Biopharmaceutic In Vitro In Vivo Extrapolation (IVIV_E) Informed Physiologically-Based Pharmacokinetic Model of Ritonavir Norvir Tablet Absorption in Humans Under Fasted and Fed State Conditions

Sumit Arora
Senior Research Scientist

Simcyp Consortium Members Webinar
18.06.2020

Outline of the Presentation

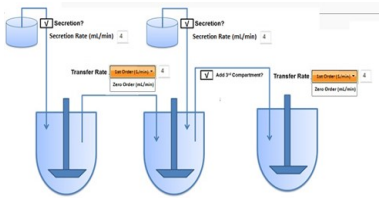
- Aim, Background and Challenges
- Model (dis)qualification in a stepwise manner
- Results and Discussion
- Conclusions and Limitations

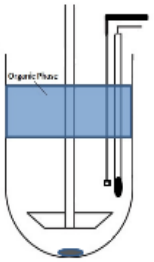
Aim of the current research work

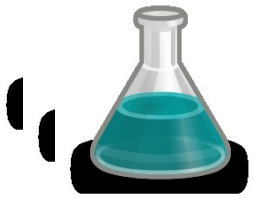
The objective of this research is to develop a mechanistic absorption and disposition model of ritonavir which could link the biopharmaceutic properties of the drug/drug product with its DDI risk potential

CERTARA

Running Theme – Biopharmaceutics *In vitro In vivo* Extrapolation (IVIVE)



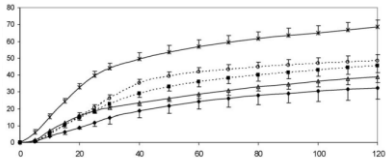




SIVA Toolkit

***In vitro* System parameters**
- RPM (Fluid velocity)
- Buffer (e.g., Phosphate)
- Media (pH, [Bile Salts] …)

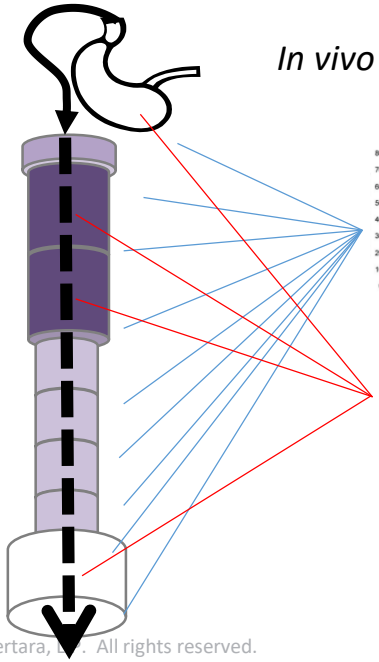
In vitro dissolution

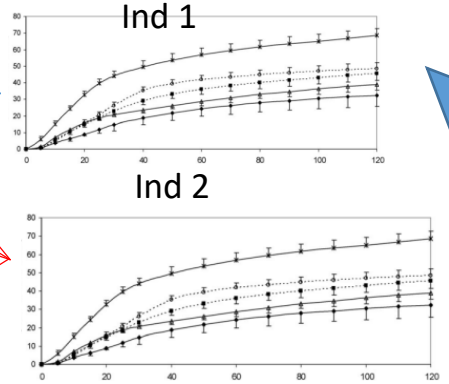


API Parameters
- S_0, pKa, SR, PRC
- $LogK_{m:w}$
- Particle size *etc.*
- DLM scalar (*S*)
- Supersaturation/Precipitation

$$DR(t) = -NS\frac{D_{eff}}{h_{eff}(t)}\,4\,\pi\,a(t)\,(\,a(t)+h_{eff}\,(t))(S_{surf}(t)-C_b\,(t)\,)$$

In vivo dissolution





***In vivo* System parameters + Pop. variability**
- Luminal fluid velocities
- Buffer (bicarbonate)
- Luminal pH, [Bile Salts] …

Simcyp Simulator

CERTARA

Biopharmaceutics IVIVE – Many Case Studies

molecular pharmaceutics

Article

In Silico Modeling Approach for the Evaluation of Gastrointestinal Dissolution, Supersaturation and Precipitation of Posaconazole

Bart Hens, Shriram M. Pathak, Amitava Mitra, Nikunjkumar Patel, Bo Liu, Sanjaykumar Patel, Masoud

molecular pharmaceutics

Article

pubs.acs.org/molecularpharmaceutics

Model-Based Analysis of Biopharmaceutic Experiments To Improve Mechanistic Oral Absorption Modeling: An Integrated *in Vitro in Vivo* Extrapolation Perspective Using Ketoconazole as a Model Drug

Shriram M. Pathak,*,† Aaron Ruff,‡ Edmund S. Kostewicz,‡ Nikunikumar Patel,† David B. Turner,† and Masoud Jamei†

ELSEVIER

Contents lists available at ScienceDirect

Journal of Pharmaceutical Sciences

journal homepage: www.jpharmsci.org

Pharmacokinetics, Pharmacodynamics and Drug Transport and Metabolism

Biopharmaceutic *IVIVE*—Mechanistic Modeling of Single- and Two-Phase *In Vitro* Experiments to Obtain Drug-Specific Parameters for Incorpora

Shriram M. Pathak [1, *], Kerstin J

molecular pharmaceutics

pubs.acs.org/molecularpharmaceutics

Article

Biopharmaceutic *In Vitro In Vivo* Extrapolation (IVIV_E) Informed Physiologically-Based Pharmacokinetic Model of Ritonavir Norvir Tablet Absorption in Humans Under Fasted and Fed State Conditions

Sumit Arora,* Amita Pansari, Peter Kilford, Masoud Jamei, Iain Gardner, and David B. Turner

Challenges with the PBPK model development with Ritonavir

- Complex pharmacokinetic profile – mechanism-based inhibitor of CYP3A4/5, inducer of CYP3A4/5, P-gp substrate and also an inhibitor
- Lack of intravenous data (parameter identifiability issues) and information regarding fraction metabolized by CYP3A4
- Commercial formulation is an amorphous solid dispersion (ASD). Dissolution of ASDs under non-sink conditions is complex with the possibility of several phase transitions within the dissolving medium and the ASD matrix
- Being a weak base, parameterization of models which adequately capture interplay between supersaturation and precipitation of ritonavir in gut lumen can be complex

We adopted a Model (dis)qualification approach in a stepwise manner until consistency with clinical data is obtained

Can we simulate ritonavir in amorphous state?

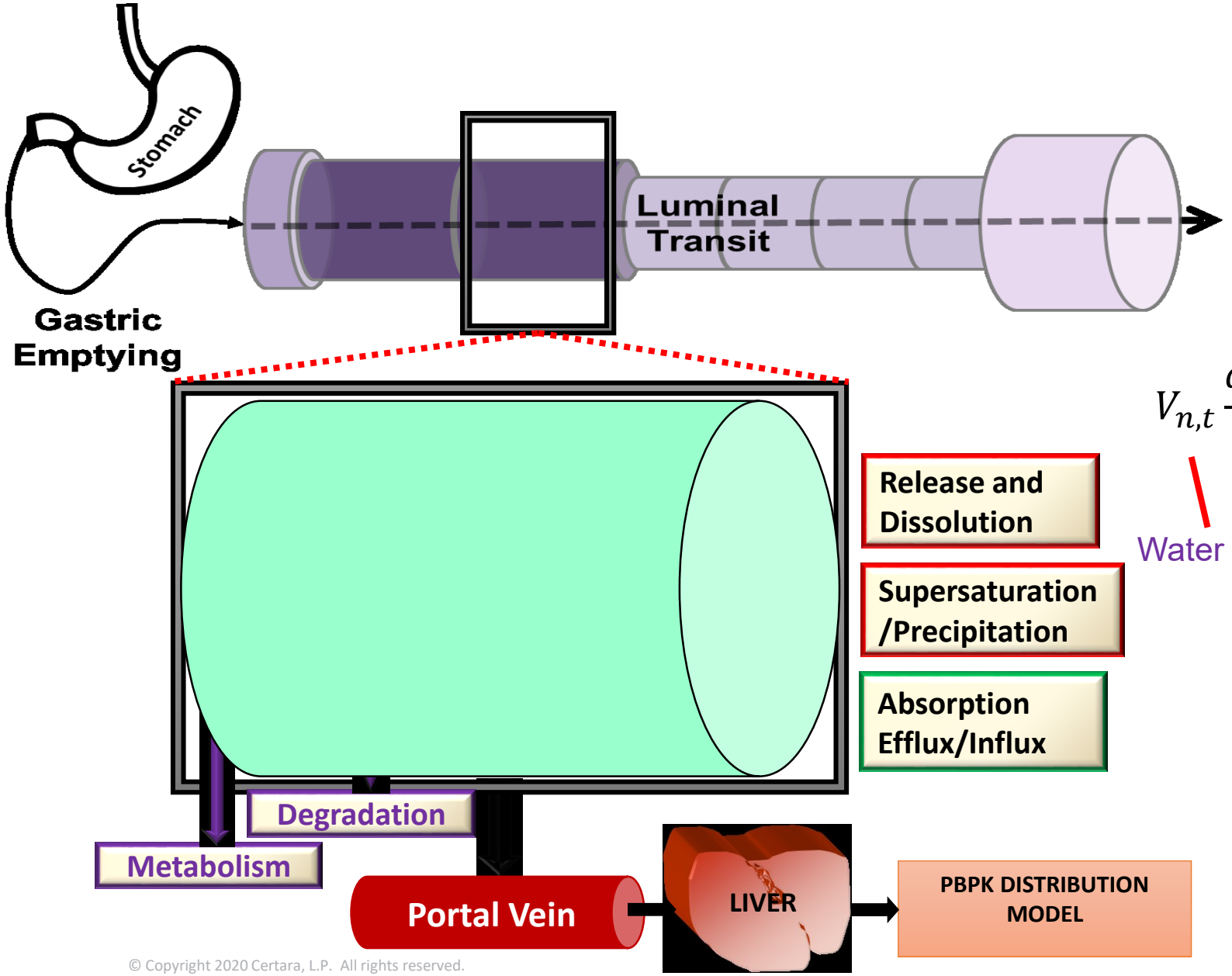
- Norvir® 100 mg tablet - amorphous solid dispersion of ritonavir with copovidone (PVP –VA) (15% drug load)
- T_g of ritonavir 50ºC[a] – indicative of inherent stability of amorphous ritonavir at room temperature
- Hancock[b] classified ritonavir as molecule with low propensity for crystallization (MW – 721 Da, 18 rotatable bonds)
- Indulkar et al[c] – Nucleation induction time (time taken to reduce initial supersaturation concentration by 2.5%) of ritonavir – 700 min in composite SIF and around 300 min in FaSSIF-V1

Hence, it is quite likely that ritonavir remains in amorphous state during its transit in gut

[a] Zhou D et al, J Pharm Sci, 2007;96(1):71-83, [b] Hancock J Pharm Sci 2017, 106(1):28-30, [c] Indulkar et al Pharm Res (2018) 35:158

CERTARA

Advanced Dissolution Absorption and Metabolism(ADAM) Model



Absorption rate from n^{th} ADAM segment

$$V_{n,t}\frac{dC_{Total,n,t}}{dt} = -A_{Total,diss,n,t}\cdot\frac{2}{R_{SI,n}}P_{eff,n,t}$$

SA to vol. ratio

ka

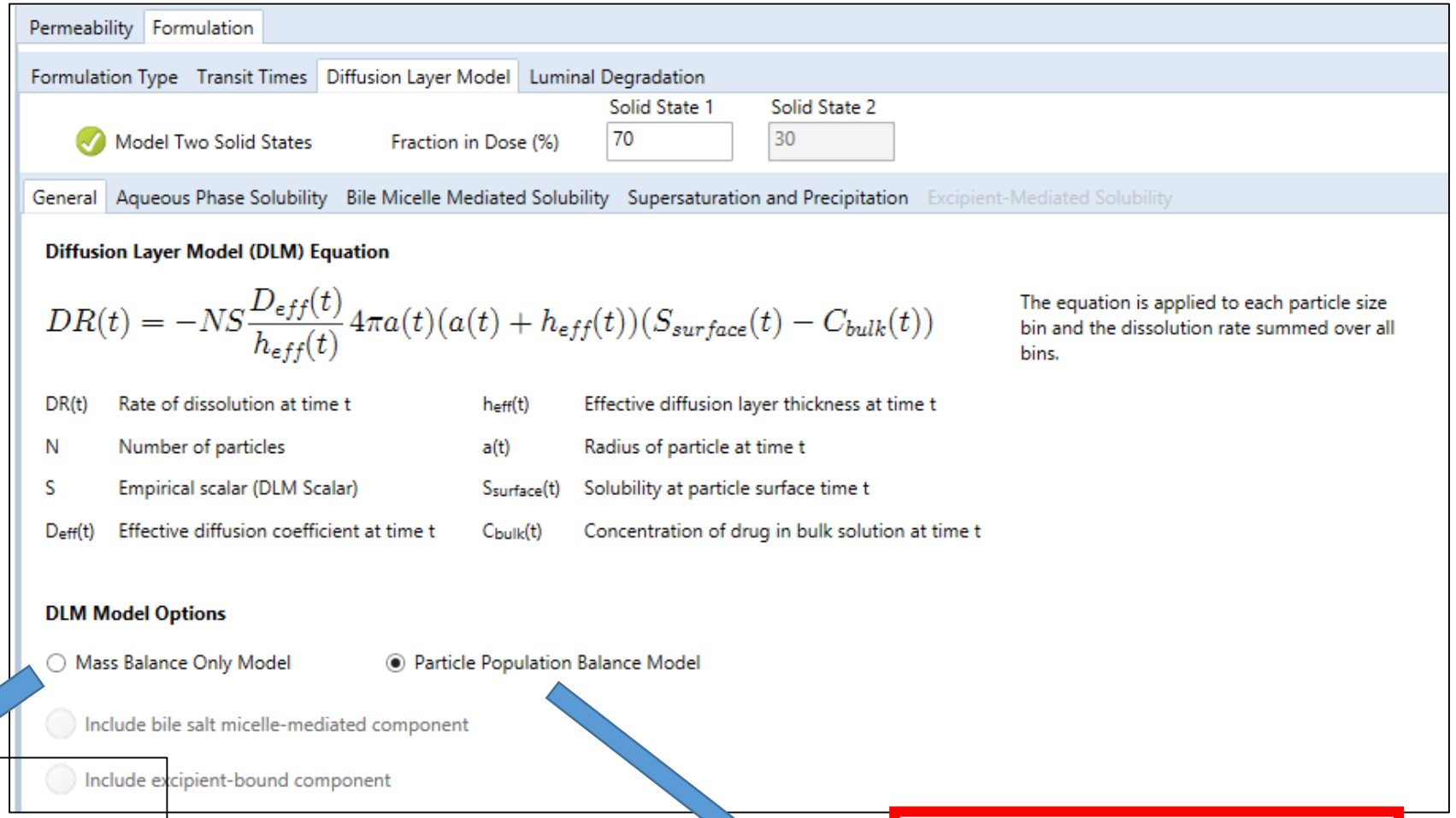
Water Volume

Total Amount dissolved

Sink assumption for enterocyte concentration

CERTARA

Two Models for Handling Dissolution of Particles



MBO
v17 and before

Retained in v18 for backward compatibility

PPB
From v18

More Mechanistic
More features to consider

Particle Population Balance (PPB) Model

- The main idea is to track the number of particles per bin.

Particles

$$\frac{dN_{i,j}(t)}{dt} = -\frac{\left[G_i(a_{j+1},t)N_{i,j+1} - G_i(a_j,t)N_{i,j}\right]}{\Delta_j} + M$$

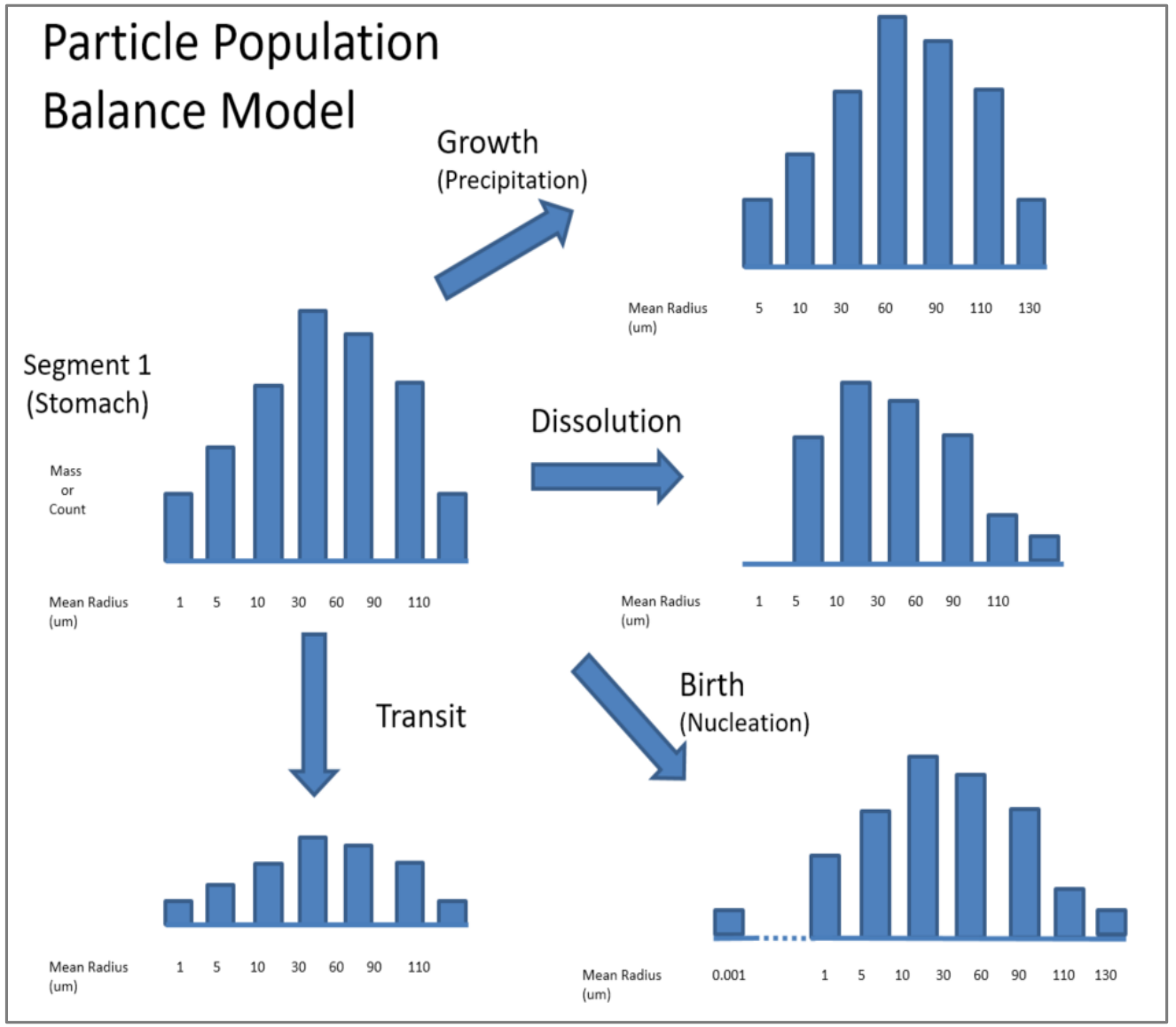
$$M = \begin{cases} -K_{t,1}N_{1,j} & i = 1 \\ K_{t_{i-1}}N_{i-1,j} - K_{t_i}N_{i,j} & i = 2 \text{ to } 8 \end{cases}$$

$$\Delta_j = a_{j+1} - a_j$$

$$N_j = n_j\Delta_j$$

$$G(a,t) = \frac{da}{dt} = -\frac{D_{eff}}{\rho h}\frac{(a+h)(C_s - C_i)}{a}$$

Different dissolution/growth function?



