Journal of
Bioequivalence & Bioavailability
ISSN: 0975-0851

Varinder Kumar et al., J Bioequiv Availab 2013, 5:1
DOI: 10.4172/jbb.1000128

Research Article Open Access

# Application of Assumed IVIVC in Product Life Cycle Management: A Case Study of Trimetazidine Dihydrochloride Extended Release Tablet

**Varinder Kumar*, Lalit Kumar Khurana, Shavej Ahmad and Romi Barat Singh**

*Ranbaxy Laboratories Limited, Gurgaon, Haryana, India*

## Abstract

The purpose of this study was to utilize IVIVC tool in the development of oral controlled release formulation of a BCS class I model drug Trimetazidine dihydrochloride (TMZ). Commercial products of TMZ are taken two to three times a day to achieve therapeutic benefit. Hence, development of once daily tablet was initiated with the development of "Assumed IVIVC". The assumed IVIVC was developed by obtaining *in vivo* data of single dose IR formulation (Vastarel® 20 mg) from literature and generating *in vitro* and *in vivo* data for Preductal® MR 35 mg modified release tablet (Reference) and TMZ extended release tablet 70 mg (Test) in house. *In vitro* dissolution ER tablet was conducted by evaluating effect of pH. The *in vitro* profile as a surrogate to *in vivo* absorption was generated in 0.1 N HCl medium. The *in vivo* absorption was calculated using deconvolution approach using the IR data for unit impulse response. A linear model with a time-scaling factor clarified the relationship between the *in vitro* and the *in vivo* data. The predictability of the final model was consistent based on internal validation. Average percent prediction errors for pharmacokinetic parameters were within ± 10% and individual values for all formulations were within ± 15%. Same model was used as a target to develop OD tablet using software WinNonlin® IVIVC toolkit™ that would be bioequivalent to 35 mg modified release reference product. The assumed IVIVC was then utilized for "Retrospective IVIVC" development and pharmacokinetic parameters of desired formulations were predicted through the IVIVC model. Predicted results for formulation F4 and F5, projected them as the most suitable for once daily use. In this work, it was demonstrated that the IVIVC can be used in the development of new dosage forms to reduce the number of human studies in product life cycle management.

**Keywords**: IVIVC modeling; Assumed IVIVC; Trimetazidine dihydrochloride; Product lifecycle management

## Introduction

Modeling and simulation methods are now commonly used in drug product development and regulatory drug review process. These applications include, but are not limited to: the development of bio-relevant specifications, determination of bioequivalence matrixes for modified release products with rapid therapeutic onset, design of *in vitro-in vivo* correlations in a mechanistic framework, and prediction of food effect [1].

Before the year 2000, the modeling and simulation methods had limited use of identification of biorelevant media for poorly soluble drugs [2,3]. Later, a correlative equation was introduced in the simulation, which correctly defined the relationship between *in vitro* and *in vivo* data. These simulation equations can be generated through traditional concepts such as Wagner Nelson and Loo Riegelman or advanced techniques like Bayesian analysis [4-8].

The simulation calculations are very complex hence software was introduced to simplify the modeling. Some commonly used software are Kinetica, IVIVC toolkit™, Gastroplus™ and STELLA®, etc. Kinetica uses the principal of deconvolution to calculate fraction absorbed [9]. The IVIVC toolkit™ uses deconvolution followed by convolution to carry out predictions of plasma concentration profiles [10]. It does not take into account absorption related factors such as gut metabolism and hepatic metabolism. The Gastroplus™ and STELLA® uses deconvolution combined with Physiological Based PharmacoKinetic (PBPK) modeling [11-14]. These have an inbuilt model to identify absorption related factors. Hence for IVIVC Toolkit™, deconvolution generated through an IR formulation is required to minimize error due to absorption related parameters. Gene expression programming (GEP) optimizes a mathematical expression tree with the help of a genetic algorithm hence takes into account variability due to genetic variation [15].

The IVIVC can be used in the development of new pharmaceuticals dosage form to reduce the number of human studies during the formulation development and for Quality by Design (QbD) based development approach [16]. The main objective of an IVIVC is to provide an *in vitro* profile as a surrogate for *in vivo* bioavailability and to support biowaiver. The IVIVC can also be employed to establish dissolution specifications and to support and/or validate the use of dissolution methods.

U.S. Food and Drug Administration (FDA) defines IVIVC as "a predictive mathematical model describing the relationship between an *in vitro* property of an extended release dosage form (usually the rate or extent of drug dissolution or release) and a relevant *in vivo* response, e.g., plasma drug concentration or amount of drug absorbed)" [17].

There are three main level of IVIVC, based on different parameters of relationship.

### Level A correlation

Point-to-point relationship between *in vitro* dissolution rate and *in vivo* input rate of the drug from the dosage form.

***Corresponding author:** Varinder Kumar, Ranbaxy Laboratories Limited, Gurgaon, Haryana, INDIA, E-mail: varinder.kumar@ranbaxy.com

**Received** November 02, 2012; **Accepted** November 27, 2012; **Published** November 30, 2012

**Citation:** Varinder Kumar, Lalit Kumar K, Shavej A, Romi Barat S (2013) Application of Assumed IVIVC in Product Life Cycle Management: A Case Study of Trimetazidine Dihydrochloride Extended Release Tablet. J Bioequiv Availab 5: 006-015. doi:10.4172/jbb.1000128

### Level B correlation

In this level of correlation, the mean *in vitro* dissolution time ($MDT_{vitro}$) of the product is compared to either mean *in vivo* residence time (MRT) or the mean *in vivo* dissolution time ($MDT_{vivo}$).

### Level C correlation

It represents a single point correlation (one dissolution time point (t50%, t90%, etc.) is compared to one mean pharmacokinetic (PK) parameter such as AUC, $t_{max}$ or $C_{max}$) and does not reflect the entire shape of the plasma drug concentration curve, which is a crucial factor that is a good indicative of the performance of modified-release products.

The IVIVC development for product life cycle management is applied at the start of the project. It is started with establishing a target profile at initial stage of formulation development. Information from various sources is collected and an IVIVC model is developed. This is termed as assumed IVIVC. The information from an assumed IVIVC is used to generate an *in vitro* target profile. Formulations are developed to meet the desired *in vitro* profile. Then, pharmacokinetic study is conducted using the formulation with target *in vitro* profile and a correlation is developed. This is termed as retrospective IVIVC. If a bio-in equivalence is observed, reformulation is carried out, while a bio equivalent study moves to a prospective IVIVC. A prospective IVIVC is establishment of well-defined relationship between the *in vitro* and the *in vivo* profiles [18,19]. A successful use of assumed IVIVC and retrospective IVIVC is very challenging process. A correct interpretation of this would avoid failed pharmacokinetic studies [20].

In this study, Trimetazidine dihydrochloride (TMZ) was selected as a model drug. The TMZ is an effective, well-tolerated drug mainly used in angina pectoris. It is the first known 3-ketoacyl coenzyme A thiolase inhibitor [21]. It is a metabolic agent with anti-ischemic and anti-anginal properties. It inhibits long chain fatty acid oxidation, shifting cardiac metabolism towards glucose oxidation. This results in an improved coupling of glycolysis with glucose oxidation, which has been shown to protect the ischemic heart [22]. Trimetazidine base is neutral and lipophilic in nature with pKa1=4.45 ± 0.02 and pKa2=9.14 ± 0.02 [23,24]. The solubility of Trimetazidine base is very low in aqueous solution compared to single-protonated or double protonated form in acidic solution. Its dihydrochloride salt is freely soluble in water and sparingly soluble in alcohol. In its protonated form, TMZ is slightly hygroscopic, white or almost white crystalline powder. It has an absorption window from stomach to small intestine.

