DRUG DEVELOPMENT AND INDUSTRIAL PHARMACY, 17(9), 1157-1176 (1991)

Limitations of Presently Available *In Vitro* Release Data for the Prediction of *In Vivo* Performance

M.K. Kottke and C.T. Rhodes

University of Rhode Island

Department of Pharmaceutics

Kingston, RI 02881

Objectives

1. To determine whether any correlations can be found between in vitro release data and in vivo performance
2. To determine whether differences exist between reference and test products in terms of both in vitro release and in vivo performance
3. To compare dissolution of the same product obtained using different dissolution test methods
4. To evaluate the variability of dissolution and bioavailability data between and within products

Introduction

With the advent of the Drug Price Competition and Patent Term Restoration Act of 1984, increasing numbers of Abbreviated New Drug Applications (ANDAs) are being submitted to the Federal Food and Drug Administration (FDA) in hopes of being granted bioequivalence to brand name (reference) products. Drugs which are deemed to be

bioequivalent to the reference product are then given an AB rating by the FDA. In order to receive this rating, there must be reasonable assurance that the rate and extent of absorption of the test product do not differ significantly from those of the reference product. In addition, the Division of Bioequivalence requires the dissolution profiles of the test and reference products be similar

Due to the excessive demands placed upon the Agency and the generic companies performing the bioavailability studies, it would be beneficial if some adequate correlation was established between *in vitro* release and *in vivo* performance so that bioequivalence might be granted based solely upon *in vitro* data. Currently, subjects used in these bioavailability studies are most often "normal" young males between the ages of 18 and 25 who are carefully selected so as to reduce any intra- or inter- subject variability (i.e. non-smokers, non-medicated). As such, these subjects do not adequately reflect the entire population in which the product is likely to be used and therefore, it does not seem justifiable to view them strictly as "clinical mirrors." It may be accurate to regard the function of these subjects as being that of "human test tubes" into which the drug is administered and blood/urine samples are removed. Thus bioavailability tests are essentially quality control tests. Also, of course, the experimentation on human subjects required for these studies may be considered unethical by some, particularly when large numbers of blood samples must be drawn.

During the past twenty years or so, numerous attempts have been made to conclusively prove that correlations between *in vitro* release and *in vivo* performance can be found (1-14). Theoretically, these correlations are based upon the assumption that the dissolution properties of a product can function as an indicator of its absorption and consequent bioavailability within the blood stream. In other words, if the product's rate of dissolution is the limiting-factor/regulator of its absorption, it should serve as a true predictor of its bioavailability. Unfortunately, most of these studies are limited in the number of products evaluated, thereby casting doubt upon their universal applicability. This study was undertaken in order to evaluate existing data available within the FDA's

Division of Bioequivalence. Due to the large data base used, it was felt that any prior claims to *in vitro - in vivo* correlation could be more substantially confirmed or denied.

Methods

Data Selection and Retrieval

All products evaluated within this study were chosen because of their immediate release characteristics. Another criteria for inclusion was the relative accessibility of ANDA files for each product. Therefore, only products for which there exist a large number of ANDA applications were included. Based on these criterion, the following products were chosen: Diazepam Tablets; Doxepin HCl Capsules; Ibuprofen Tablets and Lorazepam Tablets. Table 1 lists the relevant physico-chemical and pharmacokinetic parameters for each of these drugs. With the exception of Doxepin HCl, each has good oral bioavailability and thus would not be expected to exhibit wide subject-to-subject variability. In addition, all products, excepting Doxepin HCl, have poor water solubility and therefore, their oral absorption is likely to be rate-limited by the product's dissolution rate. These characteristics of small intersubject variability and poor water solubility should be ideal for developing *in vitro - in vivo* correlations. Analysis of Doxepin HCl may then serve as a non-ideal candidate by which the accuracy of the correlations may be measured.

All data were contained within ANDA summaries that have been prepared by reviewers from the Division of Bioequivalence and represent the mean values of each parameter investigated (i.e. individual subject/tablet data were not included). Table 2 enumerates the wide variety of dissolution tests which were performed on each of the products under investigation. It is important to note that for each ANDA application submitted, only one or two of these tests were applied.