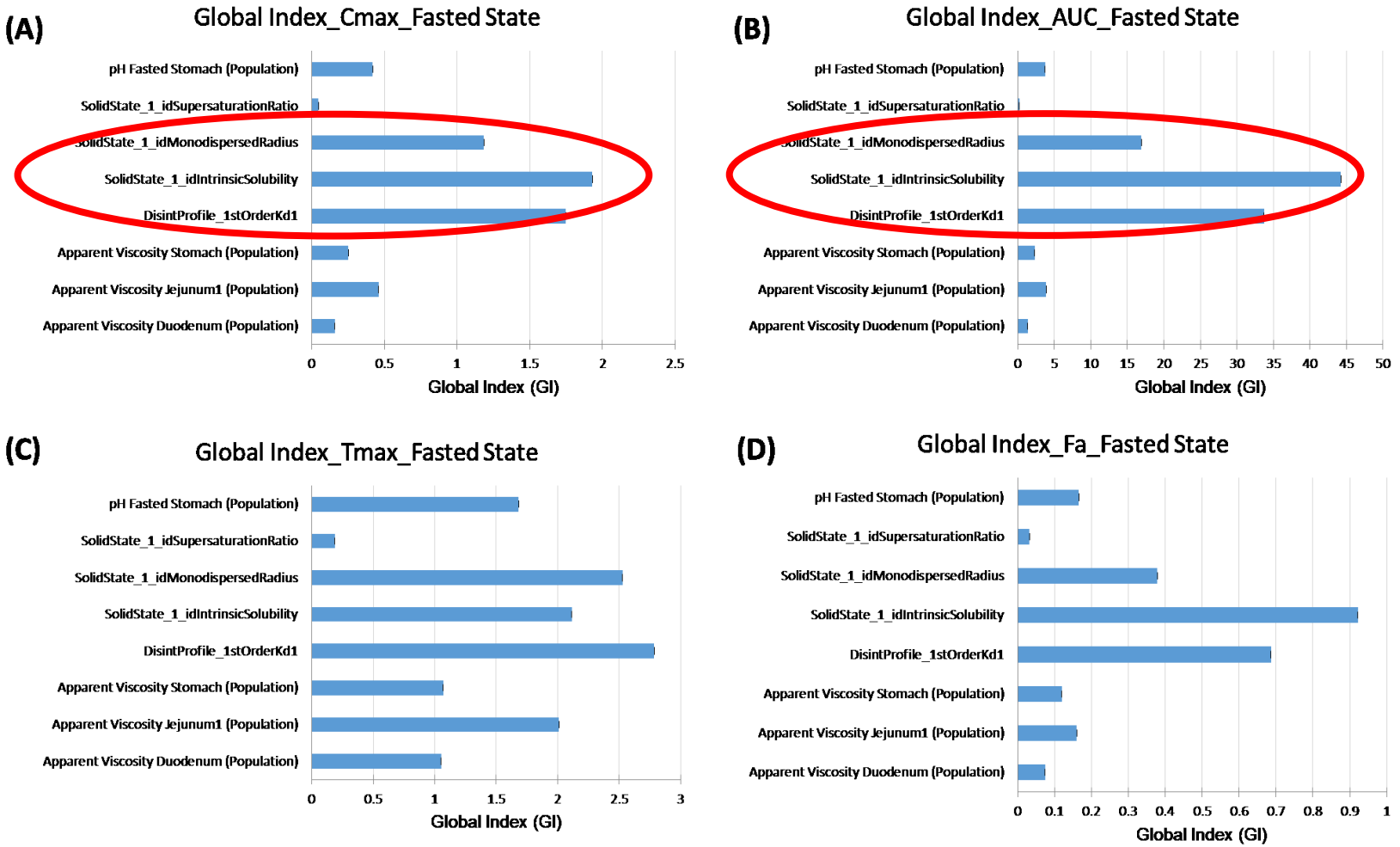
CERTARA

Main Functionalities - MBO vs PPB

	Mass Balance Only (MBO)	Particle Population Balance (PPB)
Mass balance	Y	Y
Particle count	Fixed	Particle “death” and “birth”
Particle bin handling (Polydispersed)	Discrete with gaps	Discrete with or without gaps
Maximum number of PSD bins	10 (V17), 1000 (V18)	1,000
Two PSDs	N	Y
Two solid states	N	Y (in dose and precipitating)
Excipient-mediated Solubility	Y	Y
pH-dependent & Bile Micelle Solubility	Y	Y
Particle Surface Solubility	Y	Y
Immediate/Modified/Control/Extended Release Formulations	Y	Y
Food Staggering Model	N	Y
Custom Dosing with different doses	N	Y
Segregated Transit Time model	Y	Y
First Order Precipitation	Y	Y
Mechanistic Precipitation	N	Y
Nucleation	N	Y
Luminal degradation	Y	Y

Global Sensitivity Analysis - Introduced in Simcyp V19

- The Morris method was employed which is appropriate for initial screening of parameters influencing model outcomes



GSA Analysis of chosen parameters on (A) C_{max} (B) AUC (C) T_{max} and (D) Fraction Absorbed (Fa) of ritonavir systemic exposure following administration of 100 mg Norvir® Tablet in the fasted state.

CERTARA

Sequential Modeling of *In Vitro* Biopharmaceutic Experiments

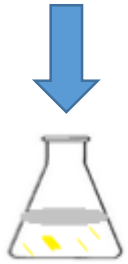
SIVA 3

Aqueous Solubility Modelling

$$S_{(BS)Tot} = \left([BS] \cdot \frac{S_0}{C_{H_2O}} \cdot K_{m:w,unionised} + S_0\right) + \left([BS] \cdot \frac{S_i}{C_{H_2O}} \cdot K_{m:w,ionised} + S_i\right)$$

Confirmed Intrinsic Solubility & Solubility Factors

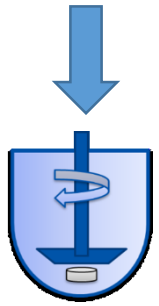
Biorelevant Solubility Modelling



$$S_{(BS)Tot} = \left([BS] \cdot \frac{S_0}{C_{H_2O}} \cdot K_{m:w,unionised} + S_0\right) + \left([BS] \cdot \frac{S_i}{C_{H_2O}} \cdot K_{m:w,ionised} + S_i\right)$$

Confirmed Bile Micelle Partition Coefficients

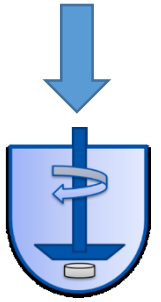
USP-2 Dissolution Modelling



$$DR(t) = -NS\frac{D_{eff}}{h_{eff}(t)}4\pi a(t)\left(a(t)+h_{eff}(t)\right)\left(S_{surface}(t)-C_{bulk}(t)\right)$$

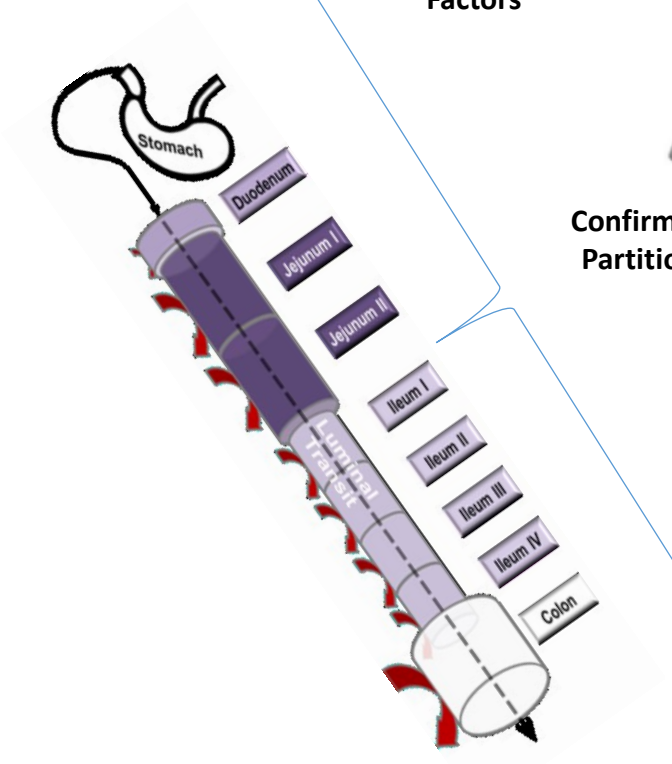
Confirmed Delayed Release and DLM Parameters

pH Shift Experiment Modelling



Critical Supersaturation Ratio[c]	1
Precipitation Rate Const. (1/h)[d]	0.001

Confirmed Precipitation Parameters



ADAM Model

Confirm and/or Estimate Parameters

CERTARA

Sequential Modeling of *In Vitro* Biopharmaceutic Experiments

Aqueous Solubility Modelling

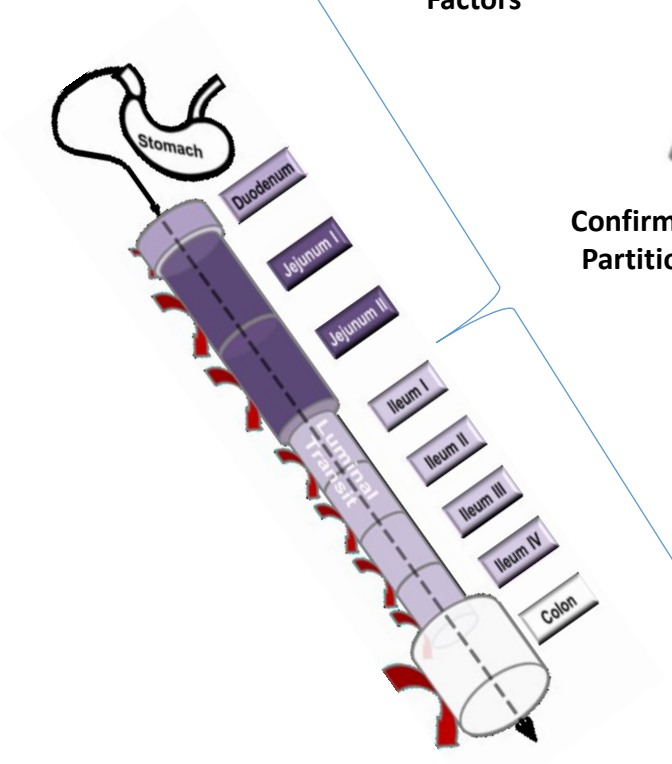
$$S_{(BS)Tot} = \left([BS] \cdot \frac{S_0}{C_{H_2O}} \cdot K_{m:w,unionised} + S_0\right) + \left([BS] \cdot \frac{S_i}{C_{H_2O}} \cdot K_{m:w,ionised} + S_i\right)$$

Confirmed Intrinsic Solubility & Solubility Factors

Biorelevant Solubility Modelling

$$S_{(BS)Tot} = \left([BS] \cdot \frac{S_0}{C_{H_2O}} \cdot K_{m:w,unionised} + S_0\right) + \left([BS] \cdot \frac{S_i}{C_{H_2O}} \cdot K_{m:w,ionised} + S_i\right)$$

Confirmed Bile Micelle Partition Coefficients



ADAM Model

Confirm and/or Estimate Parameters

Modeling of *In Vitro* Solubility Experiments

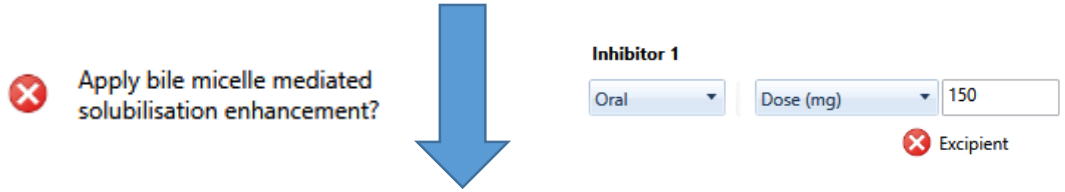
Global Solubility Equation v19

Aqueous Phase Solubility

$$S_{Tot}(t) = S_o \cdot S_{o_{scalar}}(t) \cdot \left(1 + \frac{[BS](t)}{C_{H_20}} \cdot Km{:}\,w,u\right) + S_i(t) \cdot \left(1 + \frac{[BS](t)}{C_{H_20}} \cdot Km{:}\,w,i\right) + S_{bound,excip}(t)$$

Bile Micelle Phase Solubility

Excipient "Phase" Solubility



Reduces to: $S_{Tot}(t) = S_o \cdot S_{o_{scalar}}(t) + S_i(t)$ Aqueous Phase Solubility

where

Monoprotic Base	$S_i(t) = S_o \cdot S_{o_{scalar}}(t) \cdot \left(10^{pKa-pH(t)}\right)$
Diprotic Base	$S_i(t) = S_o \cdot S_{o_{scalar}}(t) \cdot \left(10^{pKa1-pH(t)} + 10^{pKa2-pH(t)} + 10^{pKa1+pKa2-2pH(t)}\right)$
Monoprotic Acid	$S_i(t) = S_o \cdot S_{o_{scalar}}(t) \cdot \left(10^{pH(t)-pKa}\right)$
Diprotic Acid	$S_i(t) = S_o \cdot S_{o_{scalar}}(t) \cdot \left(10^{pH(t)-pKa1} + 10^{pH(t)-pKa2} + 10^{2pH(t)-pKa1-pKa2}\right)$
Ampholytes	$S_i(t) = S_o \cdot S_{o_{scalar}}(t) \cdot \left(10^{pH(t)-pKa1} + 10^{pKa2-pH(t)} + 10^{pKa2-pKa1}\right)$

where, for Ampholytes only, pKa1 is acidic and pKa2 is basic

CERTARA

Modeling of *In Vitro* Solubility Experiments

Aqueous Solubility Modelling

Confirmed Intrinsic solubility, pKa & Solubility Factors

$$S_{Tot}(t) = S_o \cdot S_{o_{scalar}}(t) \cdot \left(1 + \frac{[BS](t)}{C_{H_20}} \cdot Km{:}\,w,u\right) + S_i(t) \cdot \left(1 + \frac{[BS](t)}{C_{H_20}} \cdot Km{:}\,w,i\right) + S_{bound,excip}(t)$$

Free base amorphous solubility, pH 7.4 = 38.5 µg/mL (Xu et al 2017)
Solubility in 0.1 N HCL = 4 mg/mL (Law 2001 JPharmSci 90:1015)
Solubility Factor = 103.896 (Optimized with SIVA 3)

Biorelevant Solubility Modeling

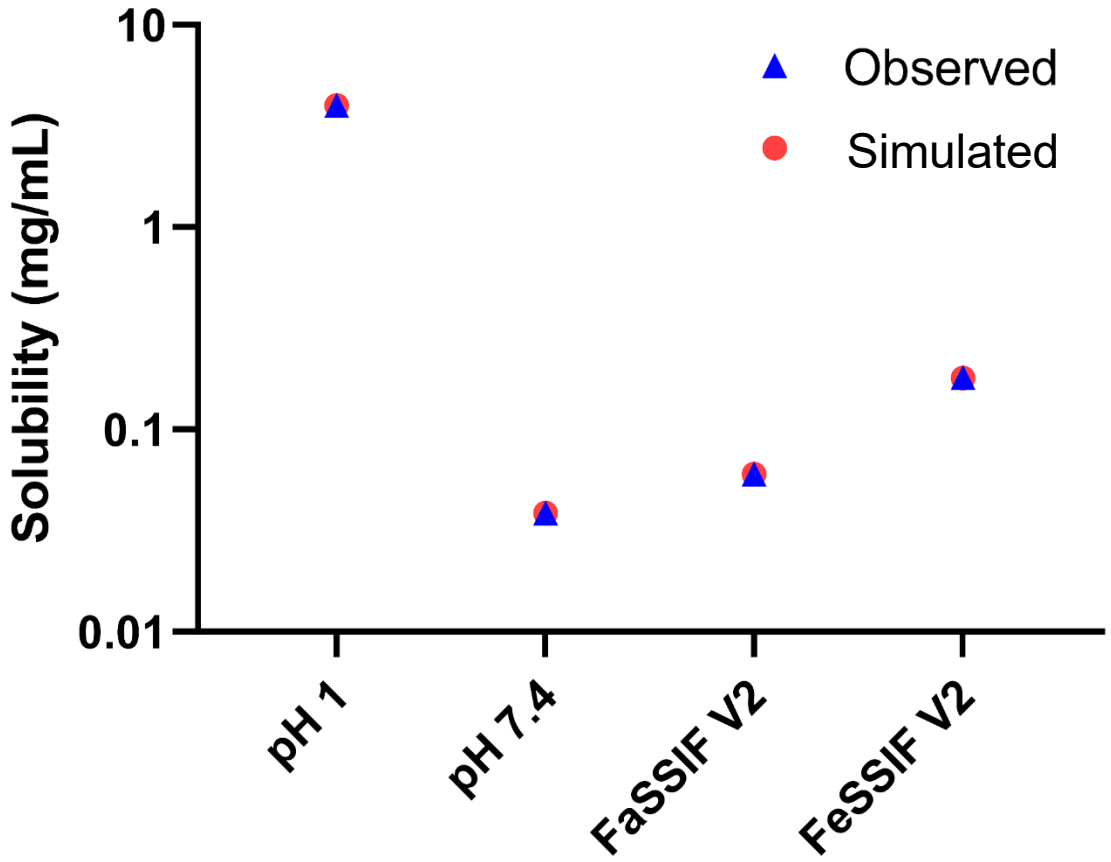
$$S_{Tot}(t) = S_o \cdot S_{o_{scalar}}(t) \cdot \left(1 + \frac{[BS](t)}{C_{H_20}} \cdot Km{:}\,w,u\right) + S_i(t) \cdot \left(1 + \frac{[BS](t)}{C_{H_20}} \cdot Km{:}\,w,i\right) + S_{bound,excip}(t)$$

