Evaluation of Different Metrics as Indirect Measures of Rate of Drug Absorption from Extended Release Dosage Forms at Steady-State

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Bioequivalence assessment of extended release (ER) dosage forms is usually carried out at steady-state, using area under the curve (AUC) to evaluate extent of absorption and maximum concentration (C_{max}) and % peak trough fluctuation ratio (%PTF) to evaluate rate of absorption. Other metrics such as C_{max}/AUC and partial AUCs have recently been proposed as alternatives for assessing the absorption rate of drugs from immediate release (IR) dosage forms under single dose conditions. The performances of these metrics were assessed using the results of two sets of simulated experiments of ER dosage forms at steady-state and 2 actual pharmacokinetic studies involving ER dosage forms of a Glaxo drug. In the first set of simulations there was no difference in bioavailability between the two formulations; in the second set of simulations the test formulation had a 50% greater absorption rate-constant (ka) than the reference formulation. The following conclusions were reached: 1. For ER dosage forms at steady-state, all the metrics, with the exception of %PTF, resulted in much smaller increases than the underlying 50% increase in ka. Although, %PTF gave the largest effect it was also the most imprecisely estimated. 2. In our studies, none of the metrics tested provided reliable information about changes in the underlying rate of absorption from ER dosage forms under steady-state conditions. 3. The current practice of comparing rate of absorption from ER dosage forms using steady-state C_{max} is inappropriate due to lack of sensitivity. The use of %PTF may require a widening in the currently accepted 80-125% permissible range set for C_{max} and AUC.

KEY WORDS: bioequivalence; extended release; steady-state; absorption rate; metrics.

INTRODUCTION

Currently, bioequivalence assessment of ER dosage forms is usually carried out at steady-state[1]. The reasons put forward for this include[2]: 1. ER dosage forms are usually intended for long-term use and steady-state studies more accurately reflect the recommended use of the drug than single dose studies. 2. Steady-state studies result in higher drug concentrations, which need only be measured over a dosing interval. 3. Smaller inter-subject variability has been found in steady-state studies, which may permit the use of fewer subjects.

In steady-state bioequivalence studies, AUC is used to evaluate extent of absorption and C_{max} and %PTF are used as indirect measures to evaluate rate of absorption[3]. C_{max} is confounded by extent of drug absorption[4] and is known to be an insensitive indicator of absorption rate[5]. Other indirect measures of rate of drug absorption (metrics) such as C_{max}/AUC and partial AUCs have recently been proposed as alternatives for assessing rate of IR dosage forms under single dose conditions[4,6,7]. Our objective was to investigate the relative performance of these metrics under steady-state conditions. We compared the different metrics at steady-state using simulated experiments of ER dosage forms and, importantly, we also assessed their performances using the results of two actual pharmacokinetic studies involving a Glaxo drug.

METHODS

Symbols. All symbols refer to steady-state conditions

- AUC_τ AUC over the dosing interval, τ.
- C_{max} Maximum concentration.
- C_τ Concentration at end of the dosing interval.
- t_{max} Time to C_{max}.
- AUC_e AUC from zero to t_{max} of reference or test formulation, whichever occurs earliest.
- AUC_r AUC from zero to t_{max} of reference formulation.
- %PTF $(\tau \times (C_{max} - C_\tau)/AUC_\tau) \times 100$.

Metrics. The following were investigated: C_{max}, C_{max}/AUC_τ, AUC_e, AUC_r, AUC_e/AUC_τ, AUC_r/AUC_τ, and %PTF. (AUC_τ was used as the measure of extent of absorption). C_{max}, and the partial area metrics are confounded by extent of absorption, whereas C_{max}/AUC_τ, and the normalised partial area metrics (AUC_e/AUC_τ, AUC_r/AUC_τ) are functions of rate-constants of absorption and elimination[4,8].

