Bioequivalence: Individual and Population Compartmental Modeling Compared to the Noncompartmental Approach

Helen S. Pentikis,[1,4] James D. Henderson,[2] Nhan L. Tran,[2] and Thomas M. Ludden[1,3]

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Purpose. The purposes of this study were to evaluate the use of individual compartmental and population compartmental methods for bioequivalence determination, and to determine their utility as adjuncts to the current methods used for bioequivalence assessment.

Methods. Data from three bioequivalence studies of chlorthalidone were analyzed with PCNONLIN using individual compartmental modeling and NONMEM for population analyses. These results were compared with results obtained from the traditional noncompartmental or SHAM (slopes, heights, areas, and moments) approach for bioequivalence assessment and the 90% confidence interval procedure.

Results. Individual compartmental modeling and population compartmental modeling techniques performed well on this routine set of bioequivalence data which displayed simple pharmacokinetic properties. A direct assessment of the analysis methods was made by comparing the final estimates and 90% confidence intervals for the test to reference ratios (T/R) of AUC and CMAX. The final estimates and 90% confidence intervals for AUC T/R and CMAX T/R were similar and suggest consistency of results, independent of the method used.

Conclusions. These results demonstrate the utility of modeling techniques as adjuncts to the traditional noncompartmental approach for bioequivalence determination.

KEY WORDS: modeling; NONMEM; bioequivalence; noncompartmental; population; compartmental.

INTRODUCTION

The Office of Generic Drugs (OGD) of the US Food and Drug Administration currently accepts the results of noncompartmental analyses of human pharmacokinetic data for bioequivalence determination; that is, to establish the therapeutic equivalence of generic drug products to other pharmaceutically equivalent products. Although commonly used to characterize the plasma concentration-time curve, the noncompartmental approach or SHAM analysis (Slopes, Heights, Area, Moment) (1) has certain limitations. These limitations may include approximations introduced by trapezoidal area under the curve (AUC) calculations, inconsistencies in estimating the terminal elimination rate constant (λ_z), and the use of "observed" CMAX values.

Modeling techniques such as individual compartmental modeling and population compartmental methods may serve as adjuncts to the traditional SHAM approach. Individual compartmental modeling and population compartmental analyses provide a mathematical equation and a set of parameter values that yield a detailed description of the plasma concentration-time curve. The predicted plasma concentrations are then determined by the model based parameters and may therefore improve upon the SHAM analyses. In addition, population compartmental methods permit the use of all the data simultaneously for model definition and are better able to estimate the interindividual variability of the pharmacokinetic parameters and the residual intraindividual (assay) variability associated with the concentration data.

The purposes of this study were to evaluate the use of individual compartmental and population compartmental modeling methods for bioequivalence determination and to determine the utility of these methods as adjuncts to the current methods used for bioequivalence assessment. Data from three bioequivalence studies of chlorthalidone were analyzed using individual compartmental modeling and mixed effect population modeling. In addition, a unique approach was used whereby Monte Carlo simulations were conducted using the variance-covariance matrix of the population parameter estimates, thereby enabling the comparison of CMAX obtained by three approaches. The results from the individual compartmental and mixed effect population modeling were compared with results obtained from the traditional noncompartmental (SHAM) approach for bioequivalence assessment, including the 90% confidence interval procedure.

METHODS

Drug Selection

Chlorthalidone was selected as the drug product in this study for three reasons: (1) several bioequivalence studies exist with AB-rated generic products; (2) the pharmacokinetic profile of chlorthalidone in whole blood is adequately described by a one-compartment model (2), thereby avoiding the difficulty of separating distribution and elimination phases for a drug with two-compartment characteristics; (3) it is slowly absorbed which enables better characterization of the absorption phase.

Study Selection

Three bioequivalence studies which were conducted as two-way crossovers under fasting conditions and which analyzed whole blood by a chromatographic method for chlorthalidone concentrations were selected.

