THE DEVELOPMENT OF FUTURE VACCINES AGAINST ENTERIC DISEASES

Duncan Steele
Bill & Melinda Gates Foundation
Correlates of enteric-vaccine induced protection
21-23 March, 2016, Fondation Mérieux, Annecy
WHAT PATHOGENS ARE CAUSING MODERATE TO SEVERE DIARRHEA IN YOUNG CHILDREN?

- Rotavirus
- Shigella
- ETEC
- Norovirus
- S Typhi

Kotloff K et al. Lancet 2013; 382(9888): 209-22

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ROTAVIRUS VACCINES – NATIONAL INTRODUCTIONS BY GEOGRAPHIC REGION*

* As of 31 January 2016
ROTAVIRUS VACCINE DEVELOPMENT IN INDIA

ROTAVAC® licensure in India

- Shown to be safe & efficacious in Phase III trial in India
  - 54% efficacy against severe rotavirus gastroenteritis over 2 years of life
  - 56% protection in the first year of life
- ROTAVAC® products could achieve major impact in India and in Gavi-eligible countries
- First-generation product to be priced at ~$1 per dose

National launch in first 4 States in India on 26th March 2016

Dr MK Bhan, former secretary, Department of Biotechnology, Dr K Vijay Raghavan, Secretary, DBT, Govt of India with Dr Krishna Ella, Chairman & Managing Director, Bharat Biotech, and Dr TS Rao, DBT at the release of Rotavac phase-III trial data in New Delhi
ROTAVIRUS VACCINE CANDIDATE PIPELINE

Discovery & preclinical

Phase 1

Phase 2

Phase 3

Liquid presentation
Bharat Biotech

Lyophilized BRV
Serum Institute

Human rotavirus
Polyvac, Vietnam.

Lamb rotavirus
Lanzhou Institute of
Biological Products

Bharat Biotech
Frozen product licensed in India

Phase 3 efficacy
RCT in 7500 infants

Immunogenicity comparator to RotaTeq

Live-attenuated, oral

Liquid BRV
Serum Institute

RV3-BB
Biofarma, Indonesia

Lyophilized BRV
Shantha Biotechnics

NRRV (P2-VP8*)
PATH

Non replicating

Inactivated Rotavirus
CDC
CHARACTERISTICS OF P2-VP8* VACCINE

Developed at US NIH by Yasutaka Hoshino
Truncated VP8* subunit
- human Wa strain (G1 P1a[8])
- fused to the tetanus toxin P2 CD4 epitope
- expressed in E. coli

No unexpected toxicity in rabbits at doses up to 60 µg
Non-pyrogenic
Liquid formulation, adsorbed to aluminum hydroxide

PHASE 1 STUDY OF P2 VP8* NON-REPLICATING ROTAVIRUS VACCINE

ANTI-P2-VP8* IgG EIA TITERS

ANTI-P2-VP8* IgA EIA TITERS

NEUTRALIZING ANTIBODY TO OTHER ROTAVIRUS STRAINS
(≥4-FOLD INCREASE 28 DAYS AFTER 3RD DOSE)

<table>
<thead>
<tr>
<th>Strain</th>
<th>10 µg % (CI)</th>
<th>30 µg % (CI)</th>
<th>60 µg % (CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wa (G1P[8])</td>
<td>67 (35, 90)</td>
<td>42 (15, 72)</td>
<td>58 (28, 85)</td>
</tr>
<tr>
<td>89-12 (G1P[8])</td>
<td>83 (52, 98)</td>
<td>67 (35, 90)</td>
<td>83 (52, 98)</td>
</tr>
<tr>
<td>P (G3P[8])</td>
<td>58 (28, 85)</td>
<td>67 (35, 90)</td>
<td>83 (52, 98)</td>
</tr>
<tr>
<td>DS1 (G2P[4])</td>
<td>0 (0, 26)</td>
<td>50 (21, 79)</td>
<td>58 (28, 85)</td>
</tr>
<tr>
<td>ST3 (G4P[6])</td>
<td>0 (0, 26)</td>
<td>8 (0, 38)</td>
<td>17 (2, 48)</td>
</tr>
</tbody>
</table>