Data Analysis

Once the appropriate ANDA summaries were retrieved, the raw data for each product was entered onto a floppy disk in the following format: ANDA number;

Table 1. Physico-Chemical and Pharmacokinetic Characteristics of the Drugs Evaluated in this Study

Drug	pKa	Solubility in Water	Oral Bioavailability	Half-Life (hours)	Active Metabolites
Diazepam	3.3	1g in 333ml	> 90%	20 - 70	Yes
Doxepin HCl	8.0	1g in 1.5ml	< 50%	8 - 25	Yes
Ibuprofen	5.2	very slightly sol.	> 90%	1 - 2	No
Lorazepam	11.5,1.3	practically insol.	> 90%	10 - 14	Yes

Table 2. Types of Dissolution Tests Utilized in this Study

Drug	Dissolution Type	Apparatus	Dissolution Media	Volume (ml)	RPM
Diazepam	1	[a] Basket (I)	0.1N HCl	900	100
	2	I	0.5M phosphate buffer	900	100
	3	I	0.5M acetate buffer	900	100
Doxepin HCl	4	I	distilled water	900	50
Ibuprofen	5	[a] I	pH 7.2 phosphate buffer	900	150
	6	[b] Paddle (II)	pH 7.2 phosphate buffer	900	50
Lorazepam	7	[a] I	distilled water	500	100
	8	II	distilled water	500	50

[a] Indicates dissolution parameters listed in USP XXII

[b] Indicates dissolution parameters suggested by the FDA

manufacturer; lot number of reference product; sample type (reference or test); dissolution type (see Table 2); dose (mg); AUC_{0-t}; $AUC_{0-\infty}$; C_{max}; cumulative percent dissolved at ten (D_{10}), fifteen (D_{15}), twenty (D_{20}), thirty (D_{30}), forty-five (D_{45}) and sixty minutes (D_{60}). The data were then evaluated using the SAS® statistical software package. It should be noted that most of the applications were incomplete in that they did not contain all of the variables listed above. Therefore, missing variables were replaced by a ".". This symbol is recognized by SAS® as a missing value and thus is not included in the analysis of that particular variable.

Prior to any statistical evaluation of the data, the applications were sorted by ANDA number and the biological parameters were dose-normalized to 1mg (i.e. all biological parameters were divided by the administered dose) so that all analyses would be based upon a 1mg dose. In addition, for each ANDA, the value of every test parameter was divided by its corresponding reference parameter value to form an additional set of data including the following variables: $r\,AUC_{0-t}$; $r\,AUC_{0-\infty}$; $r\,C_{max}$; $r\,D_t$. The purpose behind this manipulation was the reduction of any inter-ANDA variability (i.e. most bioavailability studies are conducted in different labs, in different parts of the country and may call for a different protocol).

With this completed, a preliminary search for existing correlations between *in vitro* (D_t, $r\,D_t$) and *in vivo* (AUC_{0-t}, $AUC_{0-\infty}$, C_{max}, etc.) parameters was applied through the use of the Correlation procedure (15). This procedure evaluates the Pearson product moment coefficient of correlation, r (Equation 1 of Appendix A), which serves as an indicator of the extent to which a correlation between two variables exists. In this manner, over 400 correlations were evaluated. Those correlations having an absolute value of r exceeding 0.45 were included for further analyses by the Regression and "Rsquare" procedures (16,17).

Both the Regression and Rsquare procedures perform analysis of variance on a given model and report its coefficient of determination, r^2 (Equation 3 of Appendix A). For instance, if the Correlation procedure outlined above showed that an acceptable

correlation exists between AUC_{0-t} and D_{45}, the model inputted for analysis would be:

$$AUC_{0-t} = b_0 + b_1D_{45}.$$

In those instances where more than one *in vitro* parameter was found to give a an acceptable correlation with a given *in vivo* parameter, the model was expanded to include each of these variables. An example being the case where AUC_{0-t} correlates well with D_{15}, D_{45} and D_{60}. In this case, the inputed model would be:

$$AUC_{0-t} = b_0 + b_1D_{15} + b_2D_{45} + b_3D_{60}.$$

The major difference between the Regression and Rsquare procedures is that when multiple independent (*in vitro*) parameters are modeled with a dependent (*in vivo*) parameter, the Rsquare procedure will evaluate each single and multiple correlation (i.e. AUC_{0-t} with D_{15}; AUC_{0-t} with both D_{15} and D_{45}, etc.) while the Regression procedure only evaluates the specified model.

