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Tadalafil pharmacokinetics in healthy subjects

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Aims
To characterize tadalafil plasma pharmacokinetics in healthy subjects following single and multiple doses.

Methods
Noncompartmental parameters were calculated for healthy subjects receiving a single 2.5–20-mg tadalafil dose in 13 clinical pharmacology studies. An integrated statistical analysis of results in 237 subjects provided global averages and an assessment of effects of body mass index (BMI), age, gender and smoking status. Diurnal variation, food effects and proportionality of exposure to dose were analysed in three studies. Multiple-dose pharmacokinetics were evaluated in a separate study in which parallel groups of 15 subjects received 10 or 20 mg tadalafil once daily for 10 days.

Results
Tadalafil was absorbed rapidly with mean C_{max} (378 μg l^{-1} for 20 mg) observed at 2 h; thereafter, concentrations declined nearly monoexponentially with a mean (5th, 95th percentiles) $t_{1/2}$ of 17.5 (11.5, 29.6) hours. Mean oral clearance (CL/F) was 2.48 (1.35, 4.35) l h^{-1} and apparent volume of distribution (V_z/F) was 62.6 (39.5, 92.1) l. No clinically meaningful effect of BMI, age, gender or smoking was identified. Exposure was not substantially affected by time of dosing. Food had negligible effects on bioavailability as assessed by 90% confidence intervals for C_{max} and AUC mean ratios. Parameters were proportional to dose, indicating that doubling the dose doubled exposure. Steady state was attained by day 5 following once-daily administration, and accumulation (1.6-fold) was consistent with the $t_{1/2}$.

Conclusions
Tadalafil pharmacokinetics are linear with respect to dose and time, and are not affected by food. Systemic clearance is low relative to other phosphodiesterase 5 inhibitors.

Introduction

Tadalafil (Cialis®) is used in the treatment of erectile dysfunction, and is a potent, reversible, competitive inhibitor of phosphodiesterase 5 (PDE5), an enzyme that inactivates cyclic guanosine monophosphate (cGMP) [1, 2]. Inhibition of PDE5 in the corpus cavernosum of the penis increases intracellular cGMP levels, thereby facilitating relaxation of smooth muscle leading to penile erection [3, 4]. Clinical trials with tadalafil administered orally as needed to patients with erectile dysfunction have demonstrated enhanced erectogenic response in both the clinic setting and the at-home setting [5–7]. A significant erectogenic response 16 min after a single 20-mg tadalafil dose [8] and a duration of action of up to 36 h have been demonstrated [9]. The duration of action reflects tadalafil's low systemic clear-

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ance and prolonged elimination half-life as described herein [9, 10].

Absolute bioavailability of tadalafil following oral dosing has not been determined in any clinical study. *In vitro*, within the therapeutic concentration range, 94% of tadalafil in plasma is protein bound [11, 12]. Tadalafil clearance is predominately via hepatic metabolism by CYP3A to a catechol that undergoes methylation and is extensively conjugated to form the major circulating metabolite, a methylcatechol glucuronide (a minor amount of the unconjugated metabolite is also detected in plasma) [13, 14]. Tadalafil is eliminated primarily as metabolites in faeces (61%) and urine (36%) [14]. Based on the negligible affinity for PDE5 observed *in vitro*, metabolites are not expected to be pharmacologically active at therapeutic dose levels [11, 12]. Therapeutic concentrations of tadalafil do not produce significant changes in clearance of drugs metabolized by CYP3A [15].

The purpose of the present report was to summarize basic pharmacokinetics of tadalafil, as evaluated in 13 clinical pharmacology studies in healthy male and female subjects, and one multiple-dose evaluation (14 studies total). Basic pharmacokinetics of the methylcatechol glucuronide are described also. This report does not address populations afflicted with erectile dysfunction, hypertension, diabetes, renal insufficiency, hepatic impairment and so forth, or those affected by interactions with drugs that induce or inhibit CYP3A. Some of these findings have been published in abstract form [16] or in a recent review on tadalafil [17].

Methods

Subjects

Healthy male and female subjects were eligible for enrolment based upon the following criteria: 18–65 years old (inclusive), body mass index (BMI) of 19–29 kg m^{-2}, clinical laboratory test results within normal reference range for the investigator site, and normal electrocardiogram (ECG). Pregnancy tests were performed on all women where appropriate. In general, subjects were excluded from the studies if they had a history or presence of significant cardiovascular, respiratory, hepatic, renal, gastrointestinal or haematological, neurological or endocrine disorders; history of impotence of any origin within the previous 6 months; history of testicular or prostatic surgery; a vasectomy; supine systolic blood pressure >160 mmHg at screening or supine diastolic blood pressure >90 mmHg at screening; blood donation of more than 1500 ml (subjects under 60 years) or 1000 ml (subjects over 60 years) within the previous 12 months; regular use of drugs of abuse and/or positive findings on urinary drug screening; known allergies or hypersensitivity to tadalafil or related drugs and/or history of relevant allergic reactions of any origin; presentation of positive results in HIV, hepatitis B or hepatitis C laboratory tests; or regular alcohol intake >28 units $week^{-1}$.

