Academic Knowledge & Innovation to Accelerate Successful Technological Transfer for a COVID-19 Vaccine

Texas Children’s Center for Vaccine Development

Maria Elena Bottazzi PhD
Co-director

DISCLAIMER: I am an inventor of patents for non-income generating vaccines to prevent parasitic infections. Baylor College of Medicine has granted a non-exclusive license to a COVID-19 vaccine candidate for an India-based vaccine manufacturer of which I am a developer.
Texas Children’s CVD Mission

A Product Development Partnership
Academic Health Center-based
+ 50 scientific and technical staff
> 40 Global Partnerships

Established in DC the year 2000
Moved to Texas Medical Center in 2011
Collaboration with Baylor College of Medicine

To develop and test new low-cost and effective vaccines against emerging and neglected tropical diseases

To build capacity for vaccine development locally and with foreign nations

To guide and influence vaccine policy and advocacy
PDPs as Key Accelerators of Vaccine Development

Source of innovation, cut costs and mitigate risks

Specialized knowledge and technologies

From basic research to critical path development:

- Targeted discovery, screening and engineering
- Production process development and scale-up
- Assay and formulation development
- Preclinical models for immunogenicity and efficacy
- Clinical trial networks
- Ethical, regulatory and quality assurance framework
Portfolio and Major Accomplishments (2011 - 2020)

- **Human Hookworm Infection**
  - Developed the first vaccine for human hookworm infection now entering phase 2 clinical trials

- **Intestinal Schistosomiasis**
  - Developed the first vaccine for intestinal schistosomiasis now entering phase 2 clinical trials

- **Chagas disease**
  - Developed the first vaccine for Chagas disease now entering phase 1 clinical trials

- **Coronavirus Initiative**
  - Developed innovative vaccines for emerging coronavirus infections: COVID-19, SARS and MERS

- **Tick-borne and Lyme disease**

- **Cutaneous Leishmaniasis**

- **Soil-transmitted Helminths (Ascaris, Trichuris and Toxocara)**
  - Signed and implemented historic capacity building agreements with Brazil, Mexico, Malaysia and the Kingdom of Saudi Arabia
Texas Children’s CVD Relies on Subunit Vaccine Technology

- Well-established technology
- Considered very safe with widespread use in licensed vaccines
- Suitable for adult & pediatric populations
- Ecosystem of global manufacturers with ease of scalability
- Stable and suitable cold-chain
- Ideal antigen and adjuvant combination for primary or boosting
- Affordable

Anthony Fauci, NIAID, NIH Presentation to National Academies
Coronavirus Vaccine Initiative

- Led by Texas Children’s CVD
- Develop Low-Cost Coronavirus Vaccines for Global Health by Microbial Fermentation in Yeast
- NIH/NIAID seed funding instrumental
  - SARS/MERS (2011-16)
  - COVID-19 (2020-)
- Specific coronavirus partnerships launched in 2011 and expanded in 2020
Coronavirus Vaccine Research a Catalyst for COVID-19

2000
Established infrastructure as academic-based PDP with a hybrid business model

2011
TCH CVD recruited to Baylor and Texas Children’s and launches the Coronavirus Vaccine Initiative

2014
TCH CVD initiates technology transfer of the SARS-RBD Vaccine

2016
TCH CVD manufactures SARS-RBD Vaccine with WRAIR*

2020
TCH CVD Accelerates the Development of a COVID-19 Vaccine with a MilliporeSigma Alliance

2020
TCH CVD alliances with PATH and IDRI*

2020
TCH CVD licenses vaccine to Biological E, Ltd.**

*WRAIR: Walter Reed Army Institute of Research Pilot Manufacturing Facility; PATH: Center for Vaccine Innovation & Access; IDRI: Infectious Disease Research Institute

** Biological E: India-based Industrial Vaccine Manufacturer
Cloning and Expression Strategy for the CoV-2 Vaccine Candidate

RBM region

Asparagine (N331)

Cysteine (C538)

219 WT, 219 N1 and 219 N1C1

SARS-CoV-2-RBD219 (PDD032520) 5 µg

SARS-CoV-2-RBD219-N1 (PDD041420) 5 µg

SARS-CoV-2-RBD219-N1-C1 (PDD060220) 5 µg

Purity: 98% 97% 96% 96% 98% 98%

https://www.biorxiv.org/content/10.1101/2020.11.09.373449v1
CoV-2-RBD219-N1-C1 Process Development and Scale up Production

<table>
<thead>
<tr>
<th>Molecule</th>
<th>Fermentation yield* (mg RBD/L of FS)</th>
<th>Level of impurity on SDS-PAGE</th>
<th>Hyperglycosylation on WB</th>
<th>Dimer formation on WB</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBD219-WT</td>
<td>142 ± 8</td>
<td>High</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>RBD219-N1</td>
<td>50 ± 13</td>
<td>Mid</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>RBD219-N1C1</td>
<td>280 ± 70</td>
<td>Low</td>
<td>No</td>
<td>No</td>
</tr>
</tbody>
</table>

[Image of wet cell weight graph]

[Image of SARS-CoV2-RBD219-N1C1 protein expression]

[Image of SDS-PAGE gel for reduced and non-reduced conditions]

https://www.biorxiv.org/content/10.1101/2020.11.09.373449v1
Biophysical Comparison using Dynamic Light Scattering and Circular Dichroism

https://www.biorxiv.org/content/10.1101/2020.11.09.373449v1
Continuous improvement and re-design strategies to enable robust CMC

> RBD219-N1C1

<table>
<thead>
<tr>
<th>Molecular weight observed by mass spectrometry (kDa)</th>
<th>N-terminal variants</th>
<th>C-terminal variants</th>
<th>Expected Molecular weight (kDa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>23310.6</td>
<td>EAEAEF</td>
<td>STNL</td>
<td>23310</td>
</tr>
<tr>
<td>23780</td>
<td>EAEAEF</td>
<td>STNLVK</td>
<td>23779</td>
</tr>
<tr>
<td>23537.3</td>
<td>EAEAEF</td>
<td>STNLVK</td>
<td>23537</td>
</tr>
<tr>
<td>23109.9</td>
<td>EAEF</td>
<td>STNL</td>
<td>23110</td>
</tr>
<tr>
<td>23408.8</td>
<td>EAEAEF</td>
<td>STNLV</td>
<td>23409</td>
</tr>
</tbody>
</table>

> RBD203-N1

<table>
<thead>
<tr>
<th>Molecular weight observed by mass spectrometry (kDa)</th>
<th>N-terminal variants</th>
<th>C-terminal variants</th>
<th>Expected Molecular weight (kDa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>23310.6</td>
<td>EAEAEF</td>
<td>STNL</td>
<td>23310</td>
</tr>
<tr>
<td>23780</td>
<td>EAEAEF</td>
<td>STNLVK</td>
<td>23779</td>
</tr>
<tr>
<td>23537.3</td>
<td>EAEAEF</td>
<td>STNLVK</td>
<td>23537</td>
</tr>
<tr>
<td>23109.9</td>
<td>EAEF</td>
<td>STNL</td>
<td>23110</td>
</tr>
<tr>
<td>23408.8</td>
<td>EAEAEF</td>
<td>STNLV</td>
<td>23409</td>
</tr>
</tbody>
</table>
CoV-2-RBD203-N1 Process Development and Scale up Production

![Image of 4-12% SDS-PAGE and CD profile graphs]

<table>
<thead>
<tr>
<th>Protein</th>
<th>Tm (°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBD219-N1C1</td>
<td>51.9</td>
</tr>
<tr>
<td>RBD203-N1</td>
<td>50.8</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>SARS-CoV-2-RBD-219-N1-C1 (PDD120220)</th>
<th>SARS-CoV-2-RBD203-N1 (PDD081120)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yield BEFORE Purification</td>
<td>428 mg/L FS</td>
<td>540 mg/L FS</td>
</tr>
<tr>
<td>Overall Recovery</td>
<td>39 %</td>
<td>49 %</td>
</tr>
<tr>
<td>Yield AFTER Purification</td>
<td>135 mg/L FS</td>
<td>265 mg/L FS</td>
</tr>
<tr>
<td>Purity (Non-Reduced)</td>
<td>95.1 %</td>
<td>94.7 %</td>
</tr>
</tbody>
</table>
Receptor Binding Assays and Functional Comparison

[Graph showing binding assay results for RBD219-WT and RBD219-N1-C1]
Preclinical Functional Comparison

https://www.biorxiv.org/content/10.1101/2020.11.04.367359v1
Efficacy and Safety in a NHP Model

Dr. Sudhir Kasturi

VACCINATE

Weeks: -4 0 d1 d2 1 2 4 d4 5 6 9 10 13 d1 d2 d4 d7

Blood
- Innate responses by Flow, transcriptomics
- CD4+ and CD8+ T cell responses
- Serology (binding, neutralization, ADCC)
- Plasmablast analysis by ELISPOT and FACS

Bone Marrow
- Plasma cell analysis by ELISPOT
- IgG and IgA binding antibody
- BAL collections for immune phenotyping

Rectal Swabs
- Details

Nasal Swabs
- Details

Throat Swabs
- Details

BAL
- Details

Challenge

Terminate

POST CHALLENGE

Frequency of collections TBD

- Nasal swabs (PCR)
- Throat swabs (PCR)
- Serum/plasma (SEROLOGY)
- PBMCs
- BAL (Immune cell phenotyping)
- H&E (Immunopathology at necropsy)

D7, Collection at necropsy
A seminal and strategic partnership for industrial scale

Coronavirus | U.S.-based Baylor College of Medicine ties up with India’s Biological E for COVID-19 vaccine

Bio E APPROACH

Classical vaccine with established infrastructure and manufacturing platform.

 Millions of doses of Hep B vaccine manufactured and distributed.

Collaboration with Baylor College of Medicine (Peter Hotez, Maria-Elena Bottazzi)

- The candidate cell bank and its production process were developed at Texas Children’s CVD
- The vaccine or closely related prototype induces high levels of protection in mice and NHPs
- Biological E scaling up production and advancing its Ph 1/2 clinical testing in seven sites in India
- Ease of production, scalability and storage at 4ºC
Emerging Strains of SARS-CoV-2

Residues within the Receptor Binding Domain (RBD) sequence

- B.1.1.7 ("UK strain"): N501Y, delH69, delV70, delY145, A570D, P681H, T716I, S982A D1118H


- P1/2 ("Manaus/Brazil strain")
  E484K

- L425R ("California strain")
  L425R

- 20G/677H ("Ohio strain I")
  Q677H (plus mutations in M: A85S and N: D377Y)

- 20G/501Y (Ohio strain II)
  N501Y (plus mutations in ORF1AB)

Mutations arising in SARS-CoV-2 spike on sustained human-to-human transmission and human-to-animal passage, Robert F. Garry
Do the mutations allow the virus to evade the immune system?

Mutant RBD proteins under development

High Priority:
• **UK-RBD**: RBD203-N1, N501Y
• **ZA-RBD**: RBD203-N1, K417N+E484K+N501Y
• **Brazil-RBD**: RBD203-N1, E484K

Lower Priority:
• Mink/DK RBD: RBD203-N1, N501Y+Y453F
• RBD203-N1, Y453F

Criteria for prioritization
• Prevalence
• Potential for immune evasion
• Potential for increased infectivity

Pseudovirus Generation

<table>
<thead>
<tr>
<th>Mutations</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>D614G</td>
<td>Completed</td>
</tr>
<tr>
<td>N501Y-D614G (UK)</td>
<td>Completed</td>
</tr>
<tr>
<td>Δ69-70-N501Y-D614G (UK)</td>
<td>Near completion</td>
</tr>
<tr>
<td>Δ69-70-N501Y-D614G-P681H (UK)</td>
<td>In progress</td>
</tr>
<tr>
<td>E484K-N501Y-D614G (ZA)</td>
<td>Near completion</td>
</tr>
<tr>
<td>K417N-E484K-N501Y-D614G (ZA)</td>
<td>Near completion</td>
</tr>
<tr>
<td>E484Q-D614G</td>
<td>Near completion</td>
</tr>
<tr>
<td>K417T-E484K-N501Y-D614G</td>
<td>Under consideration</td>
</tr>
<tr>
<td>L452R (CA)</td>
<td>Starting</td>
</tr>
</tbody>
</table>

Courtesy Dr. Jason Kimata, BCM
Leveraged a path for a COVID-19 vaccine from prior experience

Exploring a US strategy including pediatric and maternal immunization vaccine suitability

Exploring other delivery and adjuvant systems

Interest in evaluating as boosters for OWS vaccines

Expanding to universal coronavirus vaccine development

Interest in expanding partnerships with US pharma and investors
Coronavirus Vaccines in Development Team Leads

A SARS CoV Vaccine as a potential heterologous vaccine against SARS-2 CoV

A SARS-2 CoV Vaccine leveraging the knowledge gained from SARS CoV

And many other staff and faculty behind the scenes

Collaboration with BCM Cores

Dr. Kimata and others

This work was supported by Robert J. Kleberg Jr. and Helen C. Kleberg Foundation, Fifth Generation, Inc. (Tito’s Handmade Vodka), JPB Foundation, NIH-NIAID (AI14087201) and Texas Children’s Hospital Center for Vaccine Development Intramural Funds.