Confirmed Bile Micelle Partition Coefficients

Media	pH	Conc. of Bile Salts (mM)	Solubility (µg/mL)	Experimental Data
FaSSIF V2	6.5	3	**60**	Xu et al, Mol. Pharmaceutics, 2017, 14 (11), pp 3801–3814
FeSSIF-V2	5.8	10	**180**	Xu et al, Mol. Pharmaceutics, 2017, 14 (11), pp 3801–3814
		Km/w (neutral)	**3.606**	Optimized with SIVA 3
		Km/w (ion)	**NA**	Optimized with SIVA 3

CERTARA

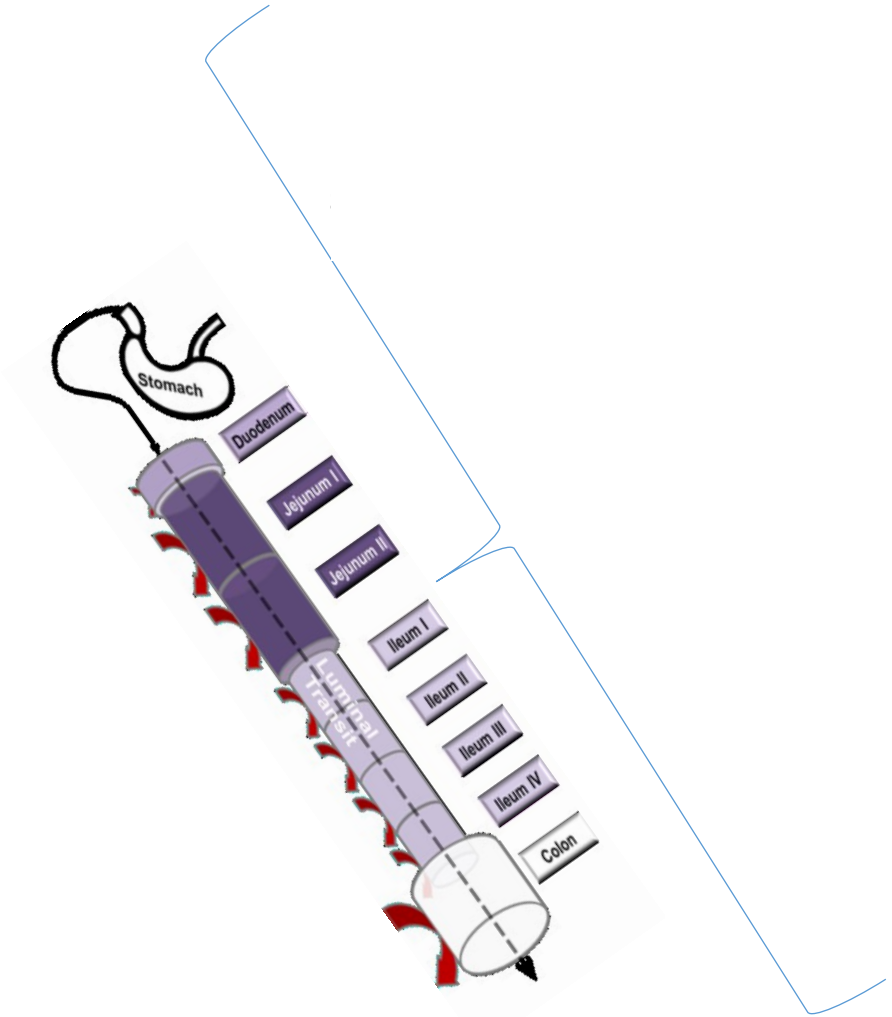
Modeling of *In Vitro* Solubility Experiments



Solubility values in aqueous and biorelevant media are nicely captured

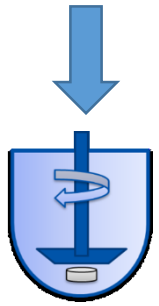
CERTARA

Sequential Modeling of *In Vitro* Biopharmaceutic Experiments



ADAM Model

USP-2 Dissolution Modelling



$$DR(t) = -NS\frac{D_{eff}}{h_{eff}(t)}4\pi a(t)\left(a(t)+h_{eff}(t)\right)\left(S_{surface}(t)-C_{bulk}(t)\right)$$

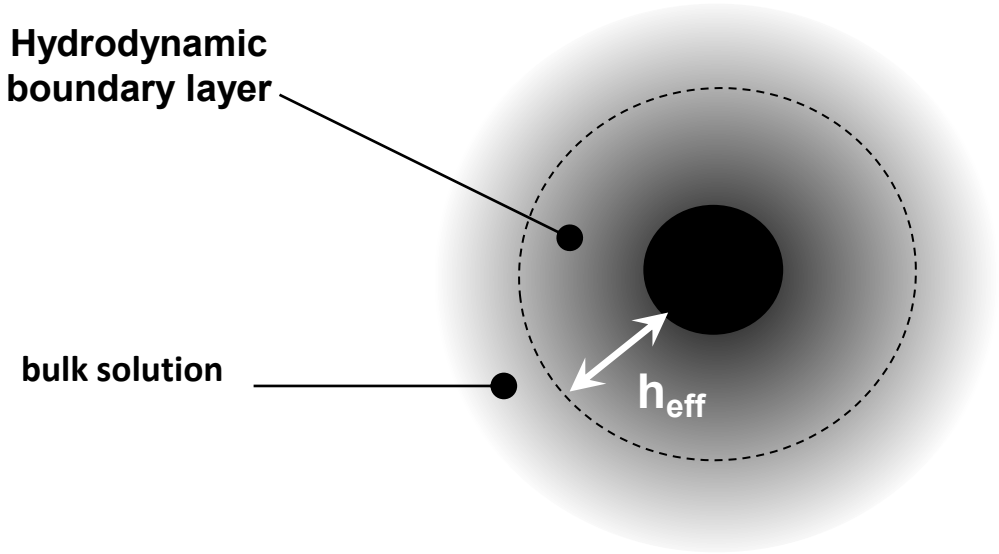
Confirmed Delayed Release and DLM Parameters

Confirm and/or Estimate Parameters

CERTARA

Mechanistic Dissolution Modelling: Diffusion Layer Models (DLM)

Assumption: Rate limiting step is diffusion across a hydrodynamic boundary layer surrounding the particles of effective thickness h_{eff}



Spherical surface - non-linear diffusion gradient*

General Form of the Dissolution Rate Equation

$$DR(t) = \sum_{NBINs}^{i=1} -N_i S_{DR} \frac{D_{eff}(t)}{h_{eff,i}(t)} 4\pi a_i(t)\left(a_i(t) + h_{eff,i}(t)\right)\left(S_{surface}(t) - C_{bulk}(t)\right)$$

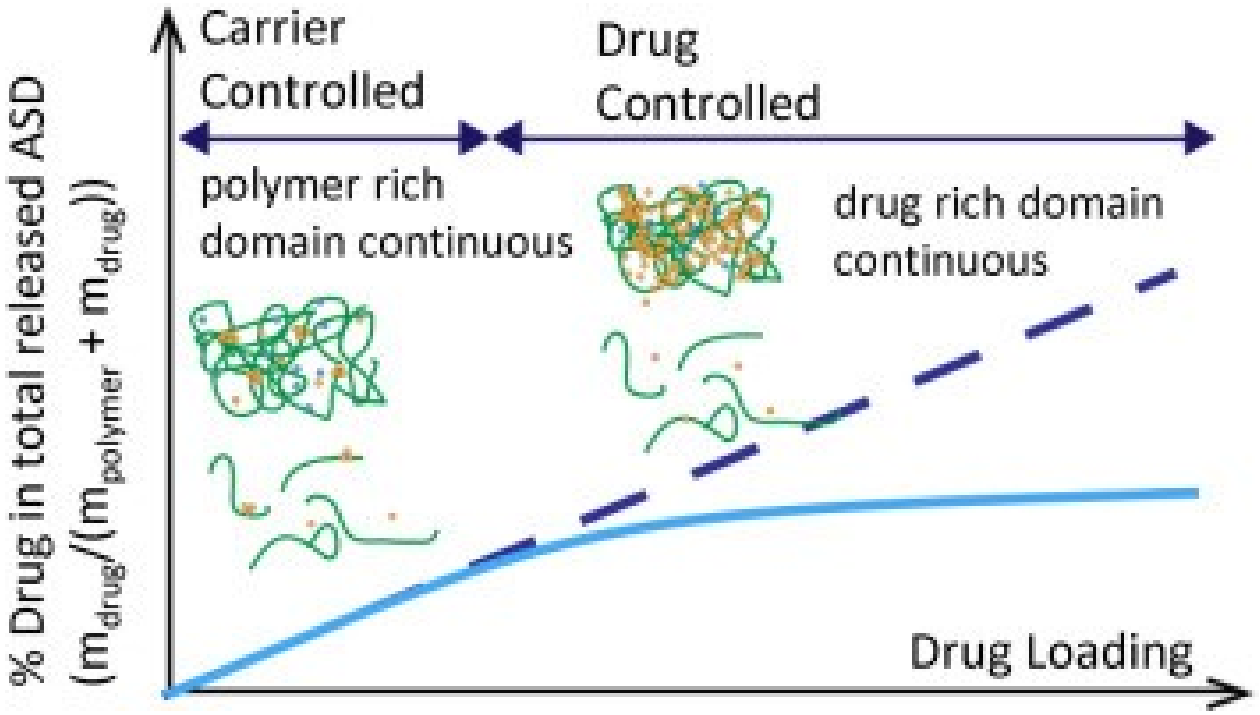
a – particle radius; $\boldsymbol{C_{bulk}}$ – bulk lumen concentration; $\boldsymbol{S_{surface}}$ – solubility at particle surface; $\boldsymbol{D_{eff}}$ – effective diffusion coefficient; ***DR*** – Dissolution rate; $\boldsymbol{h_{eff}}$ – effective diffusion layer thickness; ***N*** – number of particles in PS bin *i;* $\boldsymbol{S_{DR}}$ – dissolution DLM scalar; ***t*** - time

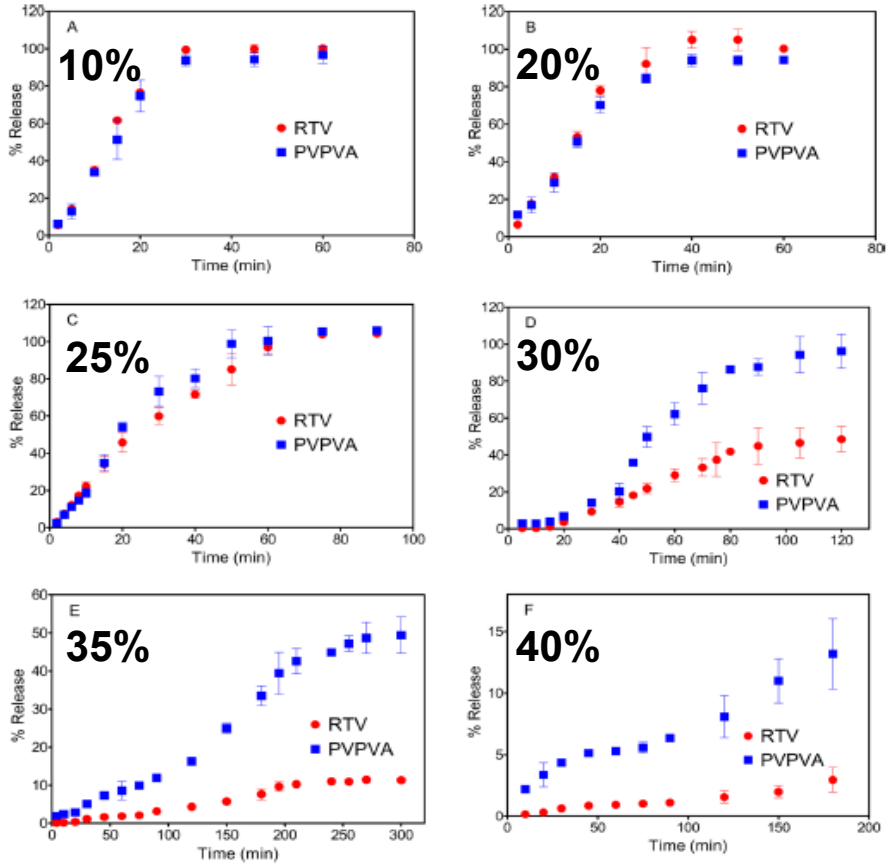
(Noyes & Whitney, 1897; *Wang & Flanagan, 1999; Further discussion: Sugano, 2010)

CERTARA

Understanding release of drug from amorphous solid dispersion

The release of the drug from the amorphous solid dispersion can be either polymer controlled or drug controlled





Dissolution is carried out in phosphate buffer pH 6.8

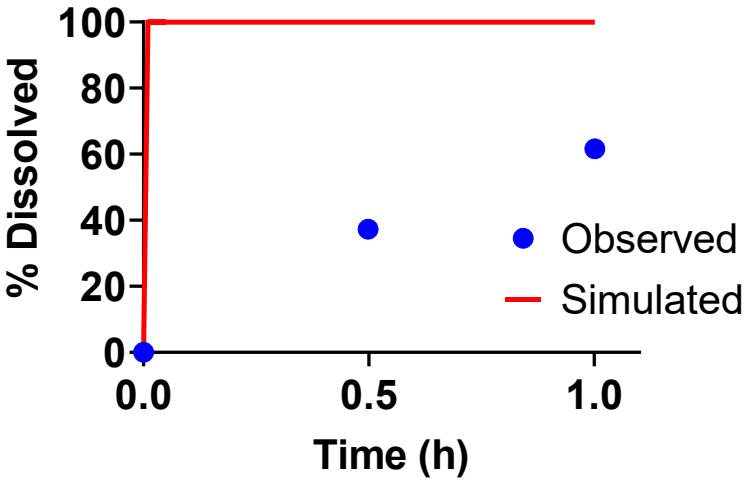
Han et al. European Journal of Pharmaceutical Sciences Volume 136, 1 August 2019
Indulkar et al. Mol. Pharmaceutics 2019, 16, 1327−1339

CERTARA

Modeling *In Vitro* Dissolution Experiments in Simulated Gastric Fluid (pH 1.2)

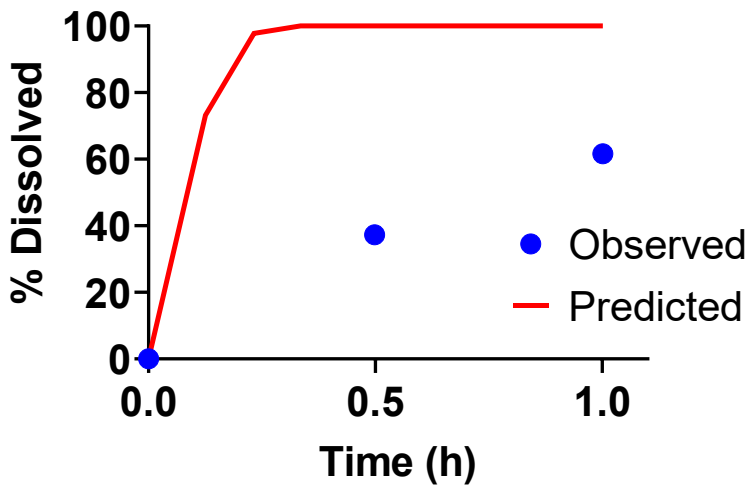
- Ritonavir is a weak base showing high solubility in the acidic pH
- Particle size of the API in commercial batches used in *in vitro* or *in vivo* studies are rarely reported in literature

Scenario 1



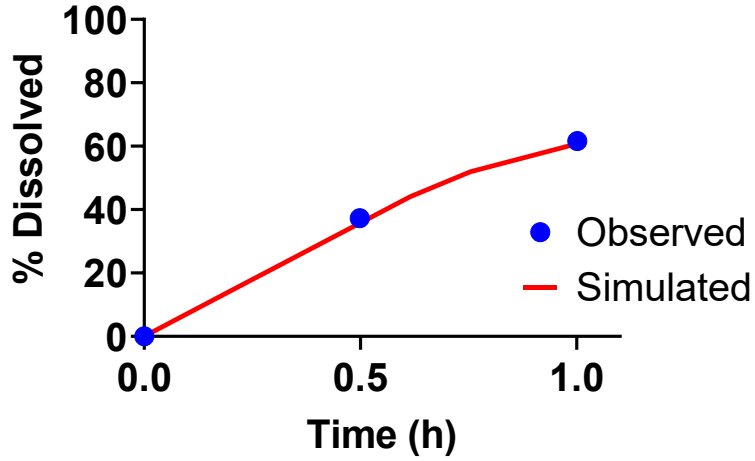
Input particle size – 10 µm
(Default in Simcyp)

Scenario 2



Input particle size – 100 µm

Scenario 3



Fitting a first order release constant
Input particle size – 10 µm

$$\%F_{release,t} = \%F_{max} \left(1 - e^{-kd1 \cdot (t - t_{lag})}\right)$$

F_{max} (%) = 100%
K_d (h^{-1}) = 0.017
T_{lag} (h) = 0

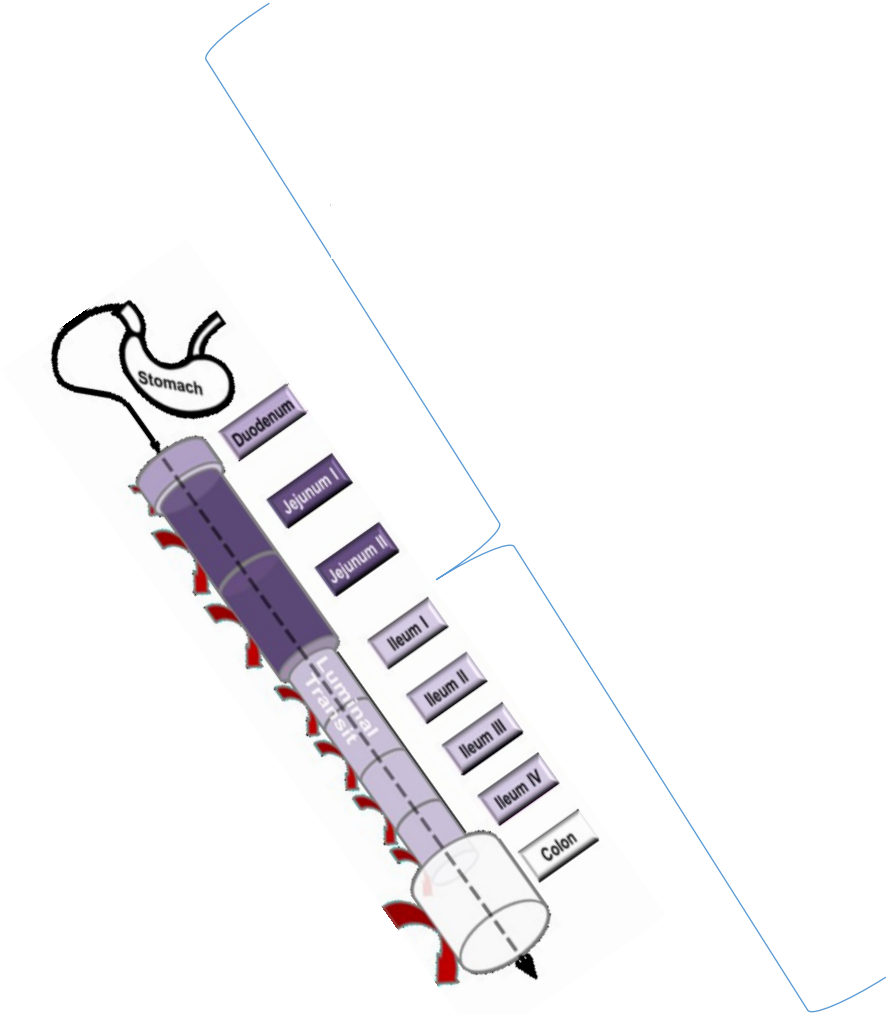
Parameters used in the PBPK model

Use of default particle size should be justified via modeling of *in vitro* experiments

Ellenberger et al, AAPS PharmSciTech. 2018 Jul;19(5):1985-1997

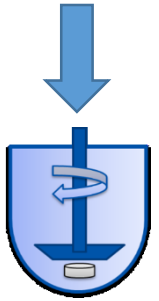
CERTARA

Sequential Modeling of *In Vitro* Biopharmaceutic Experiments



ADAM Model

pH Shift Experiment Modelling



Critical Supersaturation Ratio[c]	1
Precipitation Rate Const. (1/h)[d]	0.001

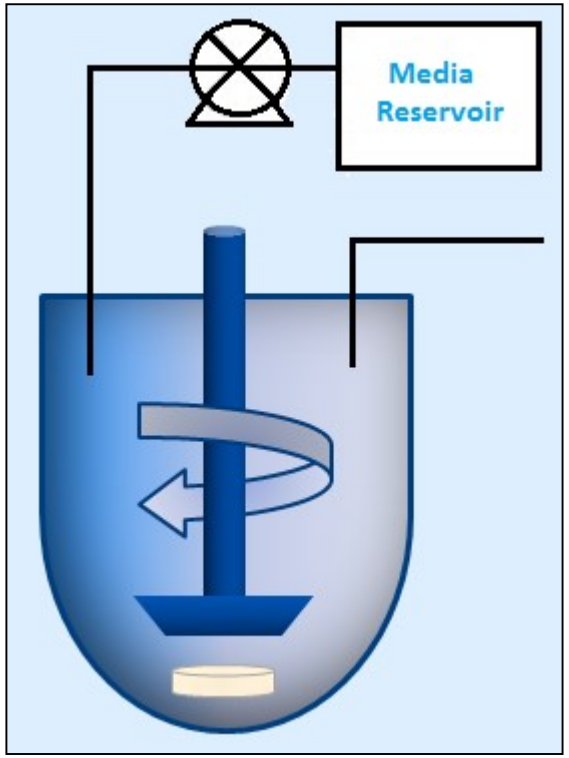
Confirmed Precipitation Parameters

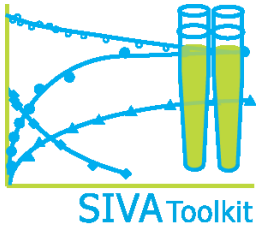
Confirm and/or Estimate Parameters

CERTARA

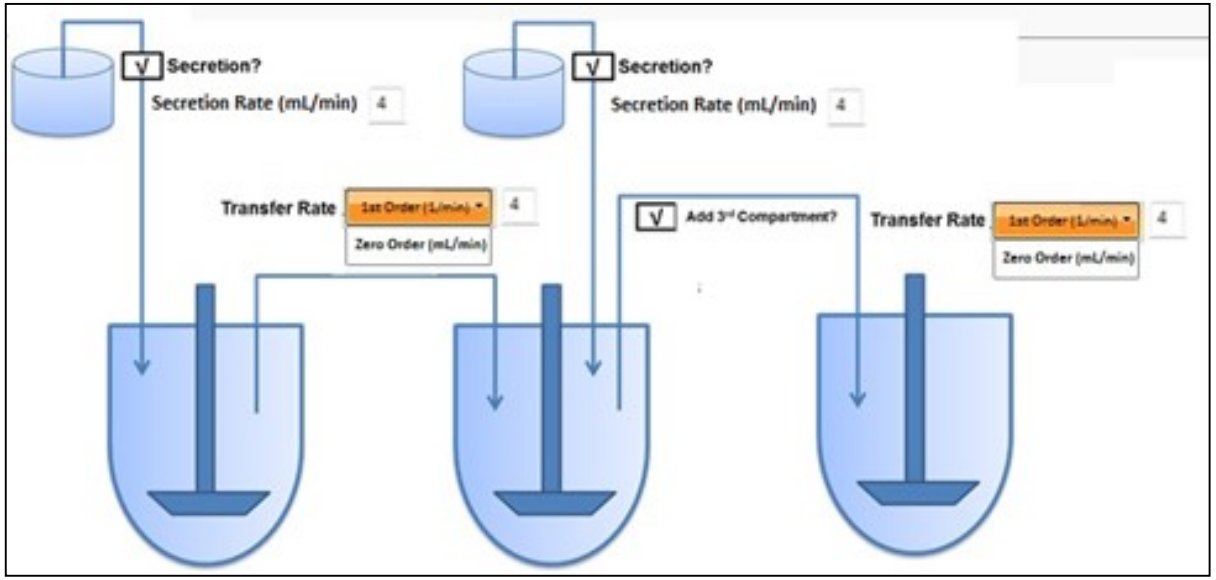
In Vitro Experiments for Assessing Supersaturation Behaviour

Static - spiked solutions, media change, pH change
solvent shift (two-phase)





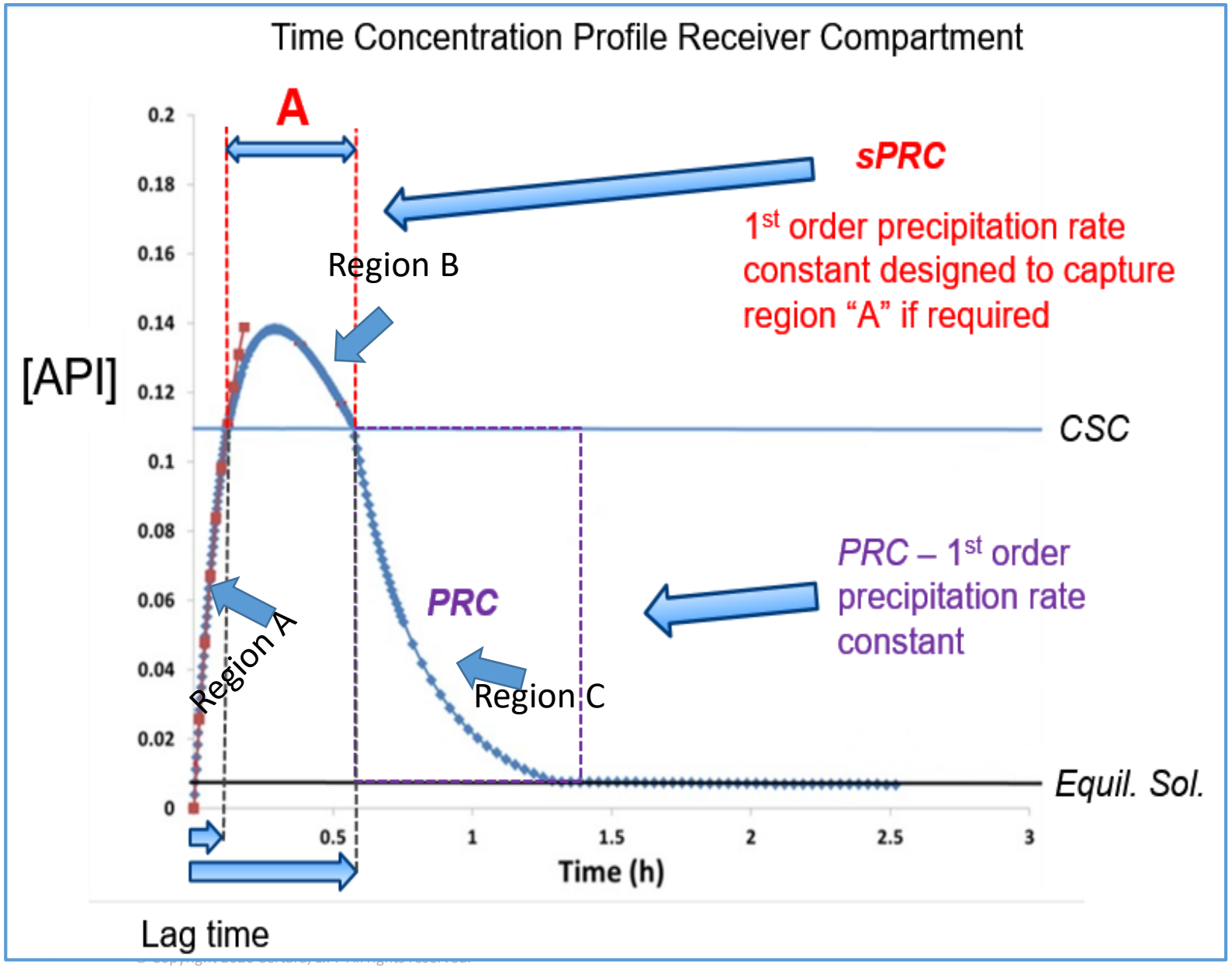
Dynamic - continuous transfer experiments



+ more complex models such as TNO TIN-1

CERTARA

Empirical Supersaturation/Precipitation Model



Empirical Model

IF supersaturated conditions encountered THEN

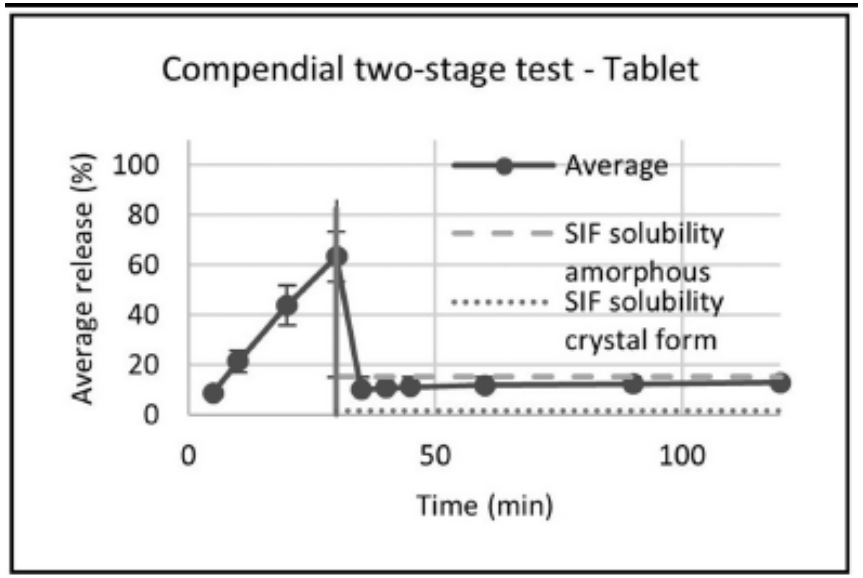
	Dissolution stops
	Precipitation can only begin
when	**CSC is reached**
	***CSC* is a critical conc. at which precipitation starts**
	[Drug] may continue to rise due to
slow	permeation of drug from skin
	Supersaturated conc. may
exceed	***CSC* (*CSR* x Eq.Sol)**

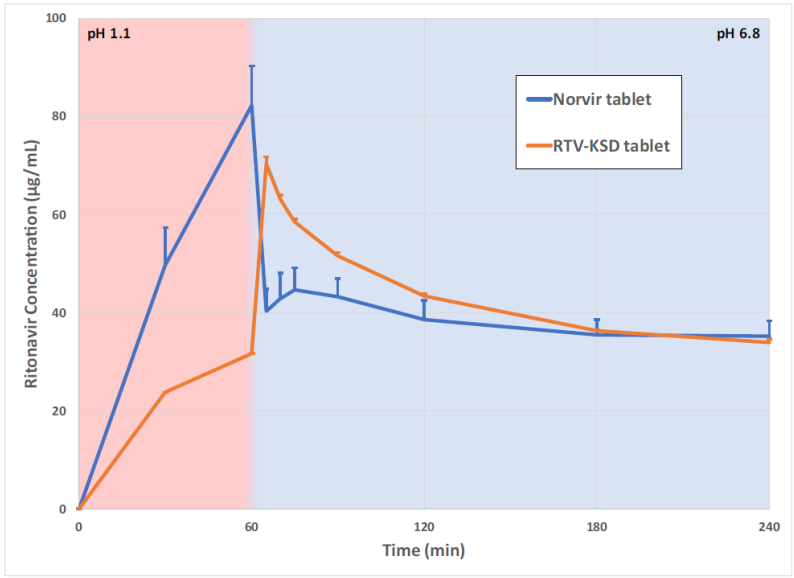
CSC – Critical Supersaturation Concentration
CSR – Critical Supersaturation Ratio
PRC – Precipitation Rate Constant (1/h)
sPRC – Secondary Precipitation Rate Constant (1/h)

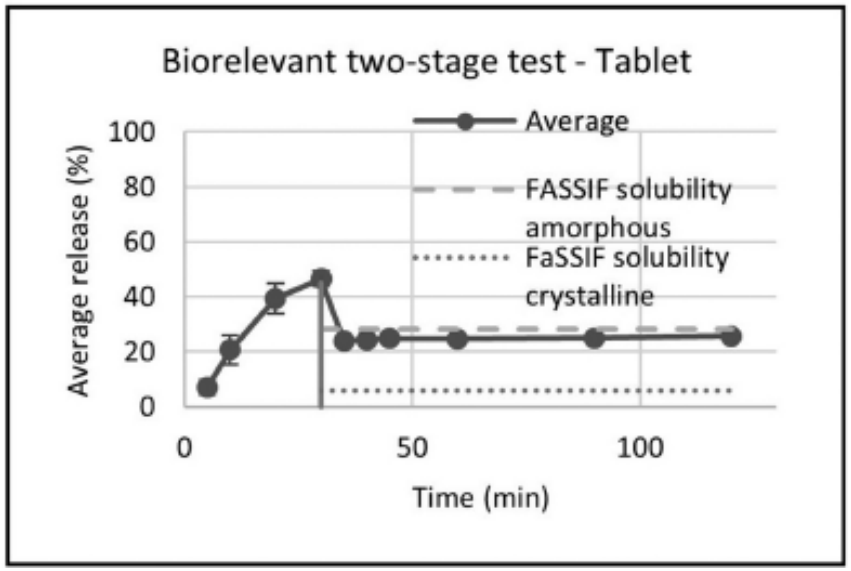
CERTARA

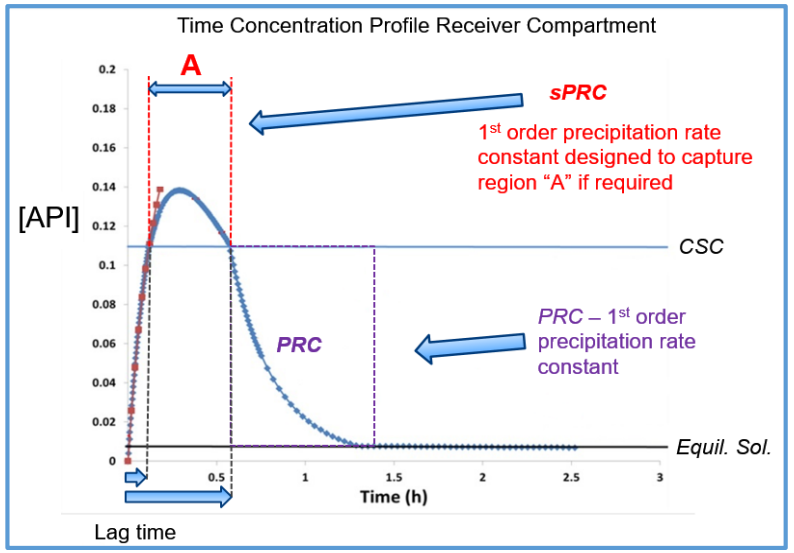
Modeling *In Vitro* Drug Supersaturation and Precipitation