Subjects were restricted to two cups of xanthine-containing drinks per day. Alcohol was not permitted from 48 h prior to each dose until leaving the end of each treatment period. Prescribed and over-the-counter medication (with the exception of paracetamol) was not permitted before the first dosing occasion until after the poststudy visit. Grapefruit-containing products (known to inhibit CYP3A) were not allowed up to 48 h prior to the first dose until after the poststudy visit.

Written informed consent was obtained in conformity with the ethical principles of the Declaration of Helsinki and the applicable European laws. The ethics committees of all participating institutions approved the final protocols. All subjects signed an informed consent document.

Study design

Time of dosing (diurnal effect) was evaluated in a 3-way crossover study in 12 subjects. A 10-mg dose was administered at approximately 08.00 h or 20.00 h following a 10-h fast or at 20.00 h after a high-fat, high-calorie meal as a preliminary assessment of food effects. The definitive assessment of food effects was performed in a two-period crossover study in which 18 subjects received a single 20-mg dose in the morning following an overnight fast, and on a separate occasion within 5 min of consuming a high-fat breakfast as defined by the US Food and Drug Administration [18]. Dose proportionality was evaluated in a study with 16 subjects who received single 2.5-, 5-, 10- and 20-mg doses in the fasted state according to a four-treatment, four-period, four-sequence, crossover design.

An integrated statistical analysis of single-dose pharmacokinetics was performed for all subjects in the aforementioned three studies and 10 other clinical pharmacology studies in which healthy subjects received a 10-mg or 20-mg tadalafil dose in the morning following an overnight fast. Data were available for 237 subjects (186 males), with 45 subjects providing data from two separate dosing occasions.

The multiple-dose safety, tolerability and pharmacokinetic study was conducted with 65 healthy subjects randomized to receive either 10 mg or 20 mg tadalafil every 24 h in the morning of 10 consecutive days. Thirty of these subjects participated in the pharmacokinetic assessment.

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Safety and tolerability assessment

For the 13 single-dose studies and the multiple-dose study reported herein, physical examination, routine vital signs, 12-lead ECG, serology, urinary drug screen and routine medical laboratory safety tests were performed at screening, before dosing and at regular intervals throughout the studies. A poststudy visit was performed up to 14 days after the last dose. Treatment-emergent adverse events were defined as any adverse events that first occurred or worsened in severity after randomization, and were mapped to the database using the Medical Dictionary for Regulatory Activities (MedDRA). Prompted by nonleading questions, subjects reported adverse events on a volunteer basis throughout the study. The investigator's assessment of the relationship of each adverse event to the study treatment was recorded throughout the study.

Bioanalytical

Serial blood samples for pharmacokinetic determinations were collected from each subject following each treatment. Plasma tadalafil concentrations were measured using a validated HPLC-MS/MS assay with a lower limit of quantification of 0.5 μg l^{-1}, essentially as described [15]. The intra-assay accuracy (% relative error) and precision (% relative standard deviation) were <12.88% and <8.84%, respectively. The interassay accuracy and precision were <6.68% and <7.09%, respectively. The major metabolite, methylcatechol glucuronide, was quantified as total methylcatechol in hydrolysed (β-glucuronidase) plasma with a validated HPLC-MS/MS assay with a lower limit of quantification of 1 μg l^{-1} (data on file; Eli Lilly & Co.). The intra-assay accuracy (% relative error) and precision (% relative SD) were <18.64% and <3.51%, respectively. The interassay accuracy and precision were <16.23% and <3.19%, respectively.

Pharmacokinetic and statistical analyses

Noncompartmental analyses of plasma concentration and actual sampling time data were performed using WinNonlin Professional® (Pharsight Corporation, Mountain View, CA, USA). The following parameters were calculated for tadalafil: maximum observed plasma concentration (C_{max}); time of C_{max} (t_{max}), terminal elimination rate constant (λ_z) and half-life ($t_{1/2}$), area under the plasma concentration–time curve extrapolated to infinite time (AUC), AUC within a 24-h dosing interval (AUC_τ), oral clearance (CL/F), and apparent volume of distribution during the terminal elimination phase (V_z/F).

Food and time of dosing (diurnal) effects were evaluated with a mixed-effects linear model. Absence of an effect was declared if the 90% confidence interval (90% CI) for the ratio of least-squares (LS) means lay entirely within a range of 0.80–1.25 for AUC and 0.70–1.43 for C_{max}. These criteria were prespecified in the protocol based upon intrasubject coefficients of variation of 19.1% for AUC and 26.2% for C_{max}. Values of t_{max} were analysed with the Wilcoxon signed-rank test [19]. Median differences and 90% CIs [20] were calculated for each of the treatment comparisons.

