The absolute bioavailability and metabolic disposition of the novel antimigraine compound zolmitriptan (311C90)

E. Seaber, N. On, R. M. Dixon,* M. Gibbens, W. J. Leavens, J. Liptrot, G. Chittick, J. Posner, P. E. Rolan† & R. W. Peck

Division of Clinical Pharmacology, Glaxo Wellcome Research and Development, Greenford, Middlesex, UK

Aims Two open studies in healthy volunteers were conducted to determine the absolute bioavailability and metabolic disposition of zolmitriptan (311C90), a novel $5HT_{1D}$ agonist for the acute treatment of migraine.

Methods After an initial test i.v. infusion, bioavailabilty was assessed by comparison of AUC after an i.v. infusion (3.5 mg) and an oral tablet (10 mg), in six men and six women using a randomised, crossover design. Disposition was studied by administration of a 25 mg capsule, labelled with 100 μCi [^{14}C]-zolmitriptan, to five men and one woman on a single occasion.

Results Zolmitriptan was well tolerated by both i.v. and oral routes. Adverse events were mostly mild, consistent with earlier studies and characteristic of this class of drug. Reports were similar in nature and number after both oral and iv dosing. Mean $\pm$ s.d. oral bioavailability was 0.49 ± 0.24 (0.38 ± 0.16 in men and 0.60 ± 0.28 in women). After oral dosing, C_{max} and AUC values in women were approximately double those in men. Relative to zolmitriptan concentrations, metabolite concentrations were higher after oral dosing than after i.v., and higher in men compared with women. Half-life was significantly longer after oral dosing (mean 22%, 95% CI 6–35%). Mean $\pm$ s.d. values for CL, V_z and $t_{1/2,z}$ after i.v. dosing (all subjects) were 8.7 ± 1.7 ml min^{-1} kg^{-1}, 122 ± 32 l and 2.30 ± 0.59 h respectively. Following administration of 25 mg [^{14}C]-zolmitriptan, 91.5% of the dose was recovered in 7 days, 64.4 ± 6.5% in urine and 27.1 ± 6.0% in faeces. Less than 10% was recovered unchanged in urine, with 31.1 ± 6.4% recovered as the inactive indole acetic acid metabolite. Most of the faecal material was unchanged zolmitriptan, representing unabsorbed drug. Plasma concentrations of [^{14}C] were slightly higher than those of the summed concentrations of known analytes zolmitriptan, the active *N*-desmethyl metabolite (183C91), the inactive *N*-oxide (1652W92) and indole acetic acid (2161W92) metabolites, which accounted for 86% of total plasma radioactivity. No other significant metabilites were detected in plasma. Some minor additional metabolites were detected in urine, none of which contributed more than 5% of the dose.

Conclusions The data suggest that zolmitriptan undergoes first-pass metabolism and this is more extensive in men than in women. Zolmitriptan has suitable bioavailabilty for an acute oral migraine treatment and there are no significant unidentified metabolites in man.

Keywords: bioavailability, metabolism, 311C90, zolmitriptan, radiolabel, pharmacokinetics, gender

Introduction

Zolmitriptan (311C90, (S)-4[[3-[2-(dimethylamino)ethyl]-1H-indol-5-yl]methyl-2-oxazolidinone) is a potent and highly selective $5HT_{1D}$ receptor agonist in development for the acute treatment of migraine [1]. Action on '$5HT_{1D}$-like' receptors present in the cranial circulation is thought to inhibit release of the neuropeptides (CGRP and substance P) from perivascular trigeminal sensory neurones [2, 3]. $5HT_{1D}$ receptors have also been shown to modulate nociceptive input at the level of the trigeminal nucleus caudalis, suggesting that central sites may represent an additional target for migraine treatment [4]. In animal models, zolmitriptan has been shown to act at these central sites in addition to its peripheral vascular effects [5] and in this way differs from sumatriptan which does not appear to cross the blood-brain barrier [4].

Zolmitriptan is well tolerated by healthy volunteers and patients at doses up to 25 mg [6]. In double-blind,

Correspondence: Ms E. Seaber, Division Clinical Pharmacology, Glaxo Wellcome Research and Development, Greenford Rd, Greenford, Middlesex, UB6 0HE, UK
Current addresses: *Zeneca Pharmaceuticals, Mereside, Alderly Park, Macclesfield, Cheshire, SK10 4TG and †Medeval Ltd, University of Manchester, Skelton House, Manchester Science Park, Lloyd Street North, Manchester, M15 6SH

placebo-controlled studies, zolmitriptan was effective in 62-81% of migraine patients following oral doses of between 2.5 and 25 mg [7]. Zolmitriptan is well absorbed with dose-independent kinetics and is extensively metabolized [8]. The metabolites hitherto identified in human plasma and urine are the same as those seen in the rat and dog, namely the *N*-desmethyl metabolite (183C91) which has $5HT_{1D}$ agonist activity and the inactive *N*-oxide (1652W92) and indole acetic acid (2161W92) metabolites. Oral bioavailability was estimated to be 41% and 79% in the rat and dog respectively. We now report the results of studies to assess the absolute oral bioavailability of zolmitriptan using intravenous and oral zolmitriptan and its disposition using [^{14}C]-zolmitriptan in healthy volunteers.

Methods

Subjects

Both the absolute bioavailability and metabolic disposition studies were carried out in healthy volunteers at the Wellcome Clinical Investigation Unit, King's College Hospital, London. Volunteers were non-smokers with no clinically significant abnormalities at medical screening. They were excluded if they had participated in other clinical trials or donated blood during the preceding 2-3 months, showed evidence of drugs of abuse in the urine, had either a systolic or diastolic blood pressure greater than 140/90 mmHg respectively or a total plasma cholesterol level exceeding 6.5 mmol l^{-1}. All subjects gave written informed consent before participation and the protocol was approved by the independent Wellcome Protocol Review Committee and Kings Healthcare Research Ethics Committee prior to the start of the study. Approval was obtained from the Administration of Radioactive Substance Advisory Committee (ARSAC) to permit the administration of the radioactive substance to humans.

