6-13-2016

Development of Typhax, a Salmonella Typhi Vi polysaccharide protein capsular matrix vaccine

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Development of Protein Capsular Matrix Vaccine Technology

ECI Portugal 2016

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Chief Scientific Officer

Matrivax

Company Overview

- Founders/Board of Directors:
  - **John J Mekalanos, PhD**- Professor, Harvard Medical School
  - **Yichen Lu, PhD**- Professor, Harvard School Public Health; President & CEO, VTI
  - **Gerald Chan, PhD**- Director Hang Lung Group Limited and Morningside

- Matrivax is a biotechnology company incorporated in October 2007, funded by Morningside Ventures, headquartered in Boston, MA; 18 FTEs

- Protein Capsular Matrix Vaccine (PCMV) technology, 'virtual conjugation', exclusively licensed from Harvard Medical School. Potential to transform complex 'conjugate' vaccine manufacture (e.g. pneumococcal and meningococcal vaccines) by enabling simplified, **low-cost** production and **extensive market penetration**

- Strategic Alliance with Vaccine Technologies Inc. (VTI) to construct a 3,000 square meter GMP vaccine pilot manufacturing facility in Haikou China to GMP manufacture pneumococcal, meningococcal, *C. difficile*, and enteric fever PCMV

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Targeted Polysaccharide Vaccine Markets

Pneumococcal Vaccines

- Merck Pneumovax® - 23-valent vaccine - no long term efficacy
- GSK Synflorix® -10-valent - limited in vaccine coverage
- Pfizer Prevnar® $6 B in sales in 2015; **Worldwide market projected to $8 billion ($5 billion in low-to-middle income markets)**

Meningococcal Vaccines

- Sales of sanofi Menactra® - quadrivalent conjugate; ages 2-55; >$1B sales
- Novartis launched quadrivalent conjugate, Menveo®

Enteric Fever Vaccines

- Typhim Vi ® and Vivotif ® moderate protective efficacy ~70%
- Typhoid fever market alone >$200MM; combination paratyphoid A projected >$350MM

