MODELING MECHANISM BASED INACTIVATION USING PBPK

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Quantitative prediction of the magnitude of drug-drug interactions (DDI) is critical to underwriting patient safety in the clinical setting. Key mechanistic information can help to inform physiologically-based modeling and enable reasonable predictions of DDI magnitude, including complex scenarios such as inhibitory metabolites. This presentation will focus on integrating in vitro, preclinical and clinical data to develop quantitative predictions for DDIs due to mechanism-based inactivation.
OUTLINE

Introduction
- Definitions
- Assay types
- Regulatory Guidance
- Static and dynamic modeling

Case studies
- Delavirdine (determining mechanism)
- CTP-347 (isotope effects)
- Verapamil (complex DDI and pharmacogenomics)
DEFINITIONS

- **Time-dependent inhibition (TDI):** the apparent inhibitory potency of a new chemical entity (NCE) increases over time
  - Slow binders (rare), formation of inhibitory metabolites, mechanism-based inactivation

- **Metabolism-dependent inhibition (MDI):** the apparent inhibitory potency of an NCE increases over time, requiring one or more metabolic conversions
  - Formation of inhibitory metabolites, mechanism-based inactivation

- **Mechanism-based inactivation (MBI):** the apparent inhibition potency of an NCE increases over time due to a enzymatic product that irreversibly inactivates the enzyme and does not leave the active site

OVERVIEW

Two features dominate the impact of MBI on victim drugs

**Therapeutic Index**

**Contribution of clearance mechanisms ($f_m$)**

Additionally, reactive metabolites may leave the DME active site and haptenize other proteins, potentially leading to immune responses

IN VITRO ASSAYS

- Application of CYP MDI assays across the drug discovery / development continuum is dependent upon:
  - **Content:** Will this information impact decision making? How robust does the information need to be?
  - **Entry Point:** Throughput of Assay - What is the compound pressure for the assay?
  - **Follow-up:** When will a more rigorous assay be applied to further characterize the lead molecule?

Grimm, SW, et al. Drug Metab Dispos 37 1355-1370, 2009
Determining Whether an NME is a Time-Dependent Inhibitor

**FDA:** Any time-dependent loss of initial product formation rate may indicate time-dependent inhibition, and definitive in vitro studies to obtain TDI parameters (i.e., $k_{\text{inact}}$ and $K_I$ where $k_{\text{inact}}$ and $K_I$ are maximal inactivation rate constant and apparent inactivation constant, respectively) are recommended. If in vitro results suggest a TDI potential (e.g., $R>1.1$), an in vivo study is recommended. Alternatively, the sponsor can estimate the degree of drug-drug interactions using mechanistic models.

**EMA:** If the inhibition is enhanced by pre-incubations, time-dependent inhibition (TDI) is present. The increased inhibition over time may either be due to formation of an inhibitory metabolite or due to mechanism-based inactivation (MBI). For mechanism based inactivators, $k_{\text{inact}}$ (maximum inactivation rate constant) and $K_I$ (the inhibitor concentration producing half the maximal rate of inactivation) should be determined.

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[Regulatory Guidance](http://www.fda.gov/downloads/drugs/guidanceregulatoryinformation/guidances/ucm292362.pdf)


[In vitro studies](http://www.fda.gov/downloads/drugs/guidanceregulatoryinformation/guidances/ucm292362.pdf)


**FDA DDI Guidance, Figure 4**
PREDICTING THE IN VIVO SITUATION: STATIC EQUATIONS

**Definitions**

- **AUC**: Area Under the Plasma Concentration Time Curve
- **$f_m$**: Fraction metabolized by a specific CYP
- **$K_i$**: Measure of Inactivation Potency
- **$k_{inact}$**: Rate of Inactivation
- **$[I]_{invivo}$**: Inhibitor Concentration in vivo
- **$f_u$**: Fraction Unbound in Plasma
- **$k_{deg}$**:Degredation rate of the P450

**Single Inactivator**

\[
\frac{AUC_{[I]}}{AUC_{cir}} = \frac{1}{1 + \frac{k_{inact} \cdot f_u \cdot [I]}{(K_i + f_u \cdot [I])k_{deg}}} + (1 - f_m)
\]  

**Multiple Inactivators**

\[
\frac{AUC_{[I]}}{AUC_{cir}} = \frac{1}{1 + \sum_{i=1}^{n} \frac{k_{inact} \cdot f_u \cdot [I]}{(K_i + f_u \cdot [I])k_{deg}}} + (1 - f_m)
\]

The static approach assumes inactivator concentration is constant and maximal

Mayhew, BS, et al. Drug Metab Dispos 28 1031-1037, 2000
ESTIMATING CYP HALF-LIFE

**In vitro**

"Pulse-chase"

Enzyme level after induction

**In vivo**

Enzyme recovery after inactivation or induction

**CYP Half-life estimates**

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Method</th>
<th>n</th>
<th>t_{1/2} (h) *</th>
<th>Reference</th>
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<td>[87]</td>
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<td>Method 1</td>
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<td>[90]</td>
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<td>Method 2</td>
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<td>[74]</td>
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<td>80</td>
<td>[91]</td>
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<td>11</td>
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<td>[97]</td>
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<td>[85]</td>
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<td>CYP3A4</td>
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</table>

* t_{1/2} = 0.693 / k_{eq}. Values in brackets are ranges and * indicates ID
** estimated from an analysis of the reported data.

Half-life estimates may vary depending upon the method used for characterization.

Yang, J, et al. Curr Drug Metab. 9 384-394, 2008
IMPACT OF CYP HALF-LIFE ON PREDICTIONS

Azithromycin: weak TDI

Mibefradil: potent TDI

As $f_m$ increases, the impact of $k_{deg}$ increases

$k_{deg}$ of -- (1 day), – (3 days) or – (6 days)

Physiologically-based pharmacokinetic (PBPK) modeling integrates in vitro, in vivo and in silico data to simulate outcomes.

Incorporates physiological and physicochemical properties.

Treats the body as compartments connected by a circulatory system.

Integrates in vitro and clinical observations to support M&S.

PBPK is suited for modeling applications where:
- Changes in physiology or populations may impact PK variability.
- Changes in physicochemical properties or formulations may alter PK.
- Dynamic simulations of drug interactions are desired.
WHY SHOULD WE CARE ABOUT PBPK?

Regulatory Expectations

Application of PBPK to FDA IND/NDA Submissions (2008-2013, 33 Submissions)

Key regulatory questions in clinical pharmacology reviews:
• What intrinsic factors (age, gender, disease, polymorphism, etc) influence exposure?
• What extrinsic factors (drugs, herbal products, diet, etc) influence exposure?
• Based on exposure-response, what dosage regimen adjustments, if any, are recommended?

Potential for improved decision making
• Study Timing (delay until proof of concept achieved)
• Improved/abbreviated study design
• Necessity of clinical studies


PBPK enables the rational translation of in vitro or pre-clinical data to the clinical situation
MECHANISMS OF CYP INACTIVATION

Mechanisms of inactivation

Heme alkylation

Heme Destruction

Apoprotein alkylation

Metabolite Intermediate Complex (MIC) Formation

Diagnostic

Loss of heme (~400 nm in HPLC assay)

Alkylated protein

UV Spectroscopy

Hanson, KL, et al. Drug Metab Dispos 38 963-972, 2010
CASE STUDY: DELAVIRDINE

Greater than dose proportional exposure was observed in single ascending dose studies

Possible mechanisms for greater than dose proportional exposure:

• Decrease in elimination
• Metabolite inhibition
• Dose-dependent tissue distribution
• Dose-dependent absorption

What additional experiments can provide insight to the mechanism of nonlinear PK?
DELAVIRDINE: DETERMINING THE MECHANISM OF MBI

It is critical to distinguish between inhibition and inactivation for accurate simulations.

- Binding Spectrum
- Time-Dependent Inhibition of CYP3A4
- TDI Kinetics

In vitro experiments indicate apoprotein alkylation is the mechanism of inactivation.
DELAVIDINE: TRANSLATION TO IN VIVO PREDICTIONS

Marked differences in effects may be observed between the gut and liver

Predicted Single Dose PK

Predicted Effect: Gut CYP3A

Predicted Effect: Liver CYP3A

Predicted Multiple Dose PK (600 mg bid)

Predicted Effect: Gut and Liver CYP3A (600 mg bid)

Co-administration of delavirdine is contraindicated for drugs that are highly dependent on CYP3A for clearance and for which elevated plasma concentrations are associated with serious events.
PAROXETINE: REDUCING MBI IN VITRO

Predicting the effects of selective deuterium incorporation on metabolism can be difficult

CTP-347

In vitro experiments indicate CTP-347 does not inactivate CYP2D6
PAROXETINE: REDUCING MBI IN VIVO

PBPK reasonably simulates the effect of CYP2D6 inactivation on paroxetine PK

PK for paroxetine and CPT-347

Predicted CYP2D6 liver activity

Effect on urinary DM/DM

\( f_m \) paroxetine after multiple doses

\( f_m \) CPT-347 after multiple doses

PBPK modeling and simulation provide additional insight to the effects of MBI

Uttamsingh, V. et al. J Pharmacol Exp Ther 2015;354:43-54
VERAPAMIL: COMPLEX DDI

PBPK successfully simulated verapamil PK

Complex DDI: multiple species or mechanisms of DDI

Verapamil and metabolites

Inactivation through MIC

Verapamil enantiomers

Norverapamil enantiomers

Inactivation parameters (CYP3A4)

<table>
<thead>
<tr>
<th>Parameters</th>
<th>S-VPM</th>
<th>R-VPM</th>
<th>S-NVPM</th>
<th>R-NVPM</th>
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<tr>
<td>$K_i$ (uM)</td>
<td>4.9</td>
<td>33</td>
<td>4.9</td>
<td>10.7</td>
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<tr>
<td>$K_{inact}$ (min$^{-1}$)</td>
<td>0.034</td>
<td>0.038</td>
<td>0.080</td>
<td>0.048</td>
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</table>

Inactivation by D617 is an order of magnitude less

PBPK modeling

CYP3A activity in gut and liver


Wang, YH. et al. *Drug Metab Dispos* 2004; 32:259-266
VERAPAMIL: COMPLEX DDI AND PHARMACOGENETICS

CYP3A5 has been detected in 10-40% of Europeans, 33% of Japanese and 55% of African Americans

Norverapamil MIC (CYP3A4 and CYP3A5)

Inactivation parameters (CYP3A4/3A5 comparison)

<table>
<thead>
<tr>
<th>Parameters</th>
<th>S-VM (CYP3A4)</th>
<th>S-VM (CYP3A5)</th>
<th>NVPM (CYP3A4)</th>
<th>NVPM (CYP3A5)</th>
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</thead>
<tbody>
<tr>
<td>$K_i$ (uM)</td>
<td>0.87</td>
<td>8.0</td>
<td>4.6</td>
<td>38</td>
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<tr>
<td>$K_{inac}$ (min$^{-1}$)</td>
<td>0.13</td>
<td>0.03</td>
<td>0.42</td>
<td>0.05</td>
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PBPK successfully predicted the impact of CYP3A5 polymorphism on verapamil PK

Wang, YH. et al. Drug Metab Dispos 2005;33:664-671
Isoherranen, N, YH. et al. Drug Metab Dispos 2008;36:146-154
CONCLUSIONS

• Early screens can be used to determine if compounds exhibit time-dependent inhibition (TDI)
• “Hits” in the early TDI screen may be followed up by characterization experiments
• Mechanisms of CYP inactivation (MBI) include heme alkylation, heme destruction and apoprotein alkylation
• Static or dynamic models may be used to estimate the magnitude of DDI in vivo
• Modeling is sensitive to CYP half-life estimates, particularly at high $f_m$
• PBPK modeling may be used to perform dynamic simulations of drug-drug interactions
• PBPK is the preferred method for modeling and simulation of clinical situations involving “complex DDI”
ACKNOWLEDGEMENTS

Gary Skiles, Dan Rock, Rob Foti, Brooke Rock, Larry Wienkers