10TH INTERNATIONAL
CONFERENCE ON TYPHOID
& OTHER INVASIVE SALMONELLOSES
APRIL 4 - 6, 2017 | KAMPALA, UGANDA

Pre-clinical Immunogenicity of Typhoid (Vi-CRM_{197}), Paratyphoid A (O:2- CRM_{197}) and Bivalent (Vi-CRM_{197}+O:2-CRM_{197}) Conjugate vaccine

Presented by

Ravi P.N. Mishra, Ph.D.
Biological E. Limited
INDIA
05-04-2017

Biological E. Limited
Celebrating Life Everyday

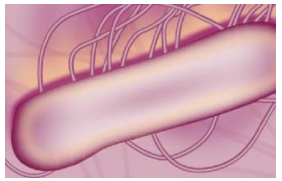
Summary of Presentation:

Biological E. Limited

- Background: Disease epidemiology and history
- Review on pre-clinical immunogenicity models used for Typhoid & Paratyphoid A vaccines
- Biological E approach to develop Bivalent (Typhoid and Paratyphoid A) conjugate vaccine
- Preclinical Immunogenicity of Bivalent TCV:
  - Immunogenicity of Bivalent Vaccine in Mice
  - Immunogenicity of Bivalent Vaccine in Rabbit
- Current status of the Bivalent conjugate Vaccine project
- Conclusion and future perspective

Salmonella Paratyphi A *: Enteric Fever*

Disease burden, rationale Paratyphi A *vaccine*



Biological E. Limited

❑ Enteric Fever

- > 27,000,000 cases in 2000
- *S.* Paratyphi A *& S.Typhi* cause similar disease
- 1:1 ratio of *S.* Typhi and *S.* Paratyphi A in some countries
- Peak age of disease 1-3 years

❑ Second leading cause of enteric fever after *S. Typhi*

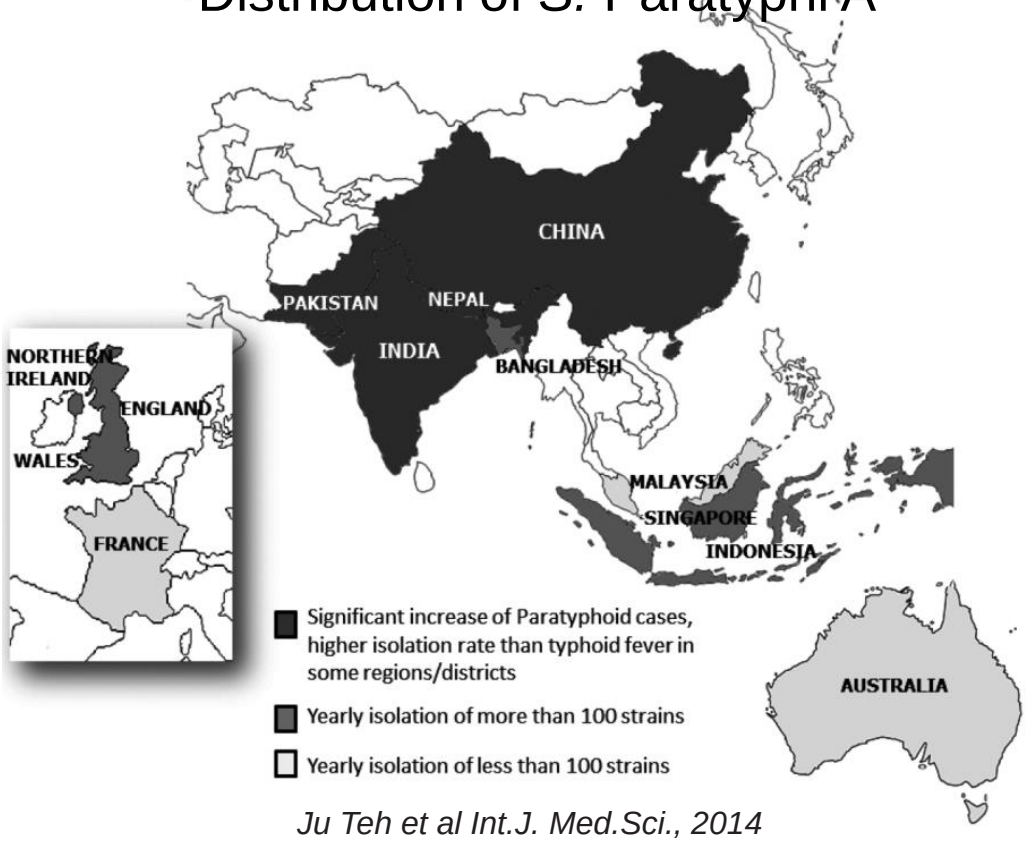
❑ Emerging Antimicrobial Resistance:

- Resistance to First-line drugs : fluoroquinolones, and third generation cephalosporins, posing therapeutic challenges.