Study design

Absolute bioavailability (Study 1) Twelve healthy volunteers (6M, 6F) aged 18–40 years participated in this open study on each of three study occasions. The study comprised two parts; a first 'test-dose' occasion was to establish the tolerability to intravenous zolmitriptan and to determine the pharmacokinetics of zolmitriptan and its metabolites when administered by this route. In the second crossover part, the same 12 subjects were randomized to receive a single zolmitriptan 10 mg oral tablet on one occasion and on another occasion, an intravenous dose predicted to give mean plasma zolmitriptan concentrations approximately equivalent to a 10 mg oral dose. The data obtained were used to estimate the absolute bioavailability of the zolmitriptan 10 mg tablet. Using data from a previous study [8], it was estimated that a bioavailability of 100% would have a confidence interval (CI) of approximately $\pm 10\%$. To take into account the possibility that pharmacokinetics would differ between the sexes, the randomisation code was stratified, such that on each of the crossover occasions half the subjects of each sex would would receive the 10 mg tablet and half the 3.5 mg i.v. dose.

Consumption of caffeine-containing beverages was not allowed on any of the study days and volunteers were fasted overnight before each study day. On the first study occasion (test-dose), subjects received a 2.5 mg dose of zolmitriptan, diluted in 250 ml normal saline, given by continuous intravenous infusion using an IVAC pump over a 2 h period. A group mean AUC was calculated and compared with data from a previous administration of oral zolmitriptan to volunteers [8]. It was estimated that an i.v. dose of 3.5 mg given over 4 h would result in a similar plasma profile to an oral 10 mg dose. Thus, on the second and third occasions, subjects were randomised to receive a single zolmitriptan 10 mg tablet or a 3.5 mg i.v. infusion. The three study occasions were separated by a minimum interval of 1 week. Blood samples were taken via an intravenous cannula pre-dose and then at 0.25, 0.5, 0.75, 1, 1.5, 2, 2.25, 2.5, 2.75, 3, 3.5, 4, 5, 6, 7, 8, 10, 12, 15 and 24 h post-dose on the first study occasion and at 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 5.5, 6, 7, 8, 9, 10, 12, 15 and 24 h on the second and third occasions, to take account of the prolonged infusion time. All urine was collected in three fractions up to 24 h, weighed, and then aliquots taken for assay of zolmitriptan and metabolites.

Metabolic disposition (Study 2) Six healthy volunteers (5M, 1F) participated in this single occasion open study, receiving a single capsule of 25 mg zolmitriptan, labelled with 100 μCi [^{14}C]-zolmitriptan. Volunteers had to be 30–55 years of age, females had to be incapable of child-bearing (post-menopausal, post-hysterectomy or sterilised) and males had to give an undertaking that he and/or his partner would use an adequate and reliable form of contraception for not less than 3 months from the study day. Plasma and whole blood samples were taken pre-dose and then at 0.25, 0.5, 0.75, 1, 1.5, 2, 2.5, 3, 4, 5, 6, 7, 8, 10, 12, 15, 24, 32 and 48 h post-dose. All urine and faeces were collected up to 168 h post-dose and weights recorded. Total radioactivity was determined in all samples of blood, plasma, urine and faeces and performed on selected plasma, urine and stool samples. Plasma and urine samples were analysed for zolmitriptan and known metabolites by a specific h.p.l.c. assay.

Clinical investigations In both studies, blood pressure and heart rate were measured at intervals in a supine position, using an automatic oscillometric device (Hewlett Packard 70354A). For the first 4 h post-dose, volunteers were confined to their beds and then rested for 10 min before each subsequent measurement. Any adverse experiences reported spontaneously or observed by the investigators were recorded. ECG was monitored continuously for 24 h by bedside monitor or telemetry and a central arrhythmia detection system with alarm (Hewlett Packard HP78560A). In addition, ECG was recorded by Holter monitoring (Hertford Medical, Hertford, UK). Detection of arrhythmias and quantitative analysis of ST segments was performed using the Reynolds Medical 'Professional' model P53 analyser. In addition, plasma biochemistry and full blood counts were performed both before and after each dosing occasion and inspected for abnormalities or trends.

Plasma and urine assays for zolmitriptan and metabolites Plasma and urine samples were analysed for the parent compound zolmitriptan and its major metabolites by solid phase extraction, followed by a validated, specific, reverse-phase h.p.l.c. method with fluorescence detection. The quantifiable ranges were between 2–150 ng ml^{-1} and 100–15000 ng ml^{-1} in plasma and urine respectively.

Plasma was separated by centrifugation of blood samples at 3000*g* for 10 min and stored at −20C° or below. Sample preparation was carried out using solid phase extraction on C_{18} Bond-Eluts from Anachem. Extraction cartridges were prewashed with 1 ml acetonitrile, 0.5 ml water and 1 ml sodium formate buffer (pH 4.0). A 0.1 ml aliquot of plasma and 25 μl of internal standard (890W92; (4S)-4-[3-(1-methyl-4-piperidyl)-1H-indol-5-yl]2-oxazolidinthione) were applied to the cartridge and drawn through under low vacuum. The cartridge was then washed with 1 ml sodium formate buffer, 0.75 ml water and 0.5 ml of 5% acetonitrile in water (v/v). Elution of the analytes of interest from the cartridge was achieved by washing the cartridge with 0.3 ml of 30% acetonitrile: 70% 0.01M di-ammonium hydrogen ortho-phosphate (pH 3.0) (v/v). This was then transferred to an h.p.l.c. vial for injection. The system comprised a Zorbax TMS column (25 cm × 4.6 mm id), a mobile phase of 15% acetonitrile: 85% 0.01M di-ammonium hydrogen ortho-phosphate pH 3.0 (v/v) and an injection volume of 175 μl with a 1 ml min^{-1} flow rate. Fluorescence detection was at 350 nm using a VG Multichrom data system.

