ORAVAX: A Bacterial DNA Vaccine to Prevent COVID-19

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Course: Vaccines, from Concept to Implementation
Course Directors: Philip LaRussa MD and Lawrence Stanberry MD, PhD
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The leadership team

Sara Sakowitz
Chief Scientific Officer

Isabella Salas-Allende
Chief Operations Officer

Peter Suwondo
Chief Medical Officer

Mona Patel
Chief Regulatory Officer

External Advisors
Revolutionizing health through bacteria
By the end of this presentation, you will understand...

- The unique advantages of our **platform and delivery system**: a bacterial DNA vaccine
- What **preclinical testing** is necessary to demonstrate safety and feasibility
- The **target product profile** we aim to deliver on
- Our plans to conduct **phase 3 clinical trials** to demonstrate efficacy
- General approach to **post-marketing surveillance**
- Anticipated **priority groups for vaccine rollout**
The Platform
A revolutionary delivery platform

Engineered *Salmonella typhimurium* containing synthetic plasmids encoding Spike (S) protein from SARS-CoV-2.
## Pros: Bacterial vectored DNA vaccine

<table>
<thead>
<tr>
<th>DNA vaccines</th>
<th>Bacterial vectors</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Defined composition</td>
<td>1. Directed delivery of target antigen to specific cells including macrophages</td>
</tr>
<tr>
<td>2. Non-replicating platform capable of inducing T-cell immunity</td>
<td>2. Large antigen-carry capacity</td>
</tr>
<tr>
<td>3. Potential application in development of therapeutic vaccines</td>
<td>3. Safety maximized by removing several genes</td>
</tr>
<tr>
<td>4. Construct may code for multiple epitopes and also inducers of innate immune responses</td>
<td>4. Potential for mucosal immunity</td>
</tr>
<tr>
<td></td>
<td>5. Oral delivery possible</td>
</tr>
</tbody>
</table>
### Cons: Bacterial vectored DNA vaccine

<table>
<thead>
<tr>
<th>DNA vaccines</th>
<th>Bacterial vectors</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Poor immunogenicity in humans</td>
<td>1. Possible genomic instability at the site of insertion giving low antigen expression levels</td>
</tr>
<tr>
<td>2. Concerns/issues regarding potential for construct to integrate with host genome</td>
<td>2. Expression of bacterial antigens may further reduce vaccine-specific immunogenicity</td>
</tr>
<tr>
<td></td>
<td>3. Efficacy decreased by existing vector immunity</td>
</tr>
<tr>
<td></td>
<td>4. Difficult to optimize engineering without conducting a number of clinical trials</td>
</tr>
</tbody>
</table>
Proposed DNA vaccine delivery system using a live bacterial vector
Mucosal immunity is the largest component of the immune system.
Oral delivery is the most desirable and patient-accepted route
Pros: A rapid, cost-effective vaccine for the globe

Ideal profile for LMIC market

Cost-effective production
Improved distribution
Mucosal immunity
Cons: a promising but largely untested technology
Preclinical Testing
Toxicology

- Stability of the plasmid
Toxicology

- Stability of the plasmid
- C57BL/6 mice
- 3 billion CFUs vs 10 billion CFUs
  - Single vs. Repeat Dose
Toxicology

- Stability of the plasmid
- C57BL/6 mice
- 3 billion CFUs vs 10 billion CFUs
  - Single vs. Repeat Dose
Single/Repeat Dose Study in Baboons

1. Vaccination
   3 or 10 CFU units

2. Monitor
   6, 24, 48, 72h

3. Evaluate
   Blood & Immune

4. Pathology
   Critical Organs
Toxicology: Reproductive/Developmental

- Preclinical testing
- Target Product Profile
- Phase 3 Testing
- Post-marketing surveillance
- Implementation Recommendations

Platform
Toxicology

- Mutagenicity
- Biodistribution
  - Integration
- Carcinogenicity
- Testing bulk plasmid products
Immunology

- Evaluate the production of pro-inflammatory cytokines in vitro
Immunology

- Evaluate the production of pro-inflammatory cytokines in vitro
- Evaluate antibody titers in vivo
  - IgA
  - IgG & Isotypes
Immunology

- Evaluate the production of pro-inflammatory cytokines in vitro
- Evaluate antibody titers in vivo
  - IgA
  - IgG & Isotypes
- Evaluate cytokines post-vaccination in vivo
Immunology

- Evaluate the production of pro-inflammatory cytokines in vitro
- Evaluate antibody titers in vivo
  - IgA
  - IgG & Isotypes
- Evaluate cytokines post-vaccination in vivo
Immunology

Bars indicate the 95% confidence interval.
Immunology

![Graph showing IgG levels with dilution and CFU comparison]

- PBS
- 10 CFU
- 3 CFU
Immunology

![Graph showing IgG1 and IgG2b levels](image)
Pre-Clinical Efficacy

- Build on our single/repeat dose study
- Baboons as our model organism
- Central idea is to “challenge”
Pre-Clinical Efficacy

Step 1: Vaccinate
- One dose of the oral vaccine
- 20 Baboons

Step 2: Challenge
- 2 weeks post vaccination

Step 3: Monitor
- Survival & disease monitored for 14 days post vaccination

Step 4: Assay
- Collect organs at 2, 5, 7, 9 DPI for virus titration
Pre-Clinical Efficacy

- Response of baboons to the challenge
- Effect of vector priming?

Future Studies!
Target Product Profile
TPP 1 - Indication for Use

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Immunization <em>protects against</em> COVID-19 infection</td>
<td>Immunization reduces the severity of COVID-19</td>
</tr>
</tbody>
</table>
# TPP 2 - Target Population

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>All ages and medical profiles, except for pregnant women</td>
<td>Adults, including the elderly, except for individuals who are pregnant or have a history of GI disease</td>
</tr>
</tbody>
</table>
## TPP 3 - Contraindications

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pregnancy</td>
<td>Pregnancy, history of GI disease, &amp; immunocompromised patients</td>
</tr>
</tbody>
</table>
### TPP 4 - Safety/Reactogenicity

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>No serious adverse events</td>
<td>Safety and reactogenicity profile whereby the vaccine benefits still outweigh the safety risks</td>
</tr>
</tbody>
</table>
## TPP 5 - Efficacy

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt;70% efficacy on a population basis</td>
<td>&gt;70% efficacy on a population basis</td>
</tr>
<tr>
<td>&gt;70% efficacy on the elderly</td>
<td>&gt;60% efficacy on the elderly</td>
</tr>
</tbody>
</table>
## TPP 6 - Dose Regimen

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single-dose regimen</td>
<td>No more than two doses, 21 days apart</td>
</tr>
</tbody>
</table>
## TPP 7 - Durability of Protection

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lifetime immunity</td>
<td>Protection for at least 12 months</td>
</tr>
</tbody>
</table>

**Platform**

- Preclinical testing
- Target Product Profile
- Phase 3 Testing
- Post-marketing surveillance
- Implementation Recommendations
### TPP 8 - Route of Administration

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral</td>
<td>Oral</td>
</tr>
</tbody>
</table>
## TPP 9 - Coverage

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Multivalent</td>
<td>Monovalent</td>
</tr>
</tbody>
</table>
### TPP 10 - Product Stability/Storage

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shelf life &gt;24 months at room temperature</td>
<td>Shelf life &gt;12 months at room temperature</td>
</tr>
<tr>
<td>Stability &gt; 6 months at room temperature</td>
<td>Stability &gt; 1 month at room temperature</td>
</tr>
</tbody>
</table>
## TPP 11 - Co-Administration

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Can be given with other vaccines without affecting immunogenicity, safety, or efficacy of the vaccines</td>
<td>Must be used as a stand-alone vaccine</td>
</tr>
</tbody>
</table>
## TPP 12 - Presentation

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Solid pill product in mono-dose presentation</td>
<td>Solid pill product in multi-dose presentation</td>
</tr>
</tbody>
</table>
TPP 13 - Production

<table>
<thead>
<tr>
<th>Preferred</th>
<th>Critical or Minimal</th>
</tr>
</thead>
<tbody>
<tr>
<td>7 billion doses</td>
<td>350 million doses</td>
</tr>
</tbody>
</table>
### TPP 14 - Registration/ Prequalification

<table>
<thead>
<tr>
<th>WHO and International Regulatory Authorities</th>
</tr>
</thead>
<tbody>
<tr>
<td>WHO prequalified and/or Emergency Use Assessment &amp; Listing Procedure (EUAL)</td>
</tr>
</tbody>
</table>
TPP 15 - Post-Marketing Surveillance

**WHO and International Regulatory Authorities**

Post-marketing surveillance will include an evaluation of all serious adverse effects, as well as vaccine effectiveness. Emergence of vaccine-resistant SARS-CoV-2 mutants will also be assessed, following the WHO’s prequalification requirements.
Phase 3 Testing
Goal for phase 3: demonstrate vaccine efficacy in large, representative populations

- Randomized, double blinded, placebo-controlled, multicenter trial
- Global consortium of sites representing target populations
- Approximately 26,000++ participants to be enrolled
- Duration follows adaptive event-driven design
  (expected 2 year follow-up for safety, but efficacy endpoints reached sooner)
Primary efficacy outcome: how effective is our vaccine at preventing moderate/severe disease?