❑ No vaccines for

- *S.* Paratyphi A (Paratyphoid Fever)
- No correlate of protection

Distribution of *S.* Paratyphi A

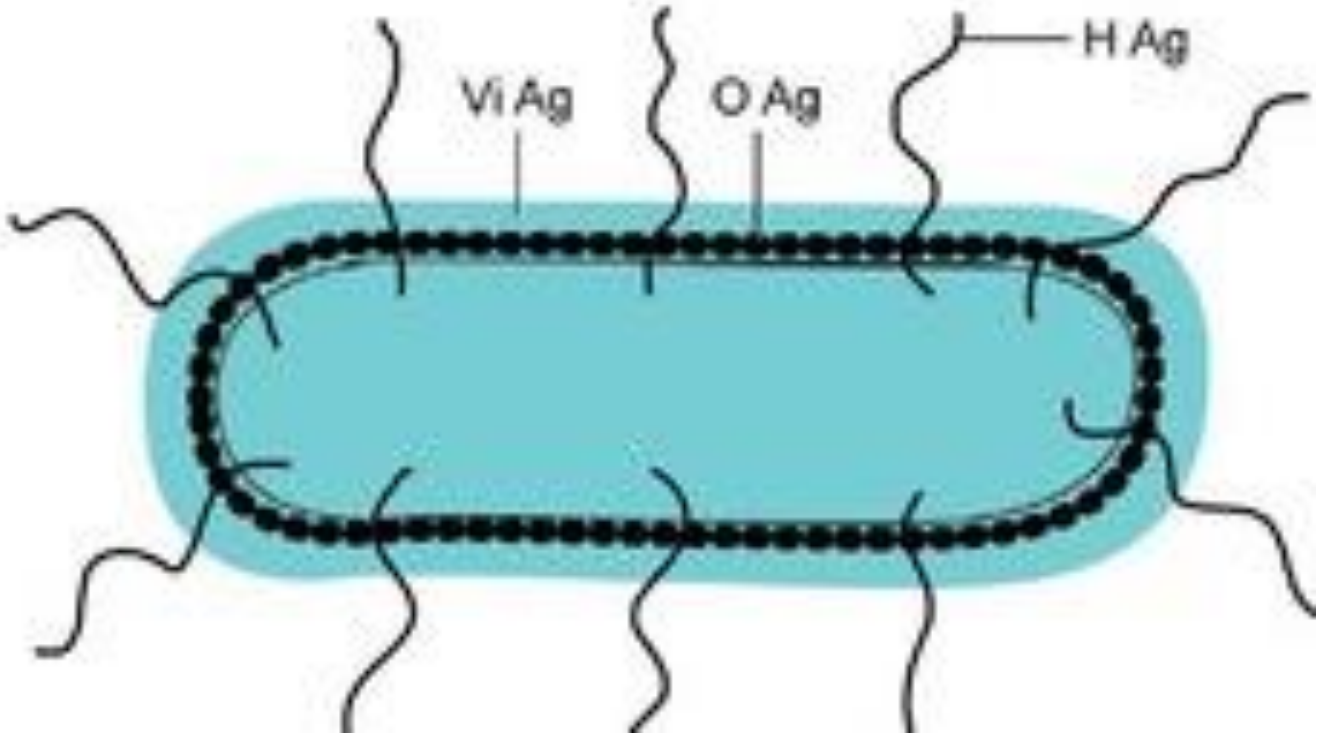


Ju Teh et al Int.J. Med.Sci., 2014

- **A bivalent conjugate vaccine for Typhoid-Paratyphoid would be ideal !**

Antigenic Structure of Salmonella

Biological E. Limited



- **Vi Antigen**: Surface polysaccharide
- **O Antigen**: Somatic antigen, Part of LPS
- **H-Antigen**: Flagellar antigen