The volume of each urine collection was determined and a 20 ml aliquot taken and frozen at −20°C or below until assay. 0.4 ml of each urine sample was shaken for 10 min with 0.4 ml 1M formic acid and 10 ml chloroform. The sample was then centrifuged at 3000*g* for 10 min and 20 μl of the urine/formic mixture (equivalent to 10 μl of urine) drawn through a Bond-Eluts cartridge prepared as described above. The cartridge was washed and eluted as above except that 2% acetonitrile was used in washing and elution was with 0.06 ml 40% acetonitrile in 0.01M sodium dihydrogen orthophosphate. The h.p.l.c. consisted of a Zorbax SIL column (8 cm × 4 mm) and a guard cartridge followed by Supelcosil LC-ABZ (25 cm × 4.6 mm) with a mobile phase of (A) 5% (v/v) acetonitrile in 0.04M soduim dihydrogen orthophosphate and (B) 60% (v/v) acetonitrile in 0.04M sodium dihydrogen orthophosphate in the ratios 0 min: A = 100%, 10 min: A = 80%, 15 min: A = 0%, 25 min: A = 100%.

A calibration curve for each analyte was constructed from the calibration sample included in the assay using the peak height ratio of analyte/internal standard. Sample peak height ratios were then calculated and concentrations determined from the appropriate standard curve. The standard curve was a linear fit with a 1/concentration weighting. The assay precision and bias was less than 20% at the limit of quantification and less than 15% at concentrations of 10 ng ml^{-1} and above in plasma and 200 ng ml^{-1} and above in urine.

Determination of radioactivity Total radioactivity in whole blood, plasma, urine and faecal samples was determined by liquid scintillation counting (LSC) using either a Beckman LS5801 or LC5000 CE liquid scintillation counter. Duplicate aliquots of plasma and urine were weighed directly into scintillation vials and mixed with scintillator cocktail (Hionic Fluor). Triplicate aliquots of blood and faecal homogenates were measured for radioactivity following combustion either in a Packard Tricarb D306 or D307 sample oxidiser.

H.p.l.c. radioprofiling Urine and faecal samples containing sufficient radioactivity were analysed by an h.p.l.c. system comprising a Waters 715 Ultra WISP autosampler, a Jasco 875-UV detector and an LDC/Milton Ray solvent delivery system. A Berthold radioactivity monitor (LB506 C-1) with a LB5035 pump to deliver liquid scintillator was used to monitor radioactivity in the h.p.l.c. element. Urine was filtered using a Millipore 0.22 μm non-sterile Ultrafree-Mc centrifuged filter unit or an Anotop 0.2 μm disposable syringe filter unit prior to radioactive component h.p.l.c. analysis. Weighed aliquots of selected faecal homogenates were extracted with methanol and centrifuged prior to h.p.l.c. analysis. Separation of the analytes was achieved with a Zorbox Rx-C8 analytical column at ambient temperature, using a binary gradient. The two mobile phases consisted of (A) 0.1% (v/v) formic acid in h.p.l.c. grade water and (B) 0.1% (v/v) formic acid in acetonitrile. A flow rate of 1 ml min^{-1} was monitored with u.v. detection at 230 nm.

Metabolite identification by liquid chromatography-mass spectrometry (LC-MS) A similar h.p.l.c. system to the above, coupled to a Finnigan-MAT TSQ 700 mass spectrometer via a pneumatically assisted electrospray source, was used for all LC-MS and LC-MS-MS metabolite identification. Argon was used as the collision gas during tandem mass spectrometry.

Pharmacokinetic and statistical analysis

Absolute bioavailability study Non-compartmental pharmacokinetic parameters were determined for zolmitriptan and its metabolites for all three treatments using EXCEL 4.0. The observed peak plasma concentration, C_{max} and the time to reach the peak concentration, t_{max} were taken directly from the plasma concentration-time profiles. The area under the plasma concentration-time curve was estimated by the trapezoidal rule up to the last measurable concentration AUC(0, *t*) and extrapolated to infinity (AUC) by addition of C_t/λ_z where λ_z is the first-order elimination rate constant, obtained by log linear regression. The corresponding area under the first moment curve (AUMC) was calculated and the mean residence time (MRT) was calculated from MRT = AUMC/AUC following oral zolmitriptan and from AUMC/AUC-Infusion period/2 following intravenous zolmitriptan. The absolute bioavailability of oral zolmitriptan was calculated from $AUC_{po}/AUC_{iv} \times Dose_{iv}/Dose_{po}$. Elimination half-life ($t_{\frac{1}{2},z}$) was calculated as $\ln 2/\lambda_Z$. Clearance following intravenous administration (CL) was calculated as CL = Dose/AUC. Renal clearance (CL_R) was calculated as CL_R = Ae/AUC where Ae is the amount excreted in urine. Volume of distribution (V_z) was calculated as $V_z = CL/\lambda_z$.