Dose proportionality of selected parameters (PK) was assessed with a power model [21, 22] of the form:

$$\ln(PK_{ij}) = \beta \cdot \ln(dose_{ij}) + s_i + t_j + \varepsilon_{ij}$$

where $\ln(PK_{ij})$ is the log of the response, $\ln(dose_{ij})$ is the log of the dose for the *i*th subject and *j*th observation, t_i is the fixed effect of study period, and ε_{ij} is random error. Ideal dose proportionality requires that $\beta = 1$ for dose-dependent parameters (AUC and C_{max}) or that $\beta = 0$ for dose-independent parameters (CL/F and V_z/F). Dose proportionality (or dose independence) was concluded if the 95% CIs for β included 1 (or included 0) [22]. The increase for doubling of the dose ($e^{\ln(2)\times\beta}$) and the associated 95% CIs were calculated for AUC and C_{max}.

In the integrated statistical analysis (ISA), a population average and 95% CI were computed for each single-dose pharmacokinetic parameter of interest. The data were analysed using the procedure MIXED of SAS to fit linear mixed effects models [23] describing the relationship between the parameters and covariates. Each parameter was evaluated separately, and was log-transformed prior to evaluation (except for t_{max}) [24]. Categorical covariates were classed as fixed effects and the subject term was included as a random effect to allow estimation of between-subject and within-subject variability. The analysis was carried out using stepwise regression methods [25]. Type III sums of squares were used so all model terms were adjusted for all other terms. The Wilcoxon rank sum test was used to compare t_{max} values between covariate subgroups.

Results

Demographics of the 237 subjects (228 caucasians) included in the ISA of pooled single-dose parameters from 13 studies are summarized in Table 1, along with demographics specific for three of these studies and for the multiple-dose study. Except for one individual in the dose-proportionality study and four in the multiple-dose study, all subjects in the four named studies were caucasian.

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Table 1
Demographics for subjects in the integrated statistical analysis (ISA) based on 13 single-dose studies (including the three named studies) and a multiple-dose study

Parameter	Diurnal effect study	Food effect study	Dose proportionality study	ISA of 13 single dose studies	Multiple dose study[b]	
Number of Subjects (Male/Female)	12/0	4/14	8/8	186/51[a]	**32/0**	**33/0**
Mean Age (yr)	35	48	30	35	32.0	34.0
(min–max)	(20–51)	(19–62)	(20–48)	(19–64)	(19–52)	(20–62)
Mean Weight (kg)	77.4	66.4	72.1	72.2	74.1	75.9
(min–max)	(58.0–98.6)	(51.2–90.8)	(54.5–96.3)	(50.9–102.3)	(64.4–85.3)	(58.7–105.6)
Doses	10 mg	20 mg	2.5, 5, 10 or 20 mg	10 or 20 mg	**20 mg every 24 h**	**10 mg every 24 h**

[a]Includes the diurnal-effect, food-effect (fasted period only), and dose-proportionality studies; 104 of 237 subjects were smokers.
[b]Age and weight data are listed for those subjects (15/0 in each cohort) represented in the pharmacokinetic evaluation.

Safety and tolerability

Tadalafil administered as single and once-daily doses of 10 and 20 mg was safe and well tolerated. No serious adverse events occurred during any of the studies, including no clinically significant changes in the 12-lead ECG or clinical laboratory data. The most frequently reported treatment-emergent adverse events in the study of once-daily dosing for 10 days were: headache (66% of 65 subjects), back pain (60%), myalgia (43%) dyspepsia (19%), nausea (19%), dizziness (17%), fatigue (12%), eye pain (11%) and spontaneous penile erection (11%). Frequencies averaged across all subjects in the single-dose diurnal-effect, food and dose-proportionality studies were: back pain (67%), headache (61%), myalgia (20%), dizziness (17%), arthralgia (15%), nausea (15%) and pharyngolaryngeal pain (15%). Frequencies for single-dose administration averaged across all subjects in the 13 ISA studies were: headache (11%), back pain (8%), myalgia (5.5%), flushing (3%), arthralgia (2.5%), eye pain (2.5%), nausea (2.5%) and pharyngolaryngeal pain (2.5%).

Morning vs. evening administration

A three-way crossover study was conducted in 12 male subjects early in clinical development to evaluate possible diurnal effects on tadalafil bioavailability. The evening/morning ratio (90% CI) of LS means was 0.81 (0.73, 0.90) for AUC and 0.79 (0.71, 0.89) for C_{max}, suggesting slightly lower bioavailability in the evening. The $t_{1/2}$ was approximately 17 h following either treatment. The third treatment was an early assessment of the effect of a high-fat, high-calorie meal on bioavailability; results were essentially similar to those for the definitive food-effect study.

Food effect

In the definitive food-effect study, 18 subjects received a single 20-mg tadalafil dose in the fasted state, and on a separate occasion following a high-fat, high-calorie breakfast. Absence of a food effect was declared since the 90% CI for the ratio (fed/fasted) of LS means lay within the prespecified equivalence limits for AUC and C_{max} (Table 2). There was no significant difference between t_{max} values.

Dose proportionality

The relationship between systemic exposure to tadalafil and dose was evaluated in 16 subjects. Inspection of the distribution of individual dose-normalized AUC values suggested that exposure was proportional to dose across the eightfold range (2.5–20 mg) (Figure 1). Proportionality was concluded since the 95% CI for the slope [$\beta = 0.97$ (0.93, 1.01)] for regression of ln(AUC) on ln(dose) was completely contained within the prespecified equivalence interval. Doubling the dose increased AUC by a factor of 1.96 (1.90, 2.02). Estimates of β for CL/F, V_z/F, and $t_{1/2}$ approximated 0, consistent with these parameters being independent of dose. Increases in C_{max} were slightly less than proportional [$\beta = 0.88$ (0.83, 0.93)], such that doubling the dose increased C_{max} by a factor of 1.84 (1.77, 1.91). Also, median t_{max} values were 1, 2, 2 and 3 h for the 2.5-, 5-, 10- and 20-mg doses, respectively, suggesting marginally faster absorption at the lowest dose.

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Table 2
Tadalafil pharmacokinetics following administration of a single 20-mg dose in the fed and fasted states to 18 healthy subjects

Parameter	Geometric mean (CV%) Fed	Fasted (fed/fasted)	Ratio of LS means (90% CI)
AUC (μg h l^{-1})	6943 (28)	6419 (32)	1.08 (1.02, 1.15)
C_{max} (μg l^{-1})	345 (27)	297 (30)	1.16 (1.07, 1.26)
t_{max} (h)[a]	2.5 (1.0–4.0)	2.0 (0.5–4.0)	0.50 (0, 1.00)
$t_{1/2}$ (h)	17.0 (26)	17.3 (24)	– –

[a]*Median (min – max) and median difference (fed – fasted).*

Table 3
Overall mean tadalafil pharmacokinetic parameter values (and individual subject percentiles) for a single 20-mg tadalafil dose based on the integrated statistical analysis

Parameter	5th percentile	Geometric mean (CV%)	95th percentile
AUC (μg h l^{-1})[a]	4597	8066 (39.3)	14844
C_{max} (μg l^{-1})[a]	239	378 (27.6)	576
t_{max} (h)[b]	–	2.0 (0.5–12.0)	–
$t_{1/2}$ (h)	11.5	17.5 (32.3)	29.6
CL/F (l h^{-1})	1.35	2.48 (39.3)	4.35
V_z/F (l)	39.5	62.6 (25.4)	92.1

[a]*Individual AUC and C_{max} values for 10-mg data were normalized to 20 mg (multiplied by 2) prior to analysis.* [b]*Median (min – max).*

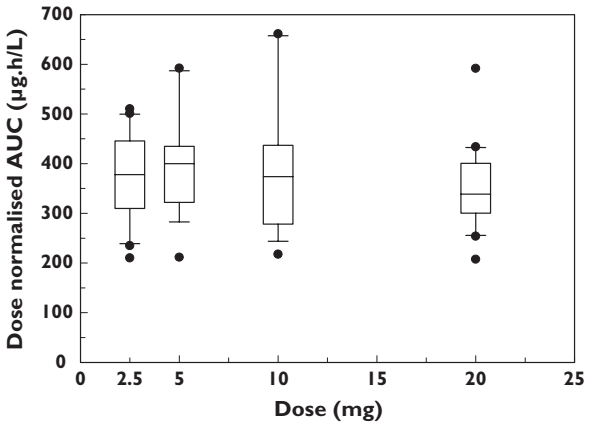


Figure 1
Distribution of dose-normalized AUC values following single-dose administration to 16 healthy subjects

Integrated statistical analysis

Tadalafil pharmacokinetics were best summarized by means of the ISA, based on parameter values for all individuals in 13 clinical pharmacology studies that had received single 10-mg or 20-mg doses in the fasted state (Table 3). Tadalafil was absorbed rapidly with a mean C_{max} (378 μg l^{-1} at 20 mg) occurring at a median t_{max} of 2 h; thereafter, mean concentrations were representative of individual profiles by declining in a nearly mono-exponential manner with an average $t_{1/2}$ of 17.5 h (Figure 2).