Once all correlations were evaluated, the Ttest procedure was employed to determine the variability of each parameter, as well as any differences existing between reference and test products or types of dissolution testing (18). This procedure computes the value of t based upon the assumptions of both equal (Equations 4 and 5 of Appendix A), t_e, and unequal, t_u variances (Equation 6 of Appendix A). Those examining the data can chose whether the variances are equal or unequal based upon the folded form of the F statistic, F', which is also computed by this procedure (Equation 7 of Appendix A). Generally, when the probability that F is greater than F' is large, the variances are assumed to be equal. For the purpose of this study, unequal variances were said to occur only when the probability was less than 0.1.

Results and Discussion

An arbitrary lower limit of 0.75 for the value of r^2 was used in order to determine whether or not a given correlation existed. These correlations, as well as corresponding r^2 values and number of observations by which they were determined, are summarized in Table 3. One can easily see that most of these correlations apply to test/reference values

Table 3. Existing Correlations Between *In Vitro* and *In Vivo* Parameters

Drug	Dissolution Test	Equation	r^2	N
Diazepam	[a]1	[b]$r\ AUC_{0-t} = 2.512 - 0.5478(r\ D_{30}) + 0.0352(r\ D_{60})$	0.9752	4
	3	$AUC_{0-\infty} = -3951.2 + 92.76(D_{20})$	0.8063	4
	3	$AUC_{0-\infty} = -4342.6 - 10.6213(D_{10}) + 107.6(D_{20})$	0.8170	4
	3	$r\ AUC_{0-t} = 0.911 + 0.0924(r\ D_{30}) - 0.1601(r\ D_{45}) + 0.133(r\ D_{60})$	0.9426	6
	3	$r\ AUC_{0-\infty} = 0.8765 + 0.4162(r\ D_{30}) - 0.5479(r\ D_{45}) + 0.2535(r\ D_{60})$	0.8587	5
Ibuprofen	5	$AUC_{0-\infty} = -2.8413 + 0.0105(D_{45}) + 0.0209(D_{60})$	0.9208	4
	5	$AUC_{0-\infty} = -10.9341 - 0.0058(D_{15}) + 0.1173(D45)$	0.8488	3
	5	$r\ AUC_{0-t} = 7.3099 - 6.248(r\ D_{60})$	0.9167	5
	5	$r\ AUC_{0-\infty} = -1.3604 + 2.3786(r\ D45)$	0.9953	3
	5	$r\ C_{max} = 14.1309 - 12.8945(r\ D_{60})$	0.9996	5
Lorazepam	[c]All	$r\ C_{max} = -0.3347 - 0.2292(r\ D_{10}) + 1.6438(r\ D_{20})$	0.7785	6
	All	$r\ AUC_{0-\infty} = 2.3908 - 3.1906(r\ D_{15}) + 12.9077(r\ D45) - 11.0143(r\ D_{60})$	0.9968	5
	8	$AUC_{0-\infty} = 297.8 - 0.3472(D_{10}) - 0.8454(D45)$	0.8543	4
	8	$C_{max} = 28.416 - 0.0637(D_{10}) - 0.1233(D45)$	0.8848	4

Table 3. Existing Correlations Between *In Vitro* and *In Vivo* Parameters (continued)

