Progress in the Development of a Vi-CRM_{197} Conjugate Vaccine
BioE’s TCV Monovalent Project

*Vi source*

- Source of Vi is *Citrobacter freundii*

- Citrobacter generates Vi PS which is structurally identical to the Vi PS

- High yields, BSL1 provides significant advantages

- Purified Vi PS meets WHO TRS requirements
Vi Fermentation Lots Summary: GMP Campaign

- GMP MCB & WCB
- All regulatory approvals (DCGI) in place for Clin Mfg
- Fermentation carried out at Pilot Scale in GMP Pilot Plant
- Three consecutive GMP lots completed, lots met acceptance criteria
BioE’s TCV Monovalent Project

Carrier Protein used for Conjugation

• CRM$_{197}$ used as a carrier protein
• BioE has developed two technology platforms for CRM$_{197}$
  • Recombinant C7 based technology platform
  • Recombinant *E.coli* based technology platform
• Both technologies result in purified CRM$_{197}$ which meets predetermined quality attributes
• Both CRM$_{197}$ technologies have been presented to Indian Regulatory Agency RCGM (Review Committee on Genetic Manipulation)
• BioE is proceeding with the recombinant C7 derived CRM$_{197}$ for the TCV program
BioE’s TCV Monovalent Project

Acceleration Strategy

- BBIL has recently introduced Typbar-TCV® in the market
- Typbar-TCV® contains 25 μg Vi antigen conjugated to TT
- Typbar-TCV® is licensed children ≥ 6 months of age and adults as a single dose administered intramuscularly
- WHO TRS defines an immunological threshold value:
  - Seroconversion (4-fold)
  - 4.3 μg/ml anti-Vi antibody measured by ELISA appears to be associated with a high level of sustained protection

BioE’s TCV is targeted as a single dose with 25 μg Vi conjugated to CRM₁₉₇. It is targeted to be licensed in children > 6 months of age and adults.
Vi-CRM\textsubscript{197} Conjugates: Critical to Quality

<table>
<thead>
<tr>
<th>Bulk Conjugate</th>
<th>Formulated Bulk</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Identity</td>
<td>• Identity</td>
</tr>
<tr>
<td>• Vi Concentration</td>
<td>• Vi Concentration (25 μg/0.5 mL)</td>
</tr>
<tr>
<td>• Vi:CRM\textsubscript{197} ratio</td>
<td>• Vi:CRM\textsubscript{197} ratio</td>
</tr>
<tr>
<td>• Size</td>
<td>• Size</td>
</tr>
<tr>
<td>• % Free PS</td>
<td>• % Free PS</td>
</tr>
<tr>
<td>• O-Acetylation level</td>
<td>• Sterility</td>
</tr>
<tr>
<td>• Residual reagents</td>
<td>• Osmolarity, pH</td>
</tr>
<tr>
<td>• Endotoxin</td>
<td>• Stability</td>
</tr>
<tr>
<td>• Stability</td>
<td></td>
</tr>
</tbody>
</table>

TCV DS and DP meets Indian Pharmacopoeia (IP) and WHO TRS recommendations
Balb/c Mice Immunization Plan for TCV

Study Plan:
1. Mice:
   • Inbred Balb/C Female SPF Mice
   • ≤ 6 weeks old
   • 20 mice/per group
2. Route: Subcutaneous
3. Dose: 1/10 SHD
4. Sera collected by terminal bleeding

Samples Evaluated:
1. PBS
2. Vi PS
3. Vi-CRM
4. Typbar-TCV

Responses Evaluated:
• Anti-Vi IgG (Fold increase over Placebo and over PS only)
• Secondary antibody response
Dose 1 and Dose 2 – Anti Vi IgG, Mice

Experiment Set 1

Post dose 1

Post dose 2
Dose 1 and Dose 2 – Anti Vi IgG, Mice

Experiment Set 2

Post dose 1

Post dose 2

10th International Conference on Typhoid and Other Invasive Salmonelloses
Initial Immunogenicity Evaluation
Vi-CRM$_{197}$: Conclusions

- BE Vi-CRM$_{197}$ is highly immunogenic in mice. BE Vi-CRM$_{197}$ preclinical immunogenicity results meet WHO TRS requirements.

- BE Vi-CRM$_{197}$ elicits a secondary antibody response in mice.

- Similar immunogenicity patterns observed in Rabbits

- BE Vi-CRM conjugate has similar characteristics to other reported conjugates
  - Vi-TT by BBIL
  - Vi-rEPA by Szu et al
  - Vi-CRM by Eubiologics
  - Vi-DT by IVI
TCV Monovalent Project: Current Status

- RCGM approval for use of recombinant CRM\textsubscript{197}
- PCT completed, submitted to NRA
- Process development and pilot-plant scaleup complete
- GMP clinical material made and on stability (both DS & DP)
- Phase I dossier submitted to DCGI. Expect clinical studies to start 2Q17
- Large scale clinical manufacturing in 3Q-4Q’17
- Target NRA licensure 2018
- Target WHO PQ 2019
THANK YOU