Differences between the sexes for zolmitriptan pharmacokinetic parameters obtained from the 3.5 mg i.v. dose were estimated and 95% CI calculated using analysis of variance. All data (except t_{max}, Ae and MRT) were log-transformed

before analysis. Differences between the sexes for pharmacokinetic parameters (except t_{max}) for the metabolites, 183C91, 1652W92 and 2161W92, were also estimated (and 95% CI calculated). The half-life and AUC_m/AUC_p data for zolmitriptan from the 3.5 mg i.v. dose and the 10 mg tablet were compared using ANOVA.

Metabolic disposition study Pharmacokinetic parameters were calculated as described above for the absolute bioavailability study. C_{max} and AUC determined for plasma levels of radioactivity were expressed as nmol equivalents. The concentration of [^{14}C] in red blood cells was calculated as $C_{RBC}=(C_B\text{-}C_P(1\text{-Hct}))/\text{Hct}$, where C_{RBC} is the red blood cell concentration, C_B is the whole blood concentration, C_P is the plasma concentration and Hct is the haematocrit value. Percent recoveries of individual compounds and total radioactivity in urine and faeces were calculated.

Results

All subjects enrolled completed the studies and there were no major protocol deviations. In the bioavailability study, for females ($n=6$), mean age was 27 years (range 21–34 years), mean height was 169 cm (range 162–175 cm) and mean weight was 65 kg (range 58–72 kg); for males ($n=6$) mean age was 33 years (range 25–38 years), mean height was 177 cm (range 166–188 cm) and mean weight was 78 kg (range 61–95 kg). In the metabolic disposition study (5M, 1F) ages ranged from 32–47 years, heights from 162–198 cm and weights from 59–89 kg.

All but one subject in each study reported at least one adverse experience. None was serious and most were mild and transient (<3 h) reports of tightness, heaviness or pressure in any part of the body, but usually the face and neck. Such symptoms are characteristic of this class of drug [6]. There were more reports by women but the number and nature of reports were similar after both oral and i.v. dosing. No clinically significant changes in pulse, blood pressure, ECG or clinical/laboratory parameters were observed in either study.

Pharmacokinetics

Absolute bioavailability After i.v. dosing, C_{max} and AUC were higher in women than men with mean ratios of 79% (95% CI 59–106%) and 77% (95% CI 56–105%) respectively. Weight adjusted clearance was only slightly higher in men than women after i.v. dosing (Table 1) suggesting that most, but not all, of the concentration difference was due to differences in body weight. After dosing by the oral route, individual peak concentrations varied more widely than after iv and multiple peaks were observed, in common with earlier studies (Table 1, Figure 2). Mean C_{max} values were similar after the 3.5 mg i.v. and 10 mg oral doses. AUC values were similar in men after the oral and i.v. dose but were over 50% higher in women after oral dosing.

After oral dosing, C_{max} and AUC values were much lower in men with male : female ratio estimates of 61% (95% CI 35–107%) and 49% (95% CI 29–84%) respectively (Tables 1 and 2). After oral dosing $t_{\frac{1}{2},z}$ was 2.94 h compared with 2.30 h (all subjects) after i.v. dosing (95% CI for ratio 106–135%). The mean absolute bioavailability of the 10 mg tablet was estimated at 49%. In females, this was 60% with values ranging from 32 to 110% and in males was 38% with

Table 1 Mean (±s.d.) pharmacokinetic parameters for 311C90 and its metabolites following an intravenous infusion of 3.5 mg and an oral dose of 10 mg 311C90 to six healthy male and six healthy female volunteers.

	311C90		*183C91*		*2161W92*	
	Male (n = 6)	*Female (n = 6)*	*Male (n = 6)*	*Female (n = 6)*	*Male (n = 6)*	*Female (n = 6)*
			3.5mg i.v.			
C_{max} (ng ml^{-1})	16.6 ± 4.1	20.9 ± 4.4	3.9 ± 1.9	3.5 ± 0.7	7.1 ± 2.8	6.5 ± 1.7
t_{max} (h)[a]	4.0	3.5	4.0	4.3	4.25	4.15
$t_{\frac{1}{2},z}$ (h)	2.22 ± 0.68	2.38 ± 0.53	—	—	2.81 ± 0.71	3.35 ± 0.37
AUC (ng ml^{-1} h)	84.4 ± 24.3	108.6 ± 21.0	12.3 ± 6.6[b]	9.2 ± 7.3[b]	41.7 ± 13.0	46.9 ± 8.9
AUC_m/AUC_p	—	—	—	—	0.55 ± 0.14	0.49 ± 0.23
CL (ml min^{-1} kg^{-1})	9.2 ± 1.7	8.3 ± 1.7	—	—	—	—
V_z (l)	134 ± 38	109 ± 20	—	—	—	—
CL_R (ml min^{-1} kg^{-1})	3.0 ± 0.5	3.0 ± 0.7	—	—	7.1 ± 2.4	5.4 ± 1.2
% urine recovery	33.3 ± 4.8	37.2 ± 10.2	4.9 ± 2.7	4.6 ± 2.5	38.7 ± 4.5	31.1 ± 2.6
			10mg orally			
C_{max} (ng ml^{-1})	16.0 ± 7.8	25.2 ± 9.0	7.5 ± 2.7	8.0 ± 2.0	14.9 ± 4.0	16.0 ± 4.1
t_{max} (h)[a]	3.25	3.5	3.5	3.75	4.5	3.75
$t_{\frac{1}{2},z}$ (h)	2.62 ± 0.81	3.26 ± 0.43	2.30 ± 0.54	3.47 ± 1.40	3.75 ± 1.13	3.94 ± 1.04
AUC (ng ml^{-1} h)	88.9 ± 46.5	173.8 ± 58.1	52.2 ± 16.3	58.4 ± 17.1	107.2 ± 43.6	121.5 ± 15.0
AUC_m/AUC_p	—	—	0.56 ± 0.13	0.42 ± 0.24	1.31 ± 0.50	0.79 ± 0.36
F	0.38 ± 0.16	0.60 ± 0.28	—	—	—	—
CL/F (ml min^{-1} kg^{-1})	29 ± 15.8	16.0 ± 6.9	—	—	—	—
V_z/F (l)	463 ± 190	281 ± 80	—	—	—	—
CL_R (ml min^{-1} kg^{-1})	2.5 ± 1.0	2.5 ± 0.7	2.0 ± 0.8	1.8 ± 0.4	6.9 ± 2.6	5.5 ± 0.5
% urine recovery	9.5 ± 2.2	17.9 ± 7.2	4.4 ± 1.1	4.2 ± 0.8	32.2 ± 6.8	27.7 ± 2.3

[a]median.
[b]AUC (0, t).