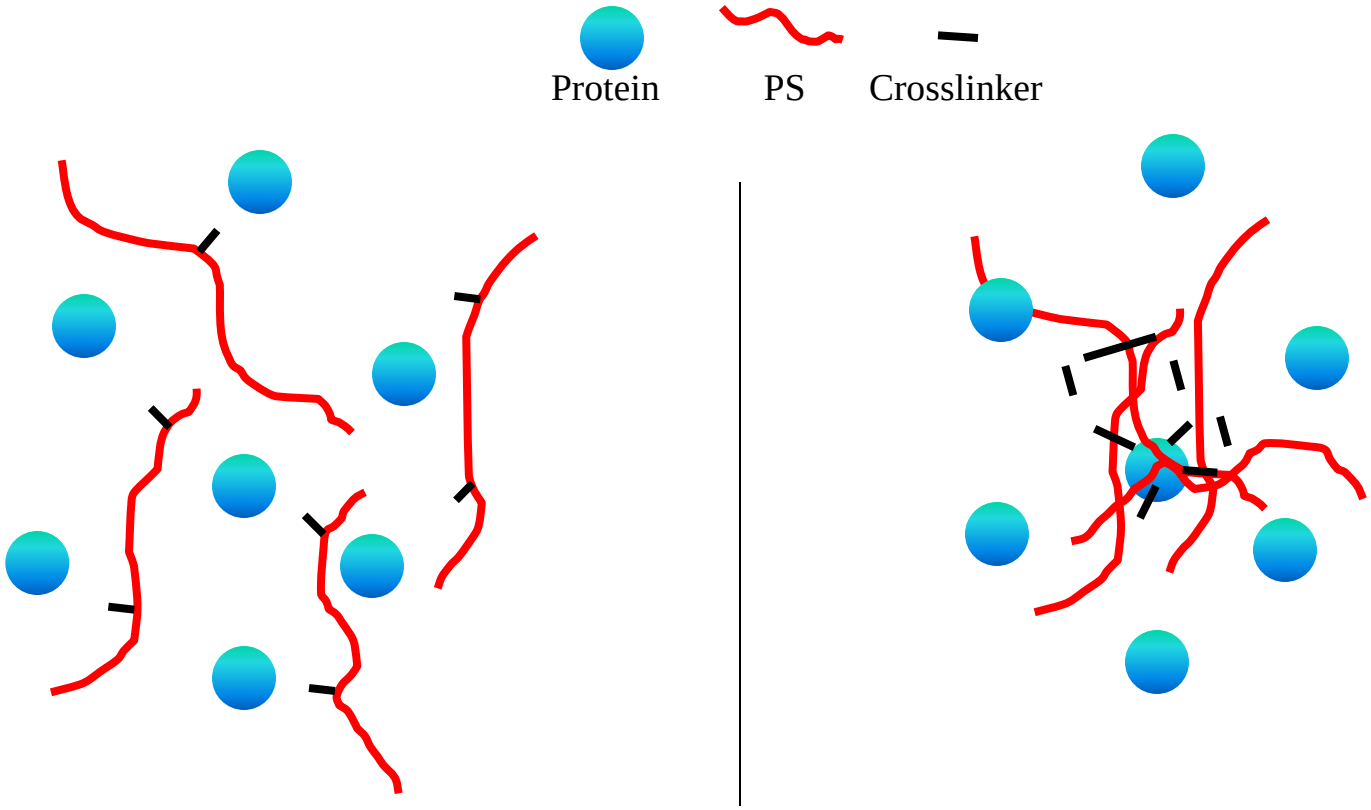
Clostridium difficile Vaccines

- No commercialized vaccine; sanofi toxoid based candidate in Phase 3
- *C. difficile* vaccine market projected > $1B sales

Conjugation versus PCMV technology

Conjugation covalently bonds PS to Protein

PCMV entraps PS in protein matrix



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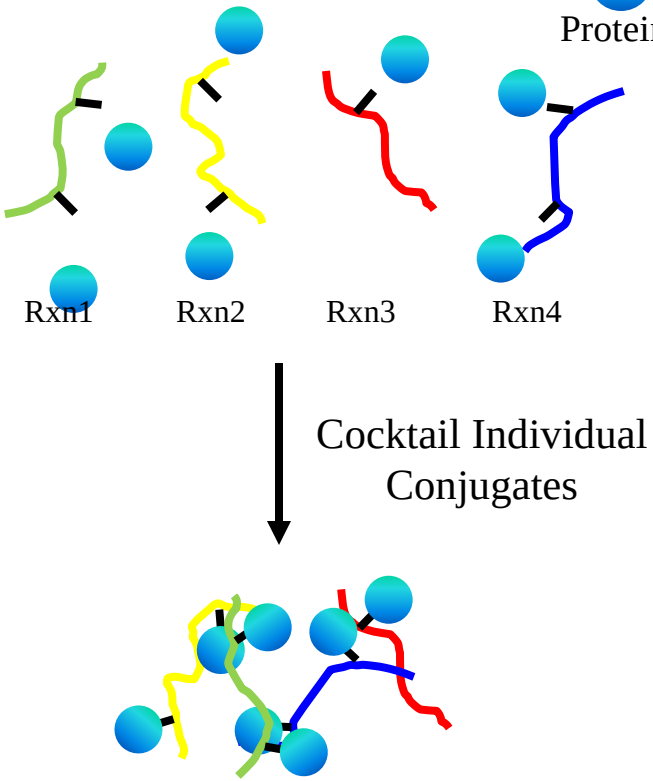
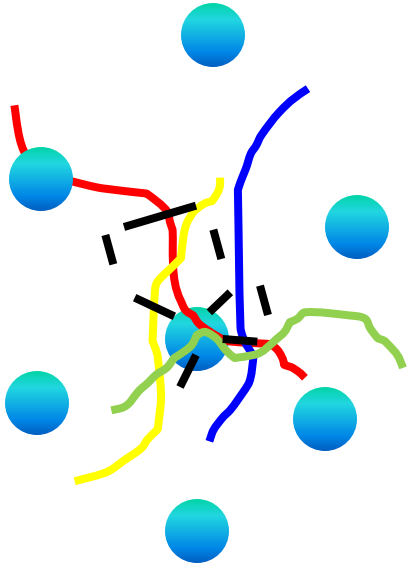
Conjugation versus PCMV technology