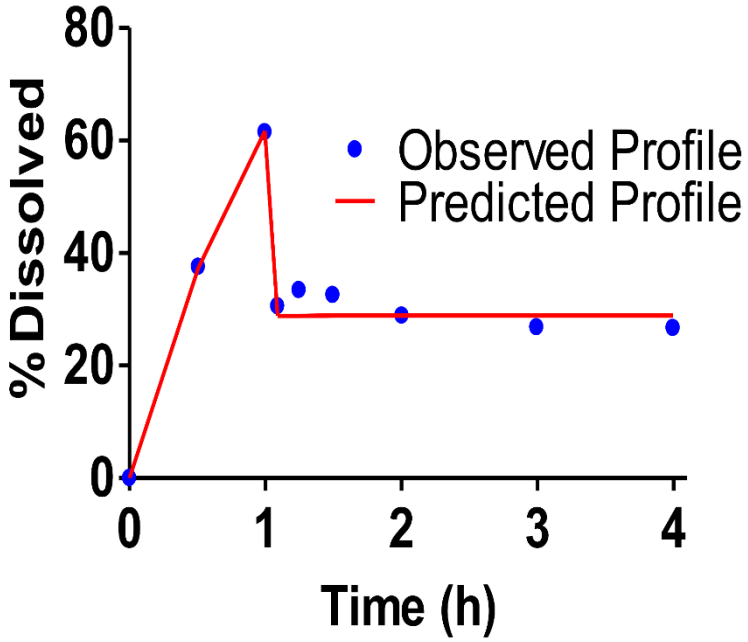
Ritonavir precipitates to its amorphous state and does not crystallize











***CSR* – 1**
***sPRC* – 1000 (1/h)**
***PRC* – 0.001 (1/h)**

Parameters used in the PBPK model

Fiolka et al, JPS 2020; Ellenberger et al, AAPS PharmSciTech. 2018 Jul;19(5):1985-1997

CERTARA

Modeling Permeability of Ritonavir in the PBPK Model – Mechanistic Permeability (MechPeff) Model

MechPeff model predicts regional Peff depending on different morphology in each gut segment

$$P_{eff,n} = \left(\left(\left(P_{Trans,0} \cdot f_{neutral,\boldsymbol{UBL},pH,n} + P_{para,n} \right) \cdot ACC_n \cdot MVE_n \cdot fu_{\boldsymbol{UBL},n} \right)^{-1} + \left(P_{\boldsymbol{UBL},n} \right)^{-1} \right)^{-1} \cdot FEp,n$$

Mechanistic modelling of *in vitro* experiments (SIVA 4)

Ionisation effects - pH of the UBL

Free fraction effects linked to bile and/or excipient solubilisation

In the present model, logP of Ritonavir was used to predict regional $P_{Trans,0}$

Input Parame …scellular

Compound type and pKa

$logP_{ow}$ or $logP_{PAMPA}$ but only if predicting $P_{trans,0}$ from these values

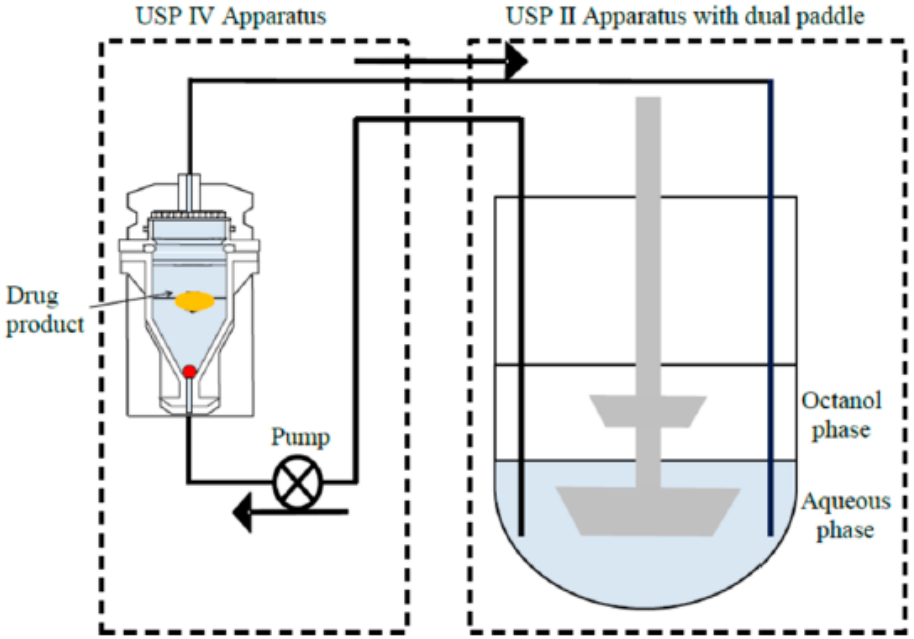
$P_{trans,0}$ – predicted, calibrated from predicted Papp, MechP (SIVA 4)

Solubility – aqueous and bile micelle mediated, calibrated/confirmed
– essential for free fraction calculations

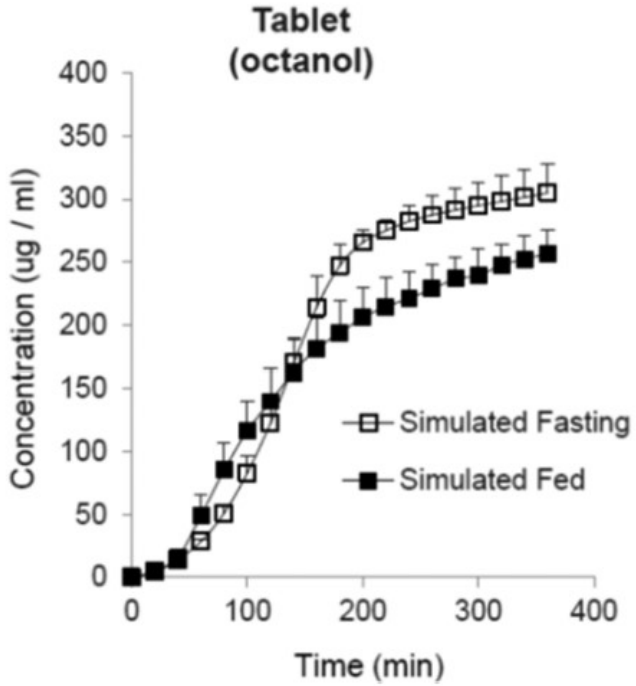
MVE – maximum villous surface area expansion
ACC – additional surface area scalar - automatically calculated (Oliver, Rowland et al 1998)
FEp – plicae circulares surface area expansion (humans)
P_{para} – paracellular permeability via Renkin function
$f_{neutral}$ – fraction unionised (aqueous phase)
fu_{UBL} – free fraction UBL

CERTARA

Why MechPeff ?



Biphasic dissolution−partition test



Cumulative RTV concentration−time profiles in octanol from the tablet associated with the simulated fasting and fed media.

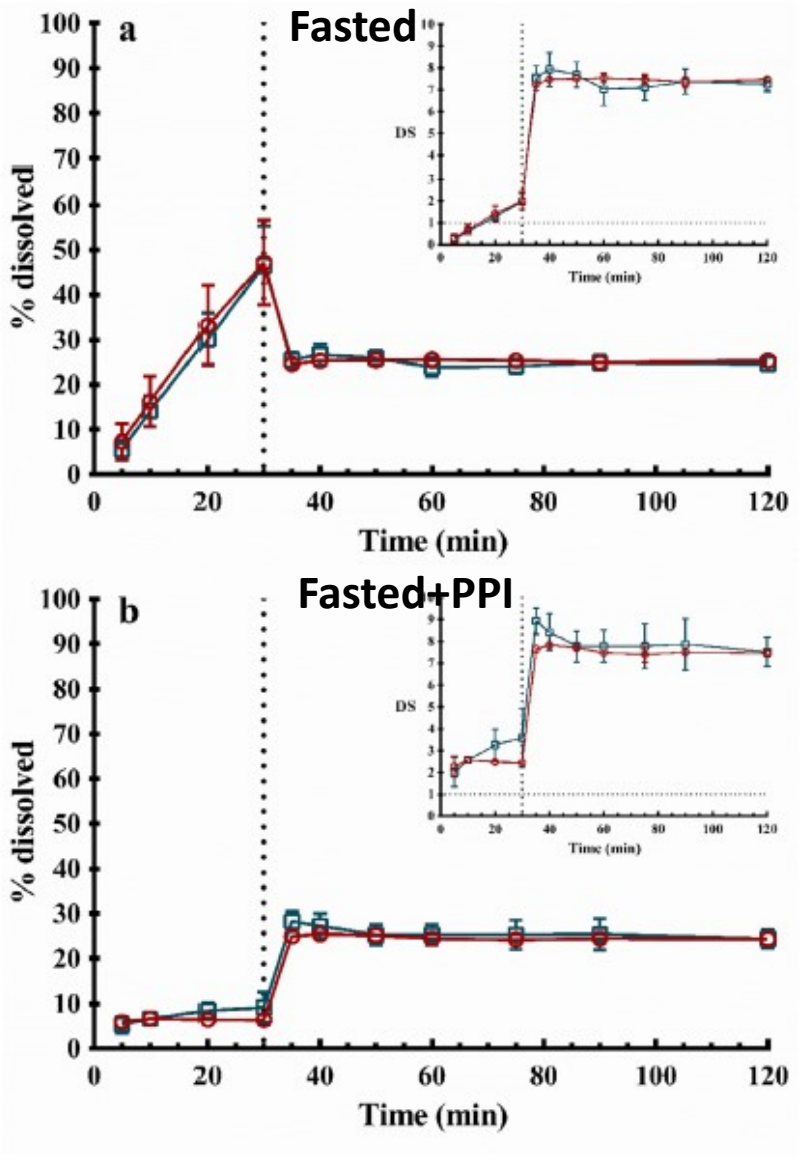
Two factors were postulated to explain the food effects –

a. Effect of gastric pH on drug dissolution

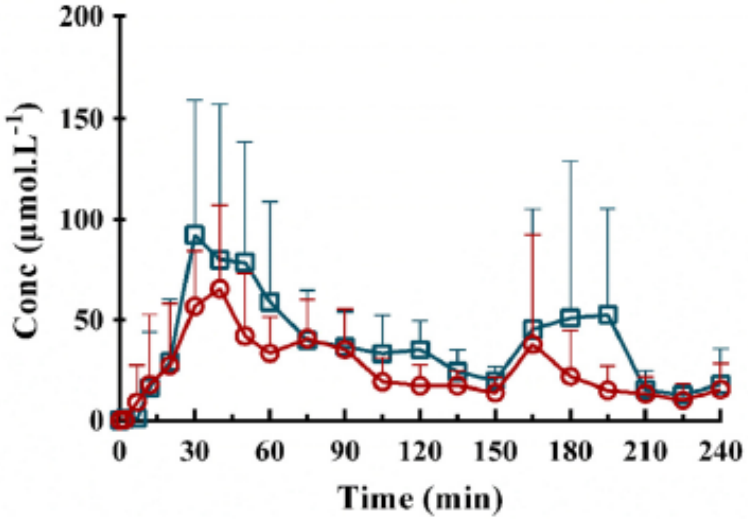
Xu et al, Mol. Pharmaceutics, 2017, 14 (11), pp 3801–3814

CERTARA

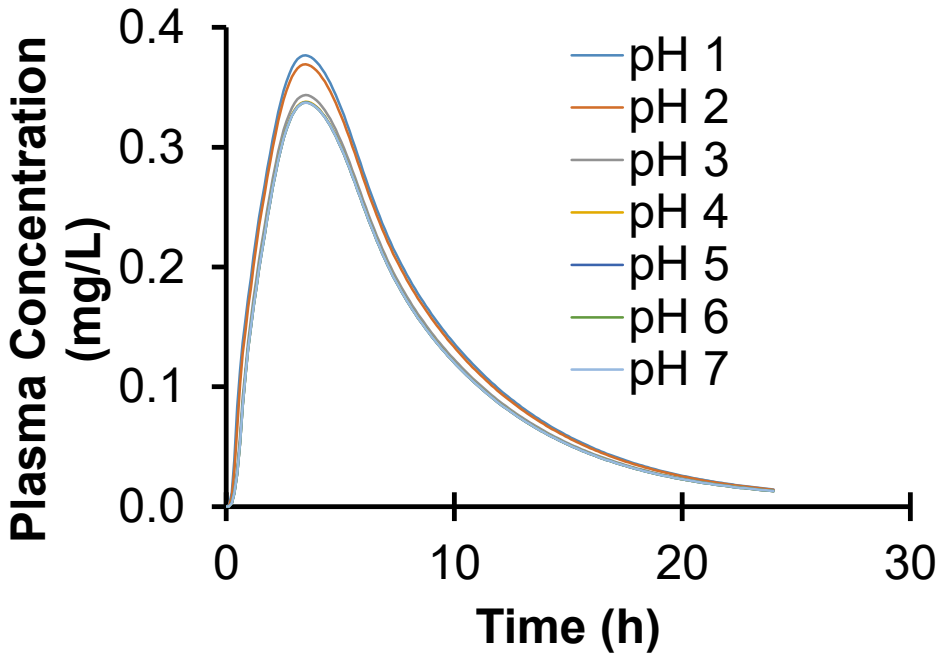
Why gastric pH may not be the cause of observed negative food effect?



pH Shift Experiment



Mean (+ S.D., n = 5) dissolved drug content - time profiles in the duodenum after oral intake of one tablet of Norvir® (100 mg ritonavir) under fasted state (bullets) and fasted + PPI conditions (squares).

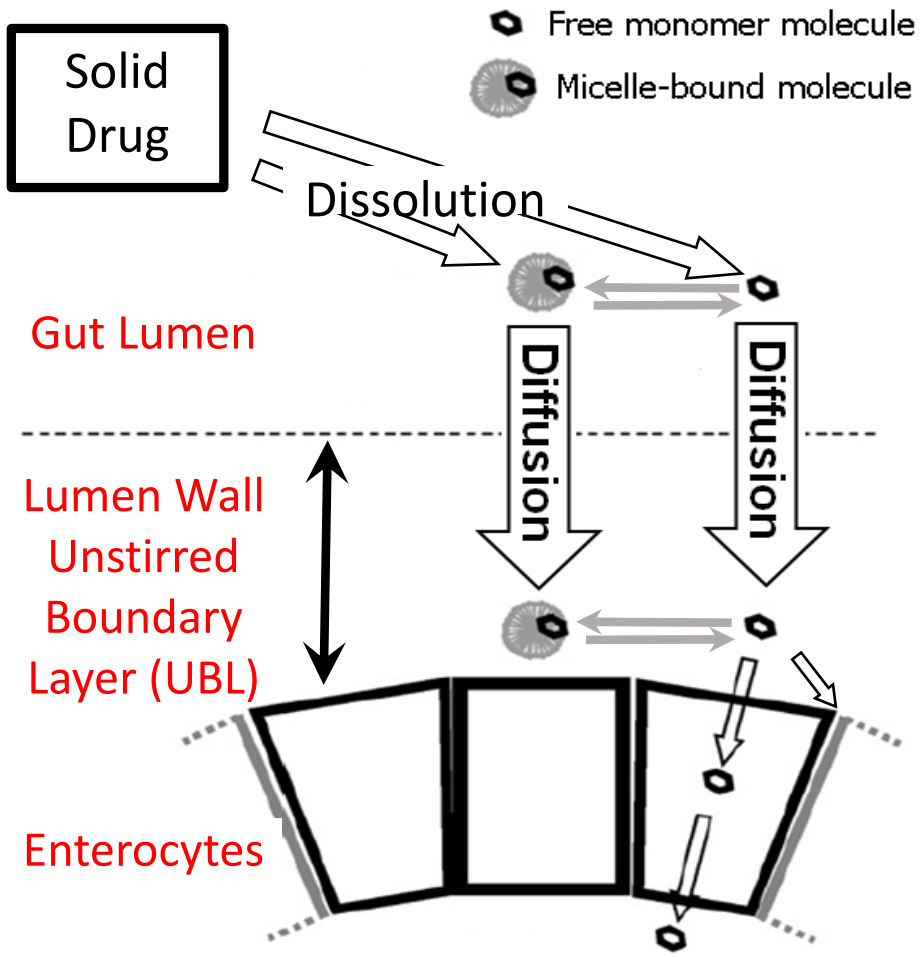


Model Sensitivity to Gastric pH

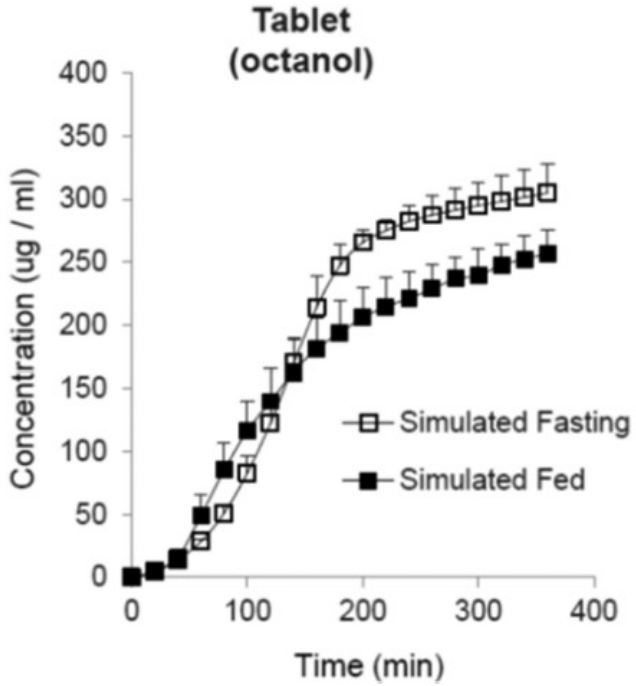
Abeele et al. Eur J Pharm Sci. 2020 May25; 151:105377

CERTARA

Why MechPeff ?



Adapted from Sugano 2009



Cumulative RTV concentration−time profiles in octanol from the tablet associated with the simulated fasting and fed media.

Two factors were postulated to explain the food effects –

a. **Effect of gastric pH on drug dissolution**
b. **Effect of free fraction of drug on its permeation**

Mechanistic Permeability (MechPeff) Model in Simcyp takes into account effect of free fraction of drug available on drug permeation

Xu et al, Mol. Pharmaceutics, 2017, 14 (11), pp 3801–3814

CERTARA

Static Viscosity Model

- In most of our models, compartments are considered as well-mixed
- Viscosity of the medium affects the diffusion coefficient of the drug, thus influencing drug dissolution and diffusion
- Viscosity of GI lumen changes drastically depending upon the prandial state

Viscosity Equation

$$D_{eff,fast\ |\ fed} = \left(\frac{1}{\mu_{fast\ |\ fed}}\right)^{0.987} D_{eff,\mu=1}$$

General | Water Physicochemical Properties | Luminal pH | Luminal Bicarbonate | Luminal Fluid Velocity | Endogenous Ions | Luminal Bile Salts | GI Morphology | Luminal Boundary | Viscosity

Fasted

		Stomach	Duodenum	Jejunum I	Jejunum II	Ileum I	Ileum II	Ileum III	Ileum IV	Colon
Apparent Viscosity	Mean (cP)	3	3	3	3	3	3	3	3	3
	CV (%)	71	71	71	71	71	71	71	71	71

Fed

		Stomach	Duodenum	Jejunum I	Jejunum II	Ileum I	Ileum II	Ileum III	Ileum IV	Colon
Apparent Viscosity	Mean (cP)	232	65	33	12	9	3	3	3	3
	CV (%)	32	27	36	14	2	71	71	71	71

Gastrointestinal compartment viscosity values used in Simcyp for fasted and fed state simulations (based on meta-analysis)

CERTARA

Distribution and Metabolism Related Parameters

- Ritonavir exhibits complex pharmacokinetic profile
- It is mainly metabolized by CYP3A4/5 with minor metabolism from CYP2D6

Parameter	Value	Comment/Reference
fu, plasma	0.015	Hsu, Clin PK, 1998
Vss (L/kg) (Full PBPK)	0.410	Umehara et al, Biopharm Drug Dispos. 2018;39:152
CL(CYP2D6) – BD Sup		Koudriakova et al. Drug Metab Dispos, 1998, 26:552
Vmax	0.93	Corrected for ISEF
Km	1	
ISEF	0.75	
CL(CYP3A4) – BD Sup		Koudriakova et al. Drug Metab Dispos, 1998, 26:552
Vmax	1.37	Corrected for ISEF
Km	0.07	
ISEF	0.24	
CL(CYP3A5) – BD Sup		Koudriakova et al. Drug Metab Dispos, 1998, 26:552
Vmax	1	Corrected for ISEF
Km	0.05	
ISEF	0.24	
Renal Clearance (L/Kg)	0.53	Denissen et al. 1997

Interaction Related Parameters

- Ritonavir exhibits competitive and mechanism based inhibition of CYP3A4/5
- Ritonavir also is competitive inhibitor of CYP2D6 and Pgp
- Ritonavir is also an inducer CYP3A4/5

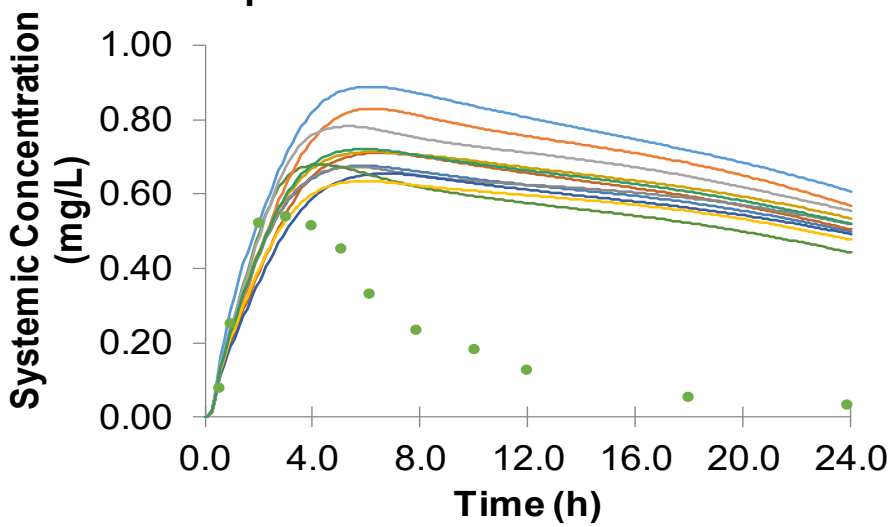
Parameter	Value	Comment/Reference
Mechanism Based Inhibition		
$K_{app\ (CYP3A4/5)}$ (μM)	0.25	Inhibition constant, Kirby et al. [13]
$k_{inact\ (CYP3A4/5)}$ (1/h)	19.8	Inactivation rate of enzyme, Kirby et al.[13]
$fu_{(mic)\ (CYP3A4/5)}$	0.71	Kirby et. al.[13]
Induction		
$IndC50_{(CYP3A4/5)}$ (μM)	1 (CV 30%)	Induction constant, Fahmi et al.[18]
$fu_{(inc)\ (CYP3A4)}$	1	Fraction unbound *in vitro,* Fahmi et al.[18]
$Indmax_{(CYP3A4)}$	68.5 (30%)	
Competitive Inhibition		
$K_{i\ (CYP3A4/5)}$ (μM)	0.0021	Inhibition constant, Shebley et al.[6]
$fu_{mic\ (CYP3A4/5)}$	0.925	Fraction unbound *in vitro*, Shebley et al.[6]
$K_{i\ (CYP2D6)}$ (μM)	0.04	Optimized (to recover DDIs)
$fu_{mic\ (CYP2D6)}$	1	
$K_{i\ (Apical\ Efflux,\ Intestine\ P\text{-}gp)}$ (μM)	0.03	Optimized (to recover DDIs)
$fu_{mic\ (Apical\ Efflux,\ Intestine\ P\text{-}gp)}$	1	
$K_{i\ (Liver\ Canalicular\ P\text{-}gp)}$ (μM)	0.03	Optimized (to recover DDIs)
$fu_{mic\ (Liver\ Canalicular\ P\text{-}gp)}$	1	

CERTARA

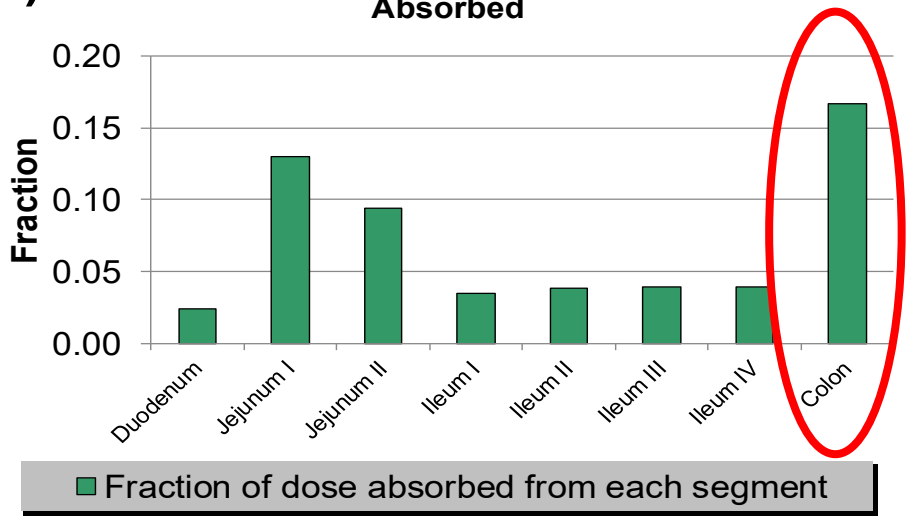
Initial Bottom Up Predictions – Ng et al 2008 Fasted State

Simulations were carried out using 10 trials of 27 individuals per the clinical study design reported by Ng et al. in fasted state

(A)



(B)



Absorption phase to 4 h was relatively well captured, giving confidence in the absorption-related parameters

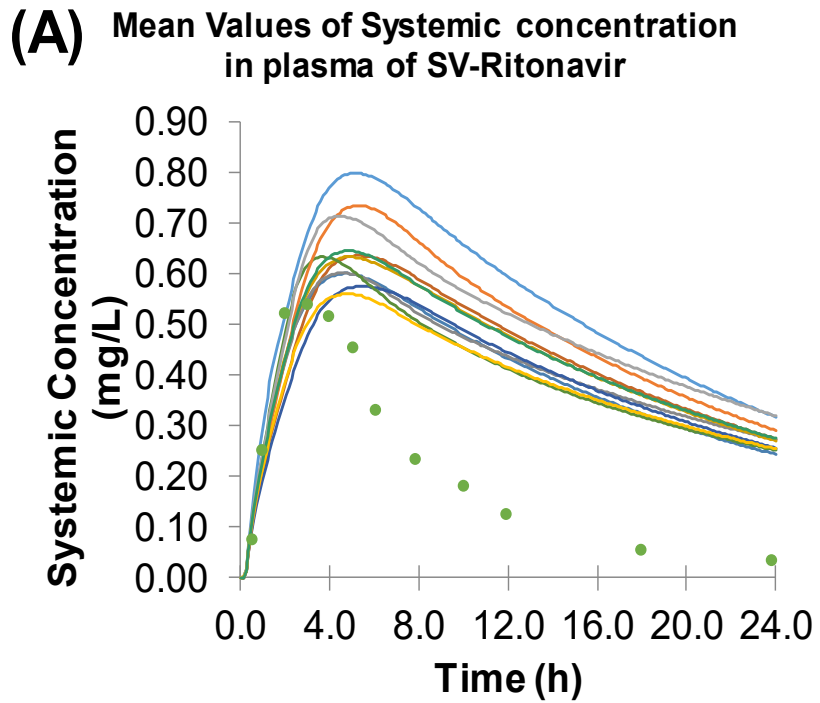
Two plausible reason for over-prediction of the terminal elimination phase:

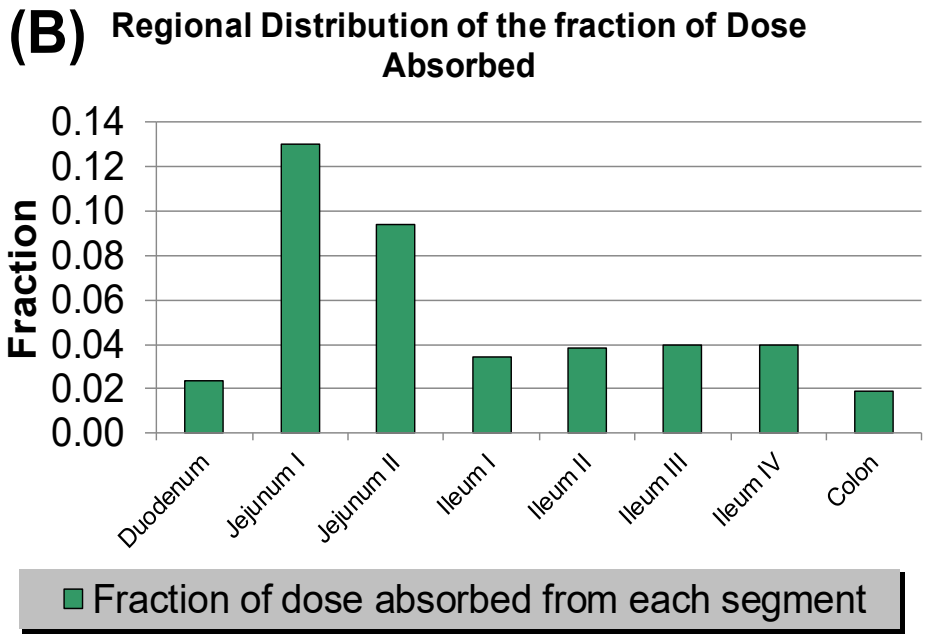
a. Over-prediction of drug absorption in the colon
b. *In vivo* clearance of the compound is not captured correctly by metabolic IVIVE (Please note - intravenous data for this compound are not available in literature).

CERTARA

First Modification - Adjustment of Colon Absorption Scalar

- Norvir is an immediate release tablet, majority of absorption should take place in the small intestine
- Understanding of drug absorption in the colon is poor, binding to faeces etc.
- Colon absorption scalar was reduced to 0.1





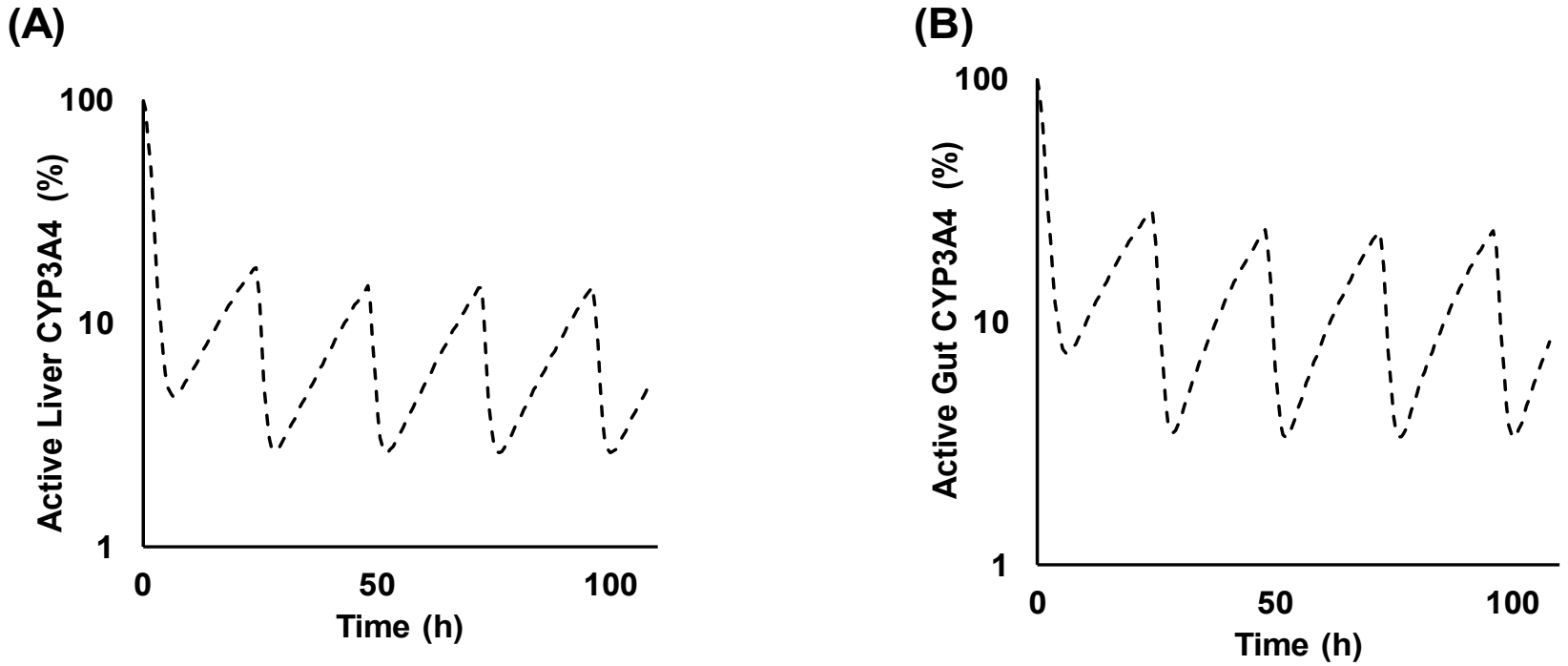
- Predictions are slightly better, C_{max} and T_{max} are relatively well captured
- Terminal phase is still overpredicted, AUC >two-fold over prediction suggesting that clearance is not captured correctly

CERTARA

Second Modification – Parameter Estimation of Additional Hepatic Intrinsic Clearance

- Ritonavir is a strong mechanism-based inhibitor of CYP3A4

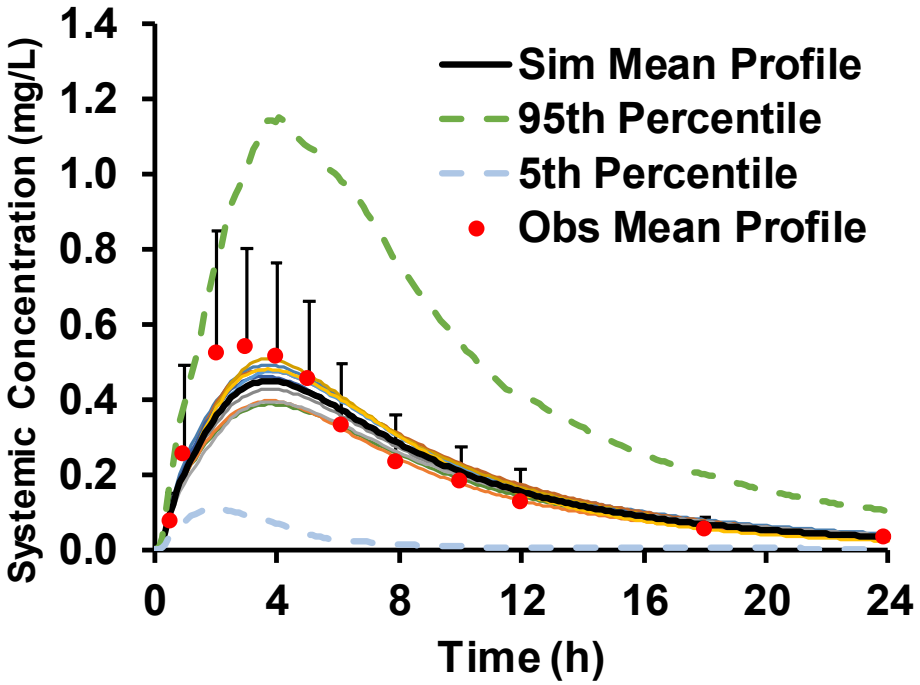
Simulated time dependent changes in hepatic (A) and gut (B) CYP3A4 abundance levels following administration of 100 mg QD oral administration of ritonavir for four days



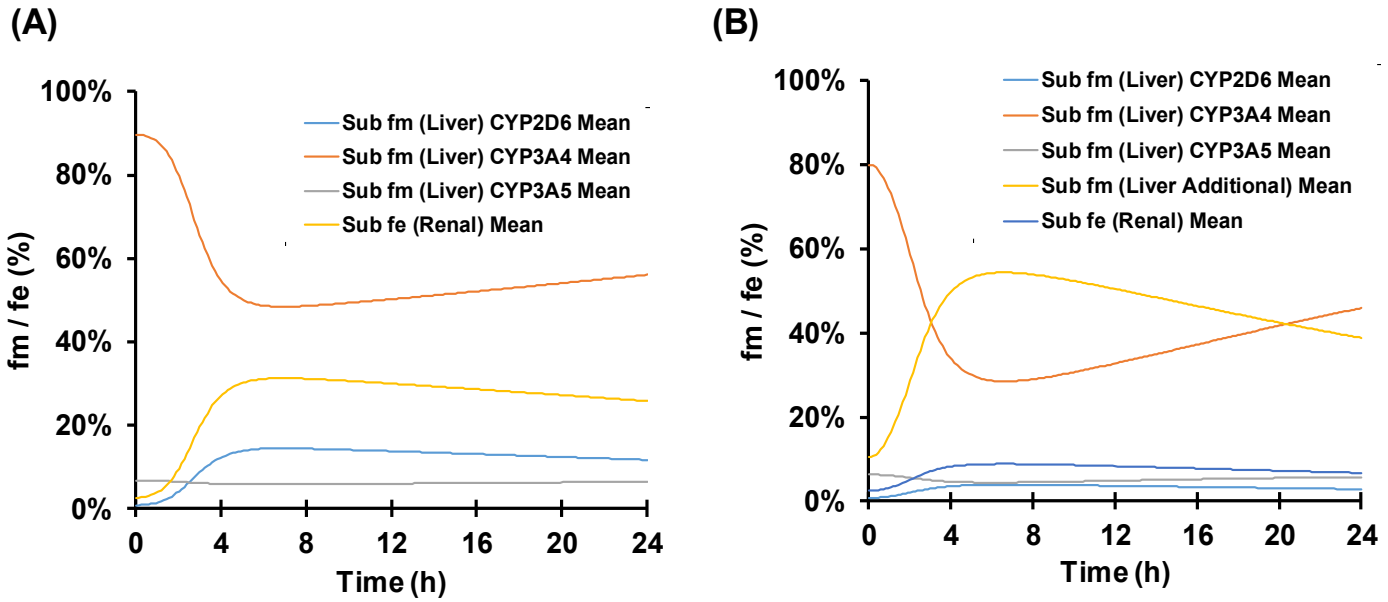
- Maximal inhibition of hepatic (4.77% activity remaining) and gut (7.59% activity remaining) CYP3A4 abundance on day 1 could be observed following administration of 100 mg of ritonavir.
- The fact that we cannot capture clearance of ritonavir suggests that there is additional clearance mechanism *in vivo*, which takes over in clearing the ritonavir from the systemic circulation once hepatic CYP3A4 is inhibited by ritonavir.

Second Modification – Parameter Estimation of Additional Hepatic Intrinsic Clearance

A value of 75 µL/min/mg additional hepatic clearance was found to capture well the observed fasted state Ng et al 2008 data (Initially analyzed by Sensitivity Analysis)



Simulated Ritonavir Concentration Time Profile of Ng et al 2008 study with additional hepatic clearance and colon absorption scalar.

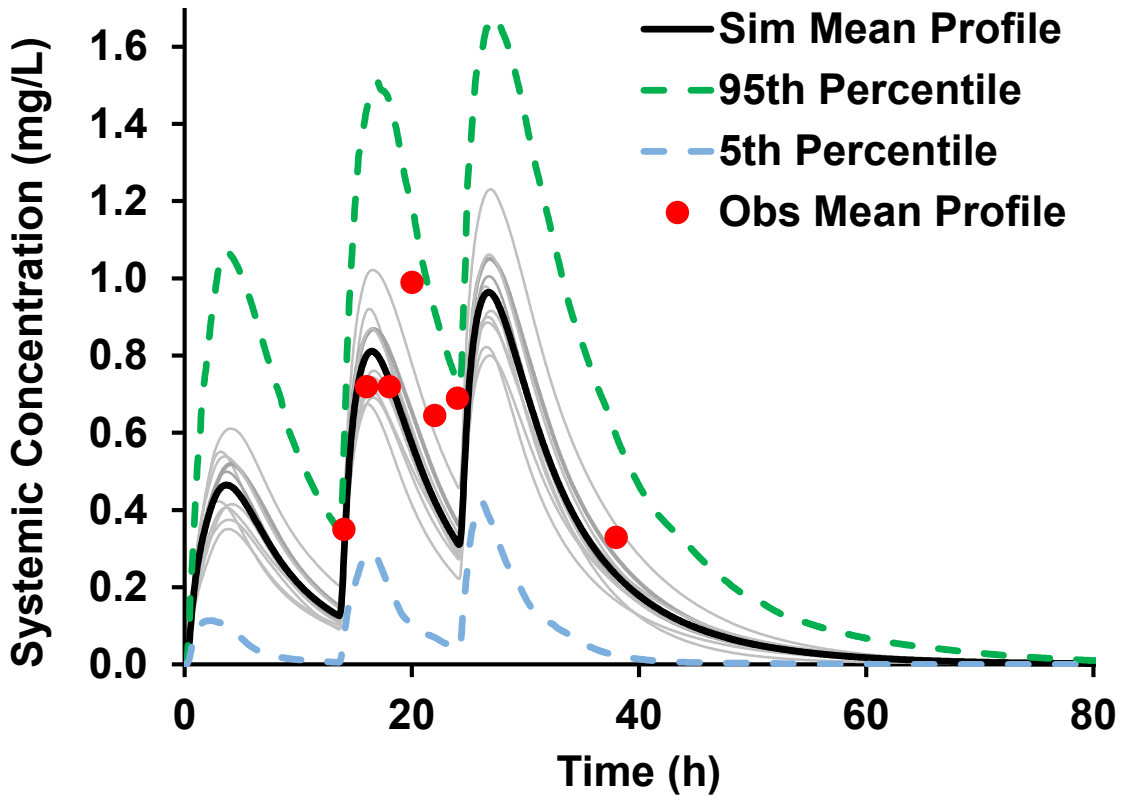


Time variant changes in percentage fraction metabolized (fm) and fraction excreted following oral administration of ritonavir (100 mg) in fasted state (A) without inclusion of additional intrinsic hepatic clearance and (B) with inclusion of additional intrinsic hepatic clearance.

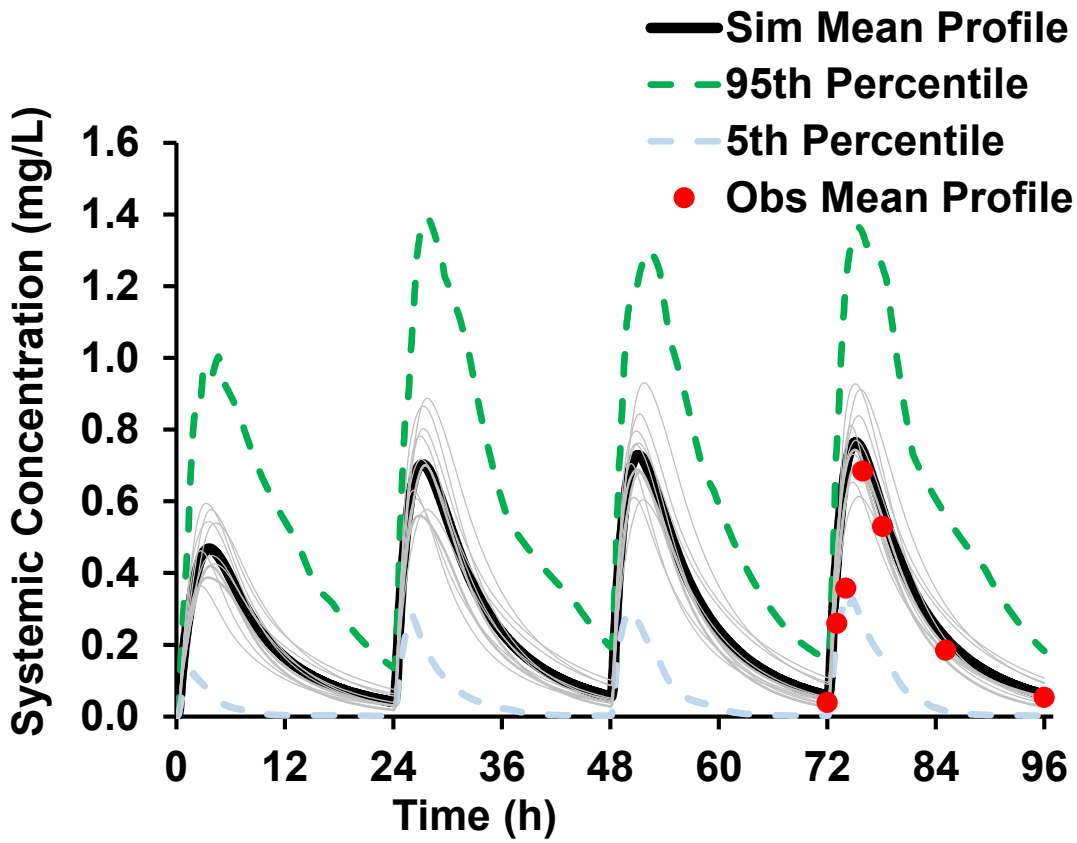
CERTARA

Verification of the Model – Multiple Dose Studies

Greenblatt et al., 2009



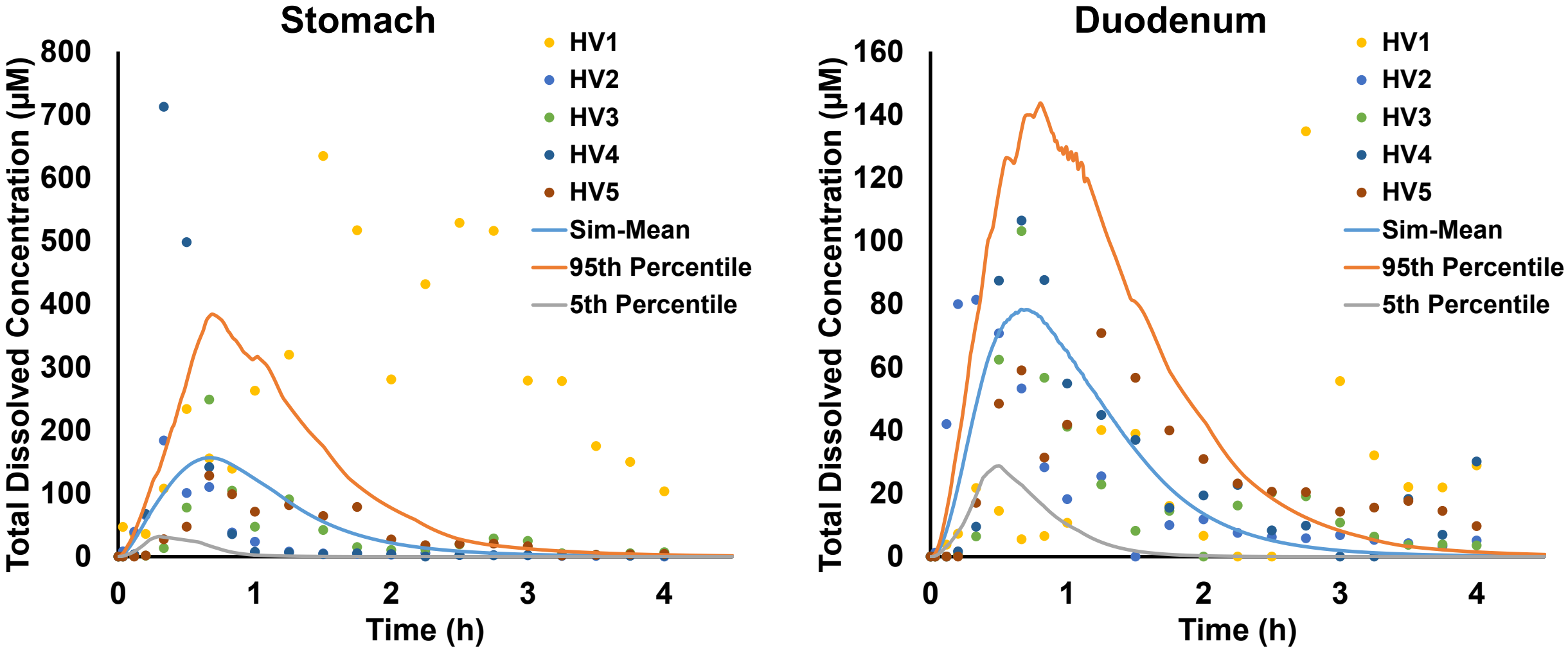
Kasserra et al., 2010



The fact that the model is able to recover multiple dose exposure following optimization of additional clearance from a single dose study also suggests that the model is able to capture the fraction metabolized by CYP3A4/5 relatively well.

CERTARA

Verification of the Model – Luminal Concentration Profiles

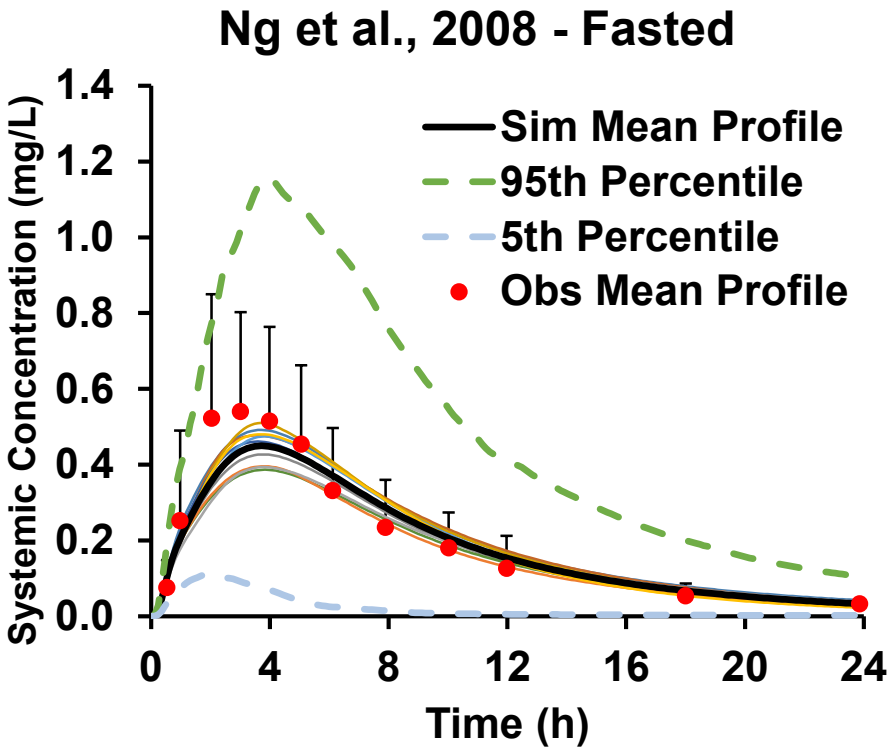


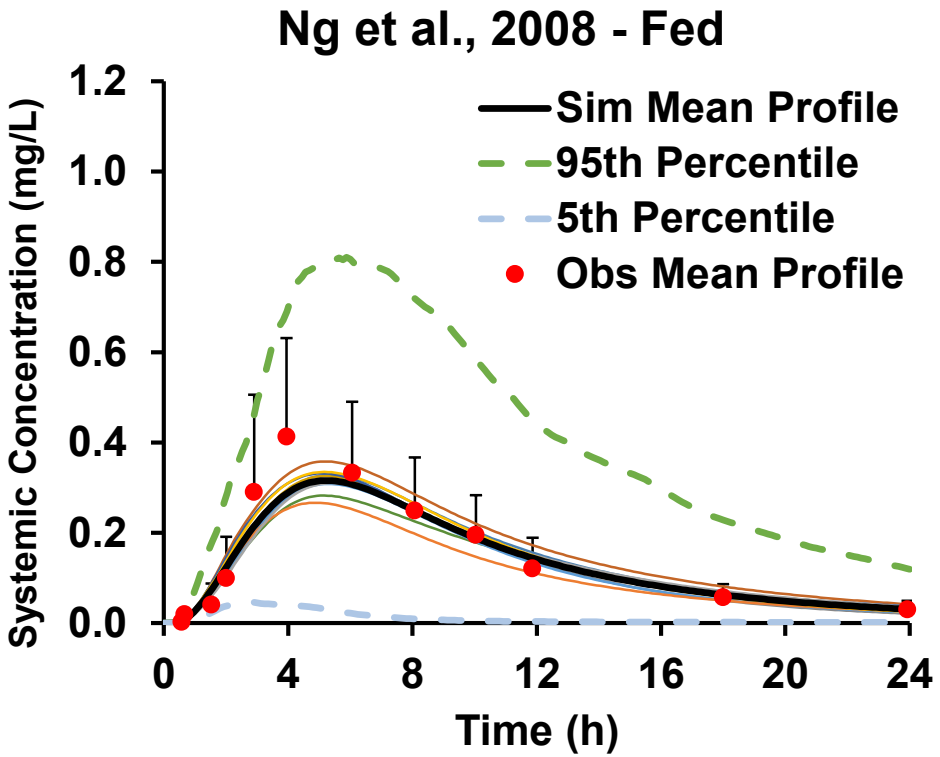
The model was able to capture both stomach and duodenal luminal profiles of ritonavir

Abeele et al. Eur J Pharm Sci. 2020 May25; 151:105377

CERTARA

Prediction of Negative Food Effects





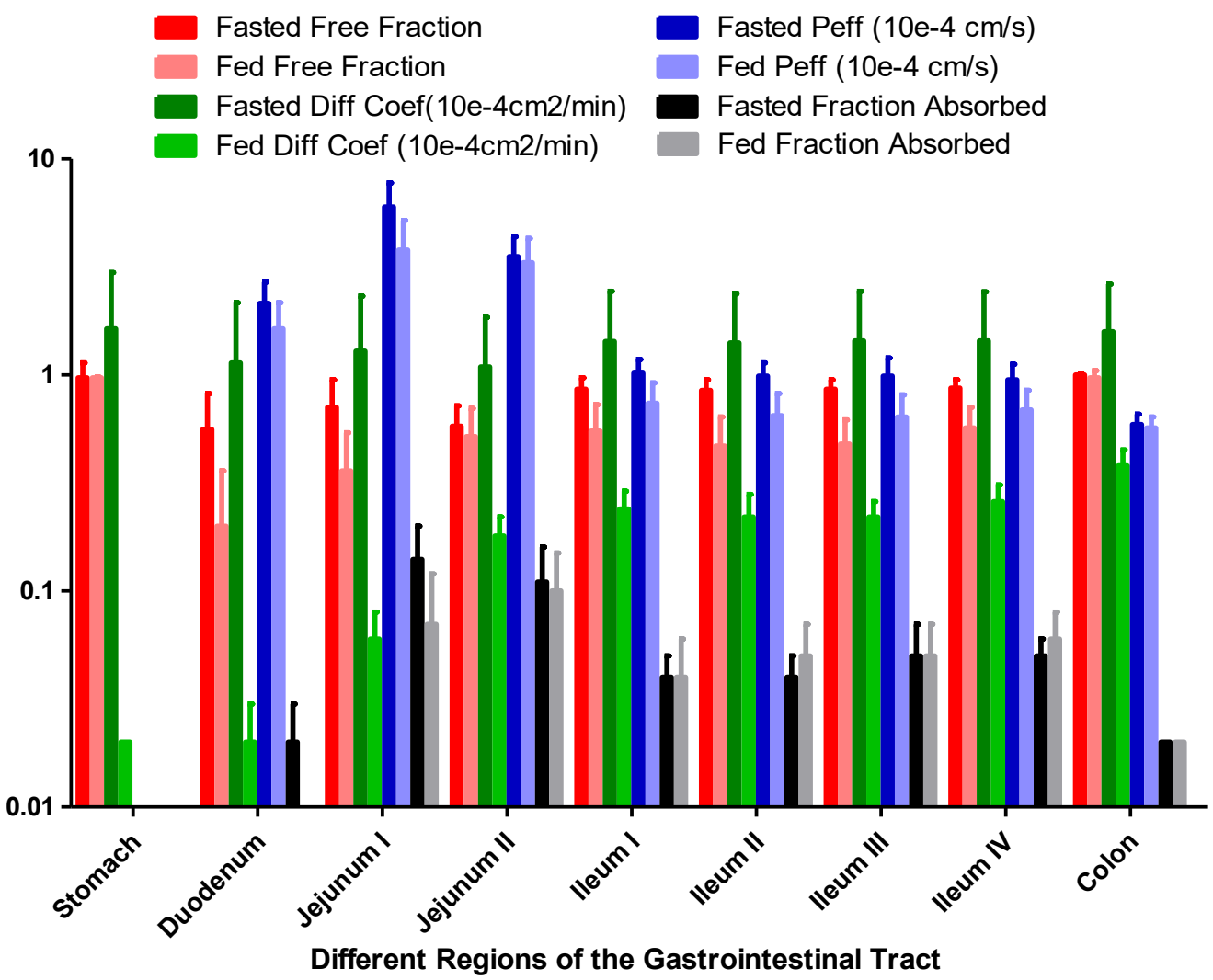
Simulated and Observed pharmacokinetic parameters (arithmetic means ± S.D.) of ritonavir following administration of 100 mg Norvir® Tablet under fasted and high-fat fed state.

PK Parameters	Observed Fasted	Simulated Fasted	Observed High-Fat Fed	Simulated High-Fat Fed	AAFE
C_{max} (µg/mL)	0.6 ± 0.31	0.49 ± 0.30***	0.44 ± 0.21	0.34 ± 0.25***	1.25
T_{max} (h)	3.2 ± 1.2	3.43 ± 1.18***	4.8 ± 1.1	4.75 ± 1.37***	1.04
AUC_{0-t} (µg/mL.h)	4.6 ± 2.0	4.46 ± 3.27***	3.5 ± 1.6	3.30 ± 2.86***	1.05

Ng *et al,* Journal of the International AIDS Society 2008, 11(Suppl 1):P247

CERTARA

Prediction of Negative Food Effects



Two factors were found to explain the observed negative food effect for ritonavir Norvir tablet –

A. Increased bile salt concentrations in fed state resulted in decrease in the free fraction of drug available as more drug is partitioned into bile salt micelles resulting in decreased Peff particularly in the duodenum and Jejunum I.

B. Decrease in effective diffusion coefficient of the drug in the fed state owing to increase in luminal viscosity – effect of dissolution rate

CERTARA

Comparison of Various Models

5 models were evaluated across 32 studies resulting in 160 workspaces

- **Model 1** – SV-Ritonavir 17 Simcyp File – First Order File
- **Model 2** – SV-Ritonavir 17 Simcyp File – Two First Order File with modified fa for fasted and fed state and Competitive Ki inhibition parameters of CYP3A4/5
- **Model 3** – ADAM Model with BD Sup ISEF Correction of V_{max} for CYP3A4/5
- **Model 4** – First order file with Fa = 0.5 (ADAM calculated) with BD Sup ISEF Correction of V_{max} for CYP3A4/5
- **Model 5** – ADAM Model with Competitive Ki inhibition parameters of CYP3A4/5

	AAFE (PK Parameters)		AAFE (DDI)	
	AUC	Cmax	AUC Ratio	Cmax Ratio
Model 1	1.32	1.38	1.83	1.42
Model 2	1.29	1.44	1.64	1.36
Model 3	1.50	1.49	1.79	1.45
Model 4	1.37	1.36	1.72	1.42
Model 5	1.49	1.53	1.52	1.31
Model 5 (with study matched formulation)	1.09	1.20	1.41	1.31
Model 3 (with study matched formulation)	1.09	1.20	1.64	1.45

AAFE – Absolute Average Fold Error
Model marked in green is the final model

CERTARA

Verification of systemic exposure of ritonavir following per oral administration of Norvir formulations

Norvir Capsule and Solution Formulation were simulated with 'Solution with Precipitation' Option

Reference	RTV Formulation	Simulation Option	Cmax (µg/mL)		$AUC_{(0-t)/(0-\infty)}$ (µg.h/mL)	
			Observed	Predicted	Observed	Predicted
Ng et al 2008	IR Tablet 100 mg	Immediate Release	0.6 ± 0.31	0.49 ± 0.30	4.6 ± 2.0	4.46 ± 3.27
Ng et al 2008	IR Tablet 100 mg	Immediate Release	0.44 ± 0.21	0.34 ± 0.25	3.5 ± 1.6	3.30 ± 2.86
NDA Application 20-945 – Capsule	Soft Elastic Capsule 6x100mg	Solution	11.98 ± 3.33	10.99 ± 3.73	108.1 ± 33	96.56 ± 44.60
Liu 2007	Capsule 400 mg BID	Solution	10.7 ± 3.19	9.57 ± 3.51	68 ± 21	67.09 ± 31.57
Liu 2007	Capsule 100 mg BID	Solution with Precipitation	1.41 ± 0.72	1.39 ± 0.63	7.81 ± 3.87	10.79 ± 5.86
Teng 2013	Soft Elastic Capsule 100 mg SD	Solution with Precipitation	0.51 ± 0.27	0.76 ± 0.42	4.29 ± 3.07	7.51 ± 4.85
Mathias 2009	Solution 100 mg SD	Solution with Precipitation	0.81 ± 0.27	0.88 ± 0.39	6.53 ± 1.76	7.70 ± 4.23
Mathias 2009	Solution 200 mg SD	Solution with Precipitation	2.46 ± 0.27	1.05 ± 0.5	16 ± 7	9.8 ± 5.53
Brennan 2013	IR 100mg SD	Immediate Release	0.69 ± 0.31	0.45 ± 0.29	4.55 ± 1.72	4.48 ± 3.61
Brennan 2015	IR 100mg SD	Immediate Release	0.44	0.45 ± 0.29	4.74	4.22 ± 3.54
Morris 2012	Capsule 100 mg SD	Solution with Precipitation	0.49 ± 0.39	0.59 ± 0.35	4.63 ± 2.89	5.98 ± 4.58
Morcosa 2014	IR 100 mg SD	Immediate Release	0.62	0.47 ± 0.31	4.74	4.13 ± 3.30
Aarnouste 2005	Capsule 100mg BID	Solution with Precipitation	0.89	1.38 ± 0.64	6.2	9.21

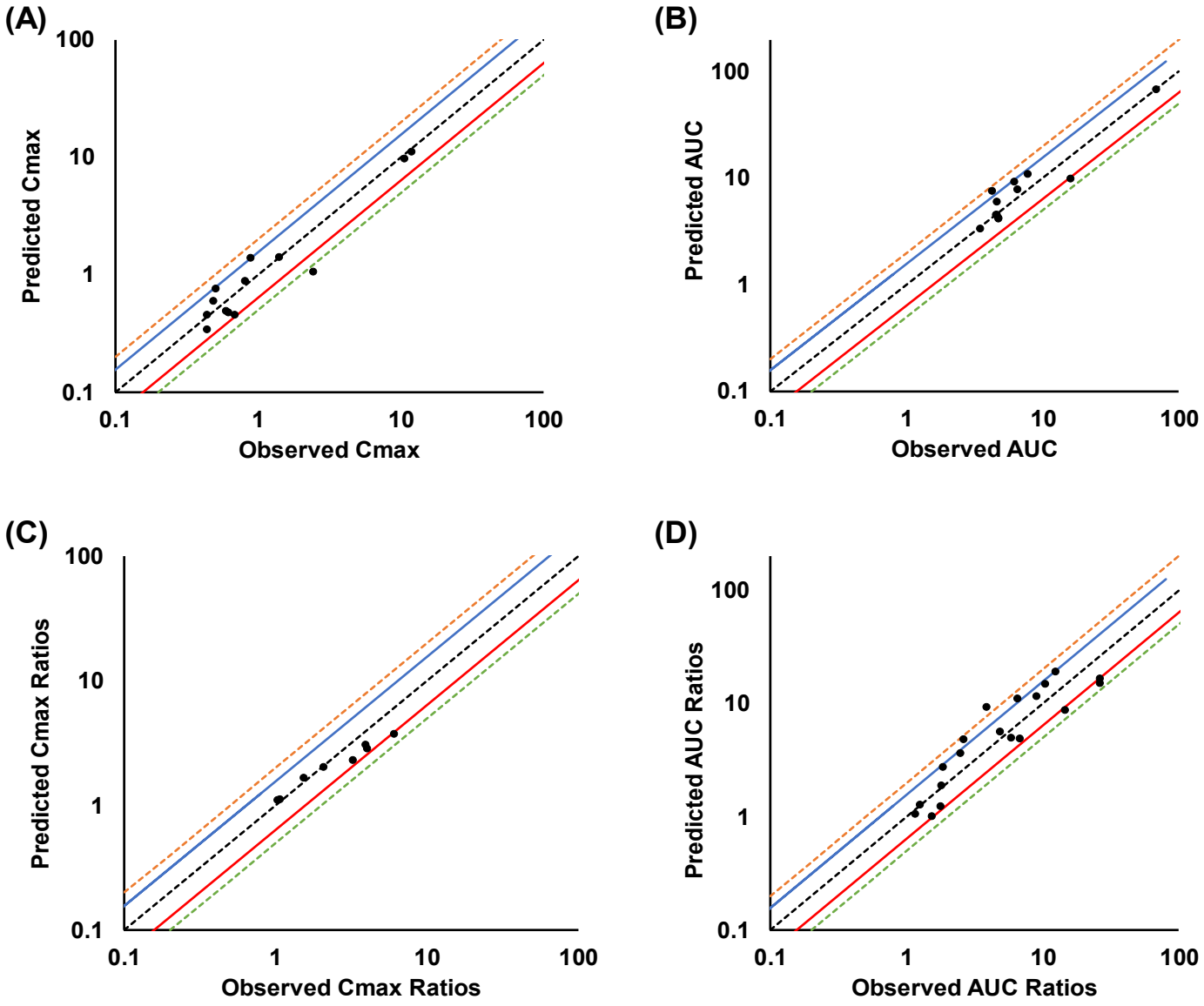
Arora *et al, Manuscript in Preparation*

DDI Verification of Ritonavir file as perpetrator of CYP3A4/5 and CYP2D6 substrate

Reference	RTV Formulation	Simulation Formulation	Cmax Ratios		$AUC_{(0-t)}$ Ratios	
			Observed	Predicted	Observed	Predicted
Eichbaum et al. 2013	Solution 0.1 mg	Solution with Precipitation	Not reported	NA	1.55	1.02 ± 0.0
Eichbaum et al. 2013	Solution 0.3 mg	Solution with Precipitation	Not reported	NA	1.17	1.07 ± 0.03
Eichbaum et al. 2013	Solution 1 mg	Solution with Precipitation	Not reported	NA	1.79	1.24 ± 0.12
Eichbaum et al. 2013	Solution 3 mg	Solution with Precipitation	Not reported	NA	1.82	1.89 ± 0.43
Eichbaum et al. 2013	Solution 10 mg	Solution with Precipitation	Not reported	NA	2.63	4.83 ± 2.27
Eichbaum et al. 2013	Solution 30 mg	Solution with Precipitation	Not reported	NA	3.89	9.38 ± 7.58
Eichbaum et al. 2013	Solution 100 mg	Solution with Precipitation	Not reported	NA	6.51	11.05 ± 9.44
Eichbaum et al. 2013	Solution 300 mg	Solution with Precipitation	Not reported	NA	9.01	11.68 ± 10.14
Mathias 2009	Solution 100 mg SD	Solution with Precipitation	Not reported	NA	6.81	4.90 ± 1.67
Mathias 2009	Solution 200 mg SD	Solution with Precipitation	Not reported	NA	4.90	5.70 ± 1.92
Ancrenaz et al. 2013	IR Tablet	IR	6.10	3.76 ± 1.78	26.5	16.67 ± 12.35
Greenblatt 2009	Solution	Solution with Precipitation	3.96	3.10 ± 1.28	26.41	15.08 ± 9.61
Leri 2013	Solution	Solution with Precipitation	2.09	2.03 ± 0.46	5.9	5.04 ± 1.97
Leri 2013	Solution	Solution with Precipitation	3.26	2.31 ± 0.63	14.7	8.79 ± 4.08
Mathias 2010	Capsule	Solution with Precipitation	4.03	2.87 ± 1.16	12.5	19.22 ± 11.21
Kirby 2011	IR tablet	Immediate Release	Not reported	NA	10.5(8.7-12.7)*	14.9 (13.41-16.55)
Greenblatt 2000	Capsule	Solution with Precipitation	1.04	1.10 ± 0.05	2.48	3.64 ± 1.25
Aarnouste 2005	Capsule 100 mg BID	Solution with Precipitation	1.08	1.13 ± 0.06	1.26	1.29 ± 0.12
Ouellet 1998	Solution 200 mg TID	Solution with Precipitation	1.54	1.68 ± 0.64	1.86	2.77 ± 2.28

Arora *et al, Manuscript in Preparation*

Model Performance Summary and Conclusions



Arora *et al, Manuscript in Preparation*

- Mechanism-based modelling of *in vitro* biopharmaceutics experiments helped to build confidence in the quality of the key input parameters which improved the predictivity of the developed PBPK model.
- The present study highlights the importance of inclusion of formulation properties within PBPK framework which significantly improved the prediction ability of the developed model both for ritonavir systemic exposure as well as DDI risk assessment.
- The compound file is available in Simcyp V19

Predicted and Observed Cmax (A) and AUC (B) of systemic exposure of ritonavir reported for different formulations following oral administration. Predicted and Observed Cmax ratios (C) and AUC ratios (D) of CYP3A4 (Midazolam, Alprazolam) and CYP2D6 (Desipramine, Clarithromycin) substrates with and without ritonavir administration. All data expressed as mean (in case of systemic exposure parameters) and mean ratios (DDI liability) plotted in logarithmic x and y-axis. Black dash line represents the line of unity. Orange and Green Dashed lines represent two –fold prediction space. Solid blue and red lines represent the bioequivalence (0.8-1.25) limits.

CERTARA

Limitations and Gaps

1. Disintegration rate of the Norvir Tablet in the fasted state is assumed to be the same in the fed state due to lack of in vitro data.

2. Use of static viscosity model – more research is needed in this area to parameterize and verify the dynamic viscosity model particularly time-dependent viscosity changes following administration of food in the small intestine.

3. Further experimental exploration of additional metabolic pathways which could enable bottom up predictions of ritonavir clearance *in vivo.*

Team

Sumit Arora

sumit.arora@certara.com

Amita Pansari

amita.pansari@certara.com

Peter Kilford

peter.kilford@certara.com

Iain Gardner

iain.gardner@certara.com

David B. Turner

david.turner@certara.com

Masoud Jamei

masoud.jamei@certara.com

Thank you for listening ☺

Questions?