Noncompartmental Analyses

Calculations were performed as described in a guidance issued by the Division of Bioequivalence (DBE) and OGD (3). In some cases, it was necessary to reanalyze the original study data using the General Linear Models (GLM) procedure of SAS v. 6.04 (4).

[1] Division of Biopharmaceutics, Center for Drug Evaluation and Research, Food and Drug Administration, Rockville, Maryland 20857.

[2] Office of Generic Drugs, Center for Drug Evaluation and Research, Food and Drug Administration, Rockville, Maryland 20857.

[3] Current Address: Department of Pharmaceutical Sciences, College of Pharmacy, University of Nebraska Medical Center, Omaha, Nebraska 68198.

[4] To whom correspondence should be addressed at: Rhône-Poulene Rorer Central Research, DMPK, NW13, 500 Arcola Road, Collegeville, Pennsylvania 19426.

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Individual Compartmental Analyses

Individual subject chlorthalidone blood concentration-time curves (test and reference products) were analyzed using a one-compartment model equation (first-order input and elimination, with or without lag time) with the nonlinear regression program PCNONLIN v. 3.0 (5). The Gauss-Newton algorithm (with Levenberg's modification) of PCNONLIN was used for nonlinear estimation with a weighting factor of 1/observed concentration. In some cases, the grid search option was used instead of initial parameter estimates. The results of the PCNONLIN outputs were evaluated as described elsewhere (5).

Population Compartmental Analyses

NONMEM (Version IV)(6) was used to develop a pharmacokinetic and statistical model for the test and reference formulations for three chlorthalidone products. All analyses were performed using the first order (FO) method. A one compartment model with first order absorption was used to describe the pharmacokinetics of chlorthalidone. The statistical model explained the interindividual variability in the pharmacokinetic parameters and the intraindividual residual error (6). The interindividual variability was modeled with a proportional error model:

$$CL_j = CL * (1 + \eta_j^{CL})$$

$$Vd_j = Vd * (1 + \eta_j^{Vd})$$

where η_jCL and η_jVd describe the interindividual variability for clearance and volume of distribution, respectively, and are assumed to be normally distributed random variables with a mean of zero. CL_j and Vd_j are the predicted values for clearance and volume of distribution for the jth individual and CL and Vd are the typical values for clearance and volume of distribution.

To determine differences in the rate of absorption and the lag time of the test and reference formulations of chlorthalidone, the following parameterization was used:

$$\text{For DF} = 0 \text{ (Reference)}$$

$$\text{KAR} = \theta_1$$

$$\text{ALAGR} = \theta_2$$

$$\text{For DF} = 1 \text{ (Test)}$$

$$\text{KAT} = \theta_3 * \text{KAR}$$

$$\text{ALAGT} = \theta_4 * \text{ALAGR}$$

$$\text{KAR (or T)}_j = \text{KAR (or T)} * (1 + \eta_j^{KA})$$

$$\text{ALAGR (or T)}_j = \text{ALAGR (or T)}$$

$$F1_j = 1 * (1 - DF) * \exp\cdot(\eta_j^F) + FT * DF * \exp(\eta_j^F)$$

Thus, when the dosage form indicator variable, DF is zero, KAR and ALAGR are equal to typical values of the absorption rate constant and lag time for the reference product, respectively. Alternatively, when DF = 1, θ_3 * KAR and θ_4 * ALAGR are equal to the absorption rate constant and lag time for the test product. By the aforementioned parameterization, θ_3 = KAT/R and θ_4 = ALAGT/R and are the test to reference ratios for the absorption rate and lag time. For relative bioavailability, F1 was set equal to one for the reference product and was estimated for the test product (FT). The interindividual variability for the absorption rate constant is estimated by η_j^{KA}, whereas the interindividual variability for lag time was not estimated. As a result of the final model building process, the intraindividual residual error for study #1 and study #2, was modeled with a combined additive and proportional error model:

$$C_{i,j} = Cm_{i,j} * (1 + \epsilon_{1_{i,j}}) + \epsilon_{2_{i,j}}$$

where Cm_{ij} is the ith model predicted concentration of the jth individual. $\epsilon_{1_{ij}}$ and $\epsilon_{2_{ij}}$ are assumed to be normally distributed random variables which describe the intraindividual residual error. The intraindividual residual error was modeled with an additive error model for study #3.

