Sabin-IPV Development

9th WHO-UNICEF Informal Consultation with OPV & IPV Manufacturers & NRAs

02 December 2010, Geneva

Wilfried Bakker

02 December 2010
Contents Presentation

- History & Rationale for Sabin-IPV development
- Sabin-IPV : Vaccine Development Project
- Seedlots and Clinical Lot production
- Technology Transfer to DCVM
- R&D-program: Process optimization & Antigen sparing
Rationale for Sabin-IPV development

- Current tool for the WHO Polio Eradication program is: OPV
- Emergence and outbreaks of cVDPVs since 2000
- Therefore use of all OPV should be stopped after PE

- Risk: after PE developing countries will stop polio vaccination
- IPV production (using wild-type polio) is not feasible in developing countries because of containment risks

- Sabin-IPV appears feasible:
  - OPV is currently produced in developing countries
  - Lower risk of production facilities related polio outbreaks
Lab-Scale Sabin-IPV Purification Scheme

Monovalent bulk (by BioFarma)

Concentration

Purification

Inactivation

Monovalent pool

Proof-of-principle project (2007):
Preparation of purified trivalent inactivated Sabin-IPV

based on:
The current NVI Salk-IPV production process
Sabin-IPV project at NVI

Planned activities (2008 – 2011):

I. Clinical lot preparation &
    Prepare for Clinical studies and Licensing

II. Process optimization and dose reduction studies

III. Training and Tech Transfer :
    – Generic workshop / training courses
    – Strive for bilateral Tech Transfer agreements with DCVM

Kreeftenberg et al. (2006) Biologicals; Kersten et al. (1999) Vaccine
Planning: Anticipated Milestones Summary

- **Seedlots**: Q1 2009
- **Monovalent pools**: Q2 2010
- **Trivalent product**: Q3 2010
- **Released product**: Q4 2010
- **Start clinical study**: 2011
- **Finish clinical study**: 2011

- **Start stability testing**: Q1 2009
- **Sabin-IPV Workshop**: Q2 2010
- **Finish stability testing**: Q3 2010
- **Start pre-clinical testing**: Q4 2010
- **Start hands-on training of TT partners**: Q1 2011
Current Salk-IPV production scheme

Upstream processing

Vero cell
Media
Virus

Downstream processing

Monovalent pool
Trivalent bulk

Inactivation

IPV DT&IPV
Before CTM production:
Lab-scale Process Development

- Multivariate Data Analysis (Salk-IPV production)
  For better process understanding
  and future improvements

- Scale-down model (USP & DSP)
  Using DOE methods for future process improvements
  ACF media; increased yields

- Set initial process specifications (Sabin-IPV production)
  MOI; virus culture temperature; SEC and IEX conditions

Thomassen et al. (2010) Biotechnol Bioeng; Bakker et al. (2010) ESACT-proceedings
Seedlot Generation

Source material:
- Type 1: WHO / Behringwerke 1976 SO+1
- Type 2: WHO / Behringwerke 1976 SO+1
- Type 3: Institut Mérieux 1963 (457-Pfizer) RSO1

Master Seed Lots (3 types): 10-L scale

Working Seed Lots (3 types): 350-L scale

Culture conditions:
MOI = 0.01 and T = 32.5°C for all types
Milestone 1:
Master (3x) & Working (3x) Seedlots made
Milestone 2: Monovalent Pools (6 lots) produced

Upstream processing:
- Vero cell
- Media
- Virus

Downstream processing:
- Monovalent pool

Inactivation:
- Process updated where appropriate: Clarification modernized

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Quality Control (QC)

- Selection of international release tests for production lots (product should meet current IPV release criteria)
- Based on EP and WHO guidelines

- General assays (e.g. Protein, TOC, Sterility, etc.)
- Polio specific assays (e.g. D-ag, Virus titer, Rat Potency, etc.)
- Sabin specific assays (Neurovirulence)
6 Monovalent Pools prepared: 2 lots per virus type (2 x 3 types = 6 lots) at 700-L bioreactor production scale

Monovalent Pool QC-testing according to Bill-of-Testing in progress:

- Current status: conform requirements (e.g. Identity Vero cells & Sabin Polio virus, Mycoplasma, Extreneous agents, Sterility, Virus titer, D-antigen content, Inactivation, Endotoxins, Formalin, Bovine serum, Protein nitrogen, Residual DNA)
- Neurovirulence testing is being outsourced
Immunogenicity: Virus neutralization titer

Type 1

Virus Neutralisation Titer (2log)

DU/shd

Salk IPV
Sabin IPV
Sabin IPV + Al(OH)₃

Type 2

Virus Neutralisation Titer (2log)

DU/shd

Type 3

Virus Neutralisation Titer (2log)

DU/shd
Sabin-IPV vaccine formulation considerations:

1. Neutralizing antibody titre should be equal or higher than that induced by the international (Salk-IPV) reference

2. At higher D-antigen doses a plateau in neutralizing antibody level is reached

<table>
<thead>
<tr>
<th>Type</th>
<th>Plain formulation (DU / single human dose)</th>
<th>Al(OH)₃ formulation (DU / single human dose)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>High</td>
<td>Target</td>
</tr>
<tr>
<td>Type 1</td>
<td>20</td>
<td>10</td>
</tr>
<tr>
<td>Type 2</td>
<td>32</td>
<td>16</td>
</tr>
<tr>
<td>Type 3</td>
<td>64</td>
<td>32</td>
</tr>
</tbody>
</table>

For reference: plain Salk-IPV formulation is (type 1 – 2 – 3): 40 – 8 – 32 DU/shd
Formulation Development

Conclusions

• On average 1 DU Sabin-IPV:
  – Type 1 is 1.5 times more potent than 1 DU type 1 Salk-IPV
  – Type 2 is 3 to 4 times less potent than 1 DU type 2 Salk-IPV
  – Type 3 is comparable potent with 1 DU type 3 Salk-IPV

• Al(OH)₃ adjuvation increases the Sabin-IPV potency 2 times

• Based on the relative potency Sabin-IPV could be formulated (expected needed dose) in:
  ➢ Plain (type 1 – 2 – 3): 10 – 16 – 32 DU/shd
  ➢ + Al(OH)₃ (type 1 – 2 – 3): 5 – 8 – 16 DU/shd

For reference: Salk-IPV formulation is (type 1 – 2 – 3): 40 – 8 – 32 DU/shd
CTM Production planning

Planned final product filling operations:

- **Milestone 3**: Pre-clinical lots (safety):
  Filled, April 2010

- **Milestone 4**: Pre-clinical lots (safety):
  Conform requirements, October 2010

- Phase I clinical lots: Q4 2010
  - One lot for European study (NVI/WHO);
  - Two lots for local study by TT partner & NVI

- Phase I clinical trial planned: Q1 2011
Regulatory Pathway / Clinical Strategy

- Dutch and/or European phase I study is preferred; local study & registration is required

- Scientific Advise by the Dutch MEB obtained in JULY 2008:
  - Sabin-IPV immunogenicity & safety should be equivalent or better than that for Salk-IPV

- Clinical Trial plan (immunogenicity & safety) and Regulatory Pathway are under development
Clinical Trial – Phase I age de-escalation approach

<table>
<thead>
<tr>
<th>Age group</th>
<th>Arm</th>
<th>Administration</th>
<th>Follow-up</th>
<th>Weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adult (19-49 yrs)</td>
<td>Normal (High dose)</td>
<td></td>
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<tr>
<td></td>
<td>Adjuvant (High dose)</td>
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<tr>
<td></td>
<td>15 arm</td>
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<tr>
<td>Toddler (4-10 yrs)</td>
<td>Normal (High dose)</td>
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<td></td>
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<tr>
<td></td>
<td>Adjuvant (High dose)</td>
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<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>15 arm</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Infants (2 mos)</td>
<td>Normal sIPV</td>
<td>Administration</td>
<td>Follow-up</td>
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<td></td>
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<tr>
<td></td>
<td>sIPV Adjuvant sIPV</td>
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<tr>
<td></td>
<td>(Low)</td>
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<tr>
<td></td>
<td>20 arm</td>
<td></td>
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<tr>
<td></td>
<td>sIPV Adjuvant sIPV</td>
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<td></td>
<td>(Middle)</td>
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<td></td>
<td>sIPV Adjuvant sIPV</td>
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<td></td>
</tr>
<tr>
<td></td>
<td>(High)</td>
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<td>WPcV</td>
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</tbody>
</table>

★ IPV
★ Blood collection
Technology transfer of Sabin-IPV to new developing country markets

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Starting from 2010:

• Sabin-IPV workshop on large-scale manufacturing done in June 2010

• Setup Sabin-IPV production & QC-testing course for TT partners

• Transfer lab/pilot-scale technology to selected DCVM partners for implementation at their own facilities in 2011

Website launched: www.sabin-ipv.nl
China Vaccine Project
1990-1998

GMP Facility Kunming

GMP Facility Lanzhou

GMP Facility Shanghai; now in use for H1N1 pandemic flu production and other vaccines
## Tech Transfer projects since 1990

<table>
<thead>
<tr>
<th>Project</th>
<th>Vaccine(s)</th>
<th>Recipient</th>
<th>Country</th>
<th>Approach</th>
<th>IP-issues</th>
</tr>
</thead>
<tbody>
<tr>
<td>World Bank Vaccine Project</td>
<td>DTP, MV, OPV</td>
<td>SIBP, LIBP, KIMB, (NCL)</td>
<td>China</td>
<td>turn-key</td>
<td>none</td>
</tr>
<tr>
<td>(1990 – 1998)</td>
<td></td>
<td></td>
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<td></td>
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</tr>
<tr>
<td>Hib – Project</td>
<td>Hib conjugate</td>
<td>Bio Farma SII, BE Ltd Glovax/SIBP</td>
<td>Indonesia, India, Korea/China</td>
<td>development and transfer of pilot process</td>
<td>non-exclusive license; fees and/or royalties</td>
</tr>
<tr>
<td>(1999 – now)</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>WHO ITPIV</td>
<td>egg-based inactivated influenza</td>
<td>VACSERA IVAC Others?</td>
<td>Egypt, Vietnam</td>
<td>1-generic, hub based 2- bilateral TT agreements</td>
<td>non-exclusive license; modest fees; no royalties</td>
</tr>
<tr>
<td>(2007 – now)</td>
<td></td>
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<tr>
<td>WHO Sabin-IPV</td>
<td>new safer polio</td>
<td>t.b.d.; potentially several</td>
<td>Shortlist of potential companies</td>
<td>1-generic, hub based 2- bilateral TT agreements</td>
<td>non-exclusive license; modest fees; royalties</td>
</tr>
<tr>
<td>(2008 – now)</td>
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Previous polio Technology Transfer experience
Continuing a tradition .... Technology Transfer

NVI pilot-plant facilities

150-L Bioreactor for Training (TT) purposes
Sabin-IPV : Vaccine Development Project

Modernization (R&D) program (preliminary results):

• Process Optimization (animal-component free)
• Characterization, Formulation & Immune response Optimization (dose reduction e.g. by using adjuvants; intradermal administration)
• Alternatives for inactivation: BPL vs. Formalin
Modernization & optimization using Animal-Component-Free media

- 750-L Production-scale
- 2.2-L Lab-scale
- Lab-scale (N = 5) using ACF-media
Batch vs recirculation cultivation

<table>
<thead>
<tr>
<th>Culture no.</th>
<th>Inoc. Cell density (x10^6 c/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>B1 (●)</td>
<td>1.00</td>
</tr>
<tr>
<td>B2 (○)</td>
<td>1.22</td>
</tr>
<tr>
<td>B3 (▲)</td>
<td>1.30</td>
</tr>
</tbody>
</table>

Inoculation cell density: 4 – 8 x 10^6 c/mL
Antigenic fingerprinting of Sabin and Salk

**Diagram:**
- **ELISA**
  - anti-Ig-HRPO
  - polio
  - MAb
  - PAb
- **Biacore**
  - RaMFc
  - polio
  - MAb

**Graph:**
- QC-ELISA
- 3-4E4
- 17C5M1
- 3-4E4
- 17C5M1
- 234
- 237
- 423 (result x2.5)
- 17C5M1
- 237
- 423 (result x2.5)

**Legend:**
- Sabin IPV 1A
- Salk IPV 1
• Salk and Sabin have different antigenic and immunogenic profiles
• D-antigen unit is less suitable from a standardisation point of view
• Antigen quantity expressed as ‘active concentration’ (amount of virus with D-antigenicity) appears to be an attractive alternative
Antigenicity of inactivated Sabin poliovirus type 1

• Alternative inactivation method using β-propiolactone (BPL)
• Antigenicity tested using 6 different monoclonal antibodies
De antigeniciteit is nauwelijks afgenomen door de inactivering met BOL of formaldehyde. Voor de meeste monoclonalen lijkt het dat BPL de epitopen iets minder aantast.
## Conclusions

<table>
<thead>
<tr>
<th>Salk-IPV</th>
<th>Sabin-IPV</th>
<th>Salk-IPV &amp; Sabin-IPV</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Routine GMP Production</strong>&lt;br&gt;MVDA for Process understanding&lt;br&gt;Lab-scale equivalent USP &amp; DSP</td>
<td><strong>Phase I Clinical lot Production (GMP)</strong>&lt;br&gt;Lab-scale equivalent used to set new specs.</td>
<td><strong>Technology Transfer for GMP Production</strong>&lt;br&gt;Scale-up to pilot-scale&lt;br&gt;Study improvements At lab-scale &amp; optimize</td>
</tr>
</tbody>
</table>

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Acknowledgements / Questions

Website:
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Sabin-IPV project team

Eric van Gerven – Facilities / Validation
Nico van den Heuvel – Production
Fred van Nimwegen – QC
Yvonne Thomassen – Process Dev.
Janny Westdijk – Assay Development
Bernard Metz – Inactivation Studies
Ahd Hamidi – Technology Transfer
Peter van ‘t Veld – QA
Lars Sundermann – QP
Monique van Oijen – Registration
Nynke Rots – Clinical Strategy
Wilfried Bakker – Project management
Peter Belt – Programme Management
And many other NVI colleagues