Rapid Responses to Novel Lethal Viruses: The Potential of Recombinant Nanoparticle Vaccines Produced in Sf9 Insect Cells

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SVP R&D
Novavax, Rockville, MD

EBOV/Makona GP Nanoparticle
Presentation Overview

- Novavax: Who are we?
- Response to H7N9 and Ebola: Case study on how the recombinant nanoparticle vaccine technology can be applied to challenges posed by novel lethal viruses?
- H7N9 Vaccine
- Ebola Vaccine
  - Background
  - Novavax Ebola/Mak Glycoprotein Vaccine
  - Novavax saponin-based Matrix-M™ Adjuvant
  - Immunogenicity and challenge data
  - Plans forward
- Conclusions
Novavax (Nasdaq: NVAX) is a clinical-stage vaccine company committed to delivering novel products to prevent a broad range of infectious diseases.

Our recombinant nanoparticles and Matrix-M™ adjuvant technology are the foundation for ground-breaking innovation that improves global health through safe and effective vaccines.

- Headquartered in Gaithersburg, MD with additional facilities in Rockville, MD and Uppsala, Sweden.
- Employ over 300 individuals dedicated to developing novel vaccines to address infectious disease.
  - 2 GMP manufacturing sites
  - Adjuvant manufacturing site
    - 4 Phase II programs
    - 2 Phase I programs
### Pipeline

<table>
<thead>
<tr>
<th>Program</th>
<th>Preclinical</th>
<th>Phase 1</th>
<th>Phase 2</th>
<th>Phase 3</th>
<th>Funding Support</th>
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<td><strong>RSV</strong></td>
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<td>Elderly</td>
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<td>Infants (Maternal Immunization)</td>
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<td>Pediatrics (6 mos – 6 yrs)</td>
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<td><strong>Influenza</strong></td>
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<td>BARDA</td>
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<td>Quadrivalent Seasonal</td>
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<td>Pandemic (H7N9 + Matrix-M)</td>
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<td>BARDA</td>
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<td><strong>New Vaccines</strong></td>
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<td>Combination Respiratory</td>
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<td>Ebola + Matrix-M</td>
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Overview

- Select and engineer genetic sequences encoding vaccine antigens
- Sf9 cellular machinery produces correctly modified and folded proteins
- Full-length antigens expressed in native conformation assemble into highly immunogenic nanoparticle
- Safety database with over 7,000 subjects
Recombinant Baculovirus-Sf9 Technology Enables a Rapid Vaccine Response to Novel Lethal Viruses

Viral Threats

• Vaccine candidate development
  • Preclinical study expertise
  • Regulatory expertise

Response

• Process & assay development
  • 1000L GMP production
  • QC and QA for testing and release

Process Development & GMP Production

Clinical study expertise
3-month Response Time from Gene Sequence to GMP Batch Release for recombinant H7N9, Ebola Vaccines

**H7N9 A/Anhui VLP**
- Highly lethal virus
- Novel
- Pandemic potential

**Ebola GP Particle**
- Highly lethal virus
- Novel
- Pandemic potential
A lethal A/Anhui wild type virus mouse challenge model was rapidly developed. Mice were immunized with VLP vaccines w/wo saponin adjuvant:
1) A(H7N9) A/Anhui/1/13, or
2) H7N3
3) An A(H5N1) VLP

After 2 vaccine doses, the mice were challenged with a lethal dose of A/Anhui virus.

Both H7 vaccines protected the mice from death, even without adjuvant, H7 VLP vaccines were cross-protective against distantly-related strains. H5N1 and controls were not protected.
Clinical: A/Anhui/1/2013 HAI Response to H7N9/saponin adjuvanted vaccine at d 28 in 225 subjects vaccinated at d 0 and 21

Day 35 A/Anhui/1/2013 HAI Titer Distributions

“Sero-protection”
Novavax Avian Influenza H7N9 Results and Outcomes

H7N9 VLP alone or with Saponin Adjuvant (2013)
- 225 subjects vaccinated 91 days post publication of gene sequence
- Achieved dose-sparing goals with 5 μg dose
- Seroconversion and seroprotection rates: 81% HAI, 97% NAI
- Achieved within 45 days of immunization

H7N9 VLP alone or with Matrix-M (Novavax Saponin Adjuvant) (2014)
- Achieved dose-sparing goals with 3.75 μg dose
- H7N9 with Matrix-M demonstrated strong HA and NA antibody responses

*Received FDA Fast Track Designation in October 2014
*Phase 2 Clinical Trial Planned for third quarter of 2015
*Funded by BARDA through September 2016
Ebola Epidemic Status

New cases continue to appear in Guinea and Sierra Leone
20+ known Filovirus outbreaks since 1976
Three studies published in 1986 documented Ebola antibody prevalence rates of 10.6, 13.4 and 14 percent in northwestern Liberia

Strategy for Development of a Recombinant Viral Vaccine: EBOV

Recombinant GP
- Full length EBOV/Makona
- Recombinant GP expressed in Sf9 cells
- Purified GP trimers form nanoparticles

EBOV mAb(s)
- Protective in NHPs
- KZ52 mAb confirms GP1,2 pre-fusion structure
- 13C6 mAb
  - GP ELISA potency
  - Competitive binding

Key Insight
- High affinity binding to vaccine by protective mAbs = epitopes intact, displayed, and predictive of protection in active immunization

Immunization Results
- High levels of anti-GP IgG (EC90) that are high affinity and neutralizing
- Polyclonal antibodies competitive with 13C6 and other neutralizing mAb
- EBOV/Makona GP vaccine induces nAb against EBOV/Mayinga
- EBOV/Makona GP vaccine induces protection in mice (mouse-adapted 1976 Mayinga) and macaques (wild type 1995 Kikwik)
Sept 12, 2014 Novavax begin development of recombinant EBOV/Makona GP subunit vaccine

Recombinant EBOV/Makona Glycoprotein (GP)
- GenBank #AIG96283
- EBOV [H.sapiens -wt/SLE/2014/Makona-G3798]; cluster 3
- Full length, unmodified GP gene
- Synthetic, codon optimized
- Cloned into a baculovirus vector rBV-GP

CLONING
Baculovirus vector rBV-GP

EXPRESSION
rBV-GP infected Sf9 cells

PURIFICATION
EBOV GP nanoparticles

Glycosylated GP1/GP2 trimers
EBOV/Mak GP nanoparticle TEM negative stain image and 2D class averaging

- Central core region with GP2/hydrophobic domains (blue)
- Six visible chalice-like GP1/mucin domain trimers extending concentrically from a central core (pink)
- 2D TEM manual coloration (NanoImaging Services, Inc.)
Matrix-M™ adjuvant: mixture A:C (85:15) saponin:cholesterol:phospholipid

EBOV GP and Matrix-M are independent particles: co-formulated
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Binding kinetics purified recombinant EBOV/Mak GP to functional EBOV mAb: High Affinity Binding

<table>
<thead>
<tr>
<th>mAb</th>
<th>EBOV GP Epitope</th>
<th>SPR $K_D$ (nM)</th>
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<tbody>
<tr>
<td>KZ52</td>
<td>aa 42-43, 513, 550-553, 556 GP1/GP2</td>
<td>Conformational Pre-fusion GP2</td>
</tr>
<tr>
<td>13C6</td>
<td>aa 1-295 GP1</td>
<td>Conformational In ZMapp</td>
</tr>
<tr>
<td>6D8</td>
<td>aa 389-405 GP1 HNTPVYKLDISEATQVE</td>
<td>Linear</td>
</tr>
<tr>
<td>13F6</td>
<td>aa 401-417 GP1 ATQVEQHHRRTDNDSTA ATQV[GQHHRA]DNDSTA$^1$</td>
<td>Linear Neutralizing</td>
</tr>
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Suggests the key epitope is present and intact
Strategy for Development of a Recombinant Viral Vaccine: EBOV

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Immunization Results
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### Baboon Immunogenicity Study: anti-EBOV/Mak GP ELISA and Competition ELISA with 13C6 mAb