Pharmacostatistical Model. The one-compartment model with first-order rate of absorption was used to generate data for cross-over studies. The log-normal distribution was used for the within-subject variability in the pharmacokinetic model parameters and residual error was introduced into the model by a multiplicative error term. A detailed description of the pharmacostatistical model employed has been reported previously[4]. The single dose pharmacokinetic model was modified as follows to provide a steady-state[9] concentration-time profile (Css(t)):

$$C_{ss}(t) = B \times \left\{ \frac{\exp(-k \times t)}{1 - \exp(-k \times \tau)} - \frac{\exp(-k_a \times t)}{1 - \exp(-k_a \times \tau)} \right\}$$

where

$$B = \frac{F \times ka \times dose}{V(ka - k)}$$

F is the absolute bioavailability, ka is the absorption rate-constant, k is the elimination-rate constant, V is the apparent volume of distribution. The expected geometric mean (GM)

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values of the model parameters, with their associated within-subject coefficients of variation (CV) were as follows:

GM of A = 80 mg/L	CV of A = 20%
GM of k = 0.250h^{-1}	CV of k = 10%
GM of ka = 0.150h^{-1}	CV of ka = 30%

where A = F × Dose/V.

The CV for the residual error was set at 20%.

Simulations. Two sets of simulated experiments were carried out. Each involved generating data from 100 separate two-period, cross-over studies, with 24 subjects per study, using a one compartment model with first-order rate of absorption. In the first set of simulations, the model parameter values were the same for both dosage forms, while in the second there was a 50% increase in the expected geometric mean ka of the test formulation (i.e., ka increasing from 0.150 to 0.225h^{-1}). The ka values were chosen to be typical of those reported in the literature for ER dosage forms[10,11]. The ka/k ratios are 0.6 and 0.9, respectively and thus flip-flop conditions apply. The simulations were designed to give each study a power of 90% to declare bioequivalence for AUC_τ. Sampling times were set as 0, 1, 2, 3, 4, 5, 6, 8, 10, 12 hours post-dose, where τ = 12 hours.

Glaxo Studies. In addition, a suitable Glaxo drug was identified (with a mean k of about 0.17h^{-1}) and two of the most recently performed two-period cross-over studies (n = 19, and n = 22, respectively) comparing two ER dosage forms at steady-state (where τ = 12 hours, and the number of doses administered were 11 and 7, respectively) were used to assess the different metrics. The mean ka of the reference ER formulation was about 0.08h^{-1}. The ka/ka ratio is approximately 0.5 and thus flip-flop conditions apply. Sampling times were as follows:

Study 1. predose, 1, 2, 3, 4, 5, 6, 8, 10, 12 hours after the last dose.

Study 2. predose, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12 hours after the last dose.

Statistical Analysis and Bioequivalence Assessment. Standard analysis of variance methods were used to obtain the treatment mean ratios of the log transformed metrics, with associated 90% confidence intervals[12]. Bioequivalence was deemed to have occurred if the 90% confidence interval for the treatment mean ratio was within the range 80 to 125%[13].

RESULTS

Results from the Two Sets of Simulated Experiments

The results obtained from the first set of simulated experiments (no difference between the formulations in terms of rate or extent of absorption) are given in Table I. Bioequivalence in terms of extent of absorption, as measured by AUC_τ, was demonstrated in 86% of cases (Table I), indicating adequate statistical power of the simulated experiments. However, it is apparent that when the two formulations are truly bioequivalent in terms of rate, the 90% confidence intervals for the %PTF mean ratio were outside the 80-125% range in the majority (62%) of studies.

Box-Whisker plots of the CVs for each of the metrics are given in Figure 1. It is apparent that %PTF was the most imprecisely estimated metric and that C_{max}/AUC_τ and the normalised partial areas were the most precisely estimated.