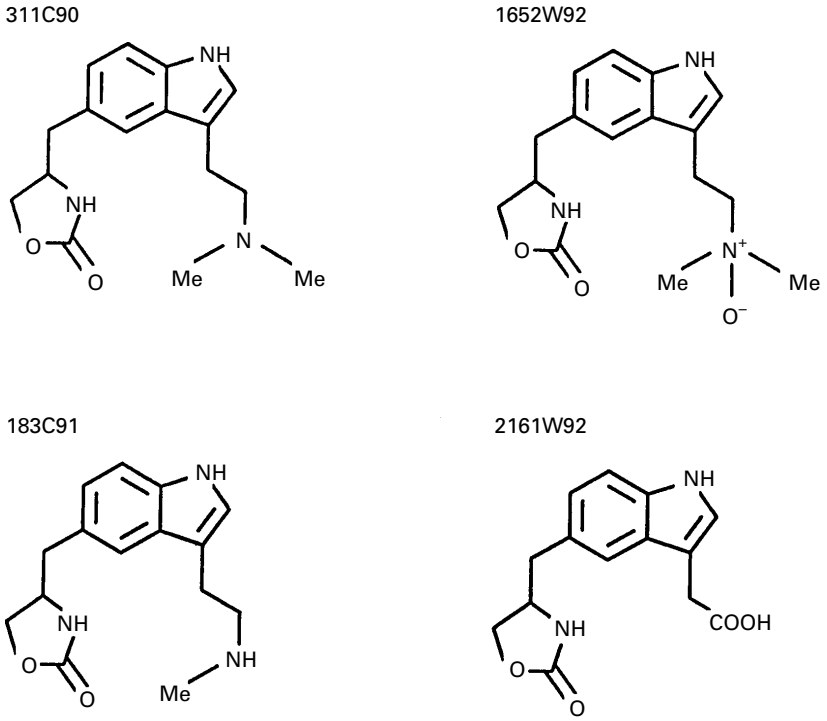

Figure 1 Chemical structures of 311C90 and its principal metabolites.

individual values ranging from 21 to 62%. This difference just failed to reach statistical significance.

Mean peak concentrations of 183C91, the pharmacologically active *N*-desmethyl metabolite, were approximately two-fold higher after oral dosing than intravenous and were 20% and 38% respectively of the peak concentrations of the parent following the 3.5 i.v. dose and 10 mg oral dose respectively (Table 1). Due to the low concentrations of 183C91 after i.v. dosing with zolmitriptan it was not possible to determine the elimination rate constant in any subjects; consequently no pharmacokinetic parameters could be estimated. After oral dosing parameters could be estimated in nine subjects. Parameters were largely consistent between sexes although, relative to peak concentrations of zolmitriptan, peak concentrations of 183C91were higher in men. Renal clearance was slightly higher after oral dosing than i.v. The t_{max} and $t_{\frac{1}{2},z}$ were similar to the parent compound.

Plasma concentrations of 1652W92, the inactive *N*-oxide metabolite, were generally too low to enable accurate determination of the elimination rate constant and other pharmacokinetic parameters. Relative to zolmitriptan peak concentrations, the mean peak concentrations of 1652W92 following the 10 mg oral dose were nearly twice those following 3.5 mg i.v. (23% and 15% of the parent respectively) with a trend for higher concentrations in males.

Similarly, peak concentrations of 2161W92, the inactive indole acetic acid, following 10 mg orally, were more than twice those seen after 3.5 mg i.v. (75% and 36% of the parent respectively) but, as with 183C91, there was little evidence of any gender difference in plasma concentrations, resulting in higher metabolite ratios in men (Tables 1 and 2). AUC was approximately three-fold larger after oral dosing with a mean (range) AUC_m/AUC_p of 1.05 (0.44–2.14) compared with 0.52 (0.33–0.83) following the 3.5 mg infusion. Renal clearance was generally lower in females even after correction for body weight but this was not significant. Other parameters were similar between the sexes.

Disposition Although there was considerable intersubject variability, the shape of individual plasma profiles for radioactivity, zolmitriptan and metabolites were similar (Figure 3). At all time points, plasma concentrations of [^{14}C] were slightly higher than the summed concentration of known analytes (zolmitriptan, 183C91, 1652W92 and 2161W92), these accounting for 76–98% of the radioactivity at each time point and 86% of total plasma radioactivity.

LC-MS analysis detected no further drug-related peaks. Half-life of radioactivity was similar to that for zolmitriptan. Calculated red blood cell [^{14}C] concentrations were 27–56% of plasma [^{14}C] at all time points 1–6 h post-dose.

By the end of the 7-day collection period, a mean ± s.d. of 91.5 ± 7.0% of the administered dose was recovered with 64.4 ± 6.5% of the dose in urine and 27.1 ± 6.0% in faeces. The majority of radioactivity, zolmitriptan and known metabolites were excreted in the first 24 h (Figure 4). The relative excretion profiles were similar for all subjects. The data indicate that mean (± s.d.) urinary recovery of zolmitriptan and known analytes accounted for 50.0 ± 5.4% of the total administered dose, representing approximately 78% of the [^{14}C] in urine. A further six radiolabelled components, not corresponding to parent and its three major metabolites were detected in some urine samples by radiochromatographic h.p.l.c. analysis. The largest of these minor peaks, accounting for 3.8–5.1% of the dose, was identified by LC-MS as an intermediate metabolite, a methylamine metabolite of the indole acetic acid. A second peak was a probable oxidation product of a known primary amine derivative of zolmitriptan and accounted for 1–2.5% of the administered dose. The remaining components were

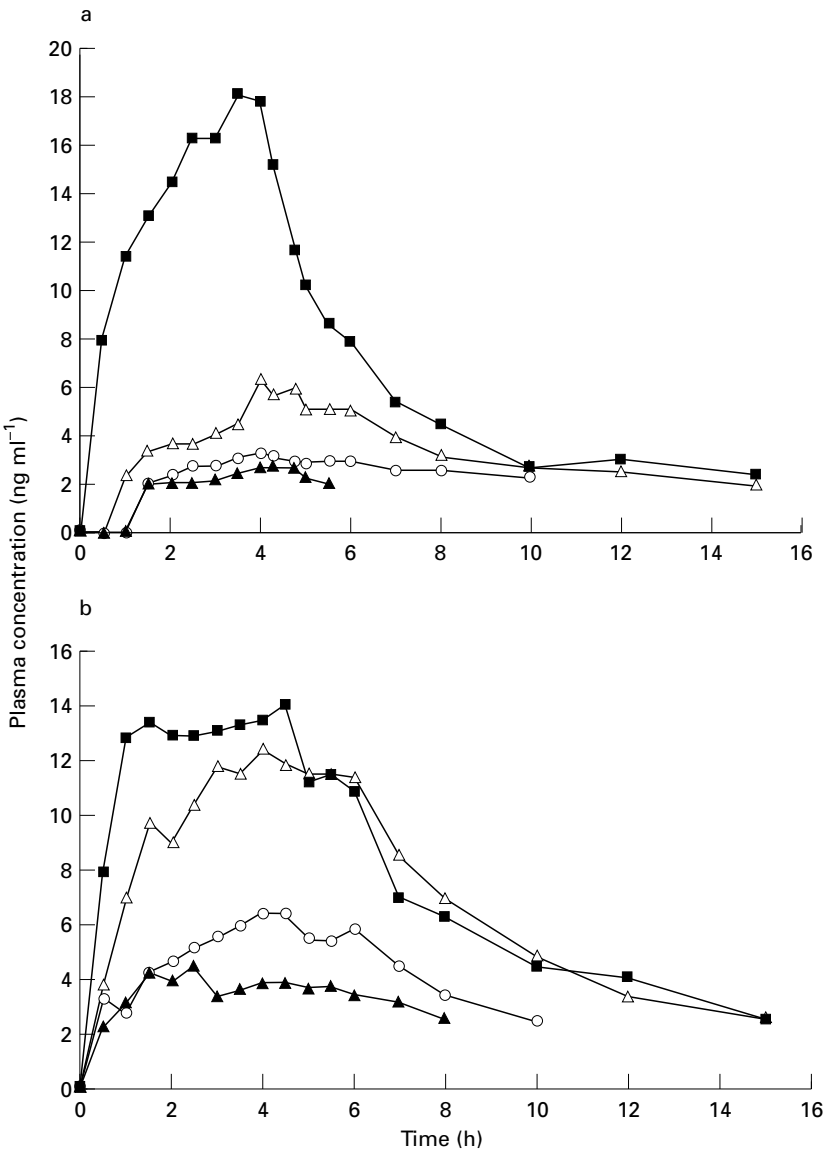

Figure 2 Median plasma concentrations of 311C90 and metabolites following: a) 3.5 mg i.v. infusion ($n=12$) and b) 10 mg tablet ($n=12$). ■ 311C90; ○ 183C91; ▲ 1652W92; △ 2161W92.

Table 2 Ratio estimates (Male : Female) and 95% confidence intervals for pharmacokinetic parameters for 311C90 and its metabolites, following an intravenous infusion of 3.5 mg and an oral dose of 10 mg 311C90.

	311C90	*183C91*	*2161W92*
	3.5 mg i/v.		
AUC (ng ml^{-1} h)	77% (56%, 105%)	160% (62%, 410%)[b]	86% (54%, 137%)
AUC_m/AUC_p ratio	—	—	116% (68%, 199%)
C_{max} (ng ml^{-1})	79% (59%, 106%)	106% (69%, 162%)	106% (68%, 164%)
$t_{\frac{1}{2},z}$ (h)	92% (65%, 129%)	—	82% (60%, 112%)
CL_R (ml min^{-1})	119% (94%, 152%)	—	148% (89%, 247%)
V_z (l)	119% (85%, 168%)	—	—
% urine recovery	−3.9[a] (−14.1, 6.3)	0.3[a] (−3.0, 3.7)	7.5[a] (2.7, 12.3)
	10mg orally		
AUC (ng ml^{-1} h)	49% (29%, 84%)	71% (38%, 129%)[b]	83% (57%, 121%)
AUC_m/AUC_p ratio	—	—	169% (102%, 281%)
C_{max} (ng ml^{-1})	61% (35%, 107%)	91% (62%, 132%)	93% (67%, 130%)
$t_{\frac{1}{2},z}$ (h)	78% (57%, 106%)	—	95% (66%, 136%)
CL_R (ml min^{-1})	116% (82%, 162%)	—	137% (90%, 210%)
% urine recovery	−8.3[a] (−15.2, −1.5)	0.2[a] (−1.0, 1.4)	4.5[a] (−2.0, 11.1)
F	64% (36%, 114%)	—	—

[a]Indicates difference between sexes (non-log transformed values)- all others are ratios of geometric means.
[b]AUC_{0-t}.