Vi and O antigen were shown to be important virulence determinant of Salmonella and therefor exploited as Immunogen for vaccine development

Vi antigen as Vaccine: Review of Prior works

Biological E. Limited

Szu et al., 1994

INFECTION AND IMMUNITY, Oct. 1994, p. 4440–4444
0019-9567/94/$04.00+0

Vol. 62, No. 10

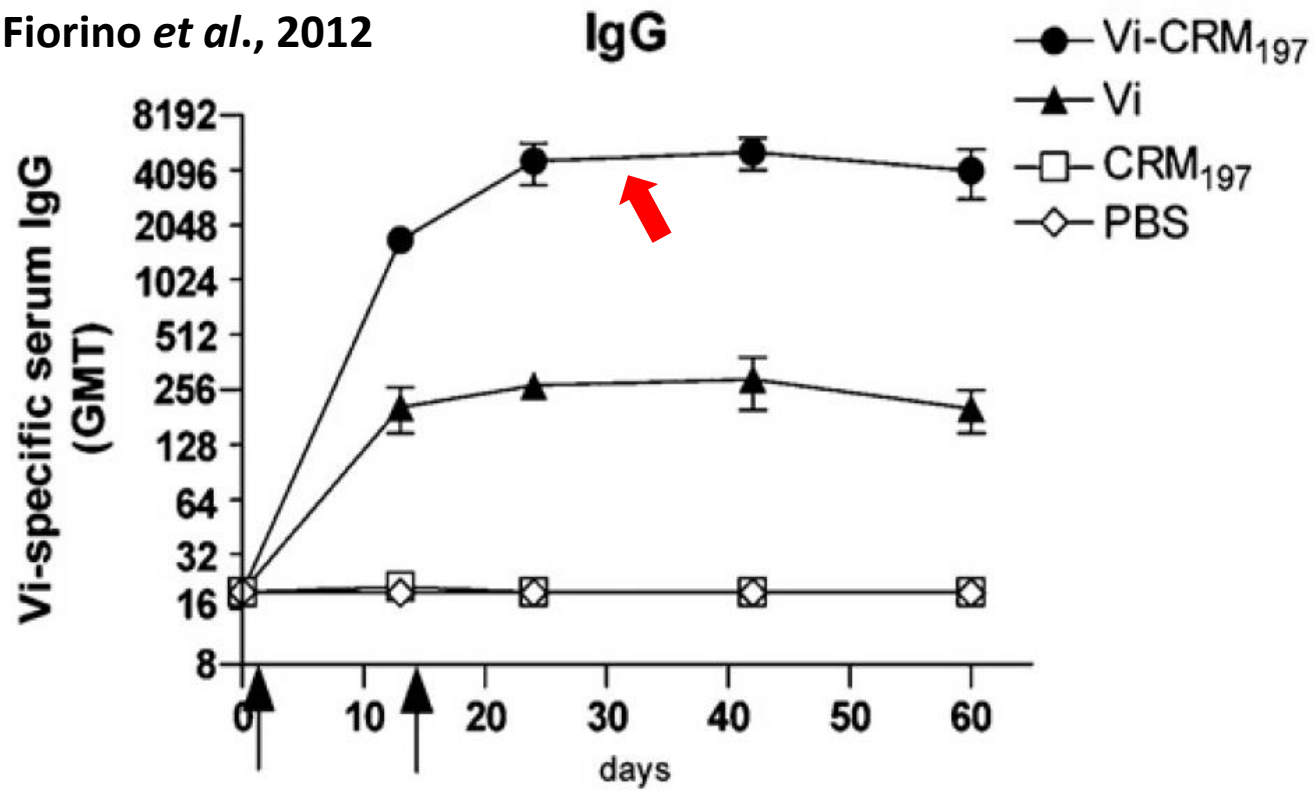
Laboratory and Preliminary Clinical Characterization of Vi Capsular Polysaccharide-Protein Conjugate Vaccines

SHOUSUN C. SZU,[1]* DAVID N. TAYLOR,[2] ANDREW C. TROFA,[2] JOHN D. CLEMENTS,[3] JOSEPH SHILOACH,[4] JERALD C. SADOFF,[2] DOLORES A. BRYLA,[1] AND JOHN B. ROBBINS[1]