Multivalent conjugate vaccine



Multivalent PCMV



Single Reaction

Pneumococcal Vaccines

Serotypes	23 valent	Prevnar	Synflorix	Prevnar13
1	X		X	X
2	X			
3	X			X
4	X	X	X	X
5	X		X	X
6A				X
6B	X	X	X	X
7F	X		X	X
8	X			
9N	X			
9V	X	X	X	X
10A	X			
11A	X			
12F	X			
14	X	X	X	X
15B	X			
17F	X			
18C	X	X	X	X
19A	X			X
19F	X	X	X	X
20	X			
22F	X			
23F	X	X	X	X
33F	X			

Vaccine	Protective Efficacy* Worldwide	China
Prevnar 7®	~65%	~64%
Synflorix®	~80%	~65%
Prevnar 13®	>85%	>80%

*predictive value

•24-valent Pneumococcal vaccine >95% predicted protection against all pneumococcal serotypes worldwide!

•An enabling technology, such as PCMV, possesses the capacity to formulate and commercialize such vaccines

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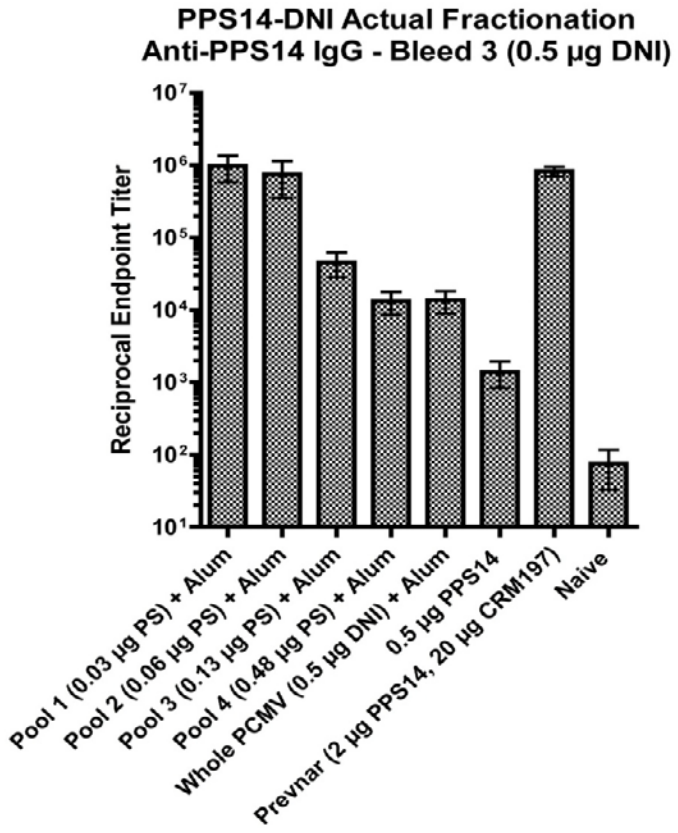
PCMV Proof-of-Principle Strategy

- Preclinical: Pneumococcal PCMV
- Clinical: Vi-CRM197 Typhoid Fever PCMV

Monovalent and Multivalent Pneumococcal PCMV

Size-Fractionated PPS14-DNI PCMV induced antibody response

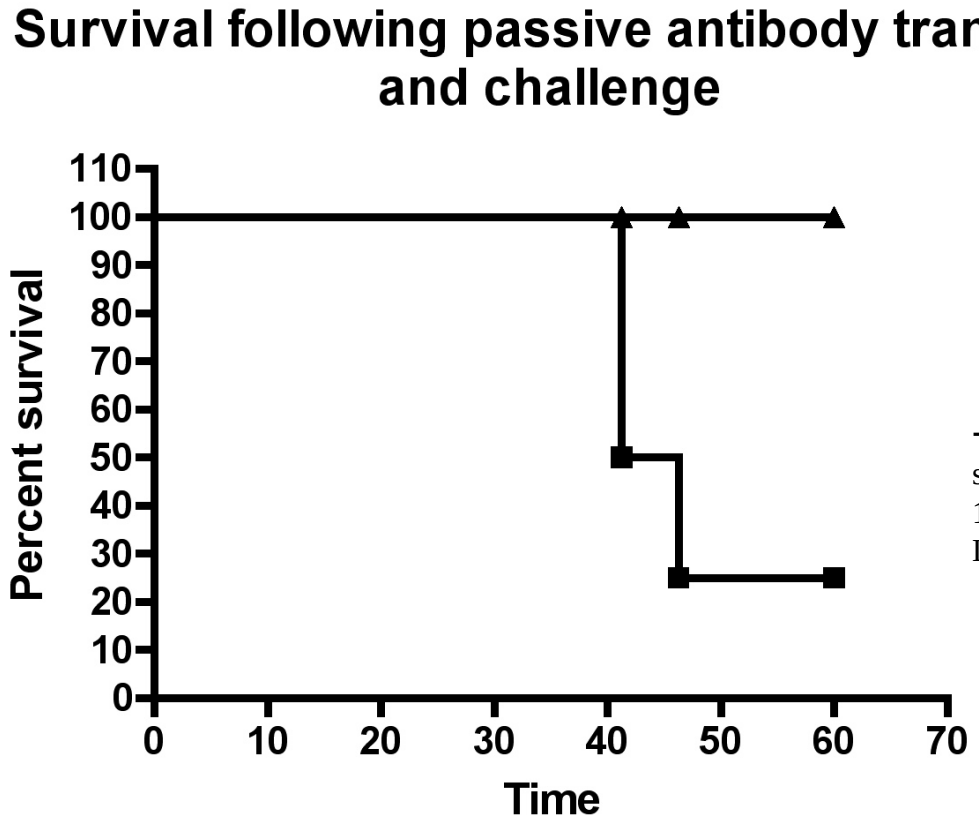


PPS14 dose (0.5 µg DNI)	Day 38 GMT
Pool 1 (0.03 µg PS)	501103
Pool 2 (0.06 µg PS)	228543
Pool 3 (0.13 µg PS)	27691
Pool 4 (0.48 µg PS)	4615
Whole PCMV (0.5 µg DNI)	9413
PPS14 only (0.5 µg)	800
Prevnar (2 µg PS, 20 µg CRM197)	776047
Naive	50

PCMV induces comparable antibody response to Prevnar at ~15% the dose

Day 39 anti-PPS14 IgG titers showing mean and standard error of the mean from mice immunized with 0.5 µg DNI + PPS14 (left panel). Day 39 anti-PPS14 GMT calculated from individual serum titers for mice immunized with 0.5 µg DNI + PPS14 (right panel)

Passive transfer of anti-PPS14 PCMV sera to mice protects against lethal IN pneumococcal challenge



-mice injected IP with 100 µl of naïve sera or pooled anti-14 sera (immunized 3x with DNI-PPS14 PCMV; ELISA titer 1:6900); Virulent *S. pneumoniae* serotype 14 (1.5 x 10^8 CFU; LD_{50}) was administered intranasally (n= 8 mice/group)

Monovalent PPS PCMV Immunogenicity

- Memory, anamnestic, response demonstrated
- In opsonophagocytic assay (OPA), mouse sera elicited by PCMV had a comparable titer to sera induced by Prevnar7®
- Passive transfer of anti-PPS14 sera elicited by PCMV to mice confers 100% protection against intranasal challenge with serotype14 *S. pneumoniae* strain

MRD517/MRD1027 Polycations

- Matrivax proprietary invention; extends PCMV patent life through May 2032
- Facilitates PCMV formation
- Increased efficiency of polysaccharide (PS) entrapment into PCMV
- Increased immunopotency of PCMV

