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Review Article

Biopharmaceutical Classification System in *In-vitro*/*In-vivo* Correlation: Concept and Development Strategies in Drug Delivery

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Abstract

Drug development is a very laborious and expensive process. One of the major reasons for failure during the clinical phases of drug development is inadequate pharmacokinetic data on the drug candidate. Therefore, it would be advantageous if the pharmacokinetic properties of drug candidates be predicted beforehand. One major obstacle in making such predictions is the inability to appropriately scale the in-vitro data to the in-vivo situation. Results from in-vitro in-vivo correlation (IVIVC) studies have been used to select the appropriate excipients and optimize the manufacturing processes for quality control purposes, and for characterizing the release patterns of newly formulated immediate release, and modified-release products relative to the references. In recent years, the concept and application of the IVIVC for pharmaceutical dosage forms have been a major focus of attention in the pharmaceutical industry, academia and regulatory agencies. Hence, this article highlights the importance of appropriate selection of IVIVC level with respect to the Biopharmaceutical Classification System (BCS) and also covers examples of BCS-based IVIVCs of drugs/products with different types of release profiles.

Keywords: *In vitro/In vivo correlation, Biopharmaceutical Classification System, Drug Delivery.*

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INTRODUCTION

In recent years, the concept and application of the *in vitro/in vivo* correlation (IVIVC) for pharmaceutical dosage forms have been a main focus of the pharmaceutical industry, academia and regulatory bodies [1].

A regulatory guidance for both immediate (IR) and modified release (MR) dosage forms has, therefore, been developed by United States Food and Drug Administration (FDA) to minimize the need for bioavailability studies as part of formulation design and optimization [2]. *In vitro* dissolution testing of solid oral dosage forms serves as a very important tool in drug development for selecting and optimizing formulations, studying drug-release mechanisms, ensuring batch-to-batch consistency, monitoring stability and demonstrating bioequivalence. Additionally, for the development of modified release (MR) dosage form. the focus of dissolution testing in the early stage of drug development is to provide predictive estimates of drug release in respect to *in vivo* performance of a drug product [3]. Thus no costly *in vivo* bioequivalence testing is required [4].

This review essentially addresses FDA guidelines for development, evaluation, and application of *in vitro/in vivo* correlations and highlights the importance of appropriate selection of IVIVC level with respect to Biopharmaceutical Classification System (BCS). The review also looks at examples of BCS based IVIVCs of drugs/products with different type of release profiles.

IN VITRO/IN VIVO CORRELATIONS

IVIVC has been defined by the FDA as "a predictive mathematical model describing the relationship between an *in-vitro* property of a dosage form and an *in-vivo* response" [4]. Generally; the *in-vitro* property is the rate or extent of drug dissolution or release while the *in-vivo* response is the plasma drug concentration or amount of drug absorbed. The United States Pharmacopoeia (USP) also defines IVIVC as "the establishment of a relationship between a biological property, and a parameter derived from a biological property produced from a dosage form, and a physicochemical property of the same dosage form" [5].

Developing the correlation

The basic requirements for developing IVIVC are:

- Data obtained from human studies are required for regulatory consideration of the correlation
- Two or more drug product formulations with different release rates are developed and their *in vitro* dissolution profiles generated using an appropriate dissolution method
- Usage of same dissolution method for all the formulations, and
- Plasma concentration data from a bioavailability study for each of the formulations [6].

Categories of *in vitro/in vivo* correlations

Level A

Level A correlation is usually estimated by a two-stage procedure: deconvolution [7] followed by comparison of the fraction of drug absorbed to the fraction of drug dissolved. Correlation of this type is generally linear. Model Level A correlation is depicted in Figure 1.

Level B

Level B uses the principles of statistical moment analysis. The mean *in vitro* dissolution time (MDT) is compared either to the mean residence time (MRT) or to the mean *in vivo* dissolution time (Figure 2). [7]

Level C

Level C establishes a single point relationship between a dissolution parameter, for example, $t_{50\%}$, percent dissolved in 4 h and a

pharmacokinetic parameter, e.g., AUC, C_{max} and T_{max} (see Figure 3).

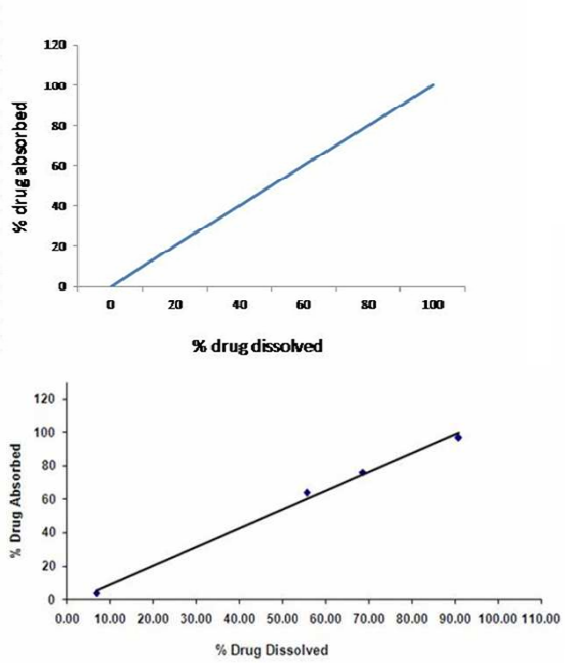

Figure 1: Model Level A IVIVC [7]

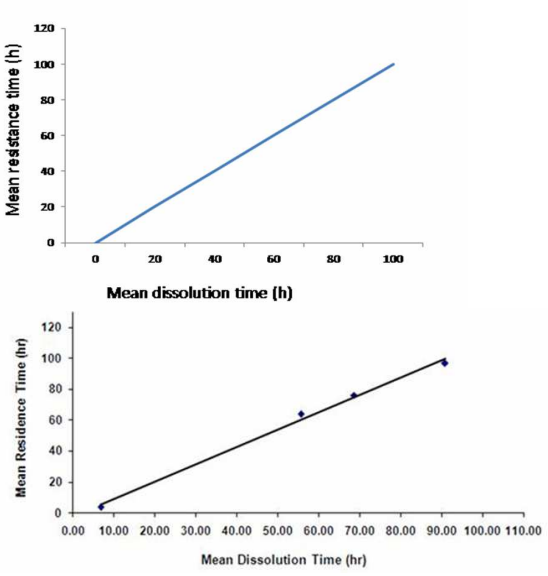

Figure 2: Model Level B for IVIVC[7]

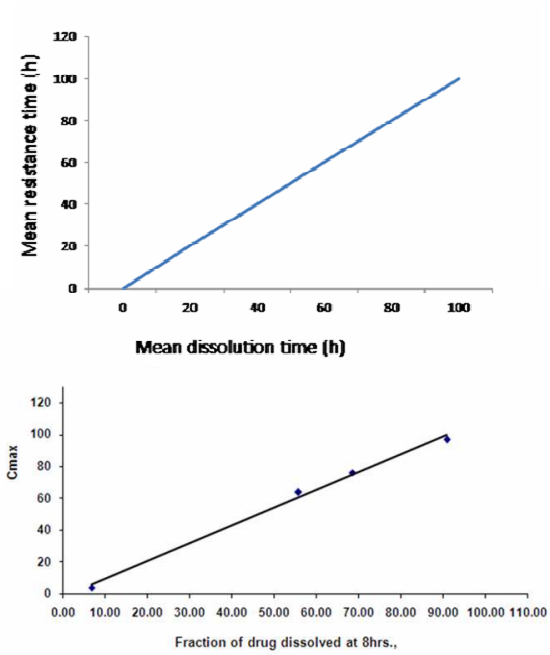

Figure 3: Model Level C for IVIVC between C $_{max}$ and percent drug dissolved at 8 hours [7]

Multiple Level C

A multiple Level C correlation relates one or several pharmacokinetic parameters of interest to the amount of drug dissolved at several time points of the dissolution profile. Various parameters used in IVIVC depends on the level, as depicted in Table1 [7].

Table 1: Various parameters used in IVIVC depending on the level [7]

Level	*In vitro*	*In vivo*
A	Dissolution curve	Input (absorption) curves
B	Statistical moments: MDT	Statistical moments: MRT, MAT, etc
C	Disintegration time, time to have 10, 50, 90% dissolved, dissolution rate and dissolution efficiency	C_{max}, T_{max}, Ka, time to have 10, 50, 90% absorbed and AUC (total or cumulative)

Key: *MDT = mean dissolution time, MRT = mean residence time, MAT = mean absorption time*

IVIVC models

The most basic IVIVC models are expressed as a simple linear equation (Eq 1) between *in vivo* drug absorption and *in vitro* drug dissolved (released).

$Y = mX + C$ …… (1)

where Y is the *in vivo* absorbed and X the *in vitro* drug dissolved, m the slope of the relationship and Cthe intercept. Ideally, m = 1 and C = 0, indicating a linear relationship. Eq 1 may be applied to most formulations with comparable *in vitro* and *in vivo* profiles. However, for dosage forms with complicated mechanisms of release, which are usually of longer duration, *in vitro* release may not be on the same time scale as the *in vivo* release. Thus, in order to model such data, it is necessary to incorporate time-shifting and time-scaling parameters within the model.[9].

Model validation

Validation is an important tool as it provides the confidence to accept a mathematical model. Here, IVIVC model validation can successfully predict the outcome (*in vivo* profile) with a given model and test condition (*in vitro* profile). Internal validation [10] serves the purpose of providing the basis for the acceptability of the model, while external validation [10] is superior and affords greater "confidence" in the model.

Internal validation

Using the IVIVC model, for each formulation, the relevant exposure parameters (C_{max} and AUC) are predicted and compared to the actual (observed) values. The prediction errors are calculated using Eq 3. The criteria set in FDA guidlines on IVIVC are as follows: For C_{max} and AUC, the mean absolute percent prediction error (% PE) should not exceed 10 %, and the prediction error for individual formulations should not exceed 15 % [10].

Prediction = [(C_{max} observed – C_{max} predicted) / C_{max} observed]*100 ……. ….. (2)

Error (%PE) = [(AUC observed – AUC predicted) / AUC observed]*100 ……. (3)

External validation [10]

For establishing external predictability, the exposure parameters for a new formulation are predicted using its *in vitro* dissolution profile and the IVIVC model whereby the predicted parameters are compared to the observed parameters. The prediction errors are computed as for the internal validation. For C_{max} and AUC, the prediction error for the external validation formulation should not exceed 10 %[11] A prediction error of 10 to 20 % indicates inconclusive predictability and illustrates the need for further study using additional data sets. For drugs with narrow therapeutic index (TI), external validation is required despite acceptable internal validation, whereas internal validation is usually sufficient for drugs with wide TI.

BIOPHARMACEUTICAL CLASSIFICATION SYSTEM

The biopharmaceutical classification system (BCS) is a scientific framework for classifying a drug substance based on its aqueous solubility and intestinal permeability.

Characteristics of drugs in BCS [5]

- Class I drugs exhibit a high absorption number and a high dissolution number.
- Class II drugs have a high absorption number but a low dissolution number.
- Class III drugs exhibit a high variation in the rate and extent of drug absorption. Permeability is rate limiting step for drug absorption.
- Class IV drugs exhibit poor and variable bioavailability.

Estimation of IVIVC using the biopharmaceutical drug classification system

BCS is a fundamental guideline for determining the conditions under which *in-vitro/in-vivo* correlations are expected. It is also used as a tool for developing *in-vitro* dissolution specification [8]. *In vitro-in vivo* correlation is normally expected for highly permeable drugs or drugs under dissolution rate-limiting conditions. This statement is supported by the biopharmaceutical classification system, which anticipates successful IVIVC for highly permeable drugs [9]. Biopharmaceutical classification system and expected IVIVC for immediate release (IR) and extended release (ER) drug products are summarized in Tables 2 and 3, respectively.

Highlights from some published works on IVIVCs in the context of BCS Classes I - IV are summarized in Table 4 - 6, respectively.

Table 2: BCS and expected IVIVC for immediate release drug products [27]

Class	Solubility	Permeability	Absorption	Expected IVIVC
1	High	High	Gastric emptying controlled	IVIVC is expected if dissolution rate is slower than gastric emptying rate, otherwise limited or no correlation
2	Low	High	Dissolution controlled	IVIVC is expected if in vitro dissolution rate is similar to in vivo dissolution rate, unless dose is very high
3	High	Low	Dissolution independent	Absorption (permeability) is rate determining; limited or no IVIVC with dissolution rate
4	Low	Low	Case-by-case evaluation	Limited or no IVIVC is expected

Table 3: BCS and expected IVIVC for extended-release drug products [6]

Class	Solubility	Permeability	Expected IVIVC
Ia	High & Site Independent	High & Site Independent	Correlation (if dissolution is rate-limiting step)
Ib	High & Site Independent	Dependent on site & Narrow absorption window	IVIVC expected
IIa	Low & Site Independent	High & Site Independent	Variable
IIb	Low & Site Independent	Dependent on site & Narrow absorption window	Little or no IVIVC
III	High Solubility	Low Permeability	Little or no IVIVC
IV	Low Solubility	Low Permeability	Little or no IVIVC
Va[a]	Variable	Variable	Little or no IVIVC
Vb[a]	Variable	Variable	IVIVC Level A expected

[a]*Drugs that have variable solubility and variable permeability are classified as Class V. Class Va includes acidic drugs and Class Vb includes basic drugs*

Table 4: Some IVIVC methods used for BCS Class I drugs

Drug	*In-vitro* evaluation	*In-vivo* evaluation	Level of IVIVC	Reference
Salbutamol Sulfate	f_2 test	Wagner-Nelson method	Level A	[12]
Buspirone hydrochloride ER Tablets	f_2 test	Wagner-Nelson method	Level A	[13]
Metoprolol ER	f_2 test	Wagner-Nelson method	Level A	[14]
Metoprolol IR			Level B	
Naproxen IR			Level C	
Niacin	Dissolution method	Wagner-Nelson Method	Level A	[15]
Salbutamol Sulphate	Dissolution method	Wagner-Nelson Method	Level A	[16]
Metoprolol Tartrate	f_2 test	Wagner-Nelson Method	Level A	[17]
Theophylline	f_2 test	Wagner-Nelson method	Level A	[18]
(S) Nicotine	Model dependent approaches	Metabolic Clearance Equation	Michaelis-Menten kinetic	[19]
Metoprolol	f_2 test	Wagner-Nelson method	A correlation between pooled FRD and pooled FRA	[20]
Tretinoin	Dissolution method	Pharmacodynamic and Dermatopharmacokinetic (DPK) method	Correlation	[21]
Theophylline, Aminophylline	f_2 test	Wagner-Nelson method	Correlation coefficient	[22]

Key: *FRD = fraction of drug dissolved; FRA = fraction of drug absorbed*

Biopharmaceutical Classification System (BCS) represents a criterion for the classification of drugs on the basis of their solubility and permeability. As per literature, several IVIVCs methodologies were followed according to the biopharmaceutical classification system for different drugs/products.

To investigate the BCS class I drugs, (salbutamol, buspirone, metoprolol, naproxen, niacin, thyroxin, theophylline and nicotine) the evaluation of *in-vitro* profile demonstrated by comparison of dissolution profile, model-dependent approaches and similarity factor (f_2-test) method. Wagner-Nelson deconvolution, convolution and dermatopharmacokinetic (DPK) methods were used for *in-vivo* evaluation of BCS class I drugs/products. Regarding the level of IVIVC, point-point correlation (level-A), followed by multiple level-C and Michaelis-Menten kinetics were used. Level A results revealed good correlations between *in vitro* drug release and *in vivo* drug absorption. Level B IVIVC also exhibited a strong linear relationship between mean residence time (MRT) and mean dissolution time (MDT).

Table 5: Some IVIVCs methods used for BCS Class II drugs

Drug	*In-vitro* evaluation	*In-vivo* evaluation	Level of IVIVC	Reference
Danazol	Flow-through dissolution test	Deconvolution	Level A	[23]
Albendazole, Danazol, Ketoconazole, Atovaquone	Noyes–Whitney equation	Not available	Correlation	[24]
Nifedipine	Two-phase dissolution test system	Wagner-Nelson method and Numerical Deconvolution	Level-A	[25]
Carbamazepine	Dissolution method	Wagner-Nelson method	Level-C	[26]
Diclofenac Sodium	f_2 test	Non-compartmental method and Student's paired *t*-test	Level-A	[27]
Diclofenac Sodium	Dissolution method	Non-Compartmental method	Correlation	[28]
Carbamazepine	Dissolution method	In vivo dissolution	Deconvolution	[29]
Piroxicam	Dissolution method	Non-compartmental method	Correlation	[30]
Phenoxymethyl-penicillin-potassium, Glimepiride, Levofloxacin	Flow-through cell dissolution method	Numerical deconvolution	Correlation	[31]
Nimesulide	Dissolution method	Non-compartmental method	Correlation	[32]
Griseofulvin	Dissolution method	Non-compartmental method	Correlation	[33]

Table 6: Some IVIVCs methods used for BCS Class III drugs

Drug	*In vitro* evaluation	*In vivo* evaluation	Level of IVIVC	Reference
Paracetamol and Carbopol	Dissolution method	Convolution and deconvultion	Level-A	[34]
Acetaminophen	Dissolution method	Non-compartmental method	Level-A	[35]
Roxatidine	f_2 test	Numerical deconvultion	Level-A	[36]
Levothyroxine	f_2 test	Correlation coefficient	Multiple Level C	[37]
Pseudoephedrine sulfate	Dissolution method	Wagner-Nelson method	Levy and Hollister	[38]
Metformin	f_2 test	Non-compartmental method	Correlation	[39]

Level C IVIVC did not show a perfect correlation, which may be due to single point correlation [40].

In the case of BCS class II drugs (danazol, nifedipine, carbamazepine, piroxicam, nimesulide and griseofulvin), *in vitro* evaluation includes, flow-through dissolution test, application of Noyes-Whitney equation and comparison of dissolution profiles. For *in vivo* evaluation deconvolution, Wagner-Nelson and non-compartmental analysis methods were successfully to determine *in vivo* performance. The *in vitro* results, compared with the *in vivo* data by means of level-A followed by level-C, correlated excellently for poorly soluble drugs/products. [41].

For BCS class III drugs (paracetamol, roxatidine, levothyroxine, pseudoephedrine, clonazepam and metformin), Similarity factor (f_2 value) [42] and comparison of dissolution profile were assessed to evaluate *in-vitro* drug release profile. *In vivo* evaluation was performed using numerical deconvolution, non-compartmental analysis and Wagner-Nelson methods [43]. On the basis of correlation, level-A, followed by multiple level-C demonstrate reliable and robust estimate of drug product performance.

In-vitro evaluation of BCS Class IV drugs, such as lopinavir and remoxipride, was predicted by comparison of dissolution profile as well as by f-test. For *in-vivo* evaluation, Wagner-Nelson method and level A correlation exhibited perfect correlation between *in-vitro* and *in-vivo* comparison.

APPLICATIONS OF IVIVC IN DRUG DELIVERY

A main objective of developing and evaluating IVIVC is to establish the dissolution test as a surrogate for human bioequivalence studies. However, for the applications outlined below, the adequacy of the *in vitro* dissolution method to act as a surrogate for *in vivo* testing should be shown

through an IVIVC for which predictability has been established [41].

Early stages of drug delivery technology development: Proof-of-concept

The selection of a drug candidate marks the most crucial stage in the life cycle of drug development. During this stage, exploring the relationship between *in vitro* and *in vivo* properties of the drug in animal models provides an idea of the feasibility of the drug delivery system for a given drug.

Future biowaivers

Frequently, drug development requires changes in formulations due to a variety of reasons, such as unexpected problems in stability, development, availability of better materials, better processing results, etc. Having an established IVIVC can help avoid bioequivalence studies by using the dissolution profile from the changed formulation and subsequently predicting the *in vivo* concentration-time profile. This predicted profile could act as a surrogate of *in vivo* bioequivalence study.

IVIVC and parenteral drug delivery

IVIVC can be developed and applied to parenteral dosage forms, such as controlled-release particulate systems, implants, etc, that are either injected or implanted. However, there are relatively fewer successes in the development of IVIVC for such dosage forms.

Formulation assessment: *in vitro* dissolution

A suitable dissolution method that is capable of distinguishing the performance of formulations with different release rates *in vitro* and *in vivo* is an important tool in product development. IVIVC facilitates the process of such method development [41].

Dissolution specifications

The process of setting dissolution specifications in the presence of an IVIVC starts by obtaining the reference (pivotal clinical batch) dissolution profile. Specifications should optimally be established such that all batches with dissolution profiles between the fastest and slowest batches are bioequivalent and less optimally bioequivalent to the reference batch [42].

Increased development of modified release dosage forms necessitates investigating the broader aspects of *in vitro in vivo* correlation (IVIVC).

CONCLUDING REMARKS

Based on data from the literature, it is evident that current IVIVC studies have focused more on the development and validation of level A IVIVC which gives more useful information on the relationship between *in vitro* release and *in vivo* absorption from dosage form. Levels B and C IVIVCs have been evaluated for several purposes in formulation development, for example, to select the appropriate excipients and optimize the manufacturing processes, for quality control purposes and for characterizing the release patterns of newly formulated immediate release and modified-release products relative to the references. Present regulatory guidelines for IVIVC is only applicable to oral conventional and modified release dosage forms; however, further research is necessary to develop IVIVCs for non-oral products, inhaled medicines and dermatological medicaments. Also, it is possible that the IVIVC can still be explored to provide a greater understanding of the factors influencing clinical quality.

REFERENCES

1. *Eva MD, Amo AT, Heikkinen JM. In vitro–in vivo correlation in p-glycoprotein mediated transport in intestinal absorption. Eur. J. Pharm. Sci. 2009; 36: 200-211*

2. Jaber E. In vitro - In vivo Correlation: From Theory to Applications. J Pharm Pharmaceut Sci, 2006; 9: 169-189
3. Ulrich H, Florian S, Rolf D. Predicting in vivo absorption behavior of oral modified release dosage forms containing pH-dependent poorly soluble drugs using a novel pH-adjusted biphasic in vitro dissolution test. Eur. J. Pharm. Biopharm. 2010; 76 (1): 105-111
4. Tom J, Stefaan R, Adrian DG. Combined Models for Data from In Vitro-In Vivo Correlation Experiments. J. Biopharm. Stat. 2008; 18: 1197-1211
5. Mohd Y, Mohd A, Ashwani K. Biopharmaceutical Classification System: An Account. International J Pharm Tech Research. 2010; 2: 1681-1690
6. Venkata R, Uppoor S. Regulatory perspectives on in vitro (dissolution) / in vivo (bioavailability) correlations. J. Control. Rel. 2001; 72: 127-132
7. Saeed AQ. In Vitro-In Vivo Correlation (IVIVC) and Determining Drug Concentrations in Blood from Dissolution Testing – A Simple and Practical Approach. Open Drug Deliv J, 2010; 4, 38-47
8. Cardot JM, Beyssac E, Alric M. In Vitro–In Vivo Correlation: Importance of Dissolution in IVIVC. Dissolution Technologies. 2007; 7: 15-19
9. Buchwald P. Direct differential-equation-based in-vitro–in-vivo correlation (IVIVC) method. J Pharm Pharmacol, 2003; 55 (4): 495–504
10. Ewout WS, Sacha EB, Henriëtte AM, Diederick EG, Karel GM. Internal and external validation of predictive models: A simulation study of bias and precision in small samples J Clin Epidemiol, 2003; 56(5): 441-447
11. Nattee S, Natalie DE. In-Vitro-In-Vivo Correlation Definitions and Regulatory Guidance. Int. J. of Generic Drugs. ISSN 0793 7776: 1-11
12. Esra D, Levent Ö. In Vitro - In Vivo Correlations. J. Pharm. Sci. 2003; 28: 215-224
13. Ghulam M, Mahmood A, Naveed A. Biowaiver study of oral tabletted ethylcellulose microcapsules of a BCS class I drug. Bull. Chem. Soc. Ethiop. 2009; 231: 175-186
14. Sevgi T, Adel S, Arthur G. Development and validation of an in vitro–in vivo correlation for buspirone hydrochloride extended release tablets. J. Control. Rel. 2003; 88: 147-157
15. Nattee S, Natalie DE. Systematic Method for the Development and Validation of an IVIVC Metoprolol and Naproxen Drug Examples. Int. J. of Generic Drugs. IVIVC Series Part III, ISSN 0793 7784 Euro.
16. Turner S, Federici C, Hite M. Formulation Development and Human in Vitro-In Vivo Correlation for a Novel, Monolithic Controlled-Release Matrix System of High Load and Highly Water-Soluble Drug Niacin. Drug Dev. Ind. Pharm. 2004; 30: 797-807
17. Fotaki N, Aivaliotis A, Butler J. A comparative study of different release apparatus in generating in vitro–in vivo correlations for extended release formulations. Eur. J. Pharm. Biopharm. 2009; 73: 115-120
18. Fatima R, Mahmood A, Ghulam M. Metoprolol Tartrate-Ethylcellulose Tabletted Microparticles: Development of a Validated In-vitro In-vivo Correlation. IJMMS 2010; 1: 4211-4214
19. Umeruqia T, Nisar-UR. Comparative bioavailability and in vitro in vivo correlation of two sustained release brands of theophylline: tablets and pellets. Pak. J. Pharm. Sci. 2008; 211: 131-138
20. Clifford E B, Sang BP, Steven AW. In Vitro-In Vivo Correlations of Human-Nicotine Metabolism. Biochem. Pharmacol. 1995; 50: 565-570
21. Nattee S, Natalie DE. The influence of first pass metabolism on the development and validation of an IVIVC for metoprolol extended release tablets. Eur. J. Pharm. Biopharm. 2002; 53: 301-309
22. Vinod PS. IV–IVC for topically applied preparations–a critical evaluation. Eur. J. Pharm. Biopharm. 2005; 60: 309-314
23. Jaleh V, Taghi G, Ahmad R. Biopharmaceutical characterization of oral theophylline and aminophylline tablets Quantitative correlation between dissolution and bioavailability studies. Eur. J. Pharm. Biopharm. 2000; 50: 301-306
24. Vibeke HS, Betty LP, Henning GK. In vivo - in vitro correlations for a poorly soluble drug, danazol, using the flow-through dissolution method with bio relevant dissolution media. Eur. J. Pharm. Sci. 2005; 24: 305-313
25. Jennifer BD, Christos R. In vitro – in vivo correlations for lipophilic, poorly water-soluble drugs. Eur. J. Pharm. Sci. 11 Suppl. 2000; 2: S 73-S 80
26. John SG, Keith EA, James AR. Studies on dissolution testing of the nifedipine gastrointestinal therapeutic system II. Improved in vitro-in vivo correlation using a two-phase dissolution test. J. Control. Rel. 1997; 489-517
27. Lake OA, Olling M, Barends DM. In vitro/in vivo correlations of dissolution data of carbamazepine immediate release tablets with pharmacokinetic data obtained in healthy volunteers. Eur. J. Pharm. Biopharm. 1999; 48: 13-19
28. Ekarat J, Vincenzo DM, Emanuela R. Application of biorelevant dissolution tests to the prediction of in vivo performance of diclofenac sodium from an oral modified-release pellet dosage form. Eur. J. Pharm. Sci. 2009; 37: 434 - 441
29. Hsiu OH, Cheng HL, Hua ML. The development of matrix tablets for diclofenac sodium based on an empirical in vitro and in vivo correlation. J. Control. Rel. 1997; 49: 149-156
30. Lisbet B, Lars H, Henning GK. A comparison between direct determination of in vivo dissolution and the deconvolution technique in humans. Eur. J. Pharm. Sci.1999; 819-827

31. *Nilufer Y, Aysegul K¸ Yalcın O. Enhanced bioavailability of piroxicam using Gelucire 44/14 and Labrasol: In vitro and in vivo evaluation. Eur. J. Pharm. Biopharm. 2003; 56: 453- 459*
32. *Annke F, Helga M, Ehrenfried W. Biopharmaceutical characterization of oral immediate release drug products. In vitro/in vivo comparison of phenoxy methylpenicillin potassium, glimepiride and levofloxacin. Eur. J. Pharm. Biopharm. 1998; 46: 305-311*
33. *Raguia AS, Iman SA, Rehab N. In vitro and in vivo evaluation of nimesulide lyophilized orally disintegrating tablets. Eur. J. Pharm. Biopharm. 2009;73: 162-171*
34. *Iman SA, Mona HA. In vitro and in vivo evaluation of a fast-disintegrating lyophilized dry emulsion tablet containing griseofulvin. Eur. J. Pharm. Sci 2007; 32: 58-68*
35. *Jelena P, Svetlana I, Zorica D. An investigation into the usefulness of generalized regression neural network analysis in the development of level A in vitro–in vivo correlation. Eur. J. Pharm. Sci 2007; 30: 264-272*
36. *Sabah S, Stephanie B, Eric B. A level 'A' in vitro/in vivo correlation in fasted and fed states using different methods: Applied to solid immediate release oral dosage form. Eur. J. Pharm. Sci 2006; 27: 72-79*
37. *Nabais T, Brouillet F, Kyriacos S. High-amylose carboxy methyl starch matrices for oral sustained drug-release In vitro and in vivo evaluation. J. Control. Rel. 2007; 65: 371-378*
38. *Annke F, Helga M, Ehrenfried W. Biopharmaceutical characterization of oral controlled/modified-release drug products. In vitro/in vivo correlation of roxatidine. Eur. J. Pharm. Biopharm.1998; 46: 313-319*
39. *Nadia MV, Regina LS, Ana PP. Multiple level C in vitro/in vivo correlation of dissolution profiles of two l-thyroxine tablets with pharmacokinetics data obtained from patients treated for hypothyroidism. Eur. J. Pharm. Sci. 2004; 21: 655-660*
40. *Parviz M, Jack R, Winston AV. In-vivo/in-vitro correlation of four extended release formulations of pseudoephedrine Sulfate. J. Pharm. Biomed. Anal. 1997; 15: 439-445*
41. *Lian DH, Yang L, Xing T. Preparation and in vitro/in vivo evaluation of sustained-release metformin hydrochloride pellets. Eur. J. Pharm. Biopharm. 2006; 64: 185-192*
42. *Mukesh CG, Maulik KP. Refinement of Lower Acceptance Value of the Similarity Factor f2 in Comparison of Dissolution Profiles. Dissolution Technologies | 2002; 47: 1-5.*
43. *John GW. Application of the Wagner-Nelson absorption method to the two-compartment open model. J Pharmacokin and Pharmacodyn. 2003; 2 (6): 469-486*
44. *Gangadhar S, Dakshina M, Chilukuri. IVIVC: An Important Tool in the Development of Drug Delivery Systems. Drug delivery technology. 2003; Vol. 3*
45. *Guidance for Industry. Extended Release Oral Dosage Forms. Development, Evaluation, and Application of In Vitro/In Vivo Correlations, U.S. Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research (CDER). September 1997*
46. Guidance for Industry. Dissolution Testing of Immediate, Release Solid Oral Dosage Forms, U.S. Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research (CDER). August 1997.