Phase 1/2 age-descending, dose-ranging study of P2 VP8* P[8] completed in South Africa
Phase 1/2 age-descending, dose-ranging study of trivalent P2 VP8* P[8]; P[6]; P[4] ongoing currently

SHIGELLA VACCINES

Four serogroups of *Shigella*
- Flexneri (6 serotypes)
- Sonnei (1 serotype)
- Bodyii (19 serotypes)
- Dysenteriae (15 serotypes)

Complex multivalent vaccine construct
SHIGELLA CANDIDATE PIPELINE

**Discovery & preclinical**

- **Live**
  - ShigEtec
    - EveliQure
  - Ty21a expressing LPS
    - Protein Potential
  - WRSf2a2 & WRSf2a3
    - NIAID, Walter Reed

- **Killed**
  - Inactivated trivalent
    - Walter Reed, PATH

- **Subunit**
  - SPS-Sf2a
    - Pasteur Institute
  - GMMA
    - NVGH
  - InavalexAR
    - Walter Reed
  - O-SPC/rBU, OSPC-rDT
    - Shriver Institute
  - DB Fusion
    - PATH
  - 34 kDa OMP
    - NICED
  - GVXN SD 133
    - GlycoVaxyn
  - Invaplex 50
    - Walter Reed

**Phase 1**

- WRSs2 & WRSs3
  - NIAID, Walter Reed
  - CVD1208S
  - U. Maryland, PATH

**Phase 2**

- WRSs1
  - Walter Reed
  - SC602
  - Walter Reed

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1. Shigetec is a combination ETEC-Shigella vaccine currently in development by a private vaccine manufacturer in Austria.
2. 2,3,4. Vaccines currently on hold/no longer in development
CURRENT LANDSCAPE OF *SHIGELLA* VACCINE DEVELOPMENT

**Cellular candidates**
- CVD1208 (live, attenuated)
- WRSS1 (live, attenuated)

**Subunit approaches**
- Conjugates: chemical, recombinant, synthetic
  - Invaplex
  - Generalized modules of membrane antigens (GMMA)
  - Outer membrane vesicle (OMV)
  - DB Fusion
  - 34 kDa OMP

**Ty21a + Shigella LPS**

**Shigella** whole cell (inactivated)

**Truncated whole cells**

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Walker RI. *Vaccine*. 2015;33:954-965
ETEC COLONIZATION FACTORS AND TOXINS

**Fimbriae**  *Intestinal adherence*
- CFA*I*
- CFA*II*  CS*1*, CS2, CS3
- CFA*IV*  CS4, CS5, CS6
- Others (CS17, CS14, PCF071)

**Toxins**  *Cause diarrhoea*
- LT (Thermal labile)
- ST (Thermal stable)
- LT/ST
CURRENT PIPELINE OF ETEC VACCINE DEVELOPMENT

Preclinical

Phase 1

Phase 2

Live

Killed

Subunit

ACE527
PATH

ETVAX
SBH

Stable Toxoid

Fimbrial Tip Adhesin

Anti-adhesin-toxoid Fusion (MEFA)

STOPENTERICS

NMRC, Sanofi, IDRI

KSU, JHBSPH
CURRENT STATUS OF ETEC VACCINE DEVELOPMENT

Whole cell approaches to ETEC vaccine development
- Killed, whole cell strains (eg. ETVAX, Scandinavian BioPharma)
- Live attenuated strains (eg. ACE 527, TD Vaccines and CNBG)

Subunit/peptide approaches
- Fimbrial tip adhesins (FTA) (US NMRC and PATH)
- 7 CFA-based Multi-epitope Fusion Antigen (MEFA)

Other innovative approaches in pipeline
- ST toxoid (CIH, Norway; STOPENTERICS)
- New conserved ETEC antigens
- Vectored combination vaccines (ETEC-Shigella; ETEC-Typhoid)
• **Vaccine characteristics**
  • All toxin and antibiotic resistance genes deleted
  • *aroC; ompC; ompF* genes deleted
  • Recombinant CS1 and LTB expressed from chromosome