The integrated dataset also was evaluated to identify any overt effects of covariates. A statistically significant decrease in CL/F and an increase in V_z/F with increasing BMI were detected. The median (upper and lower quartiles) BMI was 23.8 kg m^{-2} (22.2 and 25.9 kg m^{-2}). Age, gender and smoking status were also significant covariates; the magnitude of effects is summarized in Table 4. For example, an exposure (AUC) of 7277 μg h l^{-1} was predicted for a 20-mg dose in a male subject at the median BMI value. The corresponding AUC calculated at the minimal observed BMI value was 5606 μg h^{-1} l^{-1}. The effect of age was small (8% increase in $t_{1/2}$ from 35 to 65 years) and confounded with the effect of BMI. These factors together accounted for <12% of intersubject variability, suggesting that no single factor was

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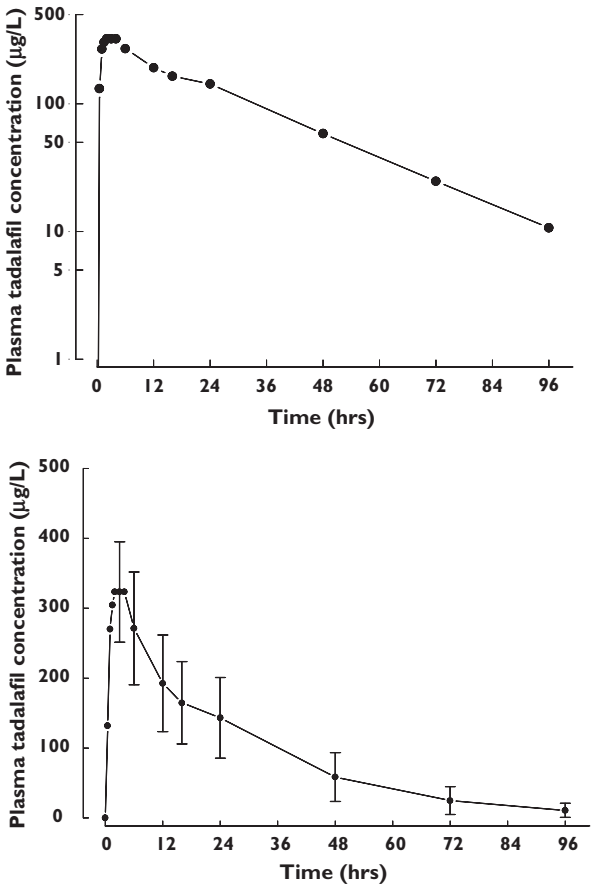


Figure 2
Arithmetic mean (± SEM) tadalafil concentrations following a single 20-mg dose to 20 healthy male subjects

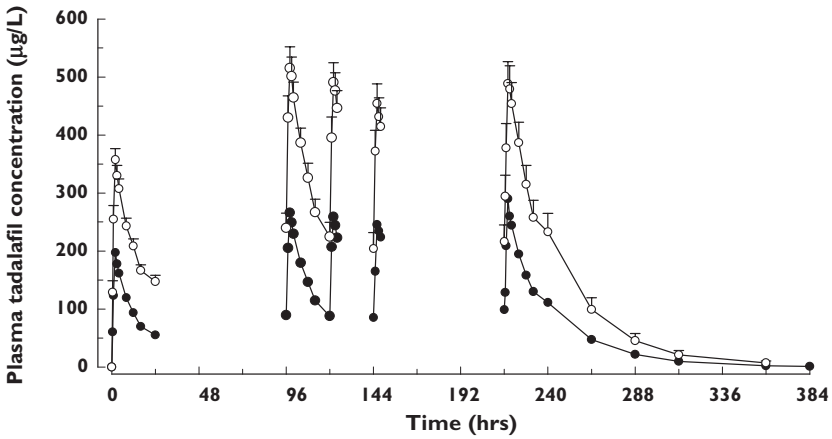
cause for dose adjustment. Within-subject variability was 13% for AUC and 16% for C_{max}, indicating that exposure within an individual was reproducible from dose to dose.

Multiple dose pharmacokinetics

Subjects were randomized to receive either 10 or 20 mg tadalafil every 24 h for 10 days. Inspection of individual and mean concentration–time profiles suggested that steady state was essentially attained by day 5 (Figure 3). This was confirmed by showing that AUC_τ and C_{max} values for day 5 were equivalent to corresponding values for day 10 as evidenced by the 90% CI for the ratio of LS geometric means lying entirely within the equivalence range of 80% to 125%. Accumulation from day 1 to day 10, calculated with either AUC or C_{max} data, was approximately 1.6-fold (Table 5). Mean CL/F, V_z/F, and $t_{1/2}$ values following the last dose were essentially similar to single-dose values, indicating linear pharmacokinetics across time. Exposure was proportional to dose; the 90% CI for the ratio (20 mg/10 mg) of dose-normalized mean AUC_τ values was (0.91, 1.30) on day 5 and (0.77, 1.17) on day 10. Corresponding 90% CI intervals for C_{max} were (0.82, 1.12) on day 5 and (0.70, 1.01) on day 10.