Pneumococcal PS-CRM197-MRD517 PCMV Murine Immunogenicity Data

Trivalent PCMV (PPS 4, 18C, 23F) administered IP induced IgG Antibody Titers

Groups	IgG GMT		
	Anti-PPS4	Anti-PPS18C	Anti-PPS23F
Batched Trivalent PCMV* (2 µg each PPS)	885,124	76,392	260
Pneumovax® (2 µg each PPS) (23 polysaccharide vaccine)	15	25	19
Prevnar 13® (2.2 µg each PPS**)	80,305	3,448	1,194
Naïve	15	10	13

* – PPS-CRM197-MRD517 PCMV with alum

** - 4.4 µg of PPS 6B

Results: PCMV superior to Prevnar 13® at eliciting IgG antibodies against 2 of 3 PPS

Pneumococcal PCMV Current/Next Steps

- Determine *S. pneumoniae* PPS bundle compatibility

- Introduce *S. pneumoniae* protective homologous matrix proteins

- Evaluate novel adjuvants (Adj-1 and Adj-2) for increased immunogenicity

- Evaluate PPS + protein antigen immunogenicity in mice and immunogenicity/efficacy in rabbits

Vi-CRM197 Typhoid Fever
PCMV

Typhoid fever vaccines

Vi polysaccharide vaccines

- Generated by fermentation of *S.* Typhi
  -Typhim Vi® (sanofi) and Typherix® (GSK)
- Single (25 µg) dose by injection
- Licensed for >2 year-olds
- Protective Efficacy ~70%
- No memory

Live attenuated vaccine

- Live, attenuated *S.* Typhi Ty2
  -Vivotif® (Crucell now PaxVax)
- 4 dose enteric coated capsules
- Protective Efficacy 53-78%

Typhax Immunogenicity in Mice

Groups	Dose (µg)	Adjuvant	Route of Imm.	Day 42 Anti-Vi IgG GMT
Typhax	0.05	Al-PO_4	IM	20,646
Typhim Vi®	0.05	NONE	IM	239
Naïve	--	NONE	IM	25

Results:

- Typhax elicits ~86-fold increase in anti-Vi antibody compared to Typhim Vi®

Typhax Preclinical Immunogenicity Summary

- Typhax was formulated at the indicated dose levels adjuvanted with 1 mg/mL of aluminum phosphate.
- Mice and rabbits were immunized intramuscularly (IM) three times at a biweekly interval.
- Non-human primates (NHP) were immunized IM twice at a four week interval.
- Blood was collected two weeks following last immunization and the anti-Vi IgG antibody titer in the sera was determined by ELISA.
- For mice and rabbits, the fold increase in anti-Vi antibody titer over Vi alone is reported. For NHP, the fold increase in titer relative to pre-immune sera is reported.

Animal Model	Typhax Dose Level	Fold increase in anti-Vi IgG antibody response
Mouse	0.05 µg	64
Rabbit	0.1 µg	2.4
	2.5 µg	28
	10 µg	34
NHP	2.5 µg	~20
	10 µg	~50

Murine Immunogenicity Study of Typhax adjuvanted with Adju-Phos®, Adj-1 or -2 compared to Typhim Vi®

Group	Vaccine	Dose (µg)	Adjuvant	Day -1	Day 21	Day 21 Fold over Typhim Vi®	Day 42	Day 42 Fold over Typhim Vi®
G1	Typhax	0.2	50 ug Al-PO_4	**25**	13,454	**21.8**	16,000	**15.4**
G2	Typhax	0.05	50 ug Al-PO_4	**25**	32,000	**87.2**	53,817	**61.7**
G3	Typhax	0.01	50 ug Al-PO_4	**25**	13,454	**420**	32,905	**658**
G4	Typhax	0.2	1 mg Adj-1	**25**	14,672	**23.8**	45,255	**43.6**
G5	Typhax	0.05	1 mg Adj-1	**25**	7,780	**21.2**	69,792	**80**
G6	Typhax	0.01	1 mg Adj-1	**25**	2,460	**76.9**	15,560	**311**
G7	Typhax	0.2	1 mg Adj-2	**25**	53,817	**87.2**	370,078	**357**
G8	Typhax	0.05	1 mg Adj-2	**25**	41,499	**113**	180,483	**207**
G9	Typhax	0.01	1 mg Adj-2	**25**	6,747	**211**	24,675	**494**
G10	Typhim Vi	0.2	--	**27**	617	--	1,037	--
G11	Typhim Vi	0.05	--	**25**	367	--	872	--
G12	Typhim Vi	0.01	--	**25**	32	--	50	--
G13	Naïve	--	--	**25**	25	--	30	--

