Population pharmacokinetic/pharmacodynamic modeling of a new antithrombotic drug, the Nanobody® ALX-0081/ALX-0681

Stefaan Rossenu, Senior Scientist, Ablynx NV

PAGE, 2011 (7-10 June, 2011)
Athens, Greece
Agenda

Introduction
- anti-von Willebrand Factor (vWF) Nanobody (ALX-0081/ALX-0681)
  - product characteristics and mode of action
  - PK characteristics

PK/PD modeling results
- structural model
- final model
- VPC (PK and PD)
- application of the model (special populations)

Conclusions
Introducing Ablynx and Nanobodies®
Conventional antibodies vs Nanobodies

Camelidae family has both forms

Conventional antibody
Heavy and light chains
Both chains required for antigen binding and stability

Heavy-chain antibody
Only heavy chains
Full antigen binding capacity and very stable

Ablynx’s Nanobody®
Based on the smallest functional fragment of a naturally occurring heavy-chain antibody
Nanobodies – beyond antibodies & small molecules

- Cavity binding
- Difficult targets (e.g. GPCRs)
- High solubility
- Low tendency to aggregate
- Resistant to heat, pH, proteases.
- Non-injectable (e.g. pulmonary)
- Robust
- Broad target applicability
- High affinity
- High selectivity
- High homology to human V\text{H}
- Tissue penetration
- Ease & low cost of manufacturing
- High potency/low inherent toxicity
- Alternative routes of administration
- Flexible formatting
- Bispecific & multivalent formats
- Tailored half-life

Ablynx’s Nanobody
12-15kD
1/10 size of a mAb

www.ablynx.com
Anti-von Willebrand Factor (vWF) Nanobody

- **Anti-vWF Nanobody product characteristics**
  - bivalent construct - high potency through formatting
  - no half-life extension - “small molecule” pharmacology with mAb specificity
  - 2 forms of administration (ALX-0081 iv and ALX-0681 sc)

- **Targets first mover in cascade of thrombosis**
  - could prevent unwanted blood clot formation
  - in animal models, demonstrated highly potent inhibition of clotting without increased bleeding – possible large therapeutic window

- **Targets pathophysiological mediator in Thrombotic Thrombocytopenic Purpura (TTP)**

- **Potentially first-in-class**

- **Manufactured in E. coli** – relatively low cost of goods

- **Phase II trials in progress in two indications** – ACS/PCI and TTP
ALX-0081/ALX-0681 pharmacokinetics

What do we see?

What are we measuring?

- total ALX-0081/ALX-0681 (free + complex)
- total vWF (free + complex)

Hypothesis

- the free drug will be cleared by the kidney
- the drug in complex will be cleared via their receptor
- PK profile total ALX-0081/ALX0681 is the superposition of the PK profiles of free drug and drug in complex
Target mediated drug disposition (TMDD)

- clearance pathway can be saturated
- depending on the vWF levels in circulation
Agenda

Introduction

- anti-von Willebrand Factor (vWF) Nanobody (ALX-0081/ALX-0681)
  - product characteristics and mode of action
  - PK characteristics

PK/PD modeling results

- structural model
- final model
- VPC (PK and PD)
- application of the model (special populations)

Conclusions
### Summary of included studies

<table>
<thead>
<tr>
<th>Study number</th>
<th>Type</th>
<th>Subject N° active Population</th>
<th>Route, dose regimen and formulation</th>
<th>PK PD sampling scheme</th>
<th>Number of samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALX-0081-1.2/08a</td>
<td>Phase I</td>
<td>Patients n=12</td>
<td>SD iv 60 min, 2, 4, 6, 9 mg</td>
<td>rich</td>
<td>PK=97 PD=120</td>
</tr>
<tr>
<td>ALX-0081-1.2/08b</td>
<td>Phase I</td>
<td>Patients N=6</td>
<td>MD iv 60 min, 6 mg+(3x4mg)</td>
<td>sparse</td>
<td>PK=41 PD=48</td>
</tr>
<tr>
<td>ALX-0081-1.2/08c</td>
<td>Phase I</td>
<td>Patients N=20</td>
<td>MD iv bolus, 6 mg+(3x4 mg)</td>
<td>sparse</td>
<td>PK=119 PD=151</td>
</tr>
<tr>
<td>OLE</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ALX-0081 sc a</td>
<td>Phase I</td>
<td>Healthy N=15</td>
<td>SD sc 2, 4, 8, 10, 16 mg</td>
<td>rich</td>
<td>PK=135 PD=177</td>
</tr>
<tr>
<td>ALX-0081 sc b</td>
<td>Phase I</td>
<td>Healthy N=12</td>
<td>MD sc 10 mg od</td>
<td>rich</td>
<td>PK=456 PD=234</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>65</td>
<td></td>
<td></td>
<td>PK=848 PD=730</td>
</tr>
</tbody>
</table>
ALX-0081/ALX-0681 Pop PK/PD Raw data

Individual Profiles/Dose Group

DOSE = 2 mg

DOSE = 4 mg

DOSE = 8 mg

Total ALX-0081 Plasma concentrations, ng/mL

Individual Profiles/Dose Group

DOSE = 2 mg

DOSE = 4 mg

DOSE = 8 mg

Total vWF, nM

Total ALX-0081 Plasma concentrations, ng/mL

Total vWF, nM

Bloodgroup O

Substantial variability in PK and PD
10 mg multiple dose (14 days)

Total ALX-0681, ng/mL
Total median vWF, nM

Time, d

PK
PD
ALX-0081/ALX-0681 Pop PK/PD
Structural model

Dose

Assumptions:
- bioavailability is 100%
- $K_{off}$ is negligible

<table>
<thead>
<tr>
<th>Model</th>
<th>OFV</th>
<th>Delta</th>
</tr>
</thead>
<tbody>
<tr>
<td>3 CMT with 1 Ka</td>
<td>11373.5</td>
<td>-</td>
</tr>
<tr>
<td>3 CMT with zero-first order input</td>
<td>11255.6</td>
<td>-117.9</td>
</tr>
<tr>
<td>4 CMT with zero-first order input</td>
<td>11212.5</td>
<td>-43.1</td>
</tr>
<tr>
<td>Previous + linear time dependent increase of kin</td>
<td>11047.6</td>
<td>-164.9</td>
</tr>
</tbody>
</table>
Which covariates are statistically significant:

- forward addition (plug-in all covariates at once)
- one by one backward elimination (if increase in OFV > 10.83, keep it in)

<table>
<thead>
<tr>
<th>Model</th>
<th>OFV</th>
<th>Delta</th>
</tr>
</thead>
<tbody>
<tr>
<td>Full covariate model</td>
<td>10966.9</td>
<td>-</td>
</tr>
<tr>
<td>Full – WGT (KA₁)</td>
<td>10979.1</td>
<td>12.2</td>
</tr>
<tr>
<td>Full – CRCL (CL)</td>
<td>10992.6</td>
<td>25.7</td>
</tr>
<tr>
<td>Full – DST (K_{com})</td>
<td>10997.0</td>
<td>30.1</td>
</tr>
<tr>
<td>Full – BLG (Baseline vWF)</td>
<td>10976.2</td>
<td>9.3</td>
</tr>
</tbody>
</table>
### ALX-0081/ALX-0681 Pop PK/PD

**Final model (estimated parameters)**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Estimate</th>
<th>RSE (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>KA1 (h⁻¹)</td>
<td>0.11</td>
<td>12</td>
</tr>
<tr>
<td>V3 (L)</td>
<td>5.18</td>
<td>16</td>
</tr>
<tr>
<td>CL (L.h⁻¹)</td>
<td>5.12</td>
<td>9</td>
</tr>
<tr>
<td>BASE (nM)</td>
<td>49.1</td>
<td>6</td>
</tr>
<tr>
<td>Kin (nM.h⁻¹)</td>
<td>1.23</td>
<td>10</td>
</tr>
<tr>
<td>Kcom (h⁻¹)</td>
<td>0.0488</td>
<td>10</td>
</tr>
<tr>
<td>Kon (nM⁻¹.93.h⁻¹)</td>
<td>0.00691</td>
<td>59</td>
</tr>
<tr>
<td>MOL</td>
<td>1.93</td>
<td>5</td>
</tr>
<tr>
<td>KA2 (h⁻¹)</td>
<td>0.0616</td>
<td>23</td>
</tr>
<tr>
<td>RDPR1 (d⁻¹)</td>
<td>0.0437</td>
<td>19</td>
</tr>
<tr>
<td>DUR (h)</td>
<td>2.33</td>
<td>11</td>
</tr>
<tr>
<td>V5 (L)</td>
<td>2.12</td>
<td>7</td>
</tr>
<tr>
<td>WGT (KA1)</td>
<td>-2.07</td>
<td>31</td>
</tr>
<tr>
<td>CRCL (CL)</td>
<td>1.01</td>
<td>20</td>
</tr>
<tr>
<td>DST (Kcom)</td>
<td>0.555</td>
<td>9</td>
</tr>
<tr>
<td>IIV KA1</td>
<td>39%</td>
<td>28</td>
</tr>
<tr>
<td>IIV V3</td>
<td>48%</td>
<td>50</td>
</tr>
<tr>
<td>IIV CL</td>
<td>23%</td>
<td>95</td>
</tr>
<tr>
<td>IIV BASE</td>
<td>37%</td>
<td>21</td>
</tr>
<tr>
<td>IIV Kin</td>
<td>40%</td>
<td>28</td>
</tr>
<tr>
<td>IIV Kcom</td>
<td>22%</td>
<td>62</td>
</tr>
<tr>
<td>IIV V5</td>
<td>10%</td>
<td>69</td>
</tr>
<tr>
<td>Sigma (PK)</td>
<td>15%</td>
<td>12</td>
</tr>
<tr>
<td>Sigma (PD)</td>
<td>18%</td>
<td>13</td>
</tr>
</tbody>
</table>

- Free compound distributed in blood and interstitial fluid (>2.5L)
- Free compound cleared via glomerular filtration
- 1.9 vWF molecules binds to 1 ALX molecule
- Complex only present in the blood (≈2.5L)

\[ \text{t}_{1/2} \text{ of complex in:} \]

\[ \text{HV} = 14h \]
\[ \text{PAT} = 26h \]
ALX-0081/ALX-0681 Pop PK/PD
Final model (GOF plots)
ALX-0081/ALX-0681 Pop PK/PD
Final model (GOF plots)
ALX-0081/ALX-0681 Pop PK/PD
Visual predictive check (PK)

www.ablynx.com
ALX-0081/ALX-0681 Pop PK/PD
Visual predictive check (PD)
ALX-0081/ALX-0681 Pop PK/PD
Visual predictive check

Same subject outside the 90% prediction interval for PK and PD
• vWF level is very high in this case which lead to high total ALX-0081 concentrations
ALX-0081/ALX-0681 Pop PK/PD
Effect of renal clearance on PK and PD

10 mg Multiple Dose (8 days)

Total ALX-0681, ng/mL

Time, days

30 mL/min
140 mL/min

Total vWF, %

Time, days

30 mL/min
140 mL/min
Conclusions

A popPK/PD model is developed that describes the total ALX-0081/ALX-0681 and total vWF profiles

- ALX-0081/ALX-0681 is going into the systemic circulation via two pathways i.e. directly and indirectly (e.g. lymphatic system)
- free ALX-0081/ALX-0681 is distributed outside the central compartment
- ALX-0081/ALX-0681 in complex stays in the blood
- rapid clearance of free ALX-0081/ALX-0681 via glomerular filtration
- low accumulation potential of the drug

Three covariates are clinically relevant

- weight on Ka1
- CRCL on CL
- disease status on $K_{com}$

Model can be used to guide dose and dosing regimen in special populations (Renal impaired subjects, pediatric population)
Acknowledgements

Pharmacology
• ML Sargentini-Maier
• J Baumeister
• H Ulrichts
• S Priem

Clinical
• JB Holz
• C Duby
• D Tersago
• K Vercruysse
• C Lyssens
Population pharmacokinetic/pharmacodynamic modeling of a new antithrombotic drug, the Nanobody® ALX-0081/ALX-0681

Stefaan Rossenu, Senior Scientist, Ablynx NV

PAGE, 2011 (7-10 June, 2011)
Athens, Greece