Table 3 Mean (±s.d.) pharmacokinetic parameters of 311C90, metabolites and ^{14}C following administration of 25 mg oral [^{14}C]-311C90 ($n=6$).

	311C90	*183C91*	*1652W92*	*2161W92*	^{14}C
C_{max} (ng ml^{-1})	27.5±4.1	15.2±1.6	14.3±3.4	44.7±13.0	383.8[b]±57.7
AUC (ng ml^{-1} h)	171.4±45.9	107.9±16.1	97.9±26.8	312.6±86.1	2845.66[b]±271.45
t_{max} (h)[a]	3.5 (1, 5)	3.5 (1, 5)	3.5 (1, 5)	3.5 (2, 5)	4 (1.5, 6)
$t_{\frac{1}{2}z}$ (h)	2.9±0.9	3.1±1.2	3.0±0.8	3.6±1.0	3.4±0.9
CL_R (ml min^{-1})[c]	200±23	140±17	309±51	418±139	—
% urinary recovery[c]	8.3±2.7	3.9±0.3	6.7±0.8	31.1±6.4	64.4±6.5

[a]Expressed as median and range.
[b]Expressed as nanomolar equivalents (nMeq).
[c]$n=5$, excludes one subject who admitted incomplete urine collection.

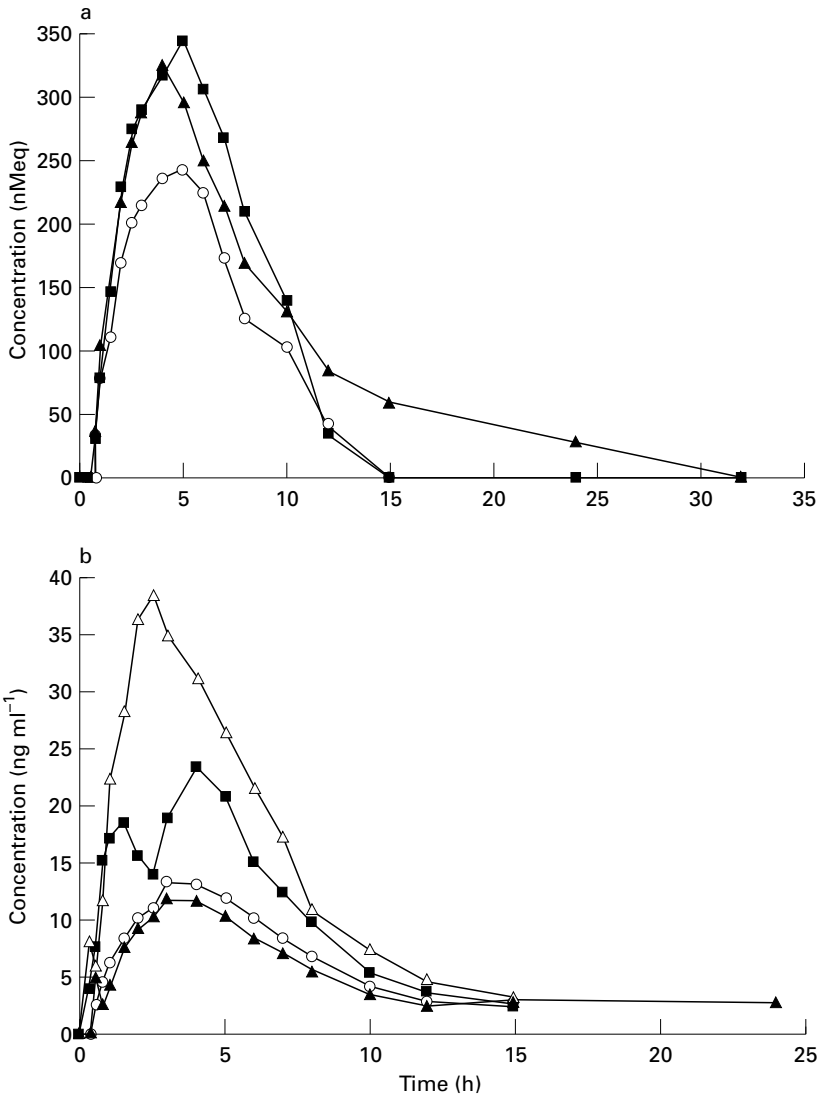

Figure 3 Median profiles, following a 25 mg oral dose of [^{14}C]-311C90, of a) ^{14}C in plasma (■), ^{14}C in whole blood (○) and summed known analytes in plasma (▲) ($n=6$) and b) 311C90 and metabolites in plasma ($n=6$). ■ 311C90; ○ 183C91; ▲ 1652W92; △ 2161W92.

not detected by LC-MS due to their low concentrations of 1.7–3.9% (one peak) and 0.3–2.2% (remaining three peaks) of the radioactive dose.

The majority of the radioactivity recovered in faeces was unchanged parent with the indole acetic acid the only other detectable metabolite.