Exposure to the major metabolite (predominately the methylcatechol glucuronide) was also dose-proportional based on 90% CI for dose-normalized mean AUC_τ and C_{max} values. Comparison of AUC and C_{max} data across days 5 and 10 indicated that steady state was achieved by day 5 (Table 5). Following cessation of dosing, plasma metabolite concentrations declined nearly in parallel with the parent-drug concentrations, suggesting formation rate-limited kinetics of the metabolite.

Figure 3
Arithmetic mean (± SEM) tadalafil concentrations following once-daily dosing of 10 or 20 mg to two cohorts of 15 subjects



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Table 4
Effects of covariates on tadalafil pharmacokinetics

Parameter	Subgroup	Estimates calculated at median BMI value (min, max)
AUC (μg h l^{-1})[a]	Male 20 mg	7277 (5606, 11401)
	Female 20 mg	8265 (6366, 12948)
	Male 10 mg	8044 (6197, 12603)
	Female 10 mg	9136 (7037, 14313)
C_{max} (μg l^{-1})[a]	Nonsmokers 20 mg	319 (319, 319)
	Smokers 20 mg	341 (341, 341)
	Nonsmokers 10 mg	387 (387, 387)
	Smokers 10 mg	414 (414, 414)
$t_{1/2}$ (h)	Male nonsmokers 20 mg	17.2 (12.0, 31.9)
	Female nonsmokers 20 mg	21.6 (15.1, 40.1)
	Male smokers 20 mg	15.6 (10.9, 28.9)
	Female smokers 20 mg	19.6 (13.7, 36.4)
	Male nonsmokers 10 mg	17.2 (12.0, 31.9)
	Female nonsmokers 10 mg	21.6 (15.1, 40.1)
	Male smokers 10 mg	15.6 (10.9, 29.0)
	Female smokers 10 mg	19.6 (13.7, 36.4)
CL/F (l h^{-1})	Male 20 mg	2.75 (3.57, 1.75)
	Female 20 mg	2.42 (3.14, 1.54)
	Male 10 mg	2.49 (3.23, 1.59)
	Female 10 mg	2.19 (2.84, 1.40)
V_z/F (l)	Male 20 mg	66.6 (58.4, 83.7)
	Female 20 mg	72.0 (63.0, 90.4)
	Male 10 mg	59.6 (52.2, 74.8)
	Female 10 mg	64.3 (56.3, 80.8)

[a]*Normalized to a 20-mg dose.*

Table 5
Pharmacokinetics of tadalafil and its major metabolite following administration of 20 mg every 24 h for 10 days to 15 healthy subjects

Parameter	Geometric mean (CV%) Day 1 $N = 15$	Day 5 $N = 15$	Day 10 $N = 13$
Tadalafil			
AUCτ (μg h l^{-1})	4950 (23)	7692 (31)	7389 (38)
C_{max} (μg l^{-1})	352 (25)	514 (27)	481 (31)
t_{max} (h)[a]	2.0 (1.0–3.0)	2.0 (1.0–4.0)	2.0 (1.0–3.0)
$t_{1/2}$ (h)	–	–	18.7 (40)
CL/F (l h^{-1})	–	2.6 (31)	2.71 (38)
V_z/F (l)	–	–	73.1 (23)
Accumulation ratio for AUCτ	–	1.58	1.55
Methylcatechol glucuronide[b]			
AUCτ (μg h l^{-1})	2863 (34)	9967 (35)	9397 (46)
C_{max} (μg l^{-1})	179 (35)	469 (34)	442 (44)
t_{max} (h)[a]	24 (24–24)	3.0 (0–8.0)	3.0 (0–24)
$t_{1/2}$ (h)	–	–	23.2 (25)
Accumulation ratio for AUCτ	–	3.58	3.38

[a]*Median (min – max).* [b]*Quantified as total methylcatechol in hydrolysed plasma.*

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Discussion

The noncompartmental pharmacokinetic analysis in healthy subjects, taken as a whole, indicates linear pharmacokinetic behaviour with respect to dose and time under the conditions of study. Food had negligible effects on rate and extent of absorption. Hence, tadalafil may be taken without regard to meal timing or fat content of the meal. Rate and extent of tadalafil absorption in the morning were slightly lower than in the evening. A similar trend has been seen for other lipophilic drugs [26, 27]. Plasma concentrations are essentially proportional to dose, i.e. doubling the dose doubles exposure to tadalafil. Plasma protein binding is constant (94%) across the therapeutic concentration range. Thus, unbound tadalafil concentrations are also dose-proportional. A total plasma concentration of 6 $\mu g\ l^{-1}$ corresponds to the unbound concentration (0.9 nM) needed for half-maximal inhibition of PDE5 *in vitro* [28]. No serious adverse events occurred in any of the 14 clinical pharmacology studies and the frequency of treatment-emergent adverse events was consistent with findings for efficacy trials with tadalafil [5, 6].