Table I. % of p-Values ≥0.05 and <0.05 for the Different Metrics, Associated Mean Ratios and % of Studies Demonstrating/Not Demonstrating Bioequivalence (BE) for the First Set of Simulations (No Difference Between the Formulations in Terms of ka)

Metric	p ≥ 0.05	p < 0.05	Mean ratio	*BE (yes)	*BE (no)
AUC_τ	95	5	1.00	86	14
C_{max}	95	5	1.00	74	26
C_{max}/AUC_τ	96	4	1.00	100	0
AUC_e	97	3	1.00	80	20
AUC_r	93	7	0.98	81	19
AUC_e/AUC_τ	92	8	1.01	100	0
AUC_r/AUC_τ	93	7	0.99	100	0
%PTF	97	3	0.99	38	62

* Using the criterion 90% confidence interval for the mean ratio to be within the range 80–125%.

The results obtained from the second set of simulated experiments (50% increase in ka from 0.150 to 0.225 h^{-1}) are given in Table II. It is apparent that %PTF was the most sensitive at detecting a true difference in the underlying rate of absorption. In 76% of studies %PTF resulted in a statistically significant difference between formulations; this compares with only 15% for C_{max} and 9% and 11% for the two partial area metrics. C_{max}/AUC_τ and the normalised partial areas had greater sensitivity than the non-normalised variables but were nonetheless considerably less sensitive than %PTF.

With the exception of %PTF, a 50% increase in ka was reflected as a much smaller increase in the other metrics (Table II). In addition, these metrics resulted in the 90% confidence intervals for the mean ratios being within the range 80-125% in the majority of cases; for C_{max}/AUC_τ and the normalised partial areas it was 100% of cases.

The CVs for each of the metrics were very similar to those obtained in the first set of simulated experiments. The median CV was 35% for %PTF compared to 26% for C_{max} (Figure 1). C_{max}/AUC_τ and the normalised partial areas were

Table II. % of p-Values ≥0.05 and <0.05 for the Different Metrics, Associated Mean Ratios and % of Studies Demonstrating/Not Demonstrating Bioequivalence (BE) for the Second Set of Simulations (50% Increase in ka from 0.150 to 0.225 h^{-1})

Metric	p ≥ 0.05	p < 0.05	Mean ratio	*BE (yes)	*BE (no)
AUC_τ	95	5	0.99	91	9
C_{max}	85	15	1.06	62	38
C_{max}/AUC_τ	55	45	1.07	100	0
AUC_e	89	11	1.04	71	29
AUC_r	91	9	1.02	80	20
AUC_e/AUC_τ	73	27	1.05	100	0
AUC_r/AUC_τ	78	22	1.03	100	0
%PTF	24	76	1.30	0	100

* Using the criterion 90% confidence interval for the mean ratio to be within the range 80–125%.