<table>
<thead>
<tr>
<th>OUTCOME</th>
<th>MEASUREMENT</th>
</tr>
</thead>
<tbody>
<tr>
<td>First occurrence of confirmed moderate-to-severe (or critical) COVID-19 (&gt;21 days post-vaccination), as per recommended US FDA case definitions*</td>
<td>● Regular symptom screening by text-message monitoring system and participant phone-calls</td>
</tr>
<tr>
<td></td>
<td>● All symptom reports investigated by clinical team</td>
</tr>
<tr>
<td></td>
<td>● Cases confirmed by molecular diagnostic test (nasal swab PCR) at 2 independent labs <em>(in cases of discrepancy, central lab results will prevail)</em></td>
</tr>
<tr>
<td></td>
<td>● Cases adjudicated by a blinded central clinical committee</td>
</tr>
</tbody>
</table>
Secondary outcomes: will our vaccine prevent infection, transmission, and/or hospitalization?

<table>
<thead>
<tr>
<th>OUTCOME</th>
<th>MEASUREMENT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Asymptomatic infection</td>
<td>Positive nasal RT-PCR at any time or antibody test (performed pre-vaccination, 1 wk, 1 mo, 3 mo, 6 mo, 1 year, 2 years - Abs reactive to an antigen not included in vaccine)</td>
</tr>
<tr>
<td>Critical illness</td>
<td>Hospital admission or death with positive SARS-CoV2 RT-PCR test (see FDA recommended case definitions)</td>
</tr>
<tr>
<td>Viral shedding</td>
<td>Presence of viable SARS-CoV2 virus in saliva, NP samples, and sputum as detectable by culture and/or other assays (assessed at intervals as noted above)</td>
</tr>
</tbody>
</table>
Plan for recruitment

Inclusion criteria

- Age 12+
- Willing and able to comply with all requirements (tests, in-person visits, etc)
- Medically stable
- Able to give informed consent

Exclusion criteria (abbreviated)

- Immunosuppressed or known immune disease
- GI disease (celiac, IBD, ulcers, etc)
- Current or recent antibiotic treatment
- Pregnant or breastfeeding
- Other unstable chronic disease*
- Active or prior COVID infection
- Already vaccinated with any COVID vaccine
- Known allergy to a vaccine component
- Past serious adverse reaction to any vaccine

*i.e. requiring hospitalization or change in therapy due to deterioration in 2 mo prior to enrollment*
Proposed dosing and schedule

- Goal: single dose regimen
  - 1-10 billion cfu, dosing studies underway

- If early immunogenicity trials show poor response...
  - 2 doses (prime and boost) separated by 21 days

- Adaptive design Phase II/III design may be warranted
Measuring the immunological response

- Assessing short term **reactogenicity**:
  - 30 min observation period on-site
  - Daily, weekly, monthly symptom logging via text-message system

- Participants will undergo additional **immunogenicity** analyses at pre-specified intervals (pre-vaccination, 1 week, 1 month, 3 months, 6 months, 1 year, 2 years)
  - Antibody titers (IgM, IgG, IgA) - and mucosal vs. systemic
  - Ab neutralization assays
  - Cell-mediated immunity
  - Cytokine levels

- If constrained by budget, immunogenicity testing to be performed only on subset of first ~5000 enrolled participants (across pre-defined age and per-center quota)
### Statistical power and duration of study

<table>
<thead>
<tr>
<th>Necessary events: ~350</th>
<th>Number of participants needed: ~26,000</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Assumptions</strong></td>
<td><strong>Assumptions</strong></td>
</tr>
<tr>
<td>2-sided t-test</td>
<td>Follow-up period: 2 years</td>
</tr>
<tr>
<td>● VE ≥ 30% (relative hazard 0.7)</td>
<td>• Attrition/censoring: 10% / yr</td>
</tr>
<tr>
<td>● Significance level (α): 5%</td>
<td>• Baseline event rate (incidence): 1% / yr</td>
</tr>
<tr>
<td>● Power (β): 90%</td>
<td></td>
</tr>
<tr>
<td>● Allocation groups: 2:1 (intervention:control)</td>
<td></td>
</tr>
</tbody>
</table>

**Factors that could increase speed of trial:**
- Higher than expected **efficacy**: if VE ≥ 70%, only 32 events needed (i.e. 2 months)
- Higher than expected **attack rate**
- More enrolled **participants**
Safety & pre-specified analyses

- Independent DSMB to be commissioned
- Interim analyses to be performed at pre-specified intervals (i.e. 25, 50, 100, 200 events) for safety and efficacy stoppage
- Both intention-to-treat (ITT) and per-protocol analyses to be reported
Ethics

- If vaccine is proven safe and effective in preliminary analysis (in consultation with DSMB and applicable regulators), placebo group participants may be offered option of vaccine.

- If another vaccine (Pfizer, Moderna, Janssen) is given full biological license approval (BLA) by US-FDA before study commencement, design can be switched over to non-inferiority trial.
Post-Marketing Surveillance
Continuing pharmacovigilance

- Given the novel platform, we intend to conduct robust post-marketing surveillance activities for safety issues.

- Particular outcomes of interest:
  - Incidence of **inflammatory gastrointestinal conditions** (inflammatory bowel disease, gastroenteritis, irritable bowel syndrome, etc)
  - Incidence of **gastrointestinal cancers**, other **neoplasms**, or anything suggestive of integration into host genome
  - Incidence of **autoimmune conditions**
  - **Persistent infection** with the bacterial vector
  - Prevalence and duration of potential milder side effects including: constipation, diarrhea, abdominal pain, etc
Methods

**Safety:**

- **Passive surveillance** for any *unexpected adverse events* using provider/patient-driven reporting systems (i.e. US VAERS and similar)
- **Active surveillance** for *outcomes of special interest*—those described previously, as well as general AEs associated with COVID vaccines—using large patient data pools (i.e. VSD and similar systems)
- Specific **patient registries** to be utilized as needed to examine *special populations* (pregnant, immunocompromised)

**Effectiveness (confirmatory):**

- Large post-marketing prospective cohort study (cases identified by active surveillance)
Recommendations for implementation
Priority groups

1) **Health workers** at high to very high risk of becoming infected & transmitting disease
   - Frontline healthcare workers
   - Front desk hospital staff
   - Custodial staff
   - Security & emergency staff

2) **Adults above age 50** with risk factors for severe disease and death:
   - Hospitalized or in a long-term care facility
   - Serious comorbid chronic illness

3) **Middle age Employers**
   - >50 with moderate to high illness actively working in crowded workplace
4) **Other social/employment groups** at elevated risk of acquiring & transmitting infection

- Group of people who are unable to effectively maintain physical distance
- For example: bus drivers, truck drivers, Uber drivers, high priority school staff, Military staff who living in tight quarters

5) **Essential workers outside health and education sections**

- Child care providers, government workers, food and agriculture workers, people living in detention facility, incarcerated people, dormitories, Urban slums, dense urban neighbourhood
Rationale & other ethical considerations

Evolution and Development of Human Health
Thank you!

ORAVAX: Revolutionizing health through bacteria
References


Appendixes
Appendix I. Pros: a rapid, cost-effective vaccine for the globe

- **Enhanced immune response** due to presence of bacterial danger signals
- Constitutive, **targeted expression** of DNA construct at sites of interest
- **Multivalent potential** (multiple antigens in one payload)
- Built-in **failsafe**: bacterial vector easily killed by routine antibiotics

Ideal profile for LMIC market
Appendix II. Cons: a promising but largely untested technology

- No human DNA vaccines currently with full FDA approval
- Considerable diversity in gut + respiratory tract microbiome
- Potential issues with low absorption/transfection in gut mucosa (solved by proprietary tech)
- Adjuvants?Extent of systemic protection?
Appendix III. A revolutionary delivery platform: bacterial DNA system

- Live bacteria carrying plasmid of interest ingested via oral pill
- Bacteria transit upper GI then colonize gut epithelia
- Transgenes taken up by local host cells, causing expression of target antigens (i.e. SARS-CoV-2 spike protein)
- Systemic and mucosal immune response induced