The ISA provided robust summary statistics on basic pharmacokinetics across numerous studies that proved useful for product labelling. The ISA database was large (282 concentration–time profiles for 237 subjects), facilitating detection of small effects of age, gender, BMI and smoking status on some pharmacokinetic parameters (Table 4). These covariates together explained at most 12% of subject-to-subject variability. Neither age nor smoking status was statistically significant in the population pharmacokinetic analysis of two phase 2 trials in patients with erectile dysfunction (weight was a covariate of distribution volume) [29]. No dose adjustment is warranted based on age or smoking status [11, 12]. An early study (in the ISA database) had indicated that gender had negligible influence on C_{max} and only a small effect on AUC; this was confirmed by the ISA. Women were enrolled in some subsequent studies to facilitate recruitment and pharmacokinetic results were summarized across gender.

A first-order absorption rate constant (k_a) can be approximated from the median t_{max} (2 h) and $t_{1/2}$ (17.5 h) values, recognizing that t_{max} is merely the time after dosing at which absorption and elimination rates are equal. For a given absorption rate, t_{max} is prolonged as $t_{1/2}$ is prolonged, because it takes longer for absorption to decline far enough to match a slower elimination rate. Since tadalafil concentration–time data conform to a one-compartment model with first-order absorption, k_a can be numerically approximated with $t_{max} = \ln(k_a/\lambda_z)/(k_a - \lambda_z)$, where $\lambda_z = \ln(2)/(t_{1/2})$. This yields a k_a of 2.0 h^{-1} or a half-time for absorption of 20 min. Detectable tadalafil concentrations are obtained by this time and, in contrast to other PDE5 inhibitors, near maximal (≥80% of C_{max}) tadalafil concentrations are present from 1.5 through 6.5 h postdose.

Based on the mean oral clearance (CL/F) and a plasma/blood concentration ratio of 1.39 (data on file; Eli Lilly & Co.), CL/F indexed to blood is 3.45 $l\ h^{-1}$ (<5% of hepatic blood flow). This value represents an upper limit for blood systemic clearance (CL) since absolute bioavailability (F) cannot exceed 1.0. This calculation, coupled with extensive evidence that the liver is the predominant eliminating organ, characterizes tadalafil as a 'low hepatic clearance' drug. Low clearance, coupled with a modest volume of distribution ($V_z/F = 63$ l) results in a $t_{1/2}$ that is prolonged relative to other PDE5 inhibitors approved for treatment of erectile dysfunction. For example, sildenafil citrate (Viagra®; Pfizer) has a systemic plasma CL (40.8 $l\ h^{-1}$; blood CL not reported) that approaches hepatic plasma flow and a corresponding $t_{1/2}$ of 4 h [30].

Observed accumulation of tadalafil during once-daily dosing (1.6-fold) is essentially similar to the expected value given by $1/(1 - e^{-\lambda z \times \tau})$ for monoexponential elimination and a 24-h dosing interval. Estimates of $t_{1/2}$, CL/F, and V_z/F following multiple dose administration were essentially similar to single-dose values, providing strong evidence that tadalafil neither induces nor inhibits its own metabolism.

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References

1 Francis SH, Corbin JD. Cyclic GMP: synthesis, metabolism, and function. In Advances in Pharmacology, ed. Murad P. New York: Academic Press 1994; 115–70.

2 Padma-Nathan H, McMurray JG, Pullman WE, Whitaker JS, Saoud JB, Ferguson KM, Rosen RC. On-demand IC351 (Cialis®) enhances erectile function in patients with erectile dysfunction. Int J Impot Res 2001; 13: 2–9.

3 Thompson WJ. Cyclic nucleotide phosphodiesterases: pharmacology, biochemistry and function. Pharmacol Ther 1991; 51: 13–3.