TABLE 2. Vi antibody levels in sera of female mice injected subcutaneously with Vi alone or as a conjugate[a]

Mice and vaccine (lot no.)	GM antibody concn[b] (μg/ml)		
	1st injection	2nd injection	3rd injection
BALB/c			
Vi (104a)	1.06	1.31	ND
Vi–LT-B (50860)	3.62	6.87	ND
Vi-rEPA (51706)	0.85	17.1	12.7

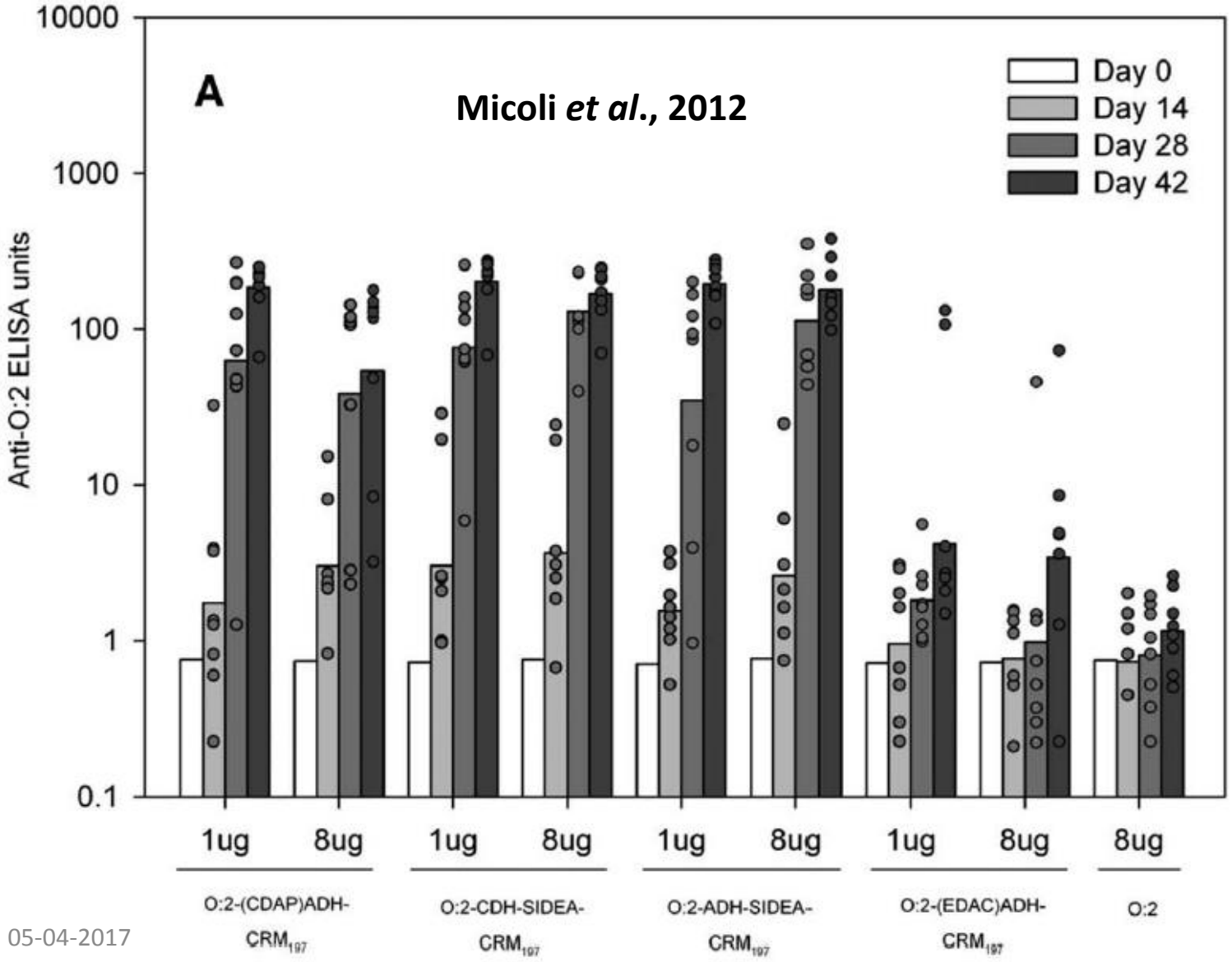
Fiorino *et al.*, 2012



Immunization: Day 0 Day 14

- Various type of Typhoid conjugate vaccines were tested in preclinical models and found to be immunogenic
- Confirms the potential of Vi antigen as vaccine for S. typhi