Drug	Dissolution Test	Equation	r^2	N
Lorazepam	8	$r\,AUC_{0-t} = 0.8351 - 0.8614(r\,D_{15}) + 0.7074(r\,D_{30}) + 0.4088(r\,D_{60})$	0.8717	6
	8	$r\,AUC_{0-t} = 0.8566 - 0.801(r\,D_{15}) + 1.2835(r\,D45) - 0.2513(r\,D_{60})$	0.9006	6
	8	$r\,AUC_{0-\infty} = 12.279 + 9.6502(r\,D_{15}) - 22.4226(r\,D_{60})$	0.9846	4
	8	$r\,C_{max} = 0.2907 - 0.7945(r\,D_{15}) + 1.9789(r\,D45) - 0.3819(r\,D_{60})$	0.7892	6

[a] see Table 2
[b] r = test parameter/ reference parameter
[c] All = all dissolution studies for Lorazepam

Note: No significant correlations were found for Doxepin HCl capsules

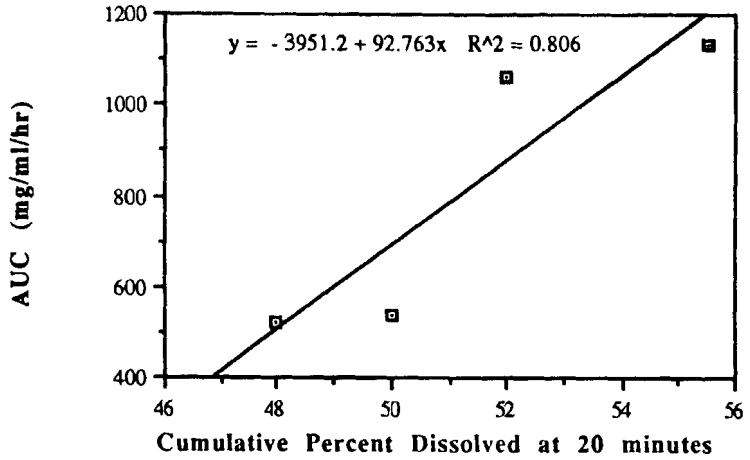

Figure 1. Diazepam - AUC vs. Cumulative Percent Released at 20 minutes, Dissolution 3

and all involve fewer than 7 observations. One of these correlations is illustrated in Figure 1. Most other correlations are based upon the use or more than one independent variable thereby complicating the manner in which they can be illustrated. Another problem encountered in the graphical representation of these correlations is the fact that most ratios of test to reference values are so close to unity that, in order to be distinguished from one another, they must be carried out to four significant figures . Unfortunately, most graphics packages only read data to a level of three significant figures.

It is difficult to determine whether the correlations found represent a true functional relationship or are simply due to the large number of correlations evaluated. We suspect that the latter version is more probable. In Table 4, the likelihood of these correlations merely being a function of probability is further exemplified. Here one can see that only 18 out of the total 402 correlations evaluated (4.5%) showed any promise.

Listed in Table 5 are all Ttest results as well as the mean Coefficient of Variation (C.V.) for each group tested. The most remarkable differences are those between the type of dissolution testing used for each particular product (see Figures 2, 3 and 4). Thus, it

Table 4. Number of *In Vivo - In Vitro* Correlations Found among the Total Number Investigated

Drug	Type of Parameters Evaluated	Number of ANDAs Evaluated	Total Number of Correlations Evaluated	Number of Correlations Found to Exist
Diazepam	All	16	96	2
	Test/Reference		72	3
Doxepin HCl	All	16	18	0
Ibuprofen	All	50	54	2
	Test/Reference		54	3
Lorazepam	All	17	54	2
	Test/Reference		54	6
TOTAL		**99**	**402**	**18**

Table 5. Ttest Results

Drug	Test Groups	Results	p-value	Mean C.V. (%)
Diazepam	Reference vs. Sample, Bioavailability	N.S.	N.S.	30.6
	Reference vs. Sample, Dissolution [a] 1	N.S.	N.S.	12.6
	Reference vs. Sample, Dissolution 2	N.S.	N.S.	13.8
	Reference vs. Sample, Dissolution 3	N.S.	N.S.	14.3
	Dissolution 1 vs. Dissolution 2	[b] 1 > 2	< 0.001	14.2
	Dissolution 1 vs. Dissolution 3	[b] 1 > 3	< 0.004	14.6
	Dissolution 2 vs. Dissolution 3	N.S.	N.S.	15.8
Doxepin HCl	Reference vs. Sample, Bioavailability	N.S.	N.S.	47.5
	Reference vs. Sample, Dissolution 4	N.S.	N.S.	9.8
Ibuprofen	Fasting vs. Non-Fasting, Bioavailability	[c] fast > non-fast	< 0.0001	19.5
	Sugar- vs. Film-Coating, Bioavailability	N.S.	N.S.	16.7
	Sugar- vs. Film-Coating, Dissolution 6	[d] film > sugar	< 0.07	44.2
	Reference vs. Sample, Bioavailability	N.S.	N.S.	20.7
	Reference vs. Sample, Dissolution 5	N.S.	N.S.	4.7
	Reference vs. Sample, Dissolution 6	N.S.	N.S.	14.9
	Dissolution 5 vs. Dissolution 6	[d] 5 > 6	< 0.02	10.5

Table 5. Ttest Results (continued)

Drug	Test Groups	Results	p-value	Mean C.V. (%)
Lorazepam	Reference vs. Sample, Bioavailability	N.S.	N.S.	23.3
	Reference vs. Sample, Dissolution 7	N.S.	N.S.	2.5
	Reference vs. Sample, Dissolution 8	N.S.	N.S.	7.2
	Dissolution 7 vs. Dissolution 8	7 > 8	< 0.03	6.9

a see Table 2
b All points except D_{45}
c C_{max} only
d All points except D_{20}