The ADVAN2 and TRANS2 subroutines from PREDPP (first order absorption and elimination model subroutines parameterized in apparent oral clearance and apparent volume of distribution) were used to calculate the predicted concentrations and parameter estimates to achieve minimization of the objective function (6). Both additive and proportional error models for the inter- and intrasubject (residual) random terms were tested and goodness of fit was assessed by examination of residual and weighted residual plots and by the relative objective function value. A model for the residual error that included both additive and proportional components was also tested. In this case, where there is a possible difference in the number of parameters between the two models a formal statistical test is possible. The difference in the objective function values between the full and reduced models is chi-squared distributed and therefore a change of 3.85 ($p < 0.05$) was used to assess a significant difference.

The likelihood profile approach (7) was used to generate 90% confidence interval estimates for KA T/R, F T/R, and ALAG T/R. In this procedure, the parameter of interest is fixed to a series of values above and below the maximum likelihood estimate and all other parameters are reestimated. A plot of the objective function values versus the fixed parameter values can be used to determine the decrease and increase in parameter values below and above the maximum likelihood estimate that correspond to a change in the objective function value of 3.85. The resulting interval is an estimate of a 90% confidence interval for the parameter.

Monte Carlo Simulations

Monte Carlo simulations were used to derive estimates of CMAX T/R and AUC T/R from the population compartmental analyses. Monte Carlo simulations were necessary to derive estimates of CMAX T/R because the final NONMEM model cannot be reparameterized to estimate the observed CMAX. The AUC T/R was also estimated in this manner. The final parameter estimates and the variance-covariance matrix of the estimates from NONMEM for the three chlorthalidone studies were used to generate 10,000 sets of mean concentration-time profiles from the equation describing one-compartment pharmacokinetics with first order input and elimination. The multivariate random terms were generated using an Splus function (9) based on a singular value decomposition of the variance-covariance matrix of the estimates. The observed CMAX was determined from each plasma concentration-time profile and AUC from the equation AUC = Dose/CL for the reference product and AUC = FT * Dose/CL for the test product. Approximate

90% confidence intervals for the mean estimates of CMAX T/R and AUC T/R were then estimated from the distribution of the 10,000 replications.

RESULTS

Noncompartmental Analyses

The test products demonstrated higher plasma concentrations for all three studies as compared to their respective reference products. Table I contains the noncompartmental mean estimates and standard deviations for the pharmacokinetic parameters from the three studies. The mean parameter estimates of CMAX and AUC for the test and reference products are shown in the top panels of Figures 1 and 2, respectively. The parameter estimates from study #1 were similar, with mean test/reference ratios for AUC_{0-t}, AUCINF, and CMAX ranging from 1.03–1.06. Greater differences were seen between parameter estimates for the test and reference products from studies #2 and #3, with mean test/reference ratios for AUC_{0-t}, AUCINF, and CMAX ranging from 1.12–1.26.

Individual Compartmental Analyses

Table II contains the compartmental mean estimates and standard deviations for the pharmacokinetic parameters from the three studies. The mean parameter estimates for CMAX and AUCINF for the test and reference products are shown in the middle panels of Figure 1 and 2, respectively. The mean parameter estimates were similar in study #1, with mean test/reference ratios of 1.04 and 1.07, respectively. Greater differences were seen between products for studies #2 and #3, with mean test/reference ratios for AUCINF and CMAX ranging from 1.13–1.24. The mean parameter estimates for KA for the test and reference products were different for all three studies.

Table I. Noncompartmental Mean Estimates and Standard Deviations for Parameters

	Study #1 25 mg	Study #2 50 mg	Study #3 50 mg
AUCT[a]			
T[b]	134 (25)	279 (47)	173 (30)
R[b]	128 (25)	241 (60)	148 (29)
AUCINF (mg-hr/L)			
T	166 (37)	328 (57)	203(39)
R	160 (35)	292 (74)	179(36)
CMAX (mg/L)			
T	2.13 (0.35)	4.62 (0.89)	2.71 (0.47)
R	2.01 (0.38)	3.93 (0.99)	2.14 (0.47)
TMAX (hr)[c]			
T	9.58 (2.43)	9.63 (4.62)	10.8 (1.9)
R	10.4 (2.6)	10.1 (2.4)	13.1 (5.0)
KE (hr^{-1})			
T	0.015 (0.002)	0.016 (0.002)	0.017 (0.002)
R	0.014 (0.002)	0.016 (0.003)	0.015 (0.002)

[a] AUCT is the area under the curve (mg-hr/L) calculated by the trapezoidal rule to the last nonzero concentration.
[b] T = test product least squares (LS) mean from the ANOVA. R = reference product LS mean.
[c] Test (T) and Reference (R) product arithmetic means.

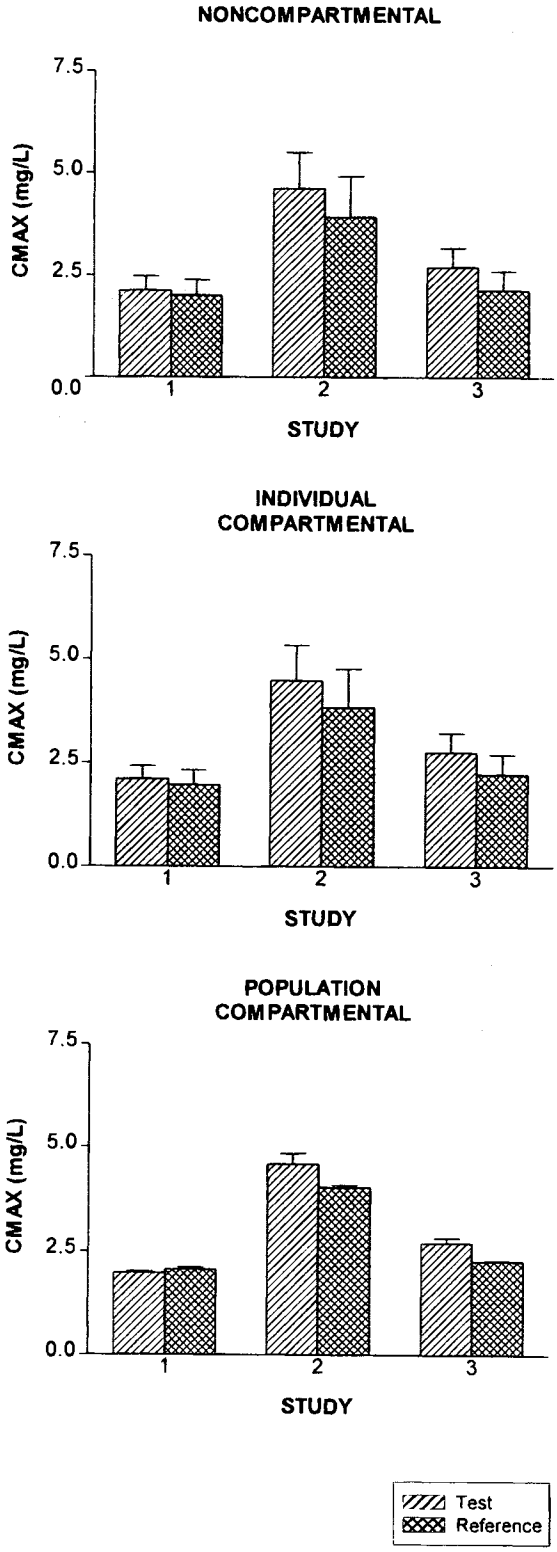

Fig. 1. Mean CMAX (mg/L) parameter estimates for test and reference products from three chlorthalidone studies derived by noncompartmental (top), individual compartmental (middle), and population compartmental (bottom) techniques. Error bars represent standard deviations for noncompartmental and individual compartmental analyses and standard errors for population compartmental analyses.
Key: ▨, Test; ▩, Reference.