<table>
<thead>
<tr>
<th>Group</th>
<th>Vaccine</th>
<th>Day 0</th>
<th>Day 21 1 dose regimen</th>
<th>Day 31 2 dose regimen</th>
<th>13C6 µg/ml</th>
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<tbody>
<tr>
<td>1</td>
<td>60µg EBOV GP</td>
<td>&lt;100</td>
<td>631</td>
<td>1,517</td>
<td>&lt;4</td>
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<tr>
<td>2</td>
<td>60µg EBOV GP 800µg AlPO4</td>
<td>&lt;100</td>
<td>19,227</td>
<td>285,206</td>
<td>20</td>
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<tr>
<td>3</td>
<td>60µg EBOV GP 50µg Matrix</td>
<td>&lt;100</td>
<td>13,115</td>
<td>6,870,339</td>
<td>159</td>
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<tr>
<td>4</td>
<td>5µg EBOV GP 50µg Matrix</td>
<td>&lt;100</td>
<td>3,242</td>
<td>11,302,798</td>
<td>129</td>
</tr>
</tbody>
</table>

13C6 mAb conformational, neutralizing, protective in NHPs, component of Zmapp
Palivizumab (RSV) protective at 30 µg/ml

**EBOV/Makona anti-GP responses to cAd3-Ebo vaccine**
- $2 \times 10^{11}$ cAd3-EBO vaccine (EC90 = 623)
- $2 \times 10^{10}$ cAd3-EBO vaccine (EC90 = 177)

## EBOV/Mak GP vaccine IFNγ-Elispot response in baboons day 31

<table>
<thead>
<tr>
<th></th>
<th>60 ug GP NHP4711</th>
<th>60ug GP/AIPO4 NHP 7311</th>
<th>60ug GP/Matrix M NHP 4411</th>
<th>5ug GP/Matrix M NHP 5910</th>
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<tbody>
<tr>
<td><strong>Medium</strong></td>
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<tr>
<td><strong>Ebola peptide pool 1 (aa1-171)</strong></td>
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<tr>
<td><strong>Ebola peptide Pool 2 (aa 172-335)</strong></td>
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<td><strong>Ebola peptide Pool 3 (336-495)</strong></td>
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<td><strong>Ebola peptide Pool 4 (496-676)</strong></td>
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<td><strong>Ebola-GP</strong></td>
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Robust IFNγ-Elispot responses to 5ug EBOV GP + Matrix-M vaccine
Ebola-GP/Matrix-M induced multifunctional T cell response in baboons

**Cytokines: interferon-γ, TNF-α and IL-2**
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RSV CONFIDENTIAL
EBOV/Makona vaccine anti-EBOV/Makona GP Antibody Responses and Challenge Results

Immunogenicity

Challenge Study: Ricardo Carrion, Jr, Rob Davey, Manu Anantpadama, Gabriela Worwa, Texas Biomedical Research Institute San Antonio, TX
DMID/NIAID: EBOV/Makona vaccine anti-EBOV/Makona GP IgG ELISA (EC50) and 100% Protection

Group G (n=2)

5µg GP + Matrix-M

- 0wk
- 3wk

Ebola/Kikwit

- wk9; 100 pfu

Group F (n=2)

5µg GP + Matrix-M

- 0wk
- 6wk

Ebola/Kikwit

- wk12; 100 pfu

Survival (day 18)

- 5µg EBOV/GP 100% (4/4)
- Saline control 0% (0/2)
• Phase 1, blinded, controlled in 230 adults 18 to < 50 y.o. in Australia
• Dose ranging, 1 and 2 dose regimens w/wo Matrix-M adjuvant
• Immune responses thru 1 year
  o GP EC50 ELISA, PRNT, PsVNA, anti-13C6, and T-cell responses
Summary

- Novavax: is a mid-size recombinant vaccine company that is uniquely capable of responding to novel lethal viral threats
- Recombinant tools/adjuvants allow NVAX scientists to solve difficult vaccine puzzles
- Demonstrated in the context of H7N9 Vaccine and Ebola Vaccines
- Path forward, regulatory approval and business models for these efforts are being assessed
- In light of the robust biological responses to the vaccines, there is a compelling case to attempt to develop and make available recombinant nanoparticle vaccines and saponin adjuvants when novel lethal viruses arise and show signs of persistence
Acknowledgement – Novavax Ebola vaccine Swat Team

Our foundation for success is rooted in the expertise and dedication of the people who performed the necessary activities. We appreciate and recognize the dedicated employees who contributed to this project:

**Adjuvant Production (Sweden):**

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