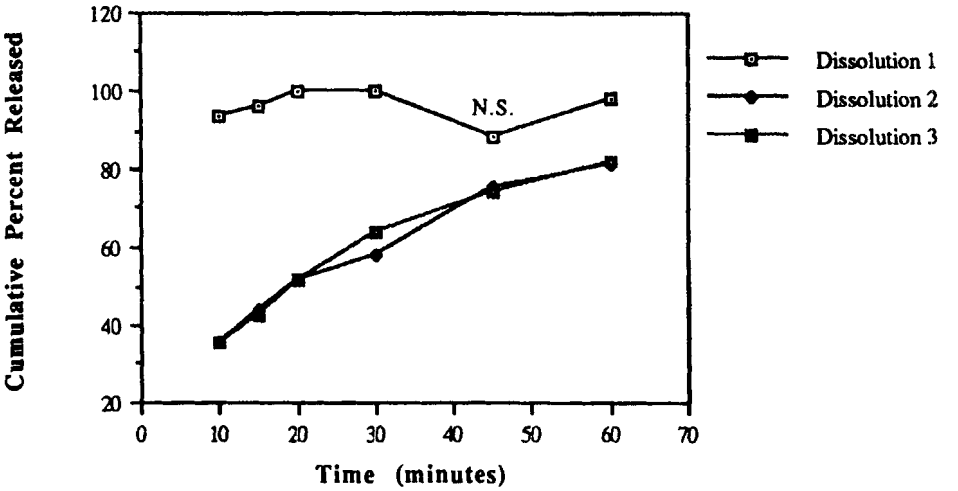

Figure 2. Diazepam Mean Dissolution Profiles

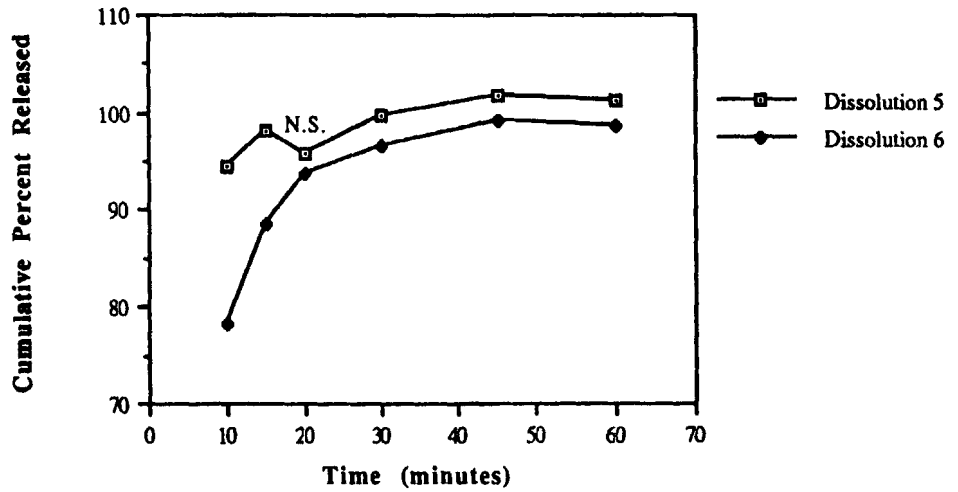

Figure 3. Ibuprofen Mean Dissolution Profiles

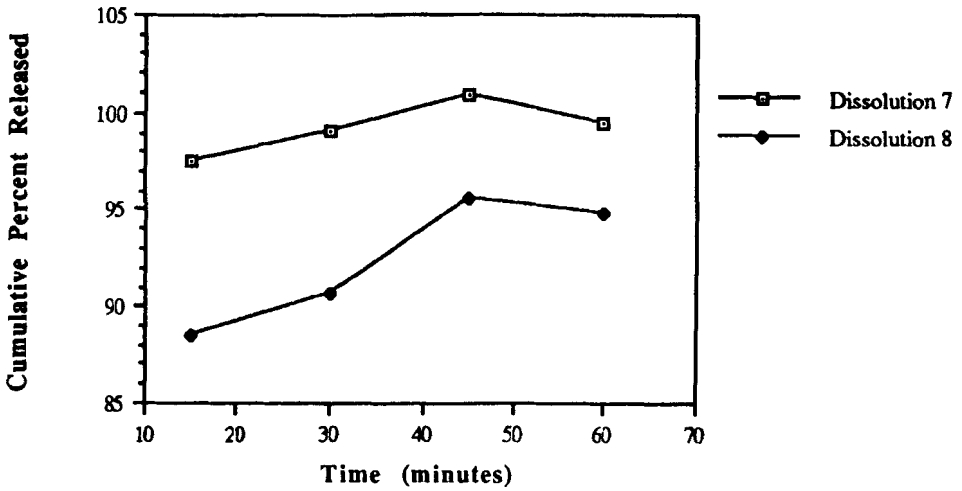

Figure 4. Lorazepam Mean Dissolution Profiles

can be postulated that correlations found using one type of dissolution test will not necessarily apply to an alternate type of dissolution test. Indeed, there has been an ongoing debate between the FDA and the United States Pharmacopeial (USP) in regards to which type of dissolution testing (type 5 being the official USP method and type 6 the method recommended by the FDA) is more meaningful to ibuprofen's bioavailability.

In particular, some have found that the conditions recommended by the FDA for the dissolution of ibuprofen tablets appear to be much more discriminating for sugar-coated tablets than film-coated tablets (19,20). However, these differences were not found to exist among corresponding bioavailability profiles (19). Thus, it may be concluded that this particular dissolution test may possibly be overly discriminating and in fact, have little bearing upon the product's bioavailability. The results from this study seem to confirm these findings.

In terms of bioavailability, significant differences were found only when ibuprofen tablets were administered under fasting and non-fasting conditions, As expected, fasting prior to administration results in higher peak blood levels. However, AUC values did not

differ and thus the bioavailability remained the same. With the exception of the dissolution testing of sugar-coated ibuprofen tablets, the bioavailability of the products studied is more variable than their dissolution. In most instances, the variation of the products' dissolution was less than 15% with the bioavailability being greater than 20%.

Conclusions

This investigation, using what is believed to be one of the largest bodies of bioavailability data available, has been depressing, in terms of potential, in allowing regulatory agencies to dispense with *in vivo* bioequivalency testing using existing official dissolution test methods. We believe that our lack of success can be attributed to several reasons. Firstly, manufacturers rarely, if ever, submit poor bioequivalence data to FDA. Thus the range of *in vitro* and *in vivo* test data is quite limited in magnitutde. This makes the definition of any functional relationship very difficult. Secondly, the lack of complete uniformity in the bioavailability protocols reduces the types of comparisons which can be made.

Thirdly, different batches of reference product were used. However, the dissolution test data for all reference products (obtained using the same type of dissolution test equipment) showed relatively little variability. This is a positive finding since it indicates that batch-to-batch variability of reference product, for the four drugs in this study, was low. Also, it implies that different analysts, at different locations, are capable of obtaining dissolution results which are quantitatively quite similar. Apparently, the propaganda concerning the use of USP calibrators has been effective.

Fourthly, in some cases, dissolution data was limited to one time point. Therefore, we were unable to evaluate the full dissolution profile and application of such methods as statistical moments and convolution/deconvolution were limited. Also, in some instances, dissolution was virtually instantaneous with almost one hundred percent of drug being released by the first sample time. Confounding this issue is that some dissolution test data

were reported to be in excess of one hundred percent (i.e. content uniformity variations are superimposed on dissolution).

Thus, at the moment, there is no justification for relaxing requirements for bioequivalency testing. However, we do suggest that use of the flow-through cell apparatus, which has already attracted interest at USP and FDA, may allow greater success to be attained in elucidating *in vitro/in vivo* correlations (6,7,9,12-14). To some extent, we feel that prolonged release products may be an especially fruitful area for this type of study since the dissolution profiles of such products are, of their very nature, much more amenable to such techniques as deconvolution.

Acknowledgements

We thank FDA for providing MKZK a COSTEP appointment which allowed her to pursue this project. We also thank Applied Analytical Industries for providing a graduate student fellowship.

References

1. Vaughan DP, Leach RH. Simple transformation method for predicting plasma drug profiles from dissolution rates, *J Pharm Sci* 65(4):601-603 (1976).

2. Smolen VF, Erb RJ. Predictive conversion of *in vitro* drug dissolution data into *in vivo* drug response versus time profiles exemplified for plasma levels of warfarin, J Pharm Sci 66(3):297-304 (1977).

3. Mislap RL et al. Comparison of two dissolution apparatuses with correlation of *in vitro - in vivo* data for prednisone and prednisolone tablets, *Biopharm Drug Disp* 1:3-17 (1979).

4. Stavchansky S et al. Correlation of *in vivo* bioavailability of erythromycin stearate tablets with *in vitro* tests, *J Pharm Sci* 69(11):1307-1310 (1980).

5. Banakar UV, Block LH. Beyond Bioavailability testing, *Pharm Tech* 7(9);107, 109,112,115,117 (1983).

6. Bradbrook ID et al. Pharmacokinetic investigation of the absorption of oxprenolol from Oros delivery system in healthy volunteers: comparison of *in vivo* and *in vitro* drug release, *Br J Clin Pharmac* 19:163S-169S (1985).

7. Lagenbucher F, Mysicka J. *In vitro* and *in vivo* deconvolution assessment of drug release kinetics from oxprenolol Oros preparations, *Br J Clin Pharm* 19:151S-162S (1985).

8. El-Yazigi A, Sawchuk RJ. *In vitro - in vivo* correlation and dissolution studies with oral theophylline dosage forms, *J Pharm Sci* 74(2):161-164 (1985).

9. Moller H. Biopharmaceutical assessment of modified release oral dosage forms, *Pharm Ind* 43(5):514-519 (1986).

10. Skelley JP et al. *In vitro* topographical characterization as a predictor of *in vivo* controlled release quinidine gluconate bioavailability, *Drug Dev Ind Pharm* 12(8&9):1177-1201 (1986).

11. Block LH, Banakar UV. Further considerations in correlating *in vitro - in vivo* data employing mean-time concept based on statistical moments, *Drug Dev Ind Pharm* 14(15-17):2143-2150 (1988).

12. Phillips JG, Chen Y, Wakeling IN. A flow-through dissolution approach to *in vitro/in vivo* correlation of adinazolam release from sustained release formulations, *Drug Dev Ind Pharm* 15(14-16):2177-2195 (1989).