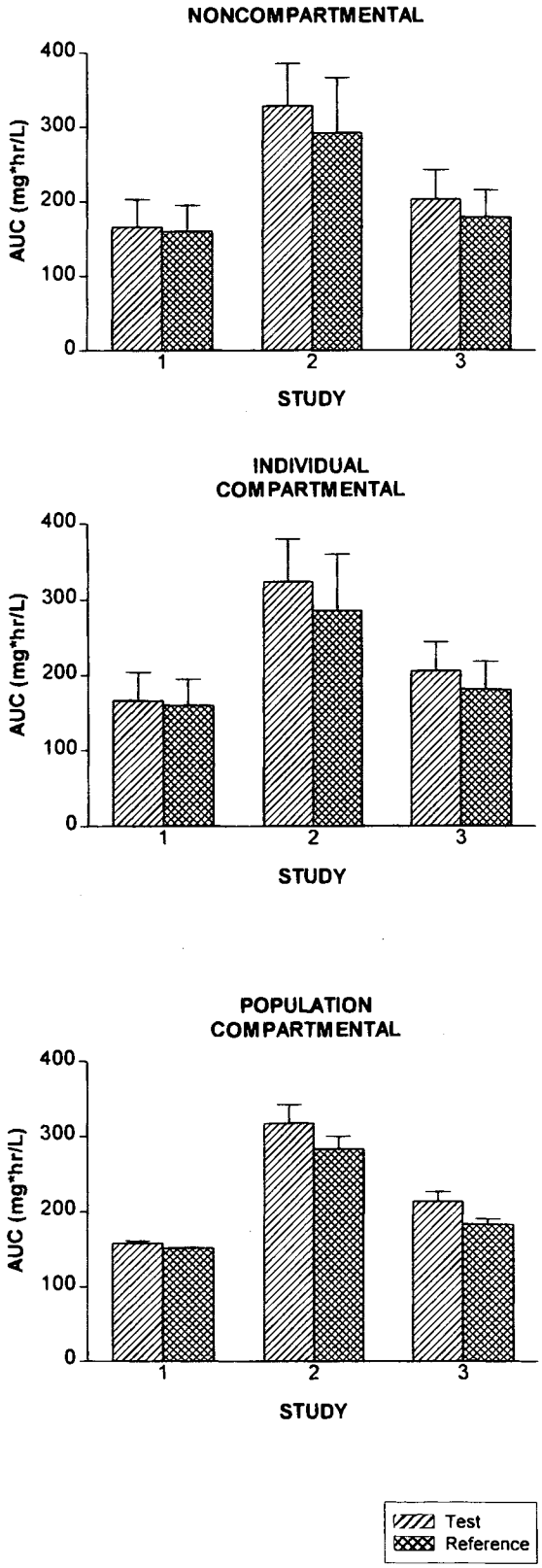

Fig. 2. Mean AUC (mg*hr/L) parameter estimates for test and reference products from three chlorthalidone studies derived by noncompartmental (top), individual compartmental (middle), and population compartmental (bottom) techniques. Error bars represent standard deviations for noncompartmental and individual compartmental analyses and standard errors for population compartmental analyses.
Key: ▨, Test; ▩, Reference.