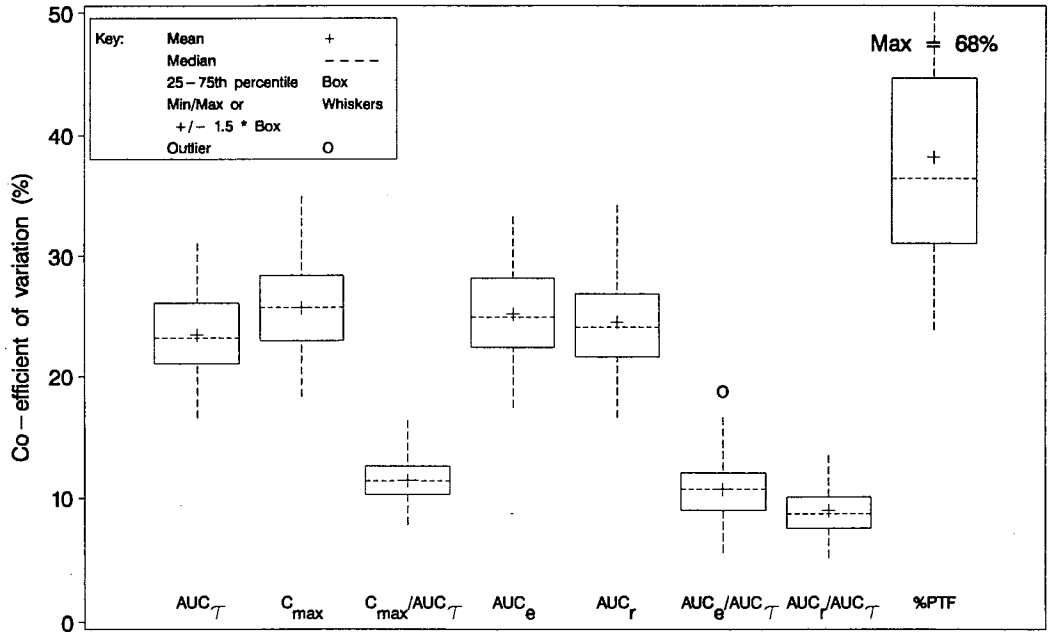


Figure 1: Box-Whisker plots of the co-efficients of variation for each of the absorption rate metrics obtained from the first set of simulated experiments.

more precisely estimated than the non-normalised variables (e.g., CV of C_{max}/AUC_τ was 12% compared to 26%).

Results from the Two Actual Studies with a Glaxo Drug

The results obtained from the two actual studies are given in Table III. Plots of the median concentration-time profiles for the two studies are given in Figures 2a and 2b, respectively. In study 1, there was a 20% difference between the formulations in extent of absorption as measured by AUC_τ. This affects interpretation of the results obtained with C_{max}, as this variable is confounded by extent of absorption[4]; such considerations also apply to the non-normalised partial area metrics. The %PTF resulted in a highly statistically significant difference between the formulations as did C_{max}/AUC_τ, although the magnitude of effect was much smaller with the latter metric, i.e. an increase in mean ratio of 10% compared to 70%.

In study 2, there was no significant difference in extent of absorption; nor was there any significant difference in rate of absorption as measured by %PTF or any of the other rate metrics, with the exception of a borderline statistically significant difference in C_{max}.

The variability in the metrics across the two studies was similar (Table III). In both the real and simulated studies, %PTF was the most imprecisely estimated variable and C_{max}/AUC_τ was more precisely estimated than C_{max}.

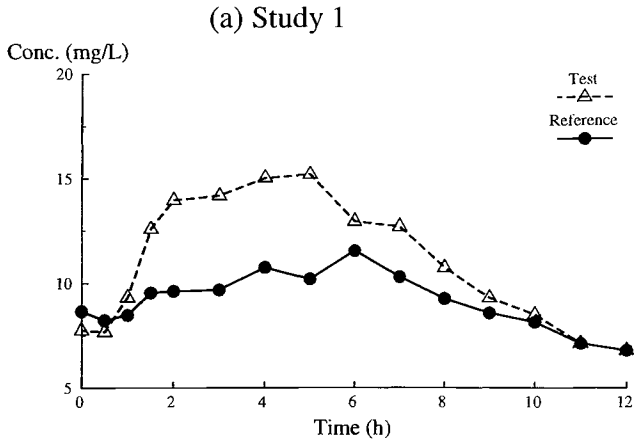
DISCUSSION

With the exception of %PTF, a 50% increase in ka was reflected as a much smaller increase in the other metrics. For example, there was only a 6% increase in C_{max}, a 2% increase in AUC_τ and a 4% increase in AUC_e (Table II). Although these metrics are only indirect measures of rate of drug absorption, their ability to detect and estimate underlying changes in rate are further attenuated after accumulation has occurred and steady-state reached. This is demonstrated by comparing the model predicted mean plasma profiles, under single dose conditions and at steady-state, for the two formulations from the second set of simulated experiments, where there was a 50% increase in ka and no change in the extent of absorption (Figure 3). The difference

Table III. Mean Ratios, 90% Confidence Intervals (CI), Associated p-Values, and CVs for the Different Metrics Obtained with Two Actual Pharmacokinetic Studies with ER Dosage Forms, Involving a Glaxo Drug

Metric	Mean ratio	90% CI	p-value	CV (%)
Study 1 (20% difference in AUC_τ)				
AUC_τ	1.20	1.13–1.28	<0.001**	11.4
C_{max}	1.32	1.25–1.40	<0.001**	10.2
C_{max}/AUC_τ	1.10	1.05–1.16	0.004**	9.0
AUC_e	1.23	1.09–1.38	0.007**	20.8
AUC_r	1.24	1.12–1.37	0.002**	18.3
AUC_e/AUC_τ	1.02	0.95–1.10	0.636	13.4
AUC_r/AUC_τ	1.03	0.97–1.10	0.379	10.8
%PTF	1.70	1.37–2.11	<0.001**	39.3
Study 2 (no significant difference in AUC_τ)				
AUC_τ	1.06	1.00–1.12	0.126	11.3
C_{max}	1.10	1.02–1.18	0.048*	14.4
C_{max}/AUC_τ	1.04	0.99–1.09	0.185	9.0
AUC_e	1.03	0.98–1.09	0.274	10.1
AUC_r	1.04	0.98–1.09	0.244	9.9
AUC_e/AUC_τ	0.98	0.95–1.01	0.298	6.3
AUC_r/AUC_τ	0.98	0.95–1.01	0.325	6.1
%PTF	1.19	0.99–1.42	0.122	35.9

CI = confidence interval for mean ratio.
* = statistically significant at $p < 0.05$.
** = statistically significant at $p < 0.01$.



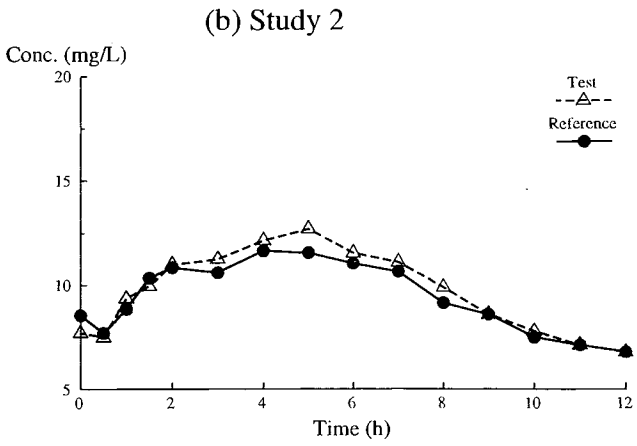
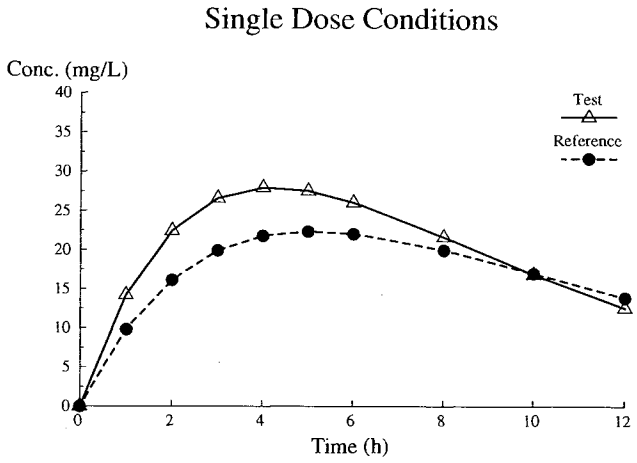


Figure 2: Plot of the median plasma concentration-time profiles obtained from: (a) Study 1 (n = 19) (20% difference in AUC_τ between formulations), and (b) Study 2 (n = 22) (no significant difference in AUC_τ between formulations).



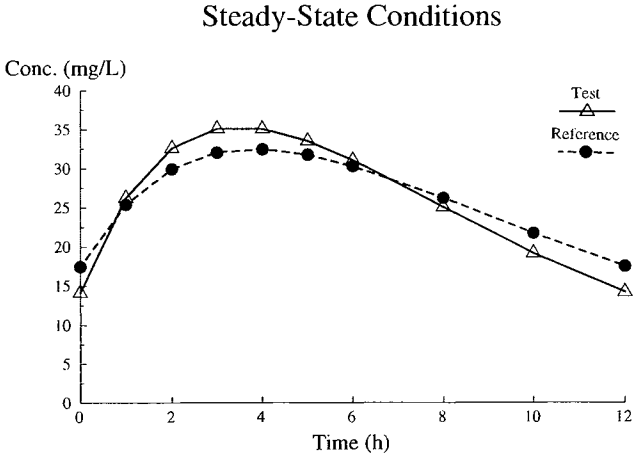


Figure 3: Plot of the model predicted mean plasma concentration-time profiles (50% increase in ka) under single dose conditions and at steady-state.

in rate of absorption is more apparent after the first dose than at steady-state.

Since ER dosage forms are usually intended for long term use, it might be argued that if a large difference in the underlying rate of absorption is not reflected in the steady-state plasma profiles then such a difference is of no clinical importance. However, this ignores the fact that the rate of absorption of ER dosage forms determines, to a great extent, the time required to reach steady-state and this may be of clinical importance. Also, although different ER dosage forms may appear to show similar steady-state profiles, there may actually be a large change in %PTF which may be clinically important. (Table II, Figure 3). Furthermore, ER dosage forms are specifically designed to control rate of drug absorption and therefore the in-vivo assessment of absorption rate must be of biopharmaceutical importance. Indeed, a recently issued guidance from the Office of Generic Drugs of the US Food and Drug Administration indicates that bioequivalence of ER dosage forms must be demonstrated under both single dose and steady-state conditions.[14]

%PTF resulted in the largest increase for a 50% increase in ka. It also had the largest variability; nonetheless, it was the most sensitive in detecting true differences in the underlying rate of absorption at steady-state. In these simulations, the large intra-subject variability in %PTF resulted in the failure to meet the 80-125% criterion in 62% of the studies, when the formulations were truly bioequivalent. The large inherent intra-subject variability in %PTF was also found in the two real studies. Therefore, the use of %PTF for bioequivalence assessment of ER dosage forms may require a widening of the currently accepted 80-125% permissible range, set for C_{max} and AUC_τ.

The current use of C_{max} as a measure of rate at steady-state for ER dosage forms is inappropriate because of its lack of sensitivity. Consequently, as shown in the second set of simulations, where there was a 50% increase in ka, use of the currently accepted range of 80-125% would have concluded bioequivalence in terms of rate of absorption for the majority (62%) of cases. The same arguments apply also to the partial areas and particularly to C_{max}/AUC_τ and the normalised partial area metrics. Such metrics would be expected to behave with greater sensitivity under single dose conditions (Figure

3). Therefore, if an indirect assessment of rate of absorption is required using these metrics, our findings suggest that a single dose design would be more appropriate. Alternatively, if a steady-state design is necessary, then the permissible range should be tightened in order to reflect the lack of sensitivity of these metrics under steady-state conditions.

When the concentration at the end of the dosing interval is much less than C_{max}, then %PTF will be approximately equal to $\tau \times C_{max}/AUC_{\tau}$, which is directly proportional to C_{max}/AUC_{τ}. Therefore, %PTF and C_{max}/AUC_{τ} are related metrics and %PTF can be considered to be a generalisation of C_{max}/AUC_{τ}. The reason why %PTF has greater intra-subject variability than C_{max}/AUC_{τ} is because a difference in concentration at two time points has a greater inherent variability than a concentration at either time point alone.

This paper only examined results from two actual studies with one drug. While there was consistency in the findings between the simulated and actual studies, our conclusions require further confirmation from additional real studies with different classes of drugs. Also, simulations of alternative scenarios (eg. non flip-flop situation, zero-order absorption, presence of a lag-time, two-compartment models etc.[15]) would provide useful additional information.

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