4 Lue TF. Erectile dysfunction. N Engl J Med 2000; 342: 1802–13.

5 Brock GB, McMahon CG, Chen KK, Costigan T, Shen W, Watkins

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V, Anglin G, Whitaker S. Efficacy and safety of tadalafil for the treatment of erectile dysfunction: results of integrated analyses. J Urol 2002; 168: 1332–6.
6 Carson CC, Rajfer J, Eardley I, Carrier S, Dennhe JS, Walker DJ, Shen W, Cordell WH. The efficacy and safety of tadalafil: an update. Br J Urol Int 2004; 93: 1276–81.
7 Saenz de Tejada I, Anglin G, Knight JR, Emmick JT. Effects of tadalafil on erectile dysfunction in men with diabetes. Diabetes Care 2002; 25: 2159–64.
8 Rosen RC, Padma-Nathan H, Shabsigh R, Saikali K, Watkins V, Pullman W. Determining the earliest time within 30 minutes to erectogenic effect after tadalafil 10 and 20 mg: a multicenter, randomized, double-blind, placebo-controlled, at-home study. J Sexual Med 2004; 1: 193–200.
9 Porst H, Padma-Nathan H, Giuliano F, Anglin G, Varanese L, Rosen R. Efficacy of tadalafil for the treatment of erectile dysfunction at 24 and 36 hours after dosing: a randomized controlled trial. Urology 2003; 62: 121–6.
10 Francis SH, Corbin JD. Molecular mechanisms and pharmacokinetics of phosphodiesterase-5 antagonists. Cur Urol Reports 2003; 4: 457–65.
11 Tadalafil (Cialis®) Summary of Product Characteristics. http://www.emea.eu.int/humandocs/Humans/EPAR/cialis/cialis.htm (Last accessed: 5 December 2005).
12 Tadalafil (Cialis®) US. prescribing information. http://pi.lilly.com/us/cialis-pi.pdf (Last accessed: 5 December 2005).
13 Ring BJ, Gillespie JS, Mullen JH, Wrighton SA. Identification of the human cytochrome P450 (CYP) responsible for the formation of the major oxidative metabolite of tadalafil (Cialis). Drug Metab Rev 2004; 36 (S1): 70.
14 Phillips DL, Smith RL, Patterson BE, Parker N, Mitchell M, Wheeler WJ, Watkins VS, Barbuch RJ. Metabolism and excretion of tadalafil in healthy men after oral administration of 100 mg [^{14}C]-tadalafil. The AAPS J 2004; 6: Abstract W5308.
15 Ring BJ, Patterson BE, Mitchell MI, Vandenbranden M, Gillespie J, Bedding AW, Jewell H, Payne CD, Forgue ST, Eckstein J, Wrighton SA, Phillips DL. Effect of tadalafil on cytochrome P450 3A4-mediated clearance: Studies in vitro and in vivo. Clin Pharm Ther 2005; 77: 63–75.
16 Patterson B, Bedding AW, Jewel H, Payne CD, Mitchell MI. Dose-normalized pharmacokinetics of tadalafil (IC351) administered as a single dose to healthy volunteers. Int J Impotence Res 2001; 13 (Suppl. 5): S32.
17 Curran MP, Keating GM. Tadalafil. Drugs 2003; 63: 2203–12.
18 Guidance for Industry, Food-Effect Bioavailability and Fed Bioequivalence Studies. Washington DC: US Food and Drug Administration, Center for Drug Evaluation and Research 2002.
19 Lehmann EL. Nonparametrics. Statistical Methods Based on Ranks, Chapters 3 and 4. New York: McGraw-Hill 1975.
20 Gardner MJ, Altman DG. Statistics with confidence: confidence intervals and statistical guidelines. British Medical Journal Books 1989; 71–8.
21 Gough K, Hutchison M, Keene O. Assessment of dose proportionality: report from the statisticians in the pharmaceutical industry/pharmacokinetics UK joint Working Party. Drug Info J 1995; 29: 1039–48.
22 Smith BP, Vandenhende FR, DeSante KA, Farid NA, Welch PA, Callaghan JT, Forgue ST. Confidence interval criteria for assessment of dose proportionality. Pharm Res 2000; 17: 1278–83.
23 Little RC, Milliken GA, Stroup WW, Wolfinger RD. The SAS System for Mixed Models. Cary NC: SAS Institute Inc.1996.
24 Keene ON. The log transformation is special. Stats Med 1995; 14: 811–9.
25 Montgomery DC, Peck EA. Introduction to Linear Regression Analysis, 2nd edn. New York: John Wiley and Sons 1992; 265–304.
26 Lemmer B. Chronopharmacokinetics: implications for drug treatment. J Pharm Pharmacol 1999; 51: 887–90.
27 Bruguerolle B. Chronopharmacokinetics: current status. Clin Pharmacokin 1998; 35: 83–94.
28 Angulo J, Gadau M, Fernandez A, Gabancho S, Cuevas P, Martins T, Florio V, Ferguson K, Saenz de Tejada I. IC351 enhances nitric oxide-mediated relaxation of human arterial and trabecular penile smooth muscle. Diabetologia 2001; 44: A295.
29 Troconiz IF, Tillmann C, Staab A, Rapado J, Mackie A, Mitchell M, Patterson B, Forgue ST. Tadalafil population pharmacokinetics in patients with erectile dysfunction. AAPS Pharm Sci 2003; 5: T2341.
30 Nichols DJ, Muirhead GJ, Harness JA. Pharmacokinetics of sildenafil citrate after single oral doses in healthy male subjects: absolute bioavailability, food effects and dose proportionality. Br J Clin Pharmacol 2002; 53: 5S–12S.

13652125, 2006, 3, Downloaded from https://bpspubs.onlinelibrary.wiley.com/doi/10.1111/j.1365-2125.2005.02553.x, Wiley Online Library on [08/04/2025]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

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