Table II. Compartmental Mean Estimates and Standard Deviations for Parameters

	Study #1 25 mg	Study #2 50 mg	Study #3 50 mg
AUCINF (mg-hr/L)			
T	166 (38)	324 (56)	206 (38)
R	160 (35)	286 (74)	181 (37)
CMAX (mg/L)			
T	2.11 (.32)	4.50 (.85)	2.76 (.46)
R	1.97 (.36)	3.84 (.94)	2.22 (.48)
TMAX (hr)			
T	7.61 (1.11)	9.45 (2.45)	10.7 (2.73)
R	8.78 (1.23)	10.36 (2.12)	11.8 (2.38)
KA (hr^{-1})			
T	0.535 (.110)	0.410 (.140)	0.35 (.130)
R	0.444 (.090)	0.359 (.110)	0.30 (.080)
KE (hr^{-1})			
T	0.014 (.002)	0.016 (.002)	0.016 (.002)
R	0.014 (.002)	0.016 (.002)	0.015 (.002)
Vd/F (L)			
T	11.0 (1.7)	9.95 (1.8)	15.8 (2.7)
R	11.7 (2.3)	11.9 (3.2)	20.4 (6.1)
CL/F[a] (L/hr)			
T	0.158	0.158	0.250
R	0.164	0.187	0.287

[a] Clearance (CL/F = KE * Vd/F).

These may well be differences in the rates of absorption that are not revealed by the CMAX estimates. The mean clearance and volume estimates were similar for the test and reference products for studies #1 and #2 but were slightly increased for study #3 and may be due to differences in the methods used to assay chlorthalidone concentrations.

Population Compartmental Analyses

For each chlorthalidone product, a proportional error model was found to best describe the interindividual variability in CL, Vd, KA, and F. A combined additive and proportional error model was found to best describe the intrasubject (residual) error for studies #1 and #2. An additive error model was found to be sufficient for the intrasubject (residual) error for study #3. The intrasubject (residual) variances corresponding to coefficients of variations of 45% at .15 ug/ml and 7% at 2.5 ug/ml for study #1, 61% at .2 ug/ml and 18% at 6 ug/ml for study #2, and 63% at .3 ug/ml and 5% at 4 ug/ml for study #3.

The population parameter estimates from the final model for the three chlorthalidone products and the standard errors of the estimates are displayed in Table III. The parameter estimates for CL and Vd from studies #1 and #2 were similar. The CL and Vd estimates from study #3 were greater than those determined from studies #1 and #2 and again, may be due to differences in the calibration of the chlorthalidone assay. The estimates for KA T/R and F T/R were 1.08 and 1.04, respectively and indicate that the absorption rate constant and bioavailability for the test and reference products were equivalent for study #1. Greater differences were observed between test and reference products for studies #2 and #3, with parameters estimates for KA T/R and F T/R being 1.23 and 1.12, respectively, for study #2, and 1.32 and 1.16, respectively, for study #3. The mean parameter estimates for AUC and CMAX for the test and refer-

Table III. Population Parameter Estimates and Standard Errors for the Final Population Model

	STUDY #1 25 mg		STUDY #2 50 mg		STUDY #3 50 mg	
	Estimate	S.E.	Estimate	S.E.	Estimate	S.E.
CL/FR (L/hr)	0.164	0.001	0.177	0.010	0.273	0.011
Vd/FR (L)	11.3	0.4	10.9	0.5	18.5	0.9
KA R[a] (hr^{-1})	0.540	0.11	0.442	0.048	0.266	0.016
ALAG R (hr)	0.627	0.038	0.341	0.038	0.586	0.082
KA T[b]/R	1.08	0.06	1.23	0.10	1.32	0.06
F T/R	1.04	0.01	1.12	0.06	1.16	0.05
ALAG T/R	0.590	0.055	0.946	0.151	1.07	0.15
CMAX T	1.99	0.04	4.60	0.25	2.72	0.12
CMAX R	2.08	0.05	4.04	0.05	2.28	0.02
AUC T	158	3	317	25	213	13
AUC R	152	1	283	17	183	7

[a] R = Reference.
[b] T = Test.

ence products are shown in Figure 2 and 3, respectively. The mean parameter estimates from study #1 were equivalent, with test/reference ratios ranging from 1.04–1.05. Greater differences were seen between products for studies #2 and #3, with test/reference ratios for AUC and CMAX ranging from 1.12–1.19.