The TMZ is marketed in a number of countries as a safe cellular anti ischemic agent devoid of hemodynamic effects [25]. TMZ is available in market as immediate release (IR) Tablet (Vastarel® IR, 20 mg and Preductal® IR, 20 mg) and modified release (MR) tablet (Vastarel® MR, 35 mg and Preductal® MR, 35 mg). Recommended dosing for IR tablet is two to three times a day and for MR tablet twice a day. The IR formulation of TMZ and other drugs with narrow absorption window has the limitations such as poor patient compliance, fluctuations in plasma level, adverse effects due to great difference in peak and trough level at steady state. Once a day formulation is desirable for better patient compliance, lesser adverse effects and lesser fluctuation in plasma level. A once a day TMZ tablet offers increased minimum plasma concentration (+31%), which is of particular interest in the early morning hours (when patients are at a greater risk of cardiovascular events), reduced plasma level fluctuations, and a prolonged plateau of desired drug concentrations [26].

Abdelbary et al. compared bioavailability of gastro retentive and immediate release conventional tablet (Vastarel® 20 mg) and found that a gastroretentive tablet has better drug bioavailability [27]. This study confirmed that TMZ has site specific absorption i.e. narrow absorption window. Commercially available MR formulations are based on non-gastroretentive technology; hence there is a need for a gastro retentive formulation. The present work is an attempt towards utilization of the available knowledge for setting quality target product profile (QTPP) and justifying the same through mathematical modeling and reducing bio study failure. This work comprises development of a gastroretentive formulation of TMZ and application of assumed IVIVC for identification of prospective bio-equivalent formulation.

## Materials and Methods

### Materials

Trimetazidine dihydrochloride (Innogent Lab Pvt. Ltd. Hyderabad), Sodium Hydrogen Carbonate (Merck KGaA, Germany), Anhydrous Citric acid (Jungbunzlauer AG, Austria), Ethyl cellulose (Dow, Germany), Xanthan gum (XANTURAL® 75; CP Kelco USA), Polyvinyl pyrrolidone (PVP K30; ISP, USA), Microcrystalline cellulose (AVICEL PH112; FMC Biopolymers, Ireland), Magnesium Stearate (COVIDIEN; Mallinckrodt, USA) and Silicon dioxide (AEROSIL 200; Evonik Industries AG, Germany) and Isopropyl alcohol (RFCL Limited; Gujarat, India) were used in the formulation development. Preductal® MR 35 mg (Servier Lab, France) was used for *in vivo* and *in vitro* study. WinNonlin® IVIVC Toolkit™ version 5.3 (Pharsight, CA, USA) software was used for IVIVC modeling. Dissolution apparatus model 2100 (Distek, New Jersey, USA) and UV-spectrophotometer (UV-2405 PC, Shimadzu Scientific Instruments, Japan) for *in vitro* evaluation. Nylon syringe filter (MILLIPORE™, USA) was used for filtration.

### Methods

IVIVC modeling requires *in vitro* as well as *in vivo* data. Once both data are available, model fitting is carried out. A step wise approach was used, as described in the flow chart (Figure 1) and explained below:

***In vivo* data collection for IR tablet:** *In-vivo* data of an IR tablet is required to generate unit impulse response. The i*n vivo* data of Vastarel® 20 mg tablet was reviewed from various sources and one data set was selected [27].

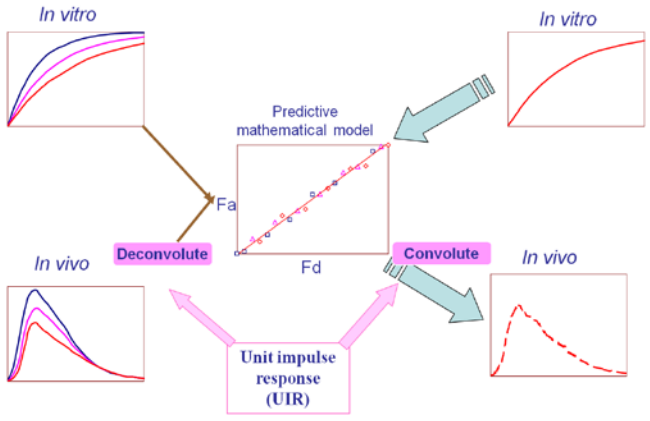


**Figure 1:** A flow chart depicting the sequence followed for IVIVC model generation by obtaining QTPP for model drug (TMZ) and selection of desired formulation based on *in vitro* characteristics.

***In vivo* and *in vitro* data generation for commercial MR tablet and in house ER tablet:**

- ***In vivo* study:** An open-label, single-dose, randomized, two period, two treatment, two sequence, cross-over study was conducted in healthy volunteers to evaluate the pharmacokinetics and bioavailability of TMZ from two products. The commercially available Preductal® MR 35 mg tablet was used as a reference for calculating the relative bioavailability of ER tablet (Test) 70 mg. The ER tablet was a non gastroretentive tablet. The study was consisted of two treatment periods separated by a wash-out period of 3 days between the clinic days. All volunteers were randomly assigned to the treatment. A total number of 24 healthy volunteers were enrolled into this study.

The $C_{max}$ and $T_{max}$ values were directly determined from the mean plasma concentaration versus time profile. The apparent terminal elimination rate constant (λz) was estimated from a regression of In (plasma concentration) versus time over the terminal log-linear drug disposition portion of the plasma concentration-time profiles. The area under curve (AUC) was calculated using the linear up/log down algorithm using software WinNonlin®.

- ***In vitro* study:** Dissolution study was performed using USP apparatus type I (Basket) at 100 rpm speed. Four dissolution media were evaluated to identify a bio relevant media: (i) 0.1 N hydrochloric acid with pH 1.2, (ii) pH 4.5 acetate buffer, (iii) pH 6.8 phosphate buffer and (iv) purified water at 37 ± 0.5°C (Table 1). Dissolution was performed on six units to obtain a statistically significant data. A sample aliquot (5 mL) was withdrawn at predetermined time interval and immediately replaced with equal volume of fresh media to maintain the total volume constant throughout the experiment. All samples were filtered through 0.45 µm membrane nylon filter and analyzed spectrophotometrically at $\lambda_{max}$=270 nm. Dissolution media at different pH condition was selected to check the relevance towards mimicking the *in vivo* conditions.

## IVIVC modeling

***In vitro* data fitting:** Different dissolution models were evaluated to fit the *in vitro* dissolution data in different media (Table 2). Various parameters like $R^2$, Akaike Information Criteria (AIC) and Schwarz's Bayesian Criterion (SBC) were used for model selection criteria (Table 3).

**Deconvolution:** Deconvolution is a calculation of cumulative *in vivo* absorption rate from plasma concentration-time data. The *in vivo* data obtained from the *in vivo* study was deconvoluted. Plots for input rate for test formulation and reference formulation are shown in (Figure 2).