• **Early Phase 1/2b studies of frozen preparation (10^{11} cfu in two doses):**
  • Majority of subjects (>50%) mounted mucosal responses to key antigens: LTB; CFA/I; CS3; CS6
  • Did not meet primary endpoint of protection against moderate/severe diarrhoea
  • Significantly impacted secondary measures of incidence and severity of disease; 41% efficacious against severe disease (p = 0.03)
  • Significantly reduced shedding of challenge strain
ACE527 ± DMLT: EFFICACY AGAINST SEVERE DIARRHOEA

Study with lyophilized preparation of ACE527 at a dose of $10^{10}$ cfu in three doses with and without 25 µg dmLT adjuvant

<table>
<thead>
<tr>
<th>Group</th>
<th>N</th>
<th>Severe Diarrhoea</th>
<th>Protective Efficacy vs. Controls (P-value)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Yes (%)</td>
<td>No (%)</td>
</tr>
<tr>
<td>Controls</td>
<td>31</td>
<td>21 (68%)</td>
<td>10 (32%)</td>
</tr>
<tr>
<td>ACE527</td>
<td>13</td>
<td>7 (54%)</td>
<td>6 (46%)</td>
</tr>
<tr>
<td>ACE527 + dmLT</td>
<td>13</td>
<td>3 (23%)</td>
<td>10 (77%)</td>
</tr>
</tbody>
</table>

**Primary Endpoint:** Prevention of severe diarrhoea defined as cumulative passage of more than 800 grams of grade 3 to 5 diarrhoea stools for episodes beginning during the 120-hour observation period post-challenge.
FIMBRIAL TIP ADHESINS (FTA)

FTA are important antigenic and functional component of the fimbria; the binding site domain

Final vaccine will have 3-5 FTA types.

• May offer broader coverage for strains of ETEC relevant to developing countries

Passive protection in humans; (active protection in non-human primates when given with mLT by ID route)

• Monovalent form (CfaE): 3 doses was safe and immunogenic in adult volunteers

• Challenge study with mLT showed value of protection against severe disease
## Enteric Controlled Human Infection Models

<table>
<thead>
<tr>
<th>Pathogen</th>
<th>Strain</th>
<th>Predominant Antigen (s)</th>
<th>Information provided by the Model</th>
<th>Vaccine Applicability (in the near term)</th>
<th>Value Proposition</th>
</tr>
</thead>
<tbody>
<tr>
<td>ETEC</td>
<td>B7A (O148:H28, LT+, ST+, CS6+)</td>
<td>LT, ST, CS6</td>
<td>Protection; Establish the role of CS6 in protection</td>
<td>FTA</td>
<td>Predictable model that can be used to evaluate vaccine efficacy</td>
</tr>
<tr>
<td></td>
<td>E24377A (O139:H:28, LT+, ST+, CS1+, CS3+)</td>
<td>LT, ST, CS1, CS3 [Non 078]</td>
<td>Protection; broader applicability to lead candidate (ETVAX) due to use a non-O78 strain;</td>
<td>ETVAX, Combination inactivated oral ETEC-Shigella vaccines</td>
<td>Identify correlates of vaccine protection</td>
</tr>
<tr>
<td></td>
<td>ST-ETEC (TBD)</td>
<td>CFA/1, ST</td>
<td>Protection, understanding role of ST;</td>
<td>ST toxoid, parenteral ETEC vaccine that incorporates ST, ST-LT constructs</td>
<td>Identify correlates of natural immunity</td>
</tr>
<tr>
<td>Pathogen</td>
<td>Strain</td>
<td>Predominant Antigen (s)</td>
<td>Information provided by the Model</td>
<td>Vaccine Applicability (in the near term)</td>
<td>Value Proposition</td>
</tr>
<tr>
<td>----------</td>
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<td>---------------------------------------------</td>
<td>-----------------------------------------------------------------------</td>
<td>-----------------------------------------</td>
<td>-----------------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Shigella</td>
<td>S. sonnei 53G</td>
<td>S. sonnei LPS, Ipa B, C &amp; D</td>
<td>Protection against Shigella species</td>
<td>GMMA</td>
<td>Predictable model that can be used to evaluate vaccine efficacy</td>
</tr>
<tr>
<td></td>
<td>S. flexneri 2a</td>
<td>S. flexneri 2a</td>
<td>Protection against Shigella species</td>
<td>GVXN Recombinant glycoconjugate, TSWC</td>
<td>Identify correlates of vaccine protection</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Identify correlates of natural immunity</td>
</tr>
</tbody>
</table>
PROGRESS IN TYPHOID CONJUGATE VACCINE DEVELOPMENT