- Balb/C mice immunized IM on day 0, 14, and 28
- Blood collection on Days -1 (pre-immune), 21, and 42
- 8 mice per group

CONCLUSIONS: Adjuvanting Typhax with Adj-2 results in more robust immune response and superior boosting capacity compared to adjuvanting with Adj-1 or Adju-Phos by day 42

Vi PCMV Adj-2 Rabbit Immunogenicity Study

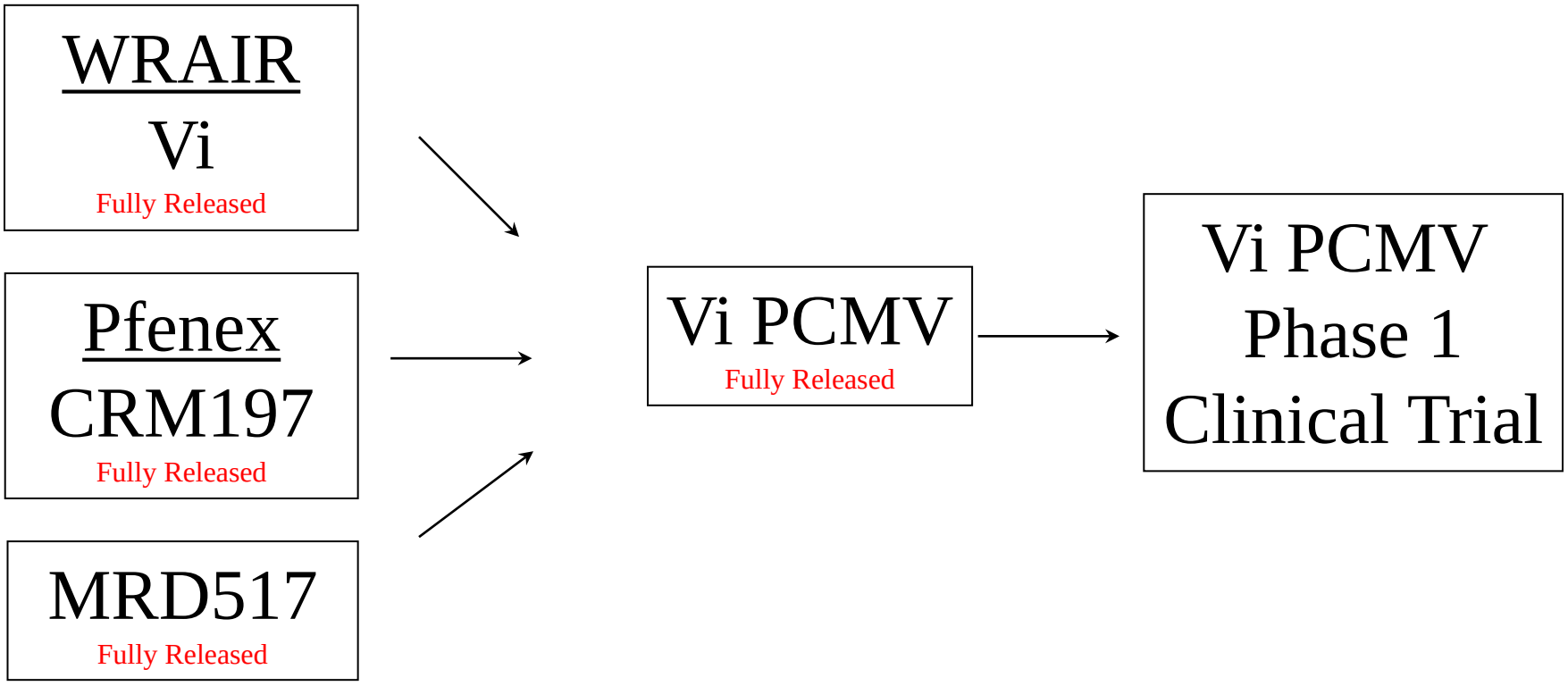
- Rabbit study initiated to test immunogenicity of Vi PCMV adjuvanted with Al-PO_4, Adj-1 and Adj-2
  - -Vi PCMV 261 synthesized at Matrivax using same Vi polysaccharide used in synthesis of Typhax
  - -3 biweekly immunizations
  - -Anti-Vi antibody titers determined in sera from blood collected on Day 7, 21 and Day 42 (SAC)
  - -Body temp, body weight and injection site monitored during study
    - No significant increase in body temp 3 or 24 hours post immunization
    - No change in body weight
    - No injection site reactivity observed

Group	Dose (µg)	Route of Imm.	Description	Adjuvant	No. of rabbits	Day 0	Day 21	Day 42
G6	5	IM	Vi PCMV 261	500 µg Al-PO_4	5	106	80	160
G7	5	IM	Vi PCMV 261	20 mg Adj-1	5	190	190	226
G8	5	IM	Vi PCMV 261	20 mg Adj-2	5	265	5072	3676
G9	25	IM	Typhim Vi	NONE	5	121	53	17

Vi PCMV DEVELOPMENT

Manufacturing

Clinical



GMP Vi PCMV Release Status

- GMP Vi PCMV synthesized March 2014
- Drug Product Vialed April 2014

  - GMP Vi PCMV **Fully Released**:

    -Passed Sterility assay

    -Passed Endotoxin content

    -Passed General Safety Test

    -Met Specifications for:

    - pH
    - Vi content
    - CRM197 content

  - Characterization of GMP Vi PCMV:

    -Residuals

    -Molecular size

    -% Crosslinking

    -Unincorporated Vi

    -Unincorporated CRM197

Vi PCMV (Typhax) Status/Pathway to Phase 1 Clinical Trial

- Typhax significantly more immunogenic than Typhim Vi® in murine, rabbit, and non-human primates studies

- Toxicology study
  - -No adverse findings
  - -Final report completed

- IND application submission 4Q2015

- Phase 1 safety and immunogenicity study *in progress*, initiated 1Q16

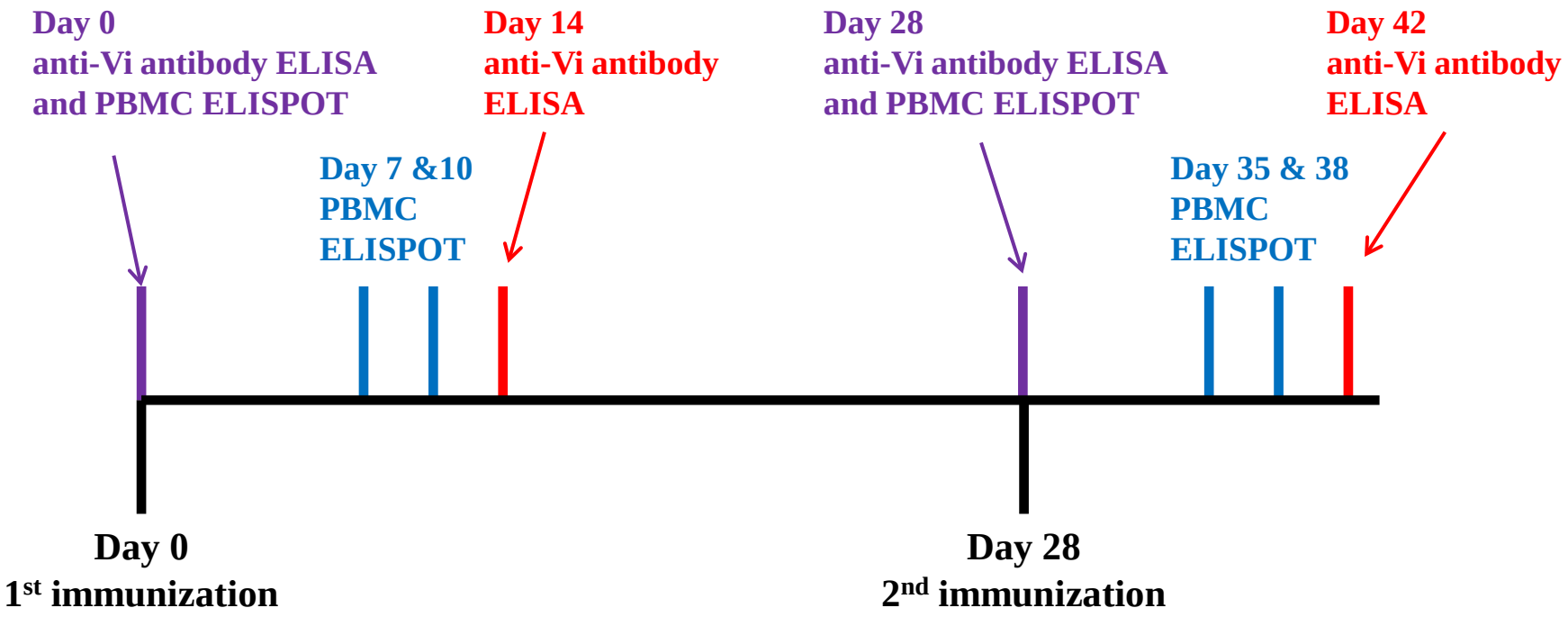
Vi PCMV Phase 1 Clinical Protocol Synopsis

- Outpatient, dose-escalation clinical trial
  - Inclusion – 18-55 year old healthy volunteers
  - Exclusion – elevated anti-Vi antibody titers
  - One site; n = 45 persons
  - Cohort size: n= 15/dose (3:1:1-Vi PCMV:Typhim Vi®:Placebo)
  - Prime-boost at day 0 and 28, IM, with 6 month follow up