90% Confidence Interval Estimation

The 90% confidence intervals for the difference between test and reference means for the noncompartmental and individual compartmental analyses were calculated as described by Schuirmann (10) and are shown in Table IV. For the population compartmental analyses, the 90% confidence intervals for KA, F, and ALAG for the three products of chlorthalidone were determined by constructing likelihood profiles (8) while the 90% confidence intervals for AUC T/R and CMAX T/R were derived by a Monte Carlo method from the NONMEM final parameter estimates and variance-covariance matrix of the estimates. These intervals are also shown in Table IV. A comparison of the 90% confidence intervals indicates a general consistency of the outcome for bioequivalence, independent of the modeling method used.

Table IV. 90% Confidence Intervals for T/R Ratios (Expressed as Percentages)

Noncompartmental Analyses[a]			
	STUDY #1	STUDY #2	STUDY #3
AUC	99.4–108	105–126	105–123
CMAX	102.7–110.3	107–133	117–141
Individual Compartmental Analyses[a]			
	STUDY #1	STUDY #2	STUDY #3
AUC	99.3–108	105–127	106–123
CMAX	104–111	107–132	115–138
Population Compartmental Analyses			
	STUDY #1	STUDY #2	STUDY #3
AUC[b]	102–106	102–122	108–122
CMAX[b]	102–107	104–124	112–126
KA[c]	101–114	110–139	115–152
F[c]	103–105	108–116	113–120
ALAG[c]	52–66	70–125	75–185

[a] Based on ln transformed parameters.
[b] Monte Carlo simulations using the variance-covariance matrix for the parameter estimates.
[c] Obtained from likelihood profile plots.

DISCUSSION

Recent reports in the literature have suggested the application of population compartmental modeling techniques to the determination of bioequivalence for clinical and experimental data (11,12). These investigators have demonstrated the utility of population compartmental modeling in affording supplemental information to bioequivalence analyses. To this end, we examined individual compartmental modeling and population compartmental modeling as methods of analyses for bioequivalence and evaluated their applicability as adjuncts to the standard noncompartmental (SHAM) approach for bioequivalence determination and the 90% confidence interval procedure. A unique method, incorporating Monte Carlo simulations for estimating CMAX from the variance-covariance matrix of the population parameters estimates, was implemented during our analyses. This enabled the derivation of CMAX from the population compartmental models. A direct assessment of modeling methodologies was then made by comparing the estimates of AUC T/R and CMAX T/R, and the 90% confidence intervals derived from the final fits of the data. The final estimates for AUC and CMAX, and the 90% confidence intervals, determined by the three methods, are similar and indicate consistency of results, independent of the modeling method used.

These results indicate the applicability of modeling techniques to the assessment of bioequivalence data. Individual compartmental modeling and population compartmental modeling may serve to better approximate the pharmacokinetics of drugs with more complicated plasma concentration-time profiles. In addition, population compartmental modeling techniques permit

the use of all the data simultaneously for model definition and provide estimates of interindividual variability, interoccasion variability (not applied in the current examples), and intraindividual residual variability. These modeling techniques use the actual observation, i.e. concentrations, instead of derived parameters as the dependent variable. The resulting analyses can be more informative and warrant further examination as adjuncts to the standard approach for bioequivalence determination. For example, the population compartmental modeling approach provides direct estimates of the test to reference ratios of KA and ALAG not available in noncompartmental analyses. In addition, the CMAX values obtained by the population compartmental approach are likely to be less biased than the observed CMAX values usually used in bioequivalence studies. A major advantage of the population approach can be the ability to assess the various components of variability, including interoccasion variability (13) and differences in variability between products (14). Neither of these issues was addressed in the current work. However, examination of the estimates for intrasubject (residual) variability suggest that study #2 exhibited more residual variability than did the other two studies. Such information may be useful in a quality control context.

The current results indicate that the various methods yield very similar conclusions regarding mean bioequivalence for data that can be described by relatively simple pharmacokinetic models. Failure to produce similar results for these data sets would have raised concerns about the usefulness of these methods. Additional analyses examining data sets that exhibit absorption complexities such as mixed rates of inputs or double peaks need to be performed.

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