<table>
<thead>
<tr>
<th>1994-2010</th>
<th>Preclinical stage</th>
<th>Phase I</th>
<th>Phase II</th>
<th>Phase III</th>
<th>National Licensure</th>
<th>Post-licensure</th>
</tr>
</thead>
<tbody>
<tr>
<td>NIH Vi-rEPA development - not commercialized (Phase I-Phase III efficacy, Infant co-admin)</td>
<td>DAVAC (Vi-DT)</td>
<td>PT Biofarma (Vi-DT)</td>
<td></td>
<td></td>
<td>PSF submission expected 2016</td>
<td></td>
</tr>
<tr>
<td>WALVAX (Vi-TT)</td>
<td>Incepta (Vi-DT)</td>
<td>SK Chemicals (Vi-DT)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Biological E (Vi-CRM197)</td>
<td>Finlay (Vi-DT)</td>
<td></td>
<td></td>
<td>Under CFDA review</td>
<td>Lanzhou CNBG (Vi-rEPA)</td>
<td></td>
</tr>
<tr>
<td>Eubiologics (Vi-CRM197)</td>
<td></td>
<td></td>
<td></td>
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<td></td>
</tr>
</tbody>
</table>
VI-rEPA CONJUGATE VACCINE (DEVELOPED BY NIH)

A double-blind, placebo-controlled and randomized efficacy study was conducted in 2 to 5 year-old children in Vietnam

- 11,091 children were injected twice, 6 weeks apart, with the Vi-rEPA vaccine or saline placebo
- Efficacy at 27 months of active surveillance was 91%
- Efficacy at 46 months after additional 19 months of passive surveillance was 89%

A second study was conducted with 301 infants who received Vi-rEPA with routine childhood vaccines at 2, 4, 6 months and Hib or Vi-rEPA at 12 months in Vietnam

- Vi-rEPA was safe in infants
- Induced protective anti-Vi levels, with robust GMTs
- Compatible with EPI vaccines and it can be used in infants.

This trial is the source of the only existing efficacy data for any typhoid conjugate vaccine, which has implications for future clinical development and regulatory pathways


<table>
<thead>
<tr>
<th>TABLE 3. Efficacy of Vi-rEPA Conjugate Vaccine.</th>
</tr>
</thead>
<tbody>
<tr>
<td>VARIABLE</td>
</tr>
<tr>
<td>Children who received two correctly labeled injections — no.</td>
</tr>
<tr>
<td>Children with typhoid fever — no.</td>
</tr>
<tr>
<td>Attack rate (cases/1000 children)</td>
</tr>
<tr>
<td>All children — no.‡</td>
</tr>
<tr>
<td>Children with typhoid fever — no.</td>
</tr>
<tr>
<td>Attack rate (cases/1000 children)</td>
</tr>
</tbody>
</table>
• Bharat Biotech vaccine licensed in India in 2013 on immunogenicity data
• Phase III - RCT in ages 2 - 45 year olds using licensed Typbar (Vi-PS) as an active comparator
• Open Label Trial in 6 months – 2 year olds
• Good immunogenicity responses across all age groups
• Post-licensure studies ongoing, including measles co-administration; evaluation of different dosing schedules; two years follow up for safety and immunogenicity data
• Human challenge study is ongoing at Oxford University to assess a clinical outcome for the vaccine

Immune Response Across Age Groups
Licensure on the basis of immunogenicity based on the Vi-rPA Vaccine Efficacy Data

1. Need a validated ELISA assay
   • Measures the concentration of anti-Vi IgG in human serum sample

2. Need a valid human reference standard to measure serum anti Vi-IgG
   • Will facilitate comparison of anti-Vi elicited from other manufactures to those of an experimental vaccine manufactured by the NIH (Vi-rPA) that has already undergone a clinical trial with efficacy outcomes
   • Allows for comparisons between laboratories
     - enables the comparison of antibodies elicited from other manufactures to those of Vi-rPA
   • Vi IgG antibody levels are currently being measured by investigators and referenced against arbitrarily-determined EU values assigned by each laboratory against its own in-house standard serum.
**TYPHOID CHALLENGE MODELS**

- **Determine the dose of Salmonella Typhi required to produce an AR of 60-75%**
- **Clinical and laboratory features**
  - Time course
  - Bacteraemia
  - Inflammatory response
- **Development of immunity**
  - Innate & humoral
  - Cell mediated immunity
  - Long-term immunity after treatment
- **Diagnostics**
- **Variation in genomic response**
- **PCR-based Mass spectrometry**
- **Research mechanisms for related pathogens**
- **Correlates of protection**

**VACCINE STUDIES**

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**FIG. 1.** Clinical summary of volunteers 9 J. C., after the ingestion of 10⁶ viable Salmonella typhi Vaccine strain. Various temperatures are recorded.

**Challenge dose**
- 10³ CFU
- 10⁴ CFU

**Cumulative percentage with typhoid diagnosis (any method)**

<table>
<thead>
<tr>
<th>Number days after challenge</th>
<th>0</th>
<th>5</th>
<th>10</th>
<th>15</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>5</td>
<td>5</td>
<td>10</td>
<td>20</td>
<td>30</td>
</tr>
<tr>
<td>10</td>
<td>20</td>
<td>30</td>
<td>40</td>
<td>50</td>
</tr>
<tr>
<td>15</td>
<td>40</td>
<td>50</td>
<td>60</td>
<td>70</td>
</tr>
</tbody>
</table>

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NEW INTRODUCTIONS WILL LEAD TO INCREASED DELIVERY COMPLEXITY AND FINANCIAL CHALLENGES

Note: All figures are based on GAVI-funded vaccines only

VACCINES IN DEVELOPMENT WILL CROWD STANDARD EPI SCHEDULES

<table>
<thead>
<tr>
<th>GAVI-supported</th>
<th>In development</th>
</tr>
</thead>
<tbody>
<tr>
<td>6/10/14 weeks</td>
<td>Up to 2 oral, 3 injections</td>
</tr>
<tr>
<td>6/7/8 months</td>
<td>Up to 5 injections!</td>
</tr>
<tr>
<td>9 months</td>
<td>(Penta, Rota, PCV, OPV/IPV)</td>
</tr>
<tr>
<td>12 months*</td>
<td>(ETEC/Shigella)</td>
</tr>
<tr>
<td>15 months</td>
<td>(Malaria)</td>
</tr>
<tr>
<td>18 months</td>
<td>(TCV)</td>
</tr>
<tr>
<td>2 years</td>
<td>(Malaria boost)</td>
</tr>
</tbody>
</table>

*cholera not yet in GAVI portfolio

- (YF, JE, Men, Measles)
- (Cholera)
- (JE (RI), MR)
- (Dengue)
The goal is to advance development of a portfolio of ETEC and *Shigella* vaccine candidates towards a combined vaccine over 5 years (2014-2018) with clear milestones to evaluate for down-selection or reprioritization.
SUMMARY

- Full, robust pipelines for vaccine development for the enteric and diarrhoeal pathogens
  - Multivalent vaccines likely necessary for ETEC and Shigella
  - Various vaccine constructs including live attenuated, killed whole cell, subunit,..
  - Various administration modes (e.g. oral, parenteral)
- Controlled human infection models available for many pathogens
- Reliable and validated assays required for comparability of vaccine constructs
- Combination vaccine constructs likely required – adding to complexity of vaccine composition

- Correlates of protection would greatly facilitate vaccine development, however timelines before large phase 3 efficacy studies are required is extremely short
THE WORK IS COMPLICATED.

WHY WE DO IT IS NOT.