Discussion

Results of the two studies reported here agree with previous findings in humans that zolmitriptan is well absorbed and predominantly eliminated by metabolism after oral administration [8]. Mean peak plasma concentrations are higher

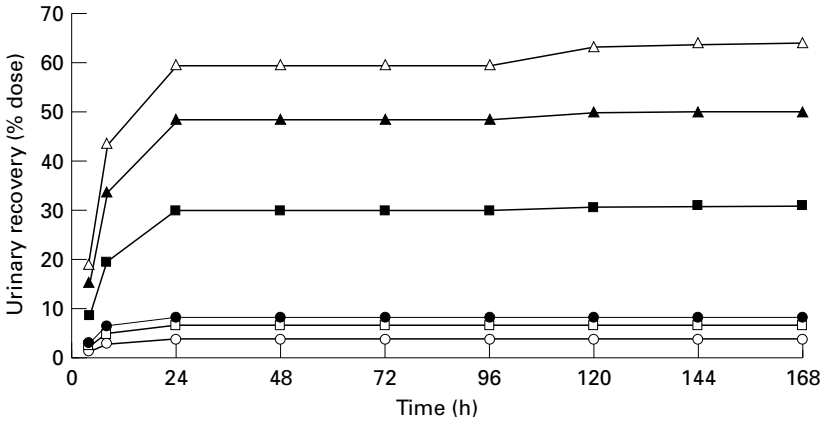

Figure 4 Cumulative mean urinary excretion of each analyte and ^{14}C following a 25 mg oral dose of ^{14}C-311C90 ($n=5$)[a]. ● 311C90; ○ 183C91; □ 1652W92; ■ 2161W92; △ ^{14}C; ▲ summed known analytes. ([a]one subject admitted to incomplete collection).

than those previously reported which can be explained by the probable higher oral bioavailabilty in women as they were excluded from this previous study [8]. In common with previous findings, several volunteers exhibited multiple-peaks in their plasma profiles for zolmitriptan and metabolites [8, 9]. This is a reported property of oral sumatriptan [10] and is almost certainly an absorption phenomenon, rather than a result of enterohepatic recycling as it was not observed following the intravenous infusions and is observed in rats following oral but not i.v. administration.

The absolute bioavailability of zolmitriptan of 49% is consistent with preclinical values and confirms that, in humans, the bioavailability is over three times greater than that of sumatriptan [11], which undergoes extensive pre-systemic metabolism in the gut wall and/or liver. The less than complete oral bioavailability of zolmitriptan is due to a combination of first-pass metabolism and incomplete absorption with over 20% of the oral dose present as unchanged drug in the faeces. The gender effect on the plasma concentrations of zolmitriptan, even allowing for differences in body weight and distribution volume, was unexpected. These differences are probably mostly due to the higher bioavailability in women. However, with the large between-subject variability and small sample size in this study, the estimate of the difference in bioavailability for men compared with women must be treated with some caution. Higher bioavailability could be due to either better absorption or reduced first-pass metabolism. The former is unlikely since gender-specific absorption differences are unusual and total urinary recovery of zolmitriptan and metabolites was similar in men and women. The evidence suggests that a difference in first-pass metabolism is likely; the higher metabolite ratios after oral as compared to iv dosing suggest that the drug does undergo first-pass metabolism and the inter-subject variability in plasma concentrations following oral dosing is greater than after intravenous dosing, typical of compounds subject to high first-pass clearance [12]. In addition, the higher metabolite ratios in men indicates that metabolism of the drug is more extensive; reduction in metabolite elimination is unlikely in men since CL_R was actually higher. Furthermore, systemic clearance, which is mostly metabolic, was also higher in men than women although the differences were small. It might be expected that systemic metabolic clearance would be higher in men if first-pass metabolism were more extensive.

Possible explanations for the gender difference include differences in hepatic enzyme activity, or in the effects of zolmitriptan on liver blood flow. Although a peripheral vasoconstrictor, zolmitriptan has been associated with increased liver blood flow in anaesthetised cats; such increases may, in turn, increase its own metabolism [Dr D. Cambridge, Glaxo Wellcome R & D, personal communication]. However, the enzymes responsible for zolmitriptan metabolism remain unidentified, hence the full explanation of the reasons for the gender difference in first-pass metabolism remain unclear. All subjects were non-smokers and none consumed excessive quantities of alcohol, therefore these inducing agents are unlikely to explain the differences. Overall, however, there was only a small difference in the range of concentrations in women compared with men after oral zolmitriptan and it is unlikely the potential gender differences in pharmacokinetics will translate to differences in treatment doses.

All three major metabolites had mean half-lives similar to that of parent. In the continued presence of zolmitriptan, these values cannot be assumed to be the true elimination half-lives; their similarity to parent drug indicating formation rate limited elimination. The significantly longer half-life after oral dosing compared with i.v. suggests that the kinetics of oral zolmitriptan may also be absorption rate limited. The indole acetic acid was found to be the most abundant metabolite, with significantly higher plasma concentrations and an AUC value twice that of the parent compound after the 25 mg dose. A previous study reported the AUC ratio of this metabolite to be 60% [8]; this earlier estimate is likely to be less accurate due to the limit of quantification being improved ten-fold before the studies described here were conducted. This analogue is also the major metabolite of sumatriptan with six-seven fold higher plasma concentrations and formation limited kinetics [13]. Renal clearance represents 20% of total clearance and is much greater than glomerular filtration rate, indicating active renal tubular secretion.

Values in excess of 60% of drug-related material in urine indicate at least this percentage is absorbed from the gastrointestinal tract. Faecal recovery of zolmitriptan was mainly as unchanged parent, suggesting that this amount of drug remains unabsorbed.

In conclusion, mean oral bioavailability of a 10 mg dose of zolmitriptan is 49% and is probably higher in women than men, with less extensive first-pass metabolism. Its major route of elimination is metabolic with over 60% of the oral

dose recovered in urine, the majority as the indole acetic acid analogue. These studies have confirmed that zolmitriptan has suitable bioavailability for an acute oral migraine treatment and there are no significant unidentified metabolites in man.

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