_{max}/AUC	12·9	36·2	38·8	38·3	39·2
$t_{1/2z}$	31·5	65·6	75·6	78·5	72·7
Study 2 (dose 5 × 10 mg clomipramine HCl tablets; $n = 20$)					
C_{max}	16·0	37·0	40·8	41·9	39·6
$AUC_{(0-\infty)}$	22·7	79·4	84·5	75·2	93·6
C_{max}/AUC	19·5	39·4	44·6	35·7	52·4
$t_{1/2z}$	43·7	86·3	103·8	98·5	109·2

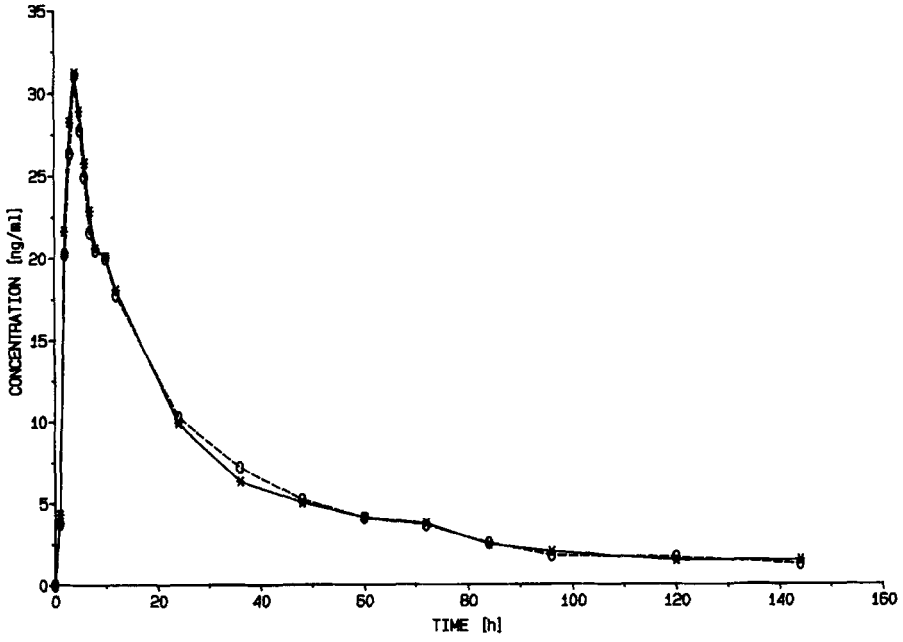

Figure 1. Geometric mean plasma clomipramine concentrations ($n = 20$) following single-dose administration of 2 × 25 mg Anafranil® tablets (x) and following single-dose administration of 2 × 25 mg clomipramine HCl tablets (○)

and 753 ng h mL^{-1} for the reference and test, respectively. In study 2, the geometric mean C_{max} values were 25·8 ng mL^{-1} and 23·9 ng mL^{-1} for the reference and test respectively; the geometric mean AUC values were 569 ng h mL^{-1} and 547 ng h mL^{-1}.

In study 1, the point estimates (90% CI) of the 'test/reference' mean ratio for C_{max} are 101% (93–109%), for AUC 102% (92–113%), and for C_{max}/AUC 98% (92–106%). In study 2, the point estimates (90% CI) of the 'test/reference' mean ratio for C_{max} are 93% (85–101%), for AUC 96% (85–109%), and for C_{max}/AUC 96% (86%–107%).

DISCUSSION AND CONCLUSION

Pharmacokinetic data on clomipramine are sparse.[1,2] According to Balant-Gorgia *et al.*[2] the only comprehensive study on the kinetics of clomipramine after a single dose in normal conditions is that by Evans *et al.*[7] The data from our two studies agree well with each other, and compare well with the data reported by Evans *et al.*[7] following oral administration of 50 mg clomipramine

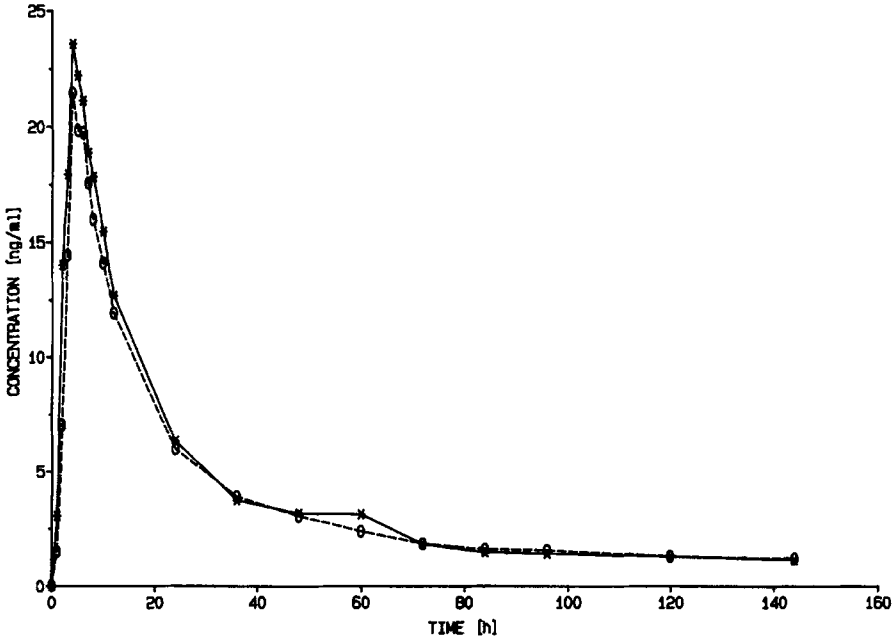

Figure 2. Geometric mean plasma clomipramine concentrations ($n=20$) following single-dose administration of 5 × 10 mg Anafranil® tablets (x) and following single-dose administration of 5 × 10 mg clomipramine HCl tablets (○)

to nine volunteers. Average C_{max}, AUC, and $t_{1/2z}$ values reported by Evans *et al.*[7] were 27·6 ng mL^{-1}, 489 ng h mL^{-1}, and 20 h respectively. Data reported by Evans *et al.*[7] included five females and four males, whereas our studies included only females. As our data are comparable to those of Evans *et al.*,[7] no gender differences in the bioavailability of clomipramine are apparent. Evans *et al.*,[7] however, did not report or compare the relative bioavailability of clomipramine in males and females.

Our data also confirm earlier reports[1] about the high intersubject variability in plasma clomipramine concentrations (Table 3).

Concerning the relative bioavailability of the respective test and reference products, the 90% CIs for the 'test/reference' mean ratios of the plasma clomipramine pharmacokinetic variables C_{max} and AUC (as measures of the rate and extent of absorption of clomipramine, respectively) all fall within the conventional bioequivalence range of 80–125%. The test products (clomipramine HCl) are therefore bioequivalent to the reference products (Anafranil®; clomipramine HCl) with respect to the rate and the extent of absorption of clomipramine for both 10 mg and 25 mg strength tablets.

ACKNOWLEDGEMENTS

The authors thank Noristan Ltd. for financial support, the nursing and technical staff of the FARMOVS Institute for assistance with the clinical study, Mr. L. W. Brown for the clomipramine assays, Mr. M. J. Saayman for the statistical analysis, and Mrs. L. van der Westhuizen for assistance in preparing this manuscript.

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