- Dosing - 0.5, 2.5, and 10 µg Vi PCMV

- Key Endpoints
  - Safety (site of injection reactogenicity, etc)
  - Seroconversion (sera collected at day 0, 14, 28, 42, 180)
  - Vi-specific ELISPOT
  - Anti-Vi IgG antibody titers/comparison of Vi PCMV with Typhim Vi®
  - Phase 3 ‘like’ data; potential to avoid efficacy trial

Schedule of Typhax Vaccination and Assessments



Salmonella PCMV

- Utilize Vi PCMV as cornerstone
  - -integrate S. *paratyphi* O-antigen
  - -integrate *S. enteriditis* O-antigen
  - -integrate *S. typhimurium* O-antigen

- Create broad based Salmonella vaccine that protects against typhoid and paratyphoid fever and food poisoning

Summary

Vi PCMV (Typhax) Project

- Vi and CRM197 expression and purification know-how technology developed at Matrivax
- GMP manufacture and release of Vi, CRM197, and MRD517 completed in 2014
- IND application submitted December 2015
- Phase 1 clinical trial started 1Q16; adjuvanted study scheduled August 2016

Pneumococcal PCMV Project

- 3-, 4-, 13-, and 23-valent PPS PCMV elicits dramatic increase in anti-PPS antibody titers compared to PPS alone; 6 of 10 PPS PCMV were immunogenically superior to Prevnar13®
- Simplified PCMV manufacturing process will enable lower COGs and inclusion of additional serotypes; project funded by BMGF
- Proprietary protein antigen evaluation underway; supplement to PS PCMV

***C. difficile* PCMV Project**

- Proprietary antigen evaluation underway
- Key immunogenicity and protection data anticipated in 2016