13. Lin S-Y, Kao Y-H, Chang H-N. Preliminary evaluation of the correlation between *in vitro* release and *in vivo* bioavailability of two aminophylline slow-release tablets, *J Pharm Sci* 79(4):326-330 (1990).

14. Abdou HM. **Dissolution, Bioavailability and Bioequivalence**, Easton, PA: Mack Publishing Co. (1989) pp. 491-596.

15. **SAS® User's Guide: Basics**, Version 5 Edition, Cary, NC: SAS Institutes (1985) pp.861-864.

16. **SAS® User's Guide: Statistics**, Version 5 Edition, Cary, NC: SAS Institutes (1985) pp. 711-724.

17. *ibid* pp. 655-709.

18. *ibid* pp. 795-800.

19. Romero AJ, Grady LT, Rhodes CT. Dissolution testing of ibuprofen tablets, *Drug Dev Ind Pharm* 14(11):1499- (1988).

20. Dash BH et al. Ibuprofen tablets: dissolution versus bioavailability, Drug *Dev Ind Pharm* 14(11):1629-1645 (1988).

Appendix A. Equations

$$r = \sum_{i=1}^{n} (x_i - \bar{x})(y_i - \bar{y}) / \sqrt{\sum_{i=1}^{n} (x_i - \bar{x})^2 (y_i - \bar{y})^2} \quad \text{(Eq. 1)}$$

$$SSE = \sum_{i=1}^{n} (y_i - x_i b)^2 \quad \text{(Eq. 2)}$$

$$r^2 = 1 - \frac{SSE}{TSS_i} \quad \text{(Eq. 3)}$$

$$t_e = (\bar{x_1} - \bar{x_2}) / \sqrt{s^2 \left(\frac{1}{n_1} + \frac{1}{n_2}\right)} \quad \text{(Eq. 4)}$$

$$s^2 = \left\{(n_1 - 1)s_1^2 + (n_2 - 1)s_2^2\right\} / (n_1 + n_2 - 2) \quad \text{(Eq. 5)}$$

$$t_u = (\bar{x_1} - \bar{x_2}) / \sqrt{\frac{s_1^2}{n_1} + \frac{s_2^2}{n_2}} \quad \text{(Eq. 6)}$$

$$F' = \left\{\text{Larger of } s_1^2 \text{ and } s_2^2\right\} / \left\{\text{Smaller of } s_1^2 \text{ and } s_2^2\right\} \quad \text{(Eq. 7)}$$

Appendix B. Abbreviations

ANDA	=	Abbreviated New Drug Application
AUC_{0-t}	=	Area Under the Plasma Concentration vs. Time Curve to time t
$AUC_{0-\infty}$	=	Area Under the Plasma Concentration vs. Time Curve to Infinity
b_0	=	y-interecept
b_i	=	Regression Coefficient Corresponding to each x_i
C_{max}	=	Peak Plasma Concentration
C.V.	=	Coefficient of Variation
D_t	=	Cumulative Percent Dissolved at Time t
FDA	=	Federal Food and Drug Administration
N.S.	=	Not Significantly Different
n	=	Number of Observations
r	=	Pearson Product Moment Coefficient of Correlation
r^2	=	Coefficient of Determination
$r\,AUC_{0-t}$	=	Test AUC_{0-t} divided by Reference AUC_{0-t}
$r\,AUC_{0-\infty}$	=	Test $AUC_{0-\infty}$ divided by Reference $AUC_{0-\infty}$
$r\ C_{max}$	=	Test C_{max} divided by Reference C_{max}
$r\,D_t$	=	Test D_t divided by Reference D_t
SSE	=	Error of Sum of Squares
s^2	=	Pooled Variance
s_i^2	=	Variance of Group i
TSS_i	=	Total Sum of Squares corrected for Mean of Dependent Variable
t_e	=	Value of t if Variances are Equal
t_u	=	Value of t if Variances are Unequal
USP	=	United States Pharmacopeia