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Research Paper

Population *In Vitro-In Vivo* Correlation Model for Pramipexole Slow-Release Oral Formulations

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Purpose. To establish an *in vitro-in vivo* level A correlation (IVIVC) for pramipexole slow-release formulations.
Methods. The IVIVC was developed based on data from an immediate-release (IR) and three slow-release (SR) formulations of pramipexole; a fourth SR formulation was used for validation purposes. *In vitro* dissolution profiles were obtained from all SR formulations. Fifteen volunteers received all pramipexole formulations in a randomized cross-over trial. Data were analyzed using the population modelling approach as implemented in NONMEM VI.
Results. Dissolution profiles of the SR formulations were described by the Weibull model. The pharmacokinetics of the IR formulation were described by a two-compartment disposition model with first-order absorption. Difference between the *in vivo* and *in vitro* estimates of the release rate constants (k_d) from the Weibull function suggests the release process occurs faster *in vivo*. Pharmacokinetic profiles for SR formulations were described based on the *in vitro* release model with k_d increased in 0.058 h^{-1} and the population pharmacokinetic model developed from the IR formulation.
Conclusion. A level A IVIVC was established and evaluated for the pramipexole SR formulations, which can be used in the future as a surrogate to avoid certain bioequivalence studies.

KEY WORDS: *in vitro-in vivo* correlation; population pharmacokinetics; pramipexole.

INTRODUCTION

Parkinson's disease is a neurodegenerative disorder manifested by rest tremor, bradykinesia, rigidity, and loss of postural reflexes (1). As the disease progresses, patients may develop clinical features partially augmented by levodopa (the conventional prescribed therapy). Dopamine agonists are currently being used either as monotherapy for the treatment of early-stage Parkinson's disease (as part of the levodopa sparing strategy, aimed at delaying the occurrence of levodopa-related motor fluctuations) or in the later phase of the disease to lessen motor complications caused by levodopa (2–4). When given as an adjuvant therapy the dopamine agonists can deliver a more continuous dopamine stimulation than levodopa, due to their longer elimination half-life, allowing a reduction in levodopa daily dose and therefore diminishing the duration and severity of levodopa-induced diskynesias (2–4). Pramipexole is a dopamine receptor agonist primarily approved for treating Parkinson's disease. It was later approved for Restless Legs Syndrome (RLS), a condition that causes discomfort in the legs and a strong urge to move the legs, especially at night and when sitting or lying down (5).

The recommend starting dose of pramipexole is 0.125 mg taken once daily (for RLS) and 0.375 mg/day given in three divided doses (for Parkinson's disease); this dose can be increased every 4–7 days up to 0.5 mg (for RLS) and every 5–7 days up to 4.5 mg/day (for Parkinson's disease).

A slow-release drug formulation of pramipexole is currently under investigation and should allow patients to treat their symptoms with a single daily dose, thereby increasing convenience and compliance. Over the past decade, increasing confidence has built on *in vitro* dissolution use as a surrogate to evaluate and predict *in vivo* performance of the modified-release drugs based on *in vitro-in vivo* correlation (IVIVC) (6). An IVIVC can be defined as a predictive mathematical model describing the relationship between an *in vitro* property of a dosage form and a relevant *in vivo* response (7). Generally, the *in vitro* performance is characterized by the rate and extent of drug release or dissolution, while the *in vivo* response is the plasma drug concentration or amount of drug absorbed. Three different levels of IVIVC have been defined: (i) level A corresponds to the case in which the entire *in vivo* time course (of the plasma drug concentration) is predicted from the *in vitro* data, (ii) level B compares the mean *in vitro* dissolution time to either the mean residence time or to the mean *in vivo* dissolution time and (iii) level C establishes a single point relationship

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between a dissolution parameter and pharmacokinetic parameter (7).

An IVIVC can potentially decrease the number of biopharmaceutical studies required in support of a drug product. Further, IVIVC can also allow setting more meaningful dissolution specifications (8,9). Therefore, level A IVIVCs for oral extended-release dosage forms have been established successfully during the last years (6,10–13).

Due to its characteristics concerning solubility and permeability, pramipexole can be considered as a Class I drug (high solubility/high permeability) according to the Biopharmaceutical Classification System, so an IVIVC can be expected for slow-release formulations of this drug (14). The objective of this study was to establish a level A IVIVC for pramipexole slow-release formulations in healthy volunteers. This IVIVC should assist in the development of an optimal dosage formulation, should be used as a surrogate for human bioavailability studies and should help to define a biorelevant dissolution method. In contrast to the general approach, in which the percentage of absorbed drug is obtained by deconvolution from the mean plasma concentration profiles, we have applied a population parametric analysis where all available data (*in vitro* and *in vivo*) were fitted sequentially to get the predicted plasma concentration time profiles based on the *in vitro* dissolution parameters. Furthermore, this approach allows considering not only mean profiles but also the individual profiles by using estimates for the interindividual variability.

MATERIALS AND METHODS

Dosage Forms

Five different oral dosage forms of pramipexole were studied in the current analysis: an immediate-release tablet (IR) containing 0.125 mg of pramipexole dihydrochloride monohydrate equivalent to 0.0873 mg of pramipexole base, and four different matrix slow-release (SR) tablets containing 0.375 mg of pramipexole dihydrochloride monohydrate equivalent to 0.262 mg of pramipexole base, identified as $SR_{1\text{-}4}$. SR tablets varied in the percentage of carbomer 941 and in the type of starch used in the formulation (SR_1, SR_2, and SR_3 used maize starch, and SR_4 used modified starch). For the IR formulation, only maize starch was used but no carbomer 941. There was no intravenous pramipexole formulation available for this analysis.

In Vitro Dissolution Test

In vitro dissolution studies were performed using an USP basket apparatus at 100 rpm. The dissolution medium was 500 mL of phosphate buffer at pH 6.8. Samples were collected through a suitable membrane filter, discarding the first 2 mL of the filtrate at the following pre-defined times: 1, 2, 3, 4, 6, 8, 9, 14, 24 h (SR formulations) and 15, 30 and 60 mi (IR formulation). Samples were stored protected from light until analysis. For each of the SR formulations, twelve replicates were obtained, whereas a single dissolution profile was performed for the IR formulation.

In Vivo Study

Data were obtained from a single-centre, open-label, single-dose, five-treatment, five-period cross-over clinical trial. In the first arm, the IR formulation was administered; the following administrations of the SR formulations were completely randomized. All participants provided written informed consent consistent with ICH-GCP (International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use—Good Clinical Practice) and local legislation, once the nature and the intention of the investigation was fully explained. The study was performed in accordance with the Declaration of Helsinki and was approved by the institutional review board of ethics committee at the study site.

Important inclusion criteria were age 18–50 years old and body mass index (BMI) within 18.5–29.9 kg/m^2. Important exclusion criteria were (i) any finding of the medical examination deviating from normal and of clinical relevance, (ii) intake of drugs with a long half-life within at least one month, or participation in another trial with an investigational drug within at least two months prior to the trial, (iii) smoker, or alcohol or drug abuse, and (iv) excessive physical activities within the last week before the trial or during the trial. Data from fifteen healthy male volunteers were available for the analysis.

Study Design

All subjects in the study received first the IR formulation. All doses were administered in the morning between 8:00 and 10:00 AM with 230 mL of non-sparkling water. Drinking water and fruit tea were allowed from 2 and 4 h after drug administration, respectively; lunch was given 4 h after the dosing. Additional meals were given 10:00 and 14:00 h after dosing, and breakfast was served at approximately 8:00 AM on day 2. The wash-out period between treatments was five days.

Sample Collection

An indwelling venous catheter was inserted into a forearm vein, and venous blood samples (2.7 mL) were withdrawn at the following times after drug administration: 0.25, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8, 10, and 14 h (IR), and 0.25, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8, 10, 14, 22, 26 and 30 h (SR). The whole blood samples were centrifuged within 30 min after sample collection at 4°C at approximately 3,000 rpm for at least 10 min. Until centrifugation, the samples were stored in ice bath, and, afterwards, samples were stored immediately in a freezer at −20°C.

Assay of Pramipexole in the Dissolution Media and in Plasma

The percentage of dissolved pramipexole was analyzed by high performance liquid chromatography (HPLC). Analytical separation was performed by an Agilent Zorbax SB Aq column (particle size of 3.5 μm, length of 5 cm; internal diameter of 4.6 mm). Detection was achieved at 262 nm with a flow rate of 1 mL/min.

Pramipexole plasma concentrations were determined by a validated HPLC-MS/MS method (high-performance liquid chromatography coupled with a triple quadrupole mass spectrometer). In brief, the analytical method involved a robotized solid phase extraction in the 96-well plate format (Oasis MCX 30 mg), followed by reversed phase liquid chromatography (gradient mode, Zorbax SB CN column, column dimension 4.6 x 50 mm, particle diameter 3.5 µm, column temperature 35°C) of the extract with tandem mass spectrometric detection. D7-pramipexole was used as internal standard. The mobile phase consisted of acetonitrile (solvent A) and 0.05 M ammonium formate pH 7.8 (solvent B), and the flow rate was 0.75 mL/min. The following ion transitions were monitored: for pramipexole m/z 212.0–153.1 and for D7-pramipexole m/z 219.1–153.1.

Both pramipexole and D7-pramipexole reference material was synthesized and certified at Boehringer Ingelheim Pharma GmbH & Co. KG.

The analytical method in human plasma (EDTA) was shown to be linear from 0.050 to 15.0 ng/mL. Concentrations were determined using the slope and the intercept of the calibration line obtained by least square regression using the appropriate weighting factor ($1/x^2$). Each batch included six quality control (QC) samples (in duplicate) at three concentration levels: one near the lower limit of quantification (QC1: 0.150 ng/mL), one in the mid-range (QC2: 2.00 ng/mL) and one near the upper limit of quantification (QC3: 12.0 ng/mL). The inaccuracy and imprecision of the data obtained was below 5.00% and 5.09%, respectively.

Data Analysis

The First Order Conditional Estimation (FOCE) method with the option INTERACTION implemented in the software NONMEM (ICON, Ellicott City, MD, USA) version VI (15) was used during the analyses.

The minimum value of the objective function provided by NONMEM, approximately equal to -2 x log likelihood [-2LL] was used to guide model development. For two nested models, a decrease in 6.63 points in -2LL for an extra parameter was considered significant at the 1% level. In the case of non-nested models, -2LL was not used directly for comparative purposes, and the value of the Akaike Information Criteria (AIC) (16) computed as -2LL+2 x Np, where Np is the number of the parameters in the model, was used instead. The model with the significant lowest -2LL (or AIC) value and acceptable parameter precision supported by the goodness-of-fit plots was finally selected. The precision of parameter estimates was expressed as coefficient of variation [(CV (%)], computed as the ratio between the standard error and the model estimate and multiplied by 100.

Inter-individual variability (IIV) was modelled exponentially. Residual variability was described using the combined error model. If during the analyses one of the components of the combined error model, the additive or the proportional element, was found to be negligible, it was deleted from the model.

A visual predictive check (17) was used to evaluate the selected final model. One thousand datasets with the same study design characteristics as the original dataset were simulated. For each dataset, the 2.5th, 50th, and 97.5th percentiles of the simulated pramipexole plasma concentrations were calculated by sampling time. Then, for each of those percentiles, the 95% prediction intervals were computed and represented graphically as a function of the visit. The same procedure was performed with the raw data, and the agreement between simulations and observations was inspected visually.

The following steps were taken to develop IVIVC for pramipexole.

Step A: Modelling the In Vitro Release of Pramipexole

The dissolution profiles for each of the SR formulations were fitted separately. Four models for *in vitro* release were tested: a zero-order and a first-order release rate model as well as models describing the release by a cubic root and the Weibull function (18). For the case of the IR formulation, 80% of pramipexole was released 30 min after the start of the experiment (data not shown), and its release was assumed to be instantaneous.

Step B: Pharmacokinetic Modelling After Administration of the IR Formulation

The absorption process after IR administration was described using a first-order rate model. The presence of a latency time was explored, and the typical population estimate for the bioavailability was arbitrarily considered to be one (in the absence of an intravenous administration), but it was allowed to vary from subject to subject. Disposition of pramipexole in plasma was characterized by compartmental models parameterized in apparent volumes of distribution and elimination and distribution clearances.

Step C: Description of Plasma Profiles of Pramipexole After Administration of the SR Formulations

The IVIVC was established using data from the $SR_{1\text{-}3}$ formulations; the slow-release formulation SR_4 was used for external validation purposes.

C.1. The disposition of pramipexole in plasma after administration of the SR formulations was simulated based on the models and model parameters selected and estimated in steps A and B, and represented graphically together with the raw data. The agreement between the observations and the simulated profiles was judged visually. For this simulation, it was assumed that the kinetics of the *in vitro* release and *in vivo* release were identical, and once the drug is dissolved in the gastrointestinal tract, it behaves as the IR formulation (without consideration of the latency time). The model is presented schematically in Fig. 1.

C.2. As shown in the results section, the simulations performed in C.1 did not adequately describe the observed pharmacokinetic profiles of the $SR_{1\text{-}3}$ formulations.

The model parameters corresponding to the release model were estimated *in vivo*, using the model selected in step A and the model (and its parameter estimates) in step B, and the pramipexole plasma concentration after the $SR_{1\text{-}3}$ administration.

Then, a common relationship between the *in vivo* and *in vitro* release model parameter estimates was established for

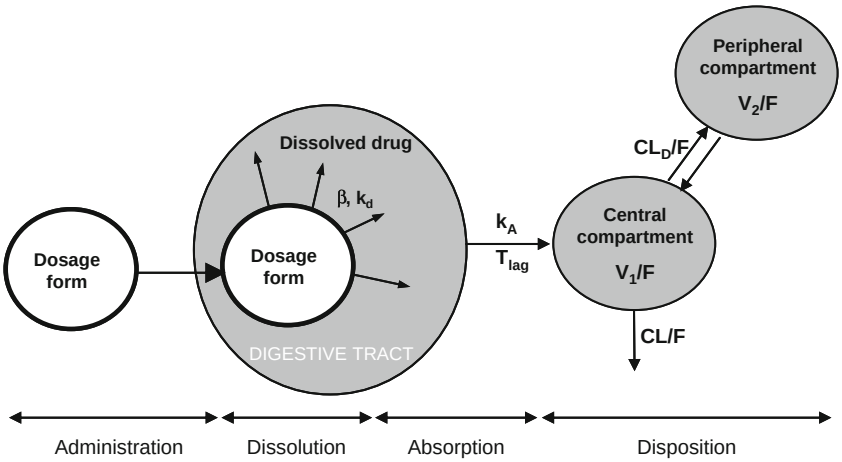

Fig. 1. Schematic representation of the biopharmaceutic/pharmacokinetic model established for the IVIVC of pramipexole. Parameters are defined in the text and in Tables I and II.

the $SR_{1\text{-}3}$ formulations, and the same simulation exercise as described above in section C.1 was performed. Differences in relative bioavailability between IR and SR formulation were also explored at this step.

Step D: Evaluation of the IVIVC

The IVIVC developed in the previous step was evaluated internally using the $SR_{1\text{-}3}$ formulations and externally using the SR_4 formulation, which was not used during the development of the IVIVC. For both evaluations, the following steps were performed.

D.1. The mean C_{MAX} (maximum concentration of pramipexole in plasma), AUC_{last} (area under the plasma *vs* time concentration curve calculated from time 0 to time 30 h), and $AUC_{0\text{-}\infty}$ (area under the plasma *vs* time concentration curve calculated from time 0 to infinity) were obtained from the individual predictions. The relative percentage of the prediction error (%PE) was then calculated using the following expression:

$$\%PE = \frac{|Predicted - Observed|}{Observed} \times 100$$

where *Predicted* is the mean C_{MAX}, AUC_{last} or $AUC_{0\text{-}\infty}$ of the predicted profiles, and *Observed* represents the corresponding mean values obtained from the raw data. The FDA has established that this %PE should not exceed 15% for each formulation and that the average percentage of the prediction error for all the formulations studied has to be less than 10% to indicate predictability of the IVIVC.

D.2. The relationship between *in vitro* release and *in vivo* absorption was evaluated. For each subject, and at the times at which the *in vitro* data were measured, the model predicted percentages of released and absorbed drug were calculated. Ideally, the relationship between the percentages dissolved and absorbed should be the same for all SR formulations, assuming immediate and complete absorption of any released pramipexole (the release rate is slow compared to the absorption rate so that the release rate dominates the absorption process).

RESULTS

Step A: Modelling the In Vitro Release of Pramipexole

For all SR formulations, the Weibull model without a latency time, represented by the following equation, provided better fits than other *in vitro* release models tested:

$$\frac{dQ}{dt} = Q_\infty \times kd^\beta \times \beta \times t^{(\beta-1)} \times e^{-(kd\times t)^\beta}$$

where dQ/dt represents the release rate of pramipexole, Q_∞ corresponds to the initial drug amount in the formulation (dose), t is the time after the start of the *in vitro* dissolution test, β is the shape parameter, and k_d corresponds to a dissolution rate constant.

As an example, the values of AIC for the case of the SR_1 formulation were 649 (Weibull model), 1936 (first-order model), 2214 (cubic-root model), and 2736 (zero-order model). For all the SR formulations, inclusion of variability between replicates was significant ($p<0.01$) for k_d but not for β ($p>0.05$), and the residual error was best described with an additive error model for all the SR formulations.

These formulations present three different release profiles: SR_1 was the fastest, SR_3 was the slowest and SR_2 and SR_4 were in between and had almost identical release profiles.

Table I shows the model parameter estimates for all four SR formulations. The low CV% indicates good precision of the parameter estimates. The results from the visual predictive check (Fig. 2) confirm that the model selected describes the data of all SR formulations adequately.

The estimate of the β parameter was similar for all the SR formulations, indicating that all the SR formulations have the same release mechanism; however, k_d was formulation-dependent, indicating that the formulations had different release rates.

Step B: Pharmacokinetic Modelling After Administration of the IR Formulation

The pharmacokinetic profiles of the immediate-release formulation were adequately described by a two-compart-

Table I. Population *In Vitro* Dissolution Parameter Estimates (RSE) for Pramipexole

Parameter	Formulation			
	SR_1	SR_2	SR_3	SR_4
β	0.692 (0.01)[a]	0.732 (0.01)	0.765 (0.01)	0.714 (0.06)
k_d (h^{-1})	0.106 (0.02)	0.0756 (0.02)	0.0568 (0.02)	0.0738 (0.01)
IIV k_d (%)	6.73 (0.30)	7.68 (0.49)	7.37 (0.46)	5.38 (0.32)
Additive error (% dissolved)	0.791 (0.08)	1.15 (0.07)	1.24 (0.07)	1.05 (0.06)

[a] Parameters are listed as estimates together with the corresponding relative standard error within parenthesis. β, Weibull shape parameter; k_d, first order rate constant of dissolution; IIV, inter-individual variability.

ment disposition model with a first-order absorption rate incorporating a latency time (t_{lag}). It was found necessary to account for IIV on the apparent volume of distribution of the central compartment (V_1/F), the apparent total plasma elimination clearance (CL/F), the first-order rate constant of absorption (K_A), and t_{lag} ($p<0.01$).

Table II shows the population model estimates after the administration of the IR formulation, where it can be seen that disposition parameters of pramipexole in plasma show low IIV in contrast to the parameters describing drug absorption.

Fig. 3 shows two standard goodness-of-fit plots confirming the adequacy of the model in describing the typical and individual concentration profiles; the latter was additionally supported by the low value of % ε-shrinkage (19). The results obtained from the visual predictive check represented in Fig. 4 also indicate that the model captures the mean tendency and dispersion of the data very well.

Step C: Description of Plasma Profiles of Pramipexole After Administration of the SR Formulations

C.1. First it was assumed that the kinetics of the *in vitro* release and *in vivo* release were identical, and once the drug is dissolved in the gastrointestinal tract, it behaves as the IR formulation. Simulations based on the models and model parameters obtained from Steps A and B were performed and presented in Fig. 5A areas covering the 95% of the confidence intervals for the median and the 2.5th and 97.5th percentile. In Fig. 5A, it is clearly shown that simulations did not adequately describe the observed pharmacokinetic profiles for any of the $SR_{1\text{-}3}$ formulations, since the plasma concentrations of pramipexole in all SR formulations were under predicted.

C.2. Based on these results, the k_d and β parameters corresponding to the release model were estimated *in vivo*

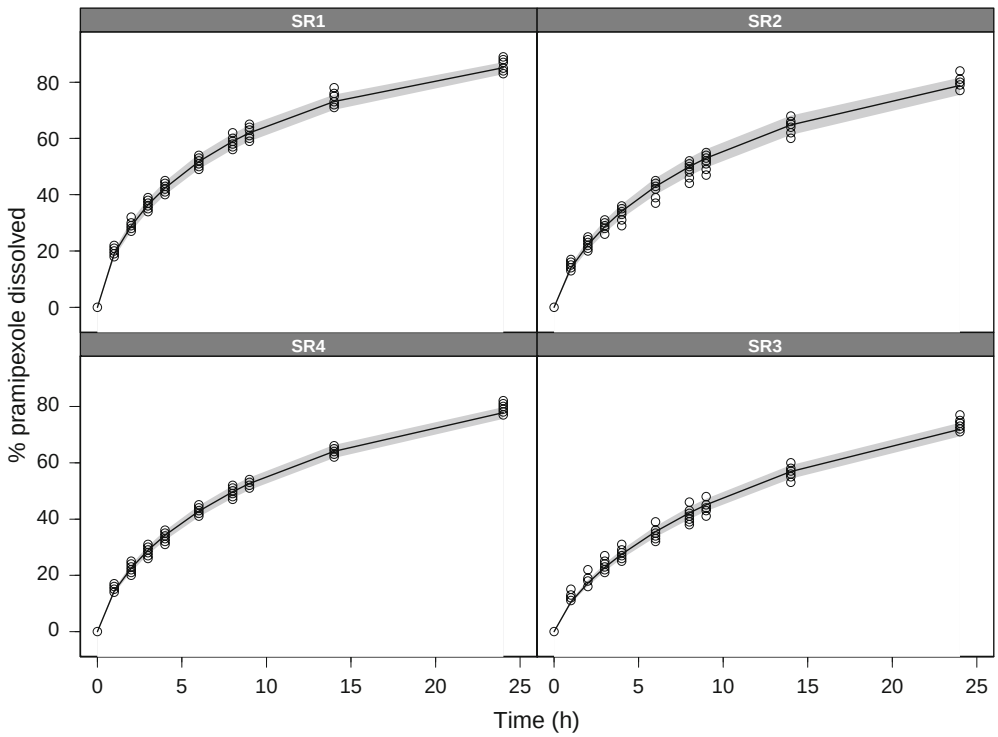

Fig. 2. Visual predictive check for the pramipexole SR formulations dissolution model. Open circles, observations; solid line, median model predictions. The grey area covers the 90% of the simulated observations.

Table II. Population Pharmacokinetic Model Parameter Estimates (RSE) After Administration of the Pramipexole IR Formulation

Parameter	Estimate	IIV (%)
V_1/F(L)	351 (0.05)	14.1 (0.31)
CL/F (L/h)	35.9 (0.03)	10.9 (0.34)
V_2/F(L)	60.9 (0.21)	–
CL_D/F (L/h)	33.2 (0.40)	–
K_A (h^{-1})	5.26 (0.32)	91.8 (0.46)
t_{lag} (h)	0.220 (0.19)	66.3 (0.46)
Proportional error (%)	15.7 (0.17)	–

Parameters are listed together with the relative standard error in parenthesis. V_1/F, V_2/F, apparent volumes of distribution in the central and peripheral compartments, respectively; CL/F, apparent total elimination plasma clearance; CL_D/F, apparent distribution clearance; K_A, first-order rate constant of absorption; t_{lag}, latency time; IIV, inter-individual variability expressed as relative standard error (%).

(based on the pramipexole plasma concentration profiles after the SR administration) using the release model selected in Step A and the pharmacokinetic model and its parameter estimates (Table II) from Step B.

The estimates of those *in vivo* release parameters together with the release parameters obtained *in vitro* are listed in Table III. The estimates corresponding to the shape parameter (β) were similar between *in vitro* and *in vivo* estimation (mean of 0.73 *vs* 0.77, respectively); however, the *in vivo* estimates of k_d were consistently higher than those obtained from the *in vitro* analysis (mean 0.08 *vs* 0.13 h^{-1}), indicating that the rate of dissolution was increased *in vivo*.

The increase in k_d was then considered in a model in which the k_d was described as $k_{d\_in\_vitro} + \theta_{SCL}$, where for each SR formulation, $k_{d\_in\_vitro}$ corresponds to the estimates shown in Table I, and θ_{SCL} represents a scale factor common to all SR formulations. The estimate [CV (%)] of θ_{SCL} was 0.0581 (13.75%). This model was able to better describe the plasma concentration data as can be observed in Fig. 5B, where the same simulation exercise as described above for Fig. 5A was performed.

Alternatively a model using the expression $k_{d\_in\_vitro}$ x θ_{SCL} was also fitted to the data, resulting in a worse fit. A similar procedure to the one applied to k_d was considered to the β parameter, but data description was not improved.

Another possible explanation for the underprediction shown in Fig. 5A could be due to a greater bioavailability of the SR formulations relative to the IR formulation. A model incorporating a scale factor (20) (and removing θ_{SCL} from the model) was then explored, but it resulted in a worse fit.

Step D: Evaluation of the IVIVC

D.1. The results shown in Table IV and Fig. 6 serve as additional support to confirm the validity of the established IVIVC. The calculated %PE values for the C_{MAX}, AUC_{last} and AUC_∞ were in both cases and for all the SR formulations lower than 10% (Table IV). The relationship between the percent of dissolved pramipexole *in vitro* and the percentage of absorbed pramipexole *in vivo* is linear and independent of the type of formulation administered (Fig. 6).

D.2. Table IV and Fig. 7 show the ability of the model to predict data from the formulation (SR_4) that was not used to develop the IVIVC model. Prediction errors for C_{MAX}, AUC_{last} and AUC_∞ were also lower than 10% for this formulation.

DISCUSSION

An IVIVC was established for pramipexole slow-release formulations by developing a model that combines the

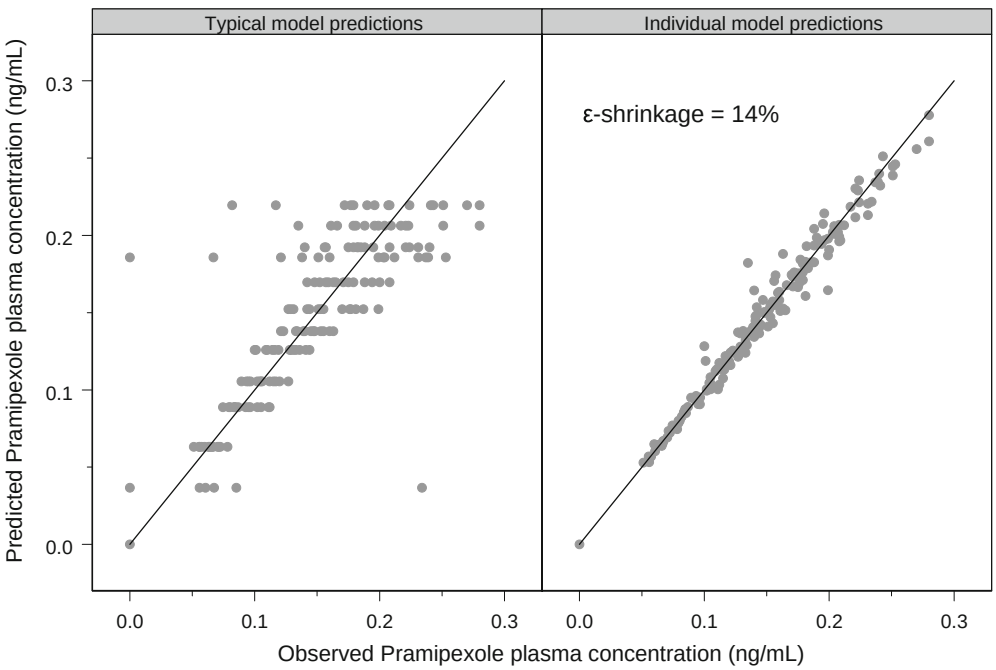

Fig. 3. Goodness-of-fit plots corresponding to the selected population pharmacokinetic model for the IR pramipexole formulation. Solid lines represent the lines of identity.

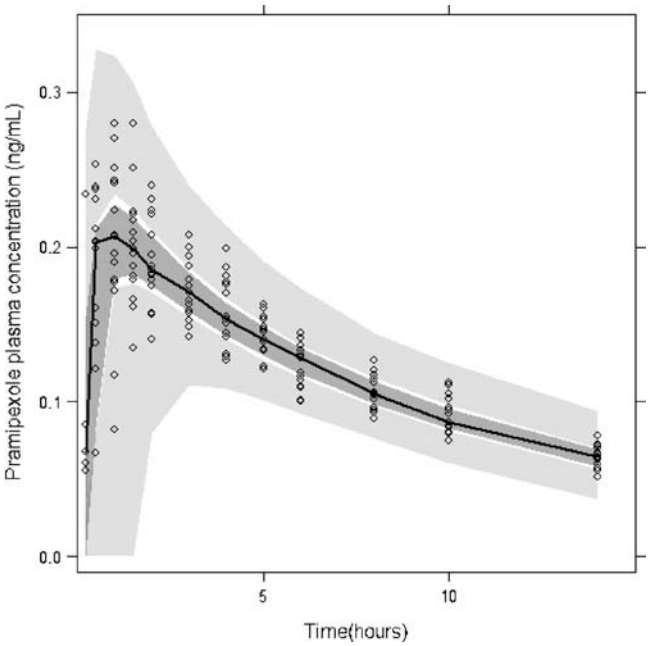

Fig. 4. Visual predictive check for the pramipexole pharmacokinetic IR model. Area covering the 95% confidence interval for the median (*dark grey*), and of the 2.5th and 97.5th percentile for the simulations is shown together with the median (*solid line*) of the observations and with the observations (*circles*).

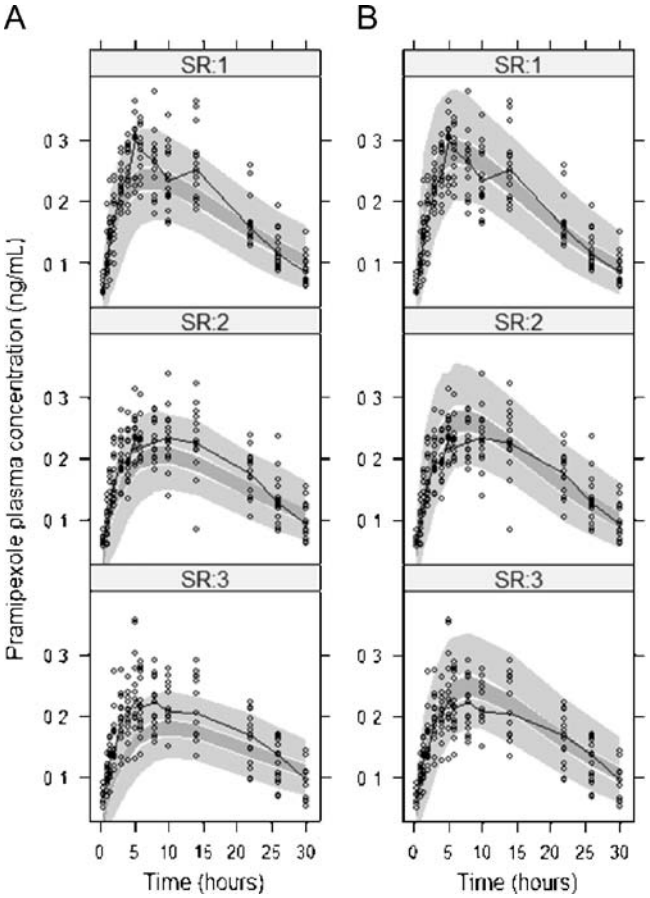

Fig. 5. Simulated plasma profiles of pramipexole after administration of the $SR_{1\text{-}3}$ formulations. Area covering the 95% confidence interval for the median (*dark grey*), and of the 2.5th and 97.5th percentile for the simulations is shown together with the median (solid line) of the observations and with the observations (*circles*). **A**, profiles corresponding to simulations performed following procedure described in Step C.1; **B**, profiles from the final selected IVIVC model obtained in Step C.2.

different dissolution mechanism of the formulations with the pharmacokinetic characteristics of pramipexole obtained after administration of an immediate-release formulation. For this approach, no deconvolution of the *in vivo* concentration profiles to obtain the percentage of absorbed drug was required. This avoids the disadvantages of deconvolution: (*i*) the process is unstable and (*ii*) when using deconvolution, the model predicts the fraction of the dosage form dissolved *in vivo*, which is not the primary focus of attention; instead, plasma drug concentration is of interest (21). The modelling approach presented here is similar to a convolution-based method, but instead of using integrals, the corresponding differential equations were used as described previously (22). Also, the typical concentration time profiles for different SR formulations and the corresponding prediction intervals (due to the consideration of the interindividual variability) can be described. The approach used in the current analysis is based on the assumption that dissolution is the limiting process for absorption, i.e. once the drug has been released from the formulation, the absorption process would be the same regardless the type of preparation. In the case of drugs for which absorption occurs at specific areas in the gastrointestinal tract, this approach would not be equally valid.

The analysis of the *in vitro* data revealed that for all SR formulations, the model selected was the Weibull model. This model has been widely used to describe (*i*) the *in vitro* release kinetics of many different dosage formulations (23–26) and (*ii*) although less frequently, the *in vivo* drug absorption process (27–30).

The estimates of the β parameter indicate that the mechanism of drug release is similar between all SR formulations. The estimate of the release rate constant is represented by the k_d parameter, which seemed responsible for the differences seen in the release profiles. These results suggest that modifications in the composition, mainly the percentage of carbomer, affect the release rate only and do not alter the overall release mechanism of the matrix tablet. The results presented in the current study indicate that low percentages of carbomer in the formulation are associated with a faster release, while higher percentages resulted in a slower release.

The disposition pharmacokinetic characteristics of pramipexole could be described by a two-compartment model. Previously, it has been reported that pramipexole has linear pharmacokinetics over the entire therapeutic range, with an apparent volume of distribution of 7 L/kg (490 L for a 70 kg

Table III. Dissolution Parameter Estimates Using *In Vitro* and *In Vivo* Data

	β		Kd	
Formulation	*In vitro*	*In vivo*	*In vitro*	*In vivo*
SR_1	0.692	0.841	0.106	0.166
SR_2	0.732	0.752	0.0756	0.127
SR_3	0.765	0.724	0.0568	0.111
Average	0.73	0.77	0.08	0.13

Table IV. Values of the Absolute Percentage of the Prediction Error (%PE) for C_{MAX} (ng/mL) and AUC_{∞} (ng x h/mL)

Formulation		Internal Validation				External Validation
		SR_1	SR_2	SR_3	Mean % PE	SR_4
Observed	**C_{MAX}** (ng/mL)	0.288	0.239	0.241	–	0.253
	AUC_{last} (ng x h/mL)	5.903	5.553	5.270	–	5.702
	AUC_{∞} (ng x h/mL)	6.864	6.628	6.287	–	6.682
Predicted	**C_{MAX}** (ng/mL)	0.284	0.264	0.250	–	0.261
	AUC_{last} (ng x h/mL)	5.846	5.667	5.509	–	5.628
	AUC_{∞} (ng x h/mL)	6.745	6.745	6.632	–	6.700
% PE	**C_{MAX}**	1.378	10.38	3.747	5.17	3.08
	AUC_{last}	0.967	2.06	5.509	2.85	1.31
	AUC_{∞}	0.365	1.77	6.632	2.92	0.27

C_{MAX} , maximum plasma concentration; AUC_{last}, the area under the plasma *vs* time concentration curve calculated from time 0 to 30 h; AUC_{∞}, the area under the plasma *vs* time concentration curve calculated from time 0 to infinity.

individual) and clearance of 30 L/hour (30). These parameter estimates are similar to the ones estimated in this study: total apparent volume of distribution of 412 L and a clearance of 35.9 L/hour. Pramipexole is well-absorbed after oral administration, with a bioavailability of more than 90% and a half life ranging from 8 to 12 h (31).

During the development of the IVIVC, it was found that the *in vitro* and *in vivo* dissolution processes occurred at different rates. There is not a 1-to-1 relationship between the percentage of dissolved drug *in vitro* and the percentage *in vivo*. Our results indicate that the dissolution is faster *in vivo*, implying that the gastrointestinal physiological conditions, which are more complex than the *in vitro* conditions, accelerate the dissolution process. Despite that discrepancy, our modelling strategy allows us to identify the parameter differing between *in vitro* and *in vivo* and scale it properly, achieving predictions consistent with the observations, as presented in Fig. 5B. In fact, our final model indicates that for the SR formulations studied, the difference between the *in vivo* and *in vitro* k_d is 0.0581. Such difference in k_d is reflected in the time scaling factor, calculated as the ratio between the time required for 50% absorption *in vivo* and the time required for the 50% dissolved *in vitro*, computed using the model developed (10,32). The results were 0.64, 0.50, 0.50 and 0.50 for the $SR_{1\text{-}4}$, respectively.

A linear relationship between the observed percentage of dissolved drug and the calculated percentage of absorbed drug was found to be independent from the type of SR formulation administered (see Fig. 6).

In this study, a level A IVIVC was successfully established, since, using our approach, the entire concentration time profile can be predicted for all the SR formulations analyzed. Internal validation was performed by calculating the percentage of the absolute prediction error for C_{MAX}, AUC_{last} and $AUC_{0\text{-}\infty}$. According to the FDA guidance, an

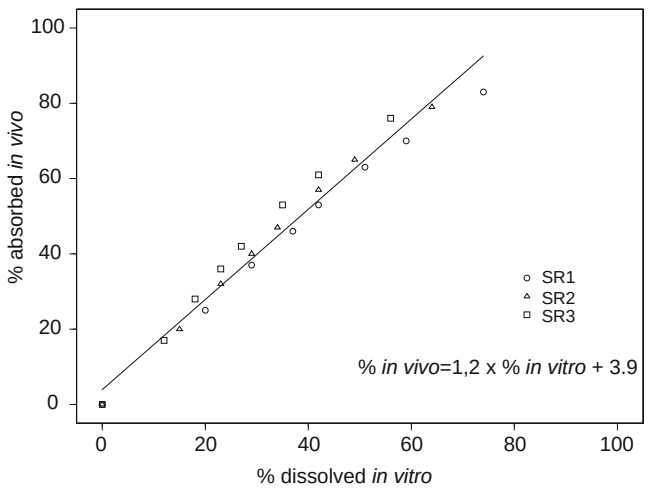

Fig. 6. Percentage of model predicted *in vivo* absorbed pramipexole *vs* percentage of *in vitro* dissolved pramipexole for the $SR_{1\text{-}3}$ formulations.

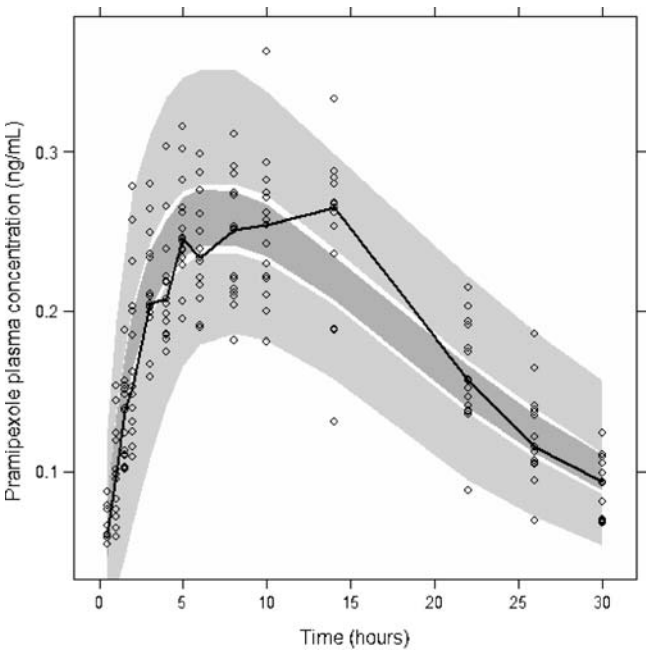

Fig. 7. External validation for the IVIVC model using data from the SR_4 formulation. Visual Predictive Check, area covering the 95% confidence interval for the median (*dark grey*), and of the 2.5th and 97.5th percentile for the simulations is shown together with the median (solid line) of the observations and with the observations (*circles*).

IVIVC is adequate when the absolute percent prediction error (%PE) for C_{MAX} or AUC for each formulation does not exceed 15%, and when the average %PE of all the formulations is lower than 10%. In our case, the %PE for C_{MAX} or AUC for each formulation was below 11%, and the average was lower than 6%, respectively. Since the FDA criteria were met for the internal validation, no external validation was required, even though the IVIVC model was used to predict SR_4 formulation, satisfying the requirements.

Considering that pramipexole is a non-narrow therapeutic index drug and that the IVIVC was developed with formulations with three different release rates, *in vitro* dissolution testing can be used as a biowaver as defined by SUPAC-MR for level 3 changes in manufacturing process, changes in the release controlling excipients and removal of or replacement of non-release controlling excipients (33) .

To summarize the results from the current study, a level A IVIVC adequately describing the *in vivo* plasma pharmacokinetic profiles of pramipexole administered in four different slow-release formulations was established based on the release properties obtained from the *in vitro* investigations and the pharmacokinetic properties obtained after administration of an immediate-release tablet. The IVIVC developed makes pramipexole dissolution profiles more meaningful, as it allows for predicting their impact on the pharmacokinetics and for the replacement of bioequivalence studies in situations defined by the SUPAC-MR guideline.

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