**IVIVC development:** Level-A IVIVC, point to point relationship between the *in vitro* dissolution and the *in vivo* input rate, was studied. The relationship (also referred as internal validation) between the *in vivo* absorption rate and the *in vitro* dissolution rates was developed between ER tablets (Test) 70 mg and Preductal® MR 35 mg (Reference), using *in vivo* data of Vastarel® 20 mg (IR formulation) for unit impulse response (UIR). The i*n vitro* data generated in different media were used for the development of an IVIVC model. The IVIVC modeling was done using different models as given below:

Model 1: Fabs=AbsScale*Diss(Tscale*Tvivo)

Model 2: Fabs=AbsScale*Diss(Tscale*Tvivo-Tshift)

Model 3: Fabs=AbsScale*[Diss(Tscale*Tvivo-Tshift) – AbsBase]

| Time (Hours) | 0.1 N HCl | | pH 4.5 AB | | pH 6.8 PB | | Water | |
|---|---|---|---|---|---|---|---|---|
| | R | T | R | T | R | T | R | T |
| 1 | 17 | 19 | 39 | 22 | 39 | 20 | 39 | 20 |
| 2 | 38 | 30 | 56 | 32 | 67 | 31 | 57 | 30 |
| 3 | 55 | 46 | 70 | 41 | 69 | 39 | 71 | 38 |
| 4 | 75 | 54 | 78 | 48 | 79 | 46 | 81 | 46 |
| 6 | 90 | 70 | 92 | 58 | 90 | 57 | 93 | 56 |
| 8 | 98 | 79 | 98 | 66 | 95 | 66 | 96 | 65 |
| 12 | 100 | 90 | 100 | 78 | 99 | 78 | 97 | 76 |
| 16 | - | 95 | - | 86 | - | 86 | - | 84 |
| 20 | - | 98 | - | 92 | - | 92 | - | 89 |

R-Reference formulation, T- Test formulation

**Table 1:** *In vitro* dissolution profile of Preductal® MR tablet 35 mg (Reference) and Extended Release tablet 70 mg (Test).

| | |
|---|---|
| Hill | $y(t) = int + \frac{(F^{inf} - int)t^b}{(MDT^b + t^b)}$ |
| Weibull | $y(t) = int + (F^{inf} - int) * (1 - e^{-(\frac{1}{MDT})^b})$ |
| Double Weibull | $y(t) = int + f1 * (F^{inf} - int)\left(1 - e^{-(\frac{1}{MDT})^{y+1}}\right) + (1 - f1) * (F^{inf} - int)(1 - e^{-\left(\frac{1}{MDT}\right)^{\frac{t}{2}}})$ |
| Makoid –Banakar | $F = k_{MB} \cdot t^n \cdot e^{-k \cdot t}$<br>$k_{MB}$, *n, and k* are empirical parameters in Makoid–Banakar model ( $k_{MB}$, *n, k>0)* |

Where $F^{inf}$ is the amount release at time infinity; MDT is mean dissolution time; int is the y-intercept; b is slope factor.

**Table 2:** Different dissolution models for model fit analysis of the *in vitro* data.

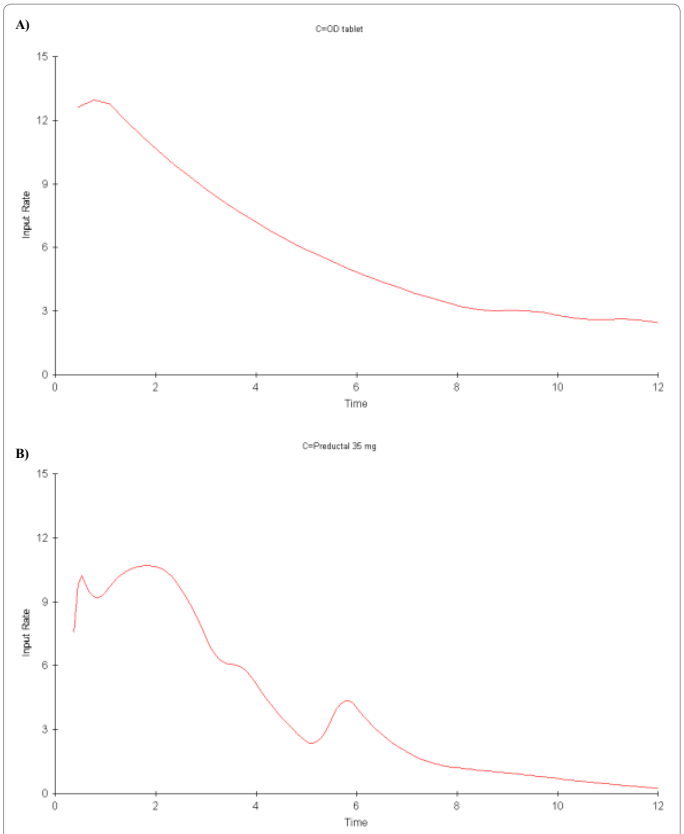


**Figure 2:** Plot for input rate for test formulation (2a) and reference formulation (2b).

The predictability of the IVIVC model was evaluated by internal validation. For this purpose, pharmacokinetic parameters ($C_{max}$ and $AUC_{0-t}$) were predicted using the *in vitro* dissolution data based on the correlation model (Table 4). The predicted pharmacokinetic parameters were compared with the observed values and prediction error was estimated using the following formula:

%PF=[(predicted value-observed value)/observed value]*100

The values of estimated parameters of mathematical correlation model are presented in table 5 and target *in vitro* profile in table 6.

## Formulation development and *in vitro* data generation

**Formulation development:** Gastro retentive tablets were prepared by wet granulation technique. Mixing and granulation of powder was carried out in a 2 Liter Rapid Mixture Granulator followed by drying in Fluidized Bed Drier. Dried granules were mixed with Magnesium stearate and Colloidal silicon dioxide and further blended for 5 min. Final lubricated blend was compressed at 16 station single rotary compression machine using suitable punch tool at optimum hardness. Various polymers and ratio of Citric acid and Sodium hydrogen carbonates were evaluated to obtain a formulation with extended release and a shorter floating lag time (Table 7).

***In-vitro* evaluation:** The *in vitro* floating behavior of tablets was studied by placing the tablet in 500 ml container filled with 300 ml 0.1 N HCl (pH 1.2) and the time taken by tablet to float on the surface was recorded as 'floating lag time' and the total duration for which tablet was floating in the medium as 'total floating time'. This was recorded by visual observation. Various parameters like dissolution profile, hardness, floating lag time and total buoyancy time for all formulation F1-F5 are presented in (Table 8).

## Release kinetics

Data obtained from in vitro drug release study was modeled to various kinetics equations to find out mechanism of drug release from matrix formulation of reference formulation (R), test formulation (T) using DDSolver software [28].

## Prediction of *in vivo* parameters of formulated batches

Using the developed IVIVC model, *in vitro* profiles of formulation F1-F5 were convoluted (Figure 4) and *in vivo* parameters (AUC, $C_{max}$) were predicted for all fabricated formulations (Table 9). A suitable formulation was selected by applying a constrain ratio of 0.95 -1.05 from the target profile for pharmacokinetic parameter $C_{max}$ and $AUC_{0-t}$.

## Results and Discussion

*In vitro* and *in vivo* parameters act as an input function for carrying out IVIVC modeling and a careful consideration should be given to the reliability of the data used in assumed IVIVC. In this study, *in vivo* data from two formulations has been taken from the same study, which may not be possible, if there is variability in the data. In such cases, genetic variability of molecule should be taken into consideration and

| Media | Makoid-Banakar | Formulation | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | ER tablet 70 mg (T) | | | | Preductal® MR 35 mg (R) | | | |
| | weighing Function | $R^2$ | Weighted $R^2$ | AIC | SBC | $R^2$ | Weighted $R^2$ | AIC | SBC |
| 0.1 N HCl | uniform | 0.9991 | 0.9991 | -58.601 | -57.995 | 0.9993 | 0.9993 | -53.219 | -52.825 |
| | 1/yhat*yhat | 0.9986 | 0.9983 | -38.026 | -37.633 | 0.9988 | 0.9995 | -41.812 | -41.653 |
| pH 4.5 AB | uniform | 0.9971 | 0.9971 | -49.874 | -49.269 | 0.9998 | 0.9998 | -66.768 | -66.374 |
| | 1/yhat*yhat | 0.9951 | 0.9978 | -39.277 | -38.883 | 0.9994 | 0.9996 | -53.864 | -53.705 |
| pH 6.8 PB | uniform | 0.9971 | 0.9971 | -49.527 | -48.922 | 0.9957 | 0.9957 | -39.955 | -39.56 |
| | 1/yhat*yhat | 0.9952 | 0.9981 | -39.482 | -39.088 | 0.9875 | 0.9865 | -26.239 | -26.08 |
| Water | uniform | 0.9963 | 0.9963 | -47.77 | -47.165 | 0.9991 | 0.9991 | -50.307 | -49.913 |
| | 1/yhat*yhat | 0.9941 | 0.9979 | -38.574 | -38.18 | 0.9981 | 0.9991 | -43.715 | -43.556 |

**Table 3:** Summary of model parameters and selection of mean *in vitro* dissolution profile of Trimetazidine dihydrochloride extended release tablet.

| Media | Formulation | Parameters | Predicted | Observed | PE | ratio |
|---|---|---|---|---|---|---|
| 0.1 N HCl | Test Formulation 70 mg Internal | $AUC_{last}$ | 1605.582 | 1656.903 | -3.1 | 0.97 |
| | | $C_{max}$ | 93.48553 | 85.443 | 9.4 | 1.09 |
| | Preductal® MR 35 mg Internal | $AUC_{last}$ | 570.8056 | 592.5588 | -3.7 | 0.96 |
| | | $C_{max}$ | 65.57775 | 69.366 | -5.5 | 0.95 |
| | Avg Internal | $AUC_{last}$ | 1088.194 | 1124.731 | 3.4 | 0.97 |
| | | $C_{max}$ | 79.53164 | 77.4045 | 7.4 | 1.03 |
| pH 4.5 AB | Test Formulation 70 mg Internal | $AUC_{last}$ | 1639.706 | 1656.903 | -1 | 0.99 |
| | | $C_{max}$ | 71.47473 | 85.443 | -16.3 | 0.84 |
| | Preductal® MR 35 mg Internal | $AUC_{last}$ | 597.2477 | 592.5588 | 0.8 | 1.01 |
| | | $C_{max}$ | 60.22211 | 69.366 | -13.2 | 0.87 |
| | Avg Internal | $AUC_{last}$ | 1118.477 | 1124.731 | 0.9 | 0.99 |
| | | $C_{max}$ | 65.84842 | 77.4045 | 14.8 | 0.85 |
| pH 6.8 PB | Test Formulation 70 mg Internal | $AUC_{last}$ | 1638.416 | 1656.903 | -1.1 | 0.99 |
| | | $C_{max}$ | 68.84238 | 85.443 | -19.4 | 0.81 |
| | Preductal® MR 35 mg Internal | $AUC_{last}$ | 601.4235 | 592.5588 | 1.5 | 1.01 |
| | | $C_{max}$ | 60.89449 | 69.366 | -12.2 | 0.88 |
| | Avg Internal | $AUC_{last}$ | 1119.92 | 1124.731 | 1.3 | 1 |
| | | $C_{max}$ | 64.86843 | 77.4045 | 15.8 | 0.84 |
| Water | Test Formulation 70 mg Internal | $AUC_{last}$ | 1624.774 | 1656.903 | -1.9 | 0.98 |
| | | $C_{max}$ | 68.50113 | 85.443 | -19.8 | 0.8 |
| | Preductal® MR 35 mg Internal | $AUC_{last}$ | 607.2892 | 592.5588 | 2.5 | 1.02 |
| | | $C_{max}$ | 61.58081 | 69.366 | -11.2 | 0.89 |
| | Avg Internal | $AUC_{last}$ | 1116.032 | 1124.731 | 2.2 | 0.99 |
| | | $C_{max}$ | 65.04097 | 77.4045 | 15.5 | 0.84 |

AB- acetate buffer, PB-phosphate buffer

**Table 4:** Summary of internal validation parameters in different media.

closeness of drug dependent pharmacokinetic parameters i.e., AUC, elimination half-life ($t_{1/2}$) should be compared from different studies.

## Development of *in vitro in vivo* correlation

An IVIVC modeling requires fitting of *in vivo* data for which pharmacokinetic behavior of drug product is needed. Pharmacokinetic properties of TMZ from various IR tablets sand MR tablets are compiled in table 10. The IR tablets showed $T_{max}$ of 2 hours while the MR tablets showed 3-6 hours. The data showed significant inter-individual variability as observed from difference in $C_{max}$ and AUC values from different studies, on dosing the same formulation. While developing assumed IVIVC, the selected data should be consistent in many studies. This will reduce probability of model failure due to inter-individual variability. On this view, *in vivo* data of IR formulation for UIR was taken from study conducted by Abdelbary et al. [27].

| Parameter | Estimate | Standard_Error | CV% |
|---|---|---|---|
| AbsScale | 1.249189 | 0.007385 | 0.59 |
| Tscale | 0.882209 | 0.016226 | 1.84 |

**Table 5:** Values of estimated parameters of mathematical correlation model.

| Time | Target profile |
|---|---|
| 1 | 15-20 |
| 2 | 29-33 |
| 3 | 40-45 |
| 4 | 50-55 |
| 6 | 65-70 |
| 8 | 75-80 |
| 12 | 87-93 |
| 16 | 94-98 |
| 20 | 97-100 |

**Table 6:** Targeted *in vitro* dissolution profile in 0.1 N HCl/500 mL, USP-1/RPM-100.

The plasma concentration profile for immediate release tablet, modified release tablet (Reference), and ER tablet (Test) of TMZ are presented in (Figure 5). The PK profile clearly demonstrated that the absorption of TMZ was prolonged, when ER tablet was administered. The average time to reach peak plasma concentration ($T_{max}$) was ≥ 6 hours with the ER tablet compared with 2-3 hours and 1 hour for the MR tablet and the IR tablet respectively.

A level-A IVIVC was conducted using WinNonlin® IVIVC Toolkit™. The procedure for developing an IVIVC consisted of the following steps: (i) calculation of cumulative *in vitro* dissolution rate, (ii) calculation of cumulative *in vivo* absorption rate from concentration-time data by deconvolution, and (iii) modeling the relationship between *in vivo* absorption rate and *in vitro* dissolution rates. Initially IVIVC was developed between the ER tablet 70 mg (Test) and Preductal® MR

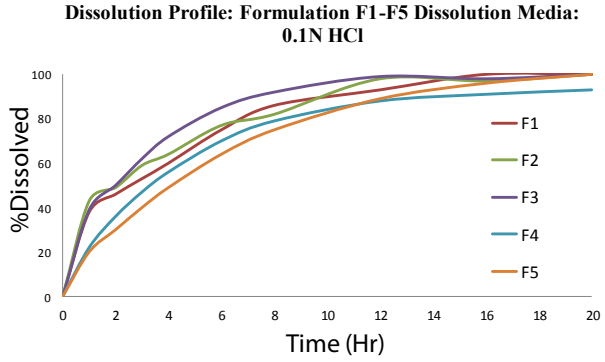


**Figure 3:** Dissolution Profile: Formulation F1-F5 Dissolution Media: 0.1 N HCl.

35 mg (Reference), using UIR from Vastarel® IR 20 mg. As absorption related parameters were constant for oral formulation, hence IR tablet (Vastarel® IR 20 mg) was used as UIR.

The *in vivo* plasma concentration time data of the ER tablet (Test) and Preductal® MR 35 mg (Reference) was analyzed using various models to identify the best fit model using UIR. Drug input rate of *in vivo* study was calculated by numerical deconvolution (Figure 2). For Test and Reference products, input rate function was maximum at 2 hours followed by a decline indicating initial burst release in first 2 hours. The difference in decline pattern in input function indicated difference in the formulations; the ER tablet showed a slow decline and maintained input function after 12 hours of dosing, while input function for Preductal® MR 35 mg showed abrupt decline behavior and insignificant drug absorption observed at 8 hours. These two formulations showed different *in vivo* profiles thus fulfilling the requirement for IVIVC modeling. A correlation was evaluated using linear and nonlinear models to establish a relationship between the *in vitro* drug release and *in vivo* drug absorption. The predictability of pharmacokinetic parameters ($C_{max}$ and $AUC_{0-t}$) was checked using convoluted plots (Figure 6).

## *In vitro* dissolution study

The dissolution data of the reference (Preductal® MR 35 mg) and the test product (ER tablet, 70 mg) (Table 1) was fitted using Hill, Weibull, and Makoid-Banakar function with uniform weighting function (Table 3). Based on the statistical parameters, i.e., $r^2$, weighted $r^2$, AIC and SBC criteria, Makoid-Banakar appeared to best fitted in all dissolution media. Makoid-Banakar function was further evaluated other weighing function. Weighing function 1/yhat*yhat appeared best. It is a model independent approach to define the dissolution behavior from controlled release dosage form. The mathematical expression of Makoid-Banakar function is shown in (Table 2). The 'n' function governs the shape of dissolution curve [29].

## Release kinetics

The curvilinear nature of the cumulative percent drug released

| Ingredients | F1 | F2 | F3 | F4 | F5 |
|---|---|---|---|---|---|
| **Intra granular Ingredients** | | | | | |
| Trimetazidine Dihydrochloride | 70 | 70 | 70 | 70 | 70 |
| Ethyl Cellulose-7cps (Ethocel) | - | 120 | - | - | - |
| Xanthan gum (Xantural 75) | 120 | 120 | 100 | 100 | 120 |
| Microcrystalline cellulose (Avicel PH 112) | 100 | 100 | 40 | 40 | 50 |
| Lactose (DCL-21) | 25 | 25 | 25 | 25 | 25 |
| Sod. Bicarbonate | 60 | 60 | 30 | 30 | 60 |
| Citric acid anhydrous | 30 | 30 | 15 | 15 | 30 |
| Polyvinyl pyrrolidone (PVP-K30) | - | - | 15 | 30 | 30 |
| Isopropyl alcohol | qs | qs | qs | qs | qs |
| **Extra granular Ingredients** | | | | | |
| Colloidal silicon di oxide (Aerosil 200) | 5 | 5 | 5 | 5 | 5 |
| Magnesium Stearate | 5 | 5 | 5 | 5 | 5 |
| **Tablet weight** | 415 | 535 | 305 | 320 | 395 |

**Table 7:** Composition for trial Formulations F1 to F5.

| Time | F1 | F2 | F3 | F4 | F5 |
|---|---|---|---|---|---|
| 1 | 38 | 43 | 39 | 22 | 20 |
| 2 | 46 | 49 | 50 | 36 | 30 |
| 3 | 53 | 59 | 62 | 47 | 40 |
| 4 | 60 | 64 | 72 | 56 | 49 |
| 6 | 75 | 77 | 85 | 70 | 64 |
| 8 | 86 | 82 | 92 | 79 | 75 |
| 12 | 93 | 98 | 99 | 88 | 89 |
| 16 | 100 | 97 | 98 | 91 | 96 |
| 20 | 100 | 100 | 100 | 93 | 100 |
| Hardness (Kp) | 13-15 | 13-14 | 13-14 | 8-9 | 12-13 |
| Floating Lag time | 1 hr | 30 min | <30 sec | <30 sec | <30 sec |
| Total Buoyancy time | 8 h | 4 hr | 20 hr | 20 hr | 20 hr |

**Table 8:** Hardness and i*n vitro* parameters in 0.1 N HCl/500 mL, USP-1/RPM-100, of formulations F1 to F5.

| Formulation | Parameter | Predicted | Observed | PE | ratio |
|---|---|---|---|---|---|
| F1 External | $AUC_{last}$ | 1435.102 | 1525.523 | -5.9 | 0.94 |
| | $C_{max}$ | 98.1932 | 92.01778 | 6.7 | 1.07 |
| F2 External | $AUC_{last}$ | 1519.28 | 1525.523 | -0.4 | 1 |
| | $C_{max}$ | 139.226 | 92.01778 | 51.3 | 1.51 |
| F3 External | $AUC_{last}$ | 1490.922 | 1525.523 | -2.3 | 0.98 |
| | $C_{max}$ | 110.8681 | 92.01778 | 20.5 | 1.2 |
| F4 External | $AUC_{last}$ | 1445.135 | 1525.523 | -5.3 | 0.95 |
| | $C_{max}$ | 91.371 | 92.01778 | -0.7 | 0.99 |
| F5 External | $AUC_{last}$ | 1410.559 | 1525.523 | -7.5 | 0.92 |
| | $C_{max}$ | 85.0059 | 92.01778 | -7.6 | 0.92 |

**Table 9:** Predicted Pharmacokinetics parameters of Formulation F1-F5.

| Formulation | Pharmacokinetic parameters | | | | | | | | Reference |
|---|---|---|---|---|---|---|---|---|---|
| | Tmax (Hours) | $C_{max}$ (ng/mL) | $C_{max}$/Dose (ng/mg ml) | $AUC_{0-t}$ (ng.hr/m) | $AUC_{0-t}$ /Dose (ng.hr/ mg. mL) | $AUC_{0-\infty}$ (ng.hr/mL) | $AUC_{0-\infty}$/Dose (ng.hr/mg. mL) | $T_{1/2}$ (Hours) | |
| Vastarel 20 mg (SD) | 2.7 ± 1 | 47 ± 9 | 2.35 | 369.8 ± 76 | 18.49 | 789.6 ± 165 | 39.48 | 4.6 ± 1.3 | 27 |
| IR 20 mg | 2.0 ± 0.008 | 35.0 ± 5.2 | 1.75 | - | - | 623.8 ± 100.8 | 31.19 | 7.39 ± 1.1 | 30 |
| IR 20 mg | 1.8 | 53.6 | 2.68 | - | - | 508.9 | 25.445 | 6 | 31 |
| IR 40 mg | 2.73 | 127.5 | 3.1875 | - | - | - | - | - | |
| Test 20 mg | 2.312 ± 0.66 | 74.85 ± 12.13 | 3.7425 | 673.1 ± 117.6 | 33.655 | 717.1 ± 120.9 | 35.855 | 4.78 ± 0.92 | 32 |
| Reference 20 mg | 2.211 ± 0.61 | 71.93 ± 14.32 | 3.5965 | 652.3 ± 121.9 | 32.615 | 692 ± 128.6 | 34.6 | 4.74 ± 0.82 | |
| Reference 35 mg MR | 3.54 | 98.57 | 2.82 | 410.01 (t-12) | | 462.78 | 13.22 | 3.45 | 33 |
| Test 35 mg MR (Metacard) | 4 | 104.78 | 2.99 | 423.81 | 12.11 | 472.51 | 13.50 | 3.69 | |
| Test 35 mg MR (Matenol) (Fasting Study) | 5 | 142 | 4.06 | 1320 | 37.71 | 1430 | 40.86 | 5.62 | 34 |
| Vastarel 35 mg MR (Fasting Study) | 4.5 | 135 | 3.86 | 1270 | 36.29 | 1360 | 38.86 | 5.32 | |
| Test 35 mg MR (Matenol) (Fed Study) | 4.5 | 183 | 5.23 | 1440 | 41.14 | 1530 | 43.71 | 4.97 | |
| Vastarel 35 mg MR (Fed Study) | 5 | 169 | 4.83 | 1450 | 41.43 | 1580 | 45.14 | 5.29 | |
| Three layer matrix tablet 50 mg | 6.1 ± 0.6 | 61.5 ± 3.3 | 1.23 | - | | 1327.9 ± 304.5 | 26.56 | 20.84 ± 3.0 | 30 |
| Test 70 mg (OD) | 6 | 85.443 | 1.22 | 1656.903 | 23.66 | - | - | 7.29 | In-House Study |
| Preductal 35 mg MR | 4.5 | 69.366 | 1.98 | 592.56 | 16.93 | - | - | 5.25 | |

**Table 10:** Pharmacokinetic properties of TMZ from literature and in-house study.

versus time plots (Figure 3) suggests that drug release from the matrix tablet does not follow zero-order kinetics. The data modeling results are summarized in table 11, and this observation is supported by the low values of correlation coefficients obtained in all cases where the dissolution data were fitted to a zero-order model. The *in vitro* dissolution studies confirmed that drug release was governed by first order model which means rate of drug release depend upon amount of unreleased drug. The Korsmeyer–Peppas release exponent, n, is less than 0.5, confirming that diffusion is the controlling factor for drug release [30,31].

## IVIVC interpretation

IVIVC modeling was evaluated in all four available media using all models. Model 1 adequately fit the reference formulation data. The values of estimated parameters of mathematical correlation model are presented in (Table 5). The prediction error was low with correlation obtained using 0.1 N HCl as dissolution media. The low prediction error indicates the reliability of model towards carrying out predictions; hence it was selected as a bio relevant tool to screen the best formulation. A reliable correlation ($r^2$=0.988) was observed between fraction dissolved *in vitro* and fraction absorbed *in vivo* (Figure 7a) and levy plot showing a relationship between $T_{in\ vitro}$ and $T_{in\ vivo}$ (time for drug dissolved and drug absorbed) is presented in (Figure 7b). Convoluted predicted profile for reference (Preductal® MR 35 mg) and test formulation (ER tablet, 70 mg) matches with observed profile confirming a level-A correlation was obtained (Figures 6a and 6b).

## Formulation development for a target *in vitro* profile

The IVIVC model suggested 0.1 N HCl medium as a bio relevant medium. The *in vivo* release from a gastro retentive system is expected in upper part of GIT at acidic pH, which also supports the identified IVIVC media. Hence, the *in vitro* evaluation of formulated batches was carried out in the same media. The other parameters like floating lag time and total buoyancy time were evaluated and it showed the

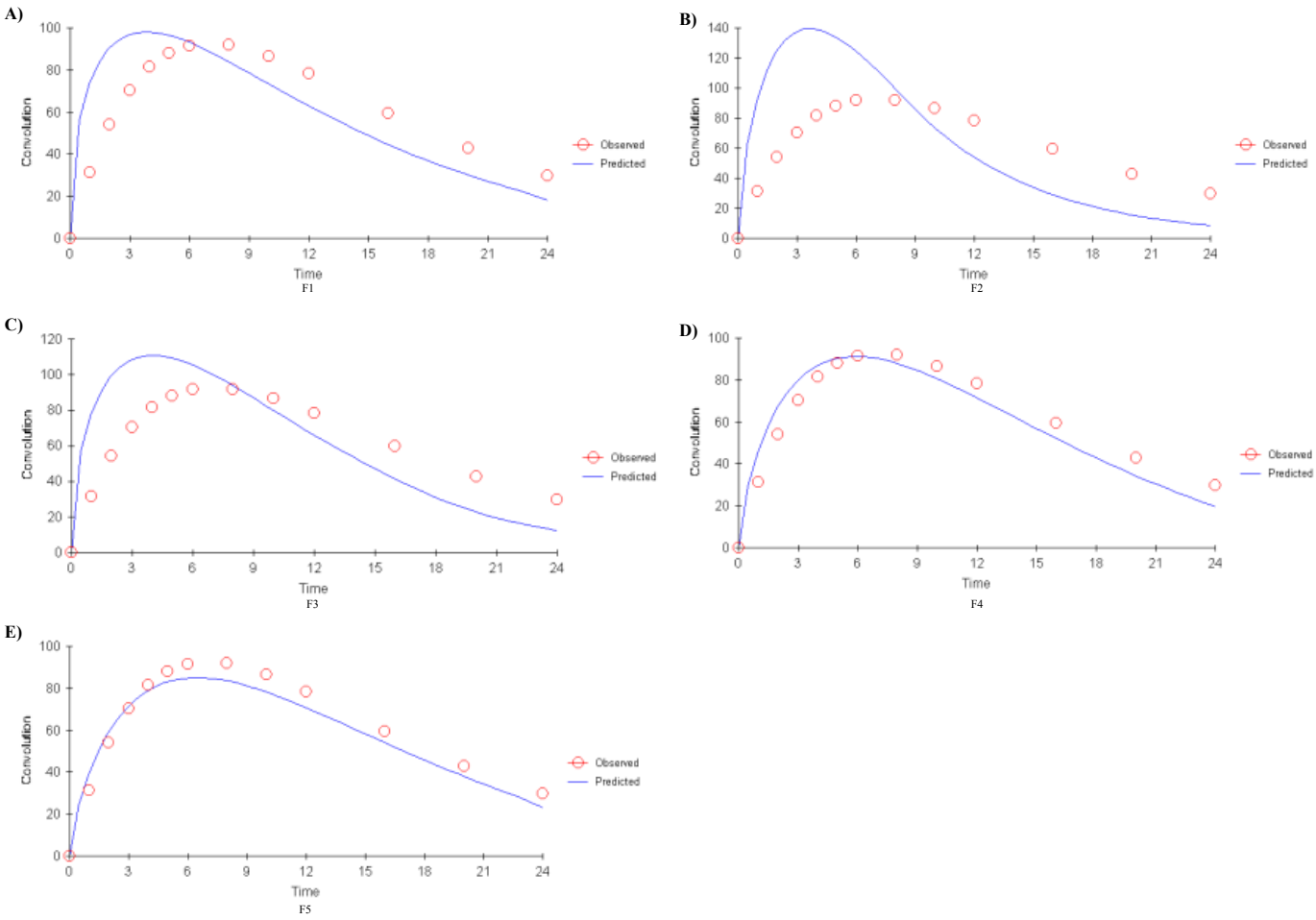


**Figure 4:** Plots of convolution output for formulation F1-F5.

| Motdel | Parameters | R | T | F1 | F2 | F3 | F4 | F5 |
|---|---|---|---|---|---|---|---|---|
| Zero-order | $R^2$ | 0.6955 | 0.532 | 0.1544 | -0.0501 | -0.1599 | 0.3686 | 0.6506 |
| | $R^2$-adj | 0.6955 | 0.532 | 0.1544 | -0.0501 | -0.1599 | 0.3686 | 0.6506 |
| First-order | $R^2$ | 0.9742 | 0.9989 | 0.9668 | 0.9466 | 0.9873 | 0.9903 | 0.9967 |
| | $R^2$-adj | 0.9742 | 0.9989 | 0.9668 | 0.9466 | 0.9873 | 0.9903 | 0.9967 |
| Higuchi | $R^2$ | 0.9389 | 0.9566 | 0.9019 | 0.8462 | 0.7975 | 0.9338 | 0.9809 |
| | $R^2$-adj | 0.9389 | 0.9566 | 0.9019 | 0.8462 | 0.7975 | 0.9338 | 0.9809 |
| Korsmeyer-Peppas | $R^2$ | 0.9392 | 0.9634 | 0.9833 | 0.9861 | 0.963 | 0.9635 | 0.9813 |
| | $R^2$-adj | 0.9305 | 0.9589 | 0.9812 | 0.9844 | 0.9584 | 0.959 | 0.9789 |
| | n | 0.5130 | 0.445 | 0.337 | 0.298 | 0.285 | 0.393 | 0.487 |
| Hixson-Crowell | $R^2$ | 0.9921 | 0.9896 | 0.937 | 0.8937 | 0.9224 | 0.9645 | 0.9919 |
| | $R^2$-adj | 0.9921 | 0.9896 | 0.937 | 0.8937 | 0.9224 | 0.9645 | 0.9919 |

**Table 11:** Results of model fitting of drug release of Reference (R), Test (T) and fabricated formulations (F1-F5) in 0.1 N HCl.

maximum buoyancy up to 20 hours indicating the formulation to be gastro retentive for sufficient time.

The IVIVC model suggested a target *in vitro* release profile in 0.1 N HCl (Table 6). A gastro retentive matrix tablet technology was evaluated. Two types of polymers in combination, hydrophobic (ethyl cellulose) and hydrophilic polymers (Polyvinyl pyrrolidone), were evaluated to control the drug release. Xanthan gum was chosen as a main release controlling polymer. Three formulation strategies were employed (i) Xanthan gum alone in the formulation (F1), (ii) with a hydrophobic polymer (ethyl cellulose) (F2) and (iii) with a hydrophilic polymer (Polyvinyl pyrrolidone) (F3). Amongst the three formulations, drug release was faster in all the formulations in comparison with the target profile. However, formulation with polyvinyl pyrrolidone (F3)

showed less floating lag time (<30 seconds) and higher buoyancy time (20 hours). Polyvinyl pyrrolidone (PVP) was selected for further study and its concentration was evaluated (F4 and F5). On increasing the concentration of PVP, desired drug release profile was achieved and buoyancy parameters were achieved [32-34].

### Prediction of *in vivo* parameters of formulated batches

The *in vitro* data for studied formulations obtained was fed into the IVIVC model and *in vivo* profiles were predicted. The predicted pharmacokinetic parameters are provided in table 9 and convoluted profiles are provided in figure 4. Predicted profiles suggested that: (i) formulations F2 and F3 would show faster *in vivo* parameters, (ii) formulations F1 would have desired $C_{max}$ value but required $t_{max}$ will not be achieved and (iii) formulations F4 and F5 would have desired $C_{max}$ and $t_{max}$ suggesting these are suitable candidates for pharmacokinetic study. Predicted pharmacokinetic parameters (predicted *vs.*observed ratios for $C_{max}$ and AUC) suggested that formulations F1, F4 and F5 passed the criteria of bioequivalence for AUC but formulation F1 failed in the $C_{max}$ ratio (1.07) and did not have floating property, hence

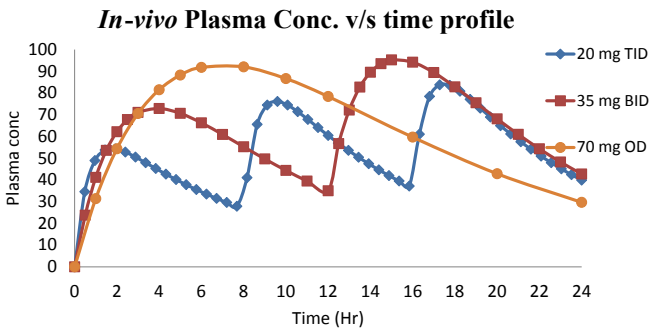


**Figure 5:** Mean *in vivo* concentration time profiles of Trimetazidine dihydrochloride profile of test formulation (70 mg OD), reference formulation (35 mg BID) and IR formulation (20 mg TID).

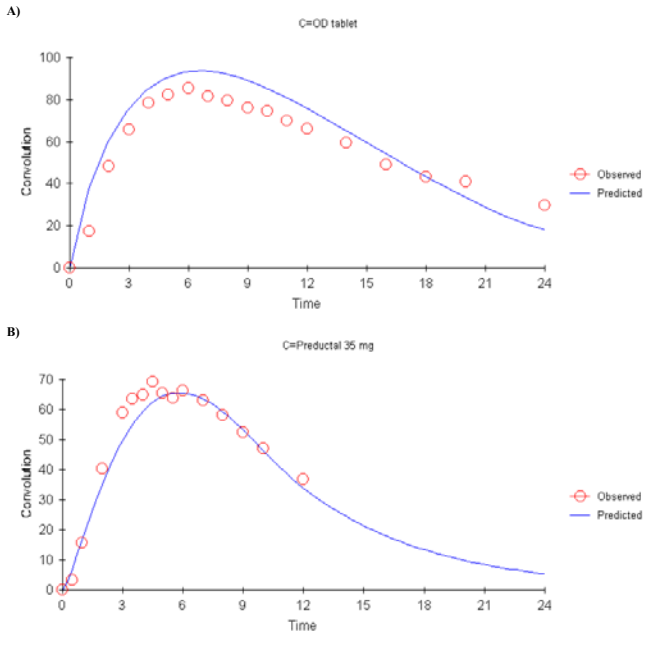


**Figure 6:** Plots of convolution output for test formulation (6a) and reference formulation (6b) .

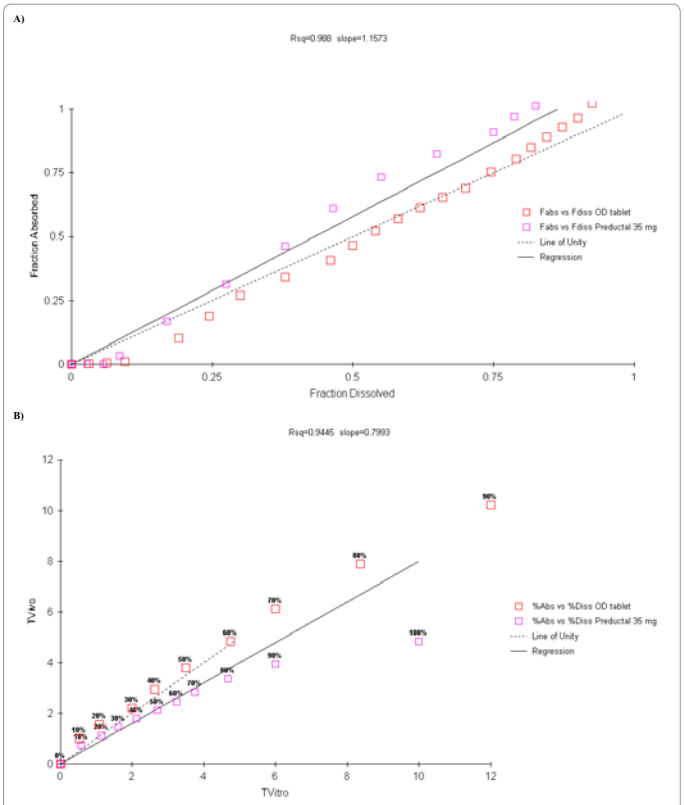


**Figure 7:** Regression (7a) and Leavy plot (7b) between fraction of absorbed and fraction of dissolved.

it was excluded from further investigation. Formulation F4 and F5 are meeting the criteria for target profile for generating a once daily formulation. These two formulations are recommended for a bioequivalence.

## Conclusion

Assumed IVIVC model for Trimetazidine dihydrochloride extended release tablet was generated using literature information and in house bio study. A target *in vitro* profile was generated from the IVIVC model. Assumed IVIVC was developed and a target profile in biorelevant media was selected for the formulation development. The prediction errors obtained were within the limits. Based on the target profile, a pharmacokinetic profile was predicted for formulation (F1-F5). It was identified that formulation F4 and F5 are the best formulations to develop once daily gastro retentive formulation of Trimetazidine dihydrochloride. Assumed IVIVC was successfully utilized in product development.

### References

1. Jiang W, Kim S, Zhang X, Lionberger RA, Davit BM, et al. (2011) The role of predictive biopharmaceutical modeling and simulation in drug development and regulatory evaluation. Int J Pharm 418: 151-160.
2. Dressman JB, Reppas C (2000) In vitro-in vivo correlations for lipophilic, poorly water-soluble drugs. Eur J Pharm Sci 2: S73-80.
3. Nicolaides E, Galia E, Efthymiopoulos C, Dressman JB, Reppas C (1999) Forecasting the in vivo performance of four low solubility drugs from their in vitro dissolution data. Pharm Res 16: 1876-1882.
4. Tang X, Tai LY, Yang XG, Chen F, Xu HM, et al. (2012) In vitro and in vivo valuation of gliclazide push–pull osmotic pump coated with aqueous colloidal polymer dispersions. Drug Dev Ind Pharm.
5. Patel AR, Spencer SD, Chougule MB, Safe S, Singh M (2012) Pharmacokinetic

evaluation and in vitro-in vivo correlation (IVIVC) of novel methylene-substituted 3,3' diindolylmethane (DIM). Eur J Pharm Sci 46: 8-16.

6. Ghosh A, Bhaumik UK, Bose A, Mandal U, Gowda V, et al. (2008) Extended release dosage form of glipizide: development and validation of a level A in vitro-in vivo correlation. Biol Pharm Bull 31: 1946-1951.

7. Gelman A, Bois F, Jiang J (1996) Physiological pharmacokinetic analysis using population modeling and informative prior distributions. J Am Stat Assoc 91: 1400-1412.

8. Kortejärvi H, Malkki J, Marvola M, Urtti A, Yliperttula M, et al. (2006) Level A in vitro-in vivo correlation (IVIVC) model with Bayesian approach to formulation series. J Pharm Sci 95: 1595-1605.

9. Balan G, Timmins P, Greene DS, Marathe PH (2001) In vitro-in vivo correlation (IVIVC) models for metformin after administration of modified-release (MR) oral dosage forms to healthy human volunteers. J Pharm Sci 90: 1176-1185.

10. Macha S, Yong CL, Darrington T, Davis MS, MacGregor TR, et al. (2009) In vitro-in vivo correlation for nevirapine extended release tablets. Biopharm Drug Dispos 30: 542-550.

11. Bhattachar SN, Perkins EJ, Tan JS, Burns LJ (2011) Effect of gastric pH on the pharmacokinetics of a BCS class II compound in dogs: utilization of an artificial stomach and duodenum dissolution model and GastroPlus,™ simulations to predict absorption. J Pharm Sci 100: 4756-4765.

12. Saibi Y, Sato H, Tachiki H (2012) Developing in vitro-in vivo correlation of risperidone immediate release tablet. AAPS PharmSciTech 13: 890-895.

13. Shono Y, Jantratid E, Kesisoglou F, Reppas C, Dressman JB (2010) Forecasting in vivo oral absorption and food effect of micronized and nanosized aprepitant formulations in humans. Eur J Pharm Biopharm 76: 95-104.

14. Shono Y, Jantratid E, Janssen N, Kesisoglou F, Mao Y, et al. (2009) Prediction of food effects on the absorption of celecoxib based on biorelevant dissolution testing coupled with physiologically based pharmacokinetic modeling. Eur J Pharm Biopharm 73: 107-114.

15. Yamashita F, Fujita A, Zhang X, Sasa Y, Mihara K, et al. (2012) Computer-based evolutionary search for a nonlinear conversion function for establishing in vitro-in vivo correlation (IVIVC) of oral drug formulations. Drug Metab Pharmacokinet 27: 280-285.

16. Selen A, Cruañes MT, Müllertz A, Dickinson PA, Cook JA, et al. (2010) Meeting Report: Applied Biopharmaceutics and Quality by Design for Dissolution/ Release Specification Setting: Product Quality for Patient Benefit. AAPS J 12: 465-72.

17. Guidance for Industry: Extended Release Oral Dosage Forms: Development, Evaluation, and Application of In Vitro/In Vivo Correlations (1997) U.S. Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research (CDER), U.S. Government Printing Office: Washington, DC.

18. Devane J and Butler J (1997) The impact of in vitro-in vivo relationships on product development. Pharm Tech 21: 146-159.

19. Emami J (2006) In vitro - in vivo correlation: from theory to applications. J Pharm Pharm Sci 9: 169-189.

20. Cook JA (2012) Development strategies for IVIVC in an industrial environment. Biopharm Drug Dispos 33: 349-353.

21. Kantor PF, Lucien A, Kozak R, Lopaschuk GD (2000) The antianginal drug trimetazidine shifts cardiac energy metabolism from fatty acid oxidation to glucose oxidation by inhibiting mitochondrial long-chain 3-ketoacyl coenzyme A thiolase. Circ Res 86: 580-588.

22. Barre J, Ledudal P, Oosterhuis B, Brakenhoff JP, Wilkens G, et al. (2003) Pharmacokinetic profile of a modified release formulation of trimetazidine (TMZ MR 35 mg) in the elderly and patients with renal failure. Biopharm Drug Dispos 24: 159-164.

23. Alemu H (2004) Voltammetry of drugs at the interface between two immiscible electrolyte solutions. Pure Appl Chem 76: 697-705.

24. Reymond F, Steyaert G, Carrupt PA, Morin D, Tillement JP, et al. (1999) The pH-partition profile of the anti-ischemic drug trimetazidine may explain its reduction of intracellular acidosis. Pharm Res 16: 616-624.

25. Sweetman SC (2005) Dose adjustment in renal impairment: response from Martindale: the Complete Drug Reference. BMJ 331: 292-293.

26. Ioana Holban (2003) Introducing Vastarel MR, the twice-daily 3-KAT inhibitor with sustained antianginal efficacy at trough. Heart Metab 20: 29-32.

27. Abdelbary A, El-Gazayerly ON, El-Gendy NA, Ali AA (2010) Floating tablet of trimetazidine dihydrochloride: an approach for extended release with zero-order kinetics. AAPS PharmSciTech 11: 1058-1067.

28. Zhang Y, Huo M, Zhou J, Zou A, Li W, et al. (2010) DDSolver: an add-in program for modeling and comparison of drug dissolution profiles. AAPS J 12: 263-271.

29. Makoid MC, Dufour A, Banakar UV (1993) Modelling of dissolution behaviour of controlled release systems. STP Pharma 3: 49-58.

30. Krishnaiah YSR, Karthikeyan RS, Bhaskar P, Satyanarayana V (2002) Bioavailability studies on guar gum-based three-layer matrix tablets of Trimetazidine dihydrochloride in human volunteers. J Control Release 83: 231-239.

31. McClellan KJ, Plosker GL (1999) Trimetazidine. A review of its use in stable angina pectoris and other coronary conditions. Drugs 58: 143-157.

32. Jiao Y, Su M, Chen M, Jia W, Chou Y, et al. (2007) LC/ESI-MS method for the determination of trimetazidine in human plasma: Application to a bioequivalence study on Chinese volunteers. J Pharm Biomed Anal 43: 1804-1807.

33. Chowdhury MM, Ullah MA, Iqbal N, Al Maruf A, Shohag MH, et al. (2011) Relative bioavailability and pharmacokinetic study of two trimetazidine modified release formulations in healthy Bangladeshi male volunteers. Arzneimittelforschung 61: 393-398.

34. Weerawadee C, Somruedee C, Tullaya R, Suthathip N, Korbtham S, et al. (2011) Bioequivalance study of two formulation of 35 mg trimetazidine modified release tablets in healthy thai volunteers under fasting and fed conditions. J Bioequiv availab 3: 052-055.