Novel Adenovirus Vector-Based Vaccines for HIV-1

Raphael Dolin
Dan Barouch
August 21, 2007
Pre-Existing Ad5 Vector-Specific Immunity

- High prevalence of pre-existing immunity to Ad5 in human populations, particularly in the developing world
- Major limitation of current rAd5 vector-based vaccines
- Ad5 Seroprevalence:
  - U.S., Western Europe: 30-50% (low to moderate titers)
  - Sub-Saharan Africa, Southeast Asia: 80-95% (high titers)
- Blunts rAd5 vaccine immunogenicity in both preclinical and clinical studies and may limit their clinical utility
Novel Adenovirus Vectors for HIV-1: Potential Applications

• To replace Ad5 vectors in regions of the world where pre-existing anti-Ad5 immunity is limiting

• To be used together with Ad5 vectors or other vectors in heterologous prime-boost regimens
Development of Novel Adenovirus Vectors

• Goal is to develop novel rAd vectors with:
  • Immunogenicity of rAd5 vectors
  • Capacity to circumvent anti-Ad5 immunity
  • Large-scale manufacturability

• Key strategies:
  ➢ Novel serotype rAd vectors
  ➢ Novel chimeric rAd vectors
Comparative Evaluation of Six Novel Serotype rAd Vectors from Ad Subgroups B and D

• Novel serotype rAd vectors evaluated:
  • rAd11, rAd35, rAd50 (subgroup B)
  • rAd26, rAd48, rAd49 (subgroup D)

• rAd26 selected as optimal rare serotype vector based on:
  • Seroprevalence studies in U.S. and sub-Saharan Africa
  • Immunogenicity studies in mice and rhesus monkeys
  • Manufacturability studies

The 7 Hexon HVRs Form the Majority of the Solvent-Exposed Surface of the Ad5 Hexon Capsid Protein

Schematic Ad5 Particle
Hexons Shown in Blue

Hexon Protein Trimer
HVRs Shown in Color
Hexon HVR-Chimeric rAd5 Vectors

Ad5 genome

Ad5 Hexon

Ad5HVR48(1) Hexon

Ad5HVR48(1-7) Hexon

Chimeric rAd5HVR48(1-7)-Gag Vector Effectively Circumvents Anti-Ad5 Immunity in C57BL/6 Mice

Naive

Anti-Ad5 Immunity

Comparative Assessment of Novel rAd Vectors in Rhesus Monkeys with Anti-Ad5 Immunity

- 30 rhesus monkeys pre-immunized with 2 injections of $10^{11}$ vp rAd5-Empty (median Ad5 NAb titers 16,384)

- Single injection of $10^{10}$ vp of the following rAd vectors expressing SIV Gag, Env, Pol, Nef (N=6/group):
  - 1) Sham
  - 2) rAd5
  - 3) rAd5HVR48
  - 4) rAd26
  - 5) rAd35
Novel rAd26 and rAd5HVR48 Vectors Elicit 5-Fold More Potent Cellular Immune Responses rAd5 Vectors in Rhesus Monkeys with Anti-Ad5 Immunity

30 Rhesus Monkeys, N=6/Group

* two-tailed Wilcoxon rank-sum test
Novel Serotype/Chimeric rAd Vectors for HIV-1: rAd26 and rAd5HVR48

- Significantly 5-fold more immunogenic than rAd5 vectors in rhesus monkeys with anti-Ad5 immunity

- More potent than five other rare serotype rAd vectors tested (Ad11, Ad35, Ad50, Ad48, Ad49)

- Immunogenic both as single modality vaccines and in the context of heterologous prime-boost regimens

- Elicit polyfunctional CD8+ and CD4+ T cell responses

- Generate both mucosal and systemic central memory T cell responses following intramuscular immunization
Heterologous rAd Prime-Boost Regimens

• DNA/rAd5 prime-boost regimens more immunogenic than rAd5 alone regimens but logistically complex

• Heterologous rAd/rAd prime-boost regimens using two serologically distinct rAd vectors offer a potentially more practical alternative

• Immunogenicity study in rhesus monkeys to compare rAd5/rAd5 with heterologous rAd/rAd prime-boost regimens
8-Fold Greater Immunogenicity of rAd26/rAd5 vs rAd5/rAd5 Vaccine Regimens in Rhesus Monkeys

Optimal Regimen: rAd26 Prime, rAd5 Boost

IFN-γ ELISPOT responses following boost immunization
Cytokine Secretion Profiles of Gag-Specific CD8 and CD4 T Lymphocyte Responses Elicited by Optimal rAd26/rAd5 Regimen in Rhesus Monkeys