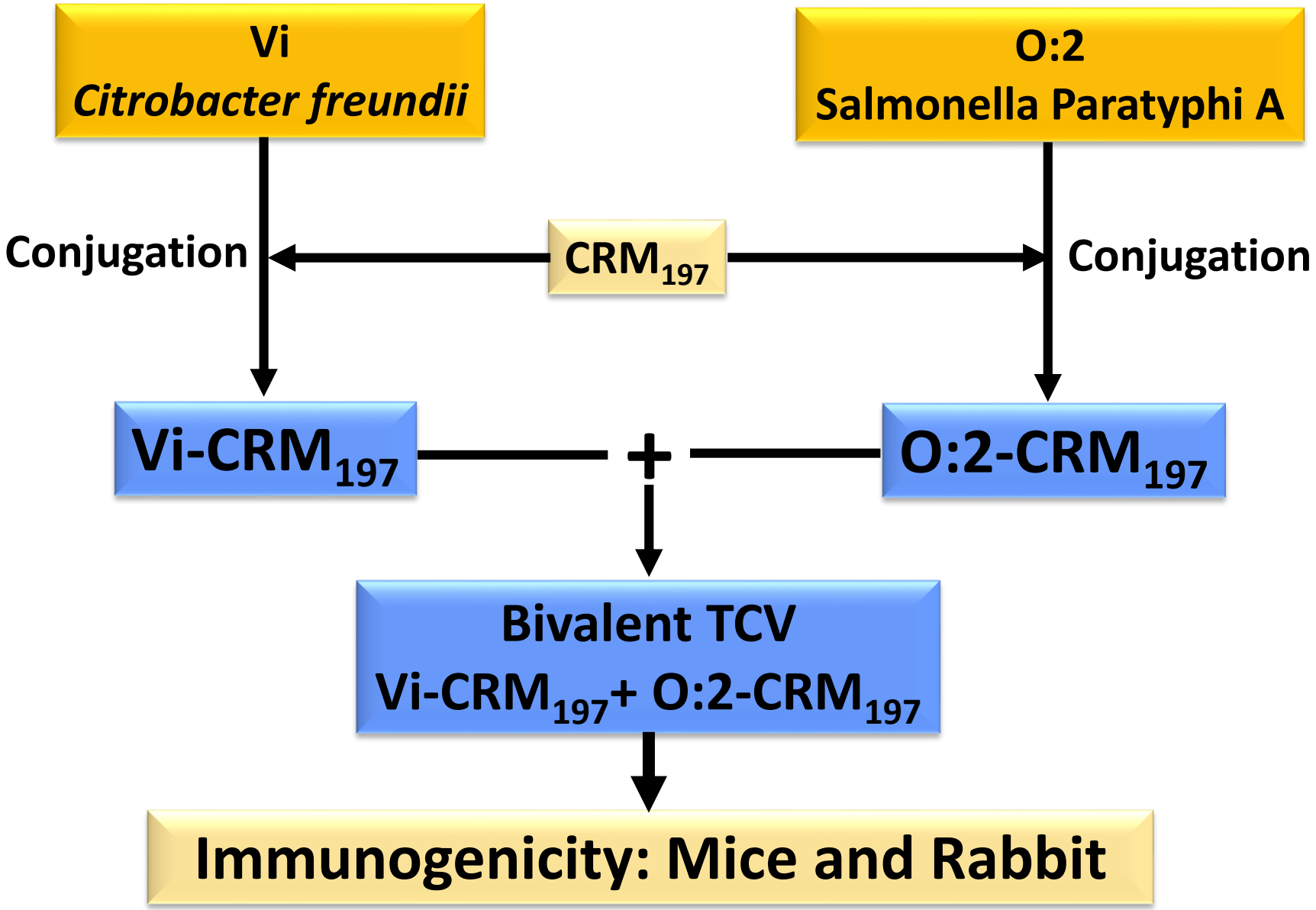
O:2 antigen as Vaccine: Review of Prior works



- