Population Pharmacokinetic Modelling of Plasma & Intracellular Once Daily Ritonavir-Boosted Darunavir in HIV-Infected Patients

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12th International Workshop on Clinical Pharmacology of HIV Therapy Miami, USA. 15 April 2011
Background

• Darunavir/ritonavir (DRV/RTV) approved for clinical use:
  o Treatment naïve: 800/100 mg once daily
  o Treatment experienced: 600/100 mg twice daily \(^{1,2}\)

• Metabolised by hepatic CYP3A4 and a substrate for efflux and influx (SLCO1A2, SLCO1B1\(^3\)) transporters \textit{in vitro}

• Co-administration with RTV (100 mg \textit{bid}) increased DRV (600 mg single dose) systemic exposure 14-fold and absolute oral bioavailability from 37% to 82% \(^{1,2}\)

\(^{1}\) Janssen-Cilag Ltd. Prezista® SPC 2010; \(^{2}\) Tibotec Inc. Prezista® US Prescribing Information 2010; \(^{3}\) Kwan WS \textit{et al.} \textit{B J Clin Pharmacol} 2009

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Background

• All antiretrovirals (except entry inhibitors) act within the cell

• May be of interest to determine intracellular concentrations (IC; concentrations within peripheral blood mononuclear cells, PBMC)

• Numerous factors may influence the distribution of drug between plasma and cellular compartments, impacting IC PK and ultimately efficacy
## Intracellular Exposure of PIs

<table>
<thead>
<tr>
<th>Regimen</th>
<th>N</th>
<th>Accumulation Ratio (IC AUC/Plasma AUC)*</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>DRV/RTV (600/100 mg qd)</td>
<td>10</td>
<td>1.3 (12.3)a 7.7 (13.0)a</td>
<td>ter Heine et al., BJCP 2010</td>
</tr>
<tr>
<td>DRV</td>
<td></td>
<td></td>
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<tr>
<td>RTV</td>
<td></td>
<td></td>
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<tr>
<td>ATV/RTV (200/100 mg qd + SQV 1600 mg qd)</td>
<td>12</td>
<td>1.2 (0.7-2.6) 1.7 (0.9-4.6)</td>
<td>Ford et al., JAC 2006</td>
</tr>
<tr>
<td>ATV</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>RTV</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SQV/RTV (1600/100 mg qd + ATV 200 mg qd)</td>
<td>12</td>
<td>4.9 (3.4-11.0) 1.7 (0.9-4.6)</td>
<td>Ford et al., JAC 2006</td>
</tr>
<tr>
<td>SQV</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>RTV</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SQV/RTV (1600/100 mg qd)</td>
<td>12</td>
<td>3.3 (1.5-6.7) 1.5 (0.8-4.2)</td>
<td>Ford et al., AVT 2004</td>
</tr>
<tr>
<td>SQV</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>RTV</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LPV/RTV (400/100 mg bid)</td>
<td>11</td>
<td>1.2 (0.7-2.1)b 4.6 (3.2-7.7)b</td>
<td>Crommentuyn et al., AVT 2004</td>
</tr>
<tr>
<td>LPV</td>
<td></td>
<td></td>
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<tr>
<td>RTV</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NFV (1250 mg bid)</td>
<td>12</td>
<td>5.3 (2.3-16.2) 2.3 (1.0-10.7)</td>
<td>Ford et al., AVT 2004</td>
</tr>
<tr>
<td>NFV</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>M8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NFV (1250 mg bid)</td>
<td>12</td>
<td>9.0 (9.6)c</td>
<td>Hennessy et al., AVT 2004</td>
</tr>
<tr>
<td>NFV</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IDV (800 mg tid)</td>
<td>10</td>
<td>0.3 (0.2)c</td>
<td>Hennessy et al., AVT 2003</td>
</tr>
<tr>
<td>IDV</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Data expressed as median (range)  
  a Data expressed as typical value (RSE%), population PK analysis  
  b Data expressed as median (IQR)  
  c Data expressed as mean (± s.d.)
Methods & Modelling Strategy
Clinical Studies

All blood sampling performed at steady-state following an observed, timed dose taken with food. All individuals were HIV-infected

- Study 1 Chelsea & Westminster Hospital, London, UK\(^1\)
  DRV/RTV 900/100 mg qd (effect on ABC PK)
  \textit{Plasma: 0, 0.5, 1, 2, 3, 4, 6, 8, 10, 12, 24 h}

- Study 2 St. Mary’s Hospital, London, UK\(^2\)
  DRV/RTV 800/100 mg qd (+TDF/FTC backbone)
  DRV/RTV 800/100 mg qd (+RGV +TDF/FTC backbone)
  DRV/RTV 800/100 mg qd (+RGV –TDF/FTC backbone)
  \textit{Plasma: 0, 0.5, 1, 2, 3, 4, 6, 8, 10, 12, 24 h}

- Study 3 Chelsea & Westminster Hospital, London, UK\(^3\)
  DRV/RTV 800/100 mg qd
  DRV/RTV 800/100 mg qd (+RGV)
  \textit{Plasma & PBMC: 0, 1, 2, 4, 6, 8, 12, 24 h}

\(^1\) Jackson A \textit{et al.} 12\textsuperscript{th} EACS 2009; \(^2\) Garvey L \textit{et al.} \textit{Antivir Ther} 2010; \(^3\) Jackson A \textit{et al.} 18\textsuperscript{th} CROI 2011

Presented at the 12\textsuperscript{th} Int. Workshop on Clin. Pharmacology of HIV Therapy, 13-15 April 2011, Miami, Fl, USA
Blood collected into Vacutainer cell preparation tubes (CPT). PBMCs separated from whole blood by centrifugation at room temperature.

Cells suspended in the plasma layer, removed from the CPT tube and isotonic saline (0.9%) added.

Cells were counted. Supernatant was removed by aspiration following centrifugation.

Cell pellet was resuspended in a lysis solution of ice cold 70% methanol.
Quantification of IC Concentrations

- Rapid elution time for 4 antiretrovirals plus internal standard within 8 minutes (LLQ DRV: 0.45 ng/ml; RTV: 0.48 ng/ml)

- Compounds were separated on an Ascentis, C18 Supelco column (flow rate: 400 µl/ min)

- Optimised gradient mobile phase [ACN: H2O (0.1% formic acid) 5:95 and 20:80]

- TSQ Quantum Ultra Triple Stage Quadrupole Mass Spectrometer (Thermo Fisher, UK) was used for detection and quantification

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Modelling

Serial blood sampling
(8-11 time points/patient)
1-3 PK profiles/patient
HPLC-MS/MS

PK Modelling
NONMEM v. VI 2.0

Structural Model

2 compartment model:
-Step-wise addition of mixed/random effects

Model fit & differences between models assessed:
-Statistical methods ($\Delta$OFV >3.84)
-Diagnostic plots

Covariate Model

Potential covariates:
RTV AUC$_{0-24}$ (plasma & IC), sex, ethnicity, age, weight, BMI, concomitant RGV

Retained if:
-Statistically significant, $\Delta$OFV >3.84
-Clinically relevant
-Backwards elimination, $\Delta$OFV >6.63

Simulations
Visual Predictive Check

3 PK studies$^{1-3}$
n=51 HIV patients
(7 female)
800/100 mg qd (n=32)
800/100 mg qd (n=24 IC)
900/100 mg qd (n=19)
n=1047 concentrations
n=384 IC concentrations

1 Jackson A et al. 12th EACS 2009; 2 Garvey L et al. Antivir Ther 2010;
3 Jackson A et al. 18th CROI 2011

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Simultaneous Modelling of Plasma & IC DRV

- IC DRV modelled as a proportion of plasma concentration estimating the DRV cellular accumulation ratio (CAR)

PLASMA: \( Y = F \times (1+\varepsilon_1) \)  

IC: \( Y = \text{CAR} \times F \times (1+\varepsilon_2) \)

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Results
Patient Demographics

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Median (range)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of patients [n (M/F)]</td>
<td>51 (44/7)</td>
</tr>
<tr>
<td>Age (yr)</td>
<td>39 (21-63)</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>74 (57-105)</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>24 (18-31)</td>
</tr>
<tr>
<td>Baseline CD4 cell count (cells/mm³)</td>
<td>500 (227-1129)</td>
</tr>
<tr>
<td>Undetectable viral load [n (%)]</td>
<td>49 (96)</td>
</tr>
<tr>
<td>Plasma DRV (mg/L)</td>
<td>2.82 (0.05-13.69)</td>
</tr>
<tr>
<td>IC DRV (mg/L)</td>
<td>10.36 (0.84-104.23)</td>
</tr>
<tr>
<td>Plasma RTV AUC₀-2₄ (mg.h/L)</td>
<td>4.35 (2.27-10.99)</td>
</tr>
<tr>
<td>IC RTV AUC₀-2₄ (mg.h/L)</td>
<td>31.01 (10.79-123.46)</td>
</tr>
</tbody>
</table>

*Unless stated otherwise
Pharmacokinetic Profiles

Plasma Darunavir

IC Darunavir

MEC1: treatment-naïve patients; 0.055 mg/L
MEC2: treatment-experienced patients; 0.550 mg/L

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# Covariates

## Potential covariates: Univariate Analysis

<table>
<thead>
<tr>
<th>Covariate</th>
<th>Equation</th>
<th>ΔOFV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plasma RTV AUC(_{0-24}) on CL/F</td>
<td>\text{CL/F} = \theta_1 \times (\text{RTV}/4.35)^{\theta_2}</td>
<td>-24.0</td>
</tr>
<tr>
<td>Age on CL/F</td>
<td>\text{CL/F} = \theta_1 \times (1+\theta_2 \times (\text{AGE}-39))</td>
<td>-19.1</td>
</tr>
<tr>
<td>IC RTV AUC(_{0-24}) on CAR</td>
<td>\text{CAR} = \theta_1 \times (\text{RTV IC}/31.01)^{\theta_2}</td>
<td>-18.5</td>
</tr>
<tr>
<td>Ethnicity on CAR</td>
<td>\text{CAR} = \theta_1 \times \theta_2^{\text{HISP}} \times \theta_3^{\text{BLK}} \times \theta_4^{\text{OTH}}</td>
<td>-6.5</td>
</tr>
<tr>
<td>Ethnicity on (k_a)</td>
<td>\text{(k_a)} = \theta_1 \times \theta_2^{\text{HISP}} \times \theta_3^{\text{BLK}} \times \theta_4^{\text{OTH}}</td>
<td>-6.1</td>
</tr>
<tr>
<td>Ethnicity on CL/F</td>
<td>\text{CL/F} = \theta_1 \times \theta_2^{\text{HISP}} \times \theta_3^{\text{BLK}} \times \theta_4^{\text{OTH}}</td>
<td>-5.4</td>
</tr>
</tbody>
</table>

- RTV plasma & IC AUC\(_{0-24}\) impact DRV PK parameters
- Suggestion of ethnic differences in some parameters but low numbers (Hispanic n=3; Black African n=11; Other n=7 vs. n=30 Caucasian)
- Low frequency of older patients, n=8 >50 years, n=1 >60 years
Diagnostic Plots: Plasma
following addition of covariates

Observed vs. Predicted

Observed vs. Individual predicted

Observed vs. Predicted

Observed vs. Individual predicted

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Diagnostic Plots: Intracellular

following addition of covariates

Observed vs. Predicted

Observed vs. Predicted

Individual predicted DRV (mg/L)

Observed DRV (mg/L)

Predicted DRV (mg/L)

R² = 0.3334

R² = 0.255

Observed DRV (mg/L)

Predicted DRV (mg/L)

R² = 0.6234

R² = 0.6209

Individual predicted DRV (mg/L)

Observed DRV (mg/L)

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# Estimated Pharmacokinetic Parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Estimate (RSE%)</th>
<th>IIV (%) (RSE%)</th>
<th>IOV (%) (RSE%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CL/F (L/h)</td>
<td>12.5 (4)(^a)</td>
<td>12 (60)</td>
<td>20 (19)</td>
</tr>
<tr>
<td></td>
<td>15.6 (6)(^b)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>V2/F (L)</td>
<td>125 (8)(^a)</td>
<td>36 (23)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>186 (11)(^b)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Q/F (L/h)</td>
<td>13.4 (6)</td>
<td>58 (43)</td>
<td></td>
</tr>
<tr>
<td>V3/F (L) fixed</td>
<td>84 (n/a)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>kₐ (h⁻¹)</td>
<td>0.9 (11)</td>
<td>33 (113)</td>
<td>54 (27)</td>
</tr>
<tr>
<td>Lag-time (h)</td>
<td>0.4 (4)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CAR</td>
<td>4.8 (5)</td>
<td>0 (fixed)</td>
<td>22 (43)</td>
</tr>
<tr>
<td>Covariates</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>( \theta_{RTV\ AUC0-24} ) on CL/F</td>
<td>-0.40 (22)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>( \theta_{AGE} ) on CL/F</td>
<td>-0.014 (22)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>( \theta_{IC\ RTV\ AUC0-24} ) on CAR</td>
<td>0.41 (27)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Residual error</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Plasma (%)</td>
<td>26 (8)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IC (%)</td>
<td>40 (13)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

\( RSE = \frac{SE_{\text{estimate}}}{\text{estimate}} \times 100 \)

\(^a\) Study 1 & 2

\(^b\) Study 3

IIV: interindividual variability

IOV: interoccasion variability

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Visual Predictive Check

90% Prediction Interval

Plasma
- Observed plasma DRV; n=1047
- 89% within prediction interval

Intracellular
- Observed IC DRV; n=384
- 97% within prediction interval

- P5, P50, P90
- MEC1: treatment-naïve patients; 0.055 mg/L
- MEC2: treatment-experienced patients; 0.550 mg/L

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Discussion

• Based on visual predictive check, model provides adequate description of plasma and IC DRV
• However, CAR of 4.8 vs. 1.3 for ter Heine et al.

<table>
<thead>
<tr>
<th></th>
<th>Present analysis</th>
<th>ter Heine et al.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cell collection</td>
<td>Vacutainer® CPT</td>
<td>Vacutainer® CPT</td>
</tr>
<tr>
<td>Cell treatment</td>
<td>No wash</td>
<td>2 x wash PBS</td>
</tr>
</tbody>
</table>

• DRV loss through washes (See poster P01, Watson et al).
• Initial modelling of RTV data suggest it is not as susceptible to the washing process (CAR: 7.9 vs. 7.7)
• Consistency needed for methods of cell collection & treatment
Conclusions

- PK modelling ideal approach to simultaneously analyse plasma & IC concentrations
- Help identify important covariates influencing distribution of drug between plasma & cellular compartments (can also link to PD endpoints)

- IC DRV modelled as a proportion of plasma DRV
- RTV plasma and IC $AUC_{0-24}$ significantly associated with DRV CL/F and CAR, respectively
- Impact of age on DRV CL/F requires clarification
- Further study required to identify covariates governing IC PK of DRV/RTV and their impact on efficacy
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