ICS responses at week 4 following boost immunization
Protective Efficacy of Heterologous rAd Prime-Boost Regimens Against SIVmac251 in Rhesus Monkeys

- Rhesus monkeys immunized with various rAd regimens expressing the single SIV Gag antigen:
  - rAd26-Gag prime, rAd5-Gag boost (N=6)
  - rAd35-Gag prime, rAd5-Gag boost (N=6)
  - rAd5-Gag prime, rAd5-Gag boost (N=4)
  - Sham (N=6)

- High-dose i.v. SIVmac251 challenge (provided by Norman Letvin) 6 months following the boost immunization

- Highly stringent challenge model:
  - Merck rAd5-Gag did not suppress SIV RNA post-challenge
  - 0.8-1.1 log decrease in peak SIV RNA observed with multiple vaccine antigens and DNA priming
Significant 1.4 Log Decrease in Peak SIV RNA Levels in rAd26/rAd5 Vaccinated Monkeys Post-Challenge

SIV RNA Levels; Day 14 Post-Challenge

* two-tailed Wilcoxon rank-sum test
Protection Correlated with Preservation of Central Memory CD4+ T Lymphocytes

CD28+CD95+CD4+/CD4+ Cells; Day 14 Post-Challenge

* two-tailed Wilcoxon rank-sum test
Protection Correlated with Preservation of CCR5+CD4+ T Lymphocytes

CCR5+CD4+/CD4+ Cells; Day 14 Post-Challenge

* two-tailed Wilcoxon rank-sum test
Protection Correlated with Preservation of Duodenal Memory CD4+ T Lymphocytes

CD4+/CD3+ Cells; Day 21 Post-Challenge

* two-tailed Wilcoxon rank-sum test
Protective Efficacy of Heterologous rAd Prime-Boost Regimens Against SIVmac251 in Rhesus Monkeys

- Results preliminary and restricted to acute infection; long-term follow-up data not yet available

- Protective efficacy correlates with immunogenicity:
  - rAd26/rAd5 > rAd35/rAd5 > rAd5/rAd5 > Sham

- Demonstrates potential of heterologous rAd prime-boost regimens, particularly rAd26/rAd5 regimen

- Protection correlated with preservation of CCR5+CD4+ T cells and duodenal memory CD4+ cells, as well as total central memory CD4+ cells
NIAID Integrated Preclinical/Clinical AIDS Vaccine Development (IPCAVD) Program

- Vectors selected for advancement into clinical trials:
  - Ad26 - optimal rare serotype Ad vector
  - Ad5HVR48 - optimal chimeric Ad vector

- Antigen selected for phase I studies of prototype vectors:
  - Clade A HIV-1 Env gp140 (Gary Nabel, VRC, NIH)
  - Plans for a complete multivalent vaccine in progress

- HER.96 cells selected for GMP manufacturing (Crucell, IAVI)
Manufacturing Clinical-Grade rAd26 and rAd5HVR48 Expressing Clade A HIV-1 Env gp140 (Crucell)

<table>
<thead>
<tr>
<th></th>
<th>2006</th>
<th></th>
<th>2007</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Q2</td>
<td>Q3</td>
<td>Q4</td>
<td>Q1</td>
</tr>
<tr>
<td>Assay development</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ad26</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Process development</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Research batches</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pre-MVS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MVS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tech Transfer</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CTM</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ad5HVR48</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Process development</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Research batches</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pre-MVS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MVS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tech Transfer</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CTM</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

- Production
- Testing
- Release

Q1 2006: Assay development for Ad26 and Ad5HVR48
Q2 2006: Process development for Ad26
Q3 2006: Process development for Ad5HVR48
Q4 2006: Research batches for both Ad26 and Ad5HVR48
Q1 2007: MVS for Ad26
Q2 2007: MVS for Ad5HVR48
Q3 2007: Tech Transfer for both Ad26 and Ad5HVR48
Q4 2007: CTM for both Ad26 and Ad5HVR48
Current Timeline to Clinical Trials

- **Ad26.ENVA.01**
  - ✓ Pre-IND package reviewed by FDA (Q4 2006)
  - ✓ GMP clinical trial material manufactured (Q1 2007)
  - ✓ GLP toxicology studies completed (Q2 2007)
  - • IND submission planned (Q3 2007)
  - • Phase I study planned (Q4 2007)

- **Ad5HVR48.ENVA.01**
  - ✓ Pre-IND package reviewed by FDA (Q1 2007)
  - ✓ GMP clinical trial material manufactured (Q2 2007)
  - ✓ GLP toxicology studies in progress (Q2-Q3 2007)
  - • IND submission planned (Q4 2007)
  - • Phase I study planned (Q1 2008)
### Phase I Study of Ad26.ENVA.01

<table>
<thead>
<tr>
<th>Group</th>
<th>Number</th>
<th>Dose</th>
<th>0 (0)</th>
<th>1 (28)</th>
<th>6 (168)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>10</td>
<td>$10^9$</td>
<td>rAd26</td>
<td>rAd26</td>
<td>rAd26</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td></td>
<td>FFB</td>
<td>FFB</td>
<td>FFB</td>
</tr>
<tr>
<td>2</td>
<td>10</td>
<td>$10^{10}$</td>
<td>rAd26</td>
<td>rAd26</td>
<td>rAd26</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td></td>
<td>FFB</td>
<td>FFB</td>
<td>FFB</td>
</tr>
<tr>
<td>3</td>
<td>10</td>
<td>$10^{11}$</td>
<td>rAd26</td>
<td>rAd26</td>
<td>rAd26</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td></td>
<td>FFB</td>
<td>FFB</td>
<td>FFB</td>
</tr>
<tr>
<td>4</td>
<td>10</td>
<td>$10^*$</td>
<td>rAd26</td>
<td></td>
<td>rAd26</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td></td>
<td>FFB</td>
<td></td>
<td>FFB</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>48 (40/8)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Conclusions

• rAd5 vectors may be limited by high levels of anti-Ad5 immunity in the developing world

• Novel serotype/chimeric rAd vectors can be constructed with:
  • Immunogenicity of rAd5 vectors
  • Capacity to circumvent anti-Ad5 immunity
  • Large-scale manufacturability

• We propose that novel rAd vectors should be advanced into clinical trials as candidate HIV-1 vaccines:
  • To replace rAd5 vectors as single modality vaccines
  • To be used with rAd5 vectors in prime-boost regimens
Acknowledgements

- Beth Israel Deaconess, Harvard Medical School
  - Peter Abbink
  - Dan Barouch
  - Matthew Denholtz
  - Bonnie Ewald
  - Jodi Hecht
  - David Kaufman
  - Jinyan Liu
  - Diana Lynch
  - Anjali Nanda
  - Joseph Nkolola
  - Kara O'Brien
  - Elizabeth Rhee
  - Diane Roberts
  - Faye Stephens
  - Betty Sun
  - Tricia Swanson
  - Anna Thorner
  - Flow Cytometry Core
- Crusell Holland BV
  - Marcel Brink
  - Nico Bunnik
  - Jerome Custers
  - Frans Delemarre
  - Jaap Goudsmit
  - Anneke Griffioen
  - Guus Hateboer
  - Menzo Havenga
  - Evert Heemskerk
  - Lennart Holterman
  - Karin Hoogendoorn
  - Peter Kares
  - Erica Kerkvliet
  - Matthijs Koorevaar
  - Fija Lagerwerf
  - Angelique Lemckert
  - Giuseppe Marzio
  - Maria Grazia Pau
  - Frank Raaphorst
  - Giulia Schirru
  - Jolande Schoemaker
  - Govert Schouten
  - Herman Van Herk
  - Mark Van Ooy
- Crusell Holland BV cont.
  - Ronald Vogels
  - Miranda Weggeman
  - Mo Weijtens
  - Sander Worst
- Children's Hospital, Harvard Medical School
  - Bing Chen
  - Stephen Harrison
- New England Primate Research Center
  - Angela Carville
  - Keith Mansfield
- IAVI
  - Jim Ackland
  - Wayne Koff
  - Nick Jackson
- SRI
  - Joan Roelands
- Bridge (GeneLogic)
  - Bin He
- CHAVI / HIVRAD
  - Bart Haynes
  - Bette Korber
  - Norman Letvin
- Merck Research Labs
  - Danny Casimiro
  - Sheri Dubey
  - John Shiver
- VRC, NIAID, NIH
  - Charla Andrews
  - Phil Gomez
  - Gary Nabel
  - Rebecca Sheets
- DAIDS, NIAID, NIH
  - Chris Butler
  - Nancy Miller
  - Michael Pensiero
- SAIC
- HVTN
- MGH
- HSPH
- Univ. Kwa-Zulu Natal
- South African MRC
- Crucell Holland BV cont.
  - Ronald Vogels
  - Miranda Weggeman
  - Mo Weijtens
  - Sander Worst
  -ikel
  - Angela Carville
  - Keith Mansfield
  - Jim Ackland
  - Wayne Koff
  - Nick Jackson
  - Joan Roelands
  - Bin He
- Children's Hospital, Harvard Medical School
  - Bing Chen
  - Stephen Harrison
  - Angela Carville
  - Keith Mansfield
  - Jim Ackland
  - Wayne Koff
  - Nick Jackson
  - Joan Roelands
  - Bin He
- Crucell Holland BV cont.
  - Ronald Vogels
  - Miranda Weggeman
  - Mo Weijtens
  - Sander Worst
  - Jaap Goudsmit
  - Anneke Griffioen
  - Guus Hateboer
  - Menzo Havenga
  - Evert Heemskerk
  - Lennart Holterman
  - Karin Hoogendoorn
  - Peter Kares
  - Erica Kerkvliet
  - Matthijs Koorevaar
  - Fija Lagerwerf
  - Angelique Lemckert
  - Giuseppe Marzio
  - Maria Grazia Pau
  - Frank Raaphorst
  - Giulia Schirru
  - Jolande Schoemaker
  - Govert Schouten
  - Herman Van Herk
  - Mark Van Ooy
  - Flow Cytometry Core
- Brigham and Women's, Harvard Medical School
  - Lindsey Baden
  - Raphael Dolin

- Children's Hospital, Harvard Medical School
  - Bing Chen
  - Stephen Harrison
- Crucell Holland BV cont.
  - Ronald Vogels
  - Miranda Weggeman
  - Mo Weijtens
  - Sander Worst
- New England Primate Research Center
  - Angela Carville
  - Keith Mansfield
- IAVI
  - Jim Ackland
  - Wayne Koff
  - Nick Jackson
- SRI
  - Joan Roelands
- Bridge (GeneLogic)
  - Bin He
- CHAVI / HIVRAD
  - Bart Haynes
  - Bette Korber
  - Norman Letvin
- Merck Research Labs
  - Danny Casimiro
  - Sheri Dubey
  - John Shiver
- VRC, NIAID, NIH
  - Charla Andrews
  - Phil Gomez
  - Gary Nabel
  - Rebecca Sheets
- DAIDS, NIAID, NIH
  - Chris Butler
  - Nancy Miller
  - Michael Pensiero
- SAIC
- HVTN
- MGH
- HSPH
- Univ. Kwa-Zulu Natal
- South African MRC

• Beth Israel Deaconess, Harvard Medical School
  - Peter Abbink
  - Dan Barouch
  - Matthew Denholtz
  - Bonnie Ewald
  - Jodi Hecht
  - David Kaufman
  - Jinyan Liu
  - Diana Lynch
  - Anjali Nanda
  - Joseph Nkolola
  - Kara O’Brien
  - Elizabeth Rhee
  - Diane Roberts
  - Faye Stephens
  - Betty Sun
  - Tricia Swanson
  - Anna Thorner
  - Flow Cytometry Core
- Crucell Holland BV
  - Marcel Brink
  - Nico Bunnik
  - Jerome Custers
  - Frans Delemarre
  - Jaap Goudsmit
  - Anneke Griffioen
  - Guus Hateboer
  - Menzo Havenga
  - Evert Heemskerk
  - Lennart Holterman
  - Karin Hoogendoorn
  - Peter Kares
  - Erica Kerkvliet
  - Matthijs Koorevaar
  - Fija Lagerwerf
  - Angelique Lemckert
  - Giuseppe Marzio
  - Maria Grazia Pau
  - Frank Raaphorst
  - Giulia Schirru
  - Jolande Schoemaker
  - Govert Schouten
  - Herman Van Herk
  - Mark Van Ooy
- Children’s Hospital, Harvard Medical School
  - Bing Chen
  - Stephen Harrison
- New England Primate Research Center
  - Angela Carville
  - Keith Mansfield
- IAVI
  - Jim Ackland
  - Wayne Koff
  - Nick Jackson
- SRI
  - Joan Roelands
- Bridge (GeneLogic)
  - Bin He
- CHAVI / HIVRAD
  - Bart Haynes
  - Bette Korber
  - Norman Letvin
- Merck Research Labs
  - Danny Casimiro
  - Sheri Dubey
  - John Shiver
- VRC, NIAID, NIH
  - Charla Andrews
  - Phil Gomez
  - Gary Nabel
  - Rebecca Sheets
- DAIDS, NIAID, NIH
  - Chris Butler
  - Nancy Miller
  - Michael Pensiero
- SAIC
- HVTN
- MGH
- HSPH
- Univ. Kwa-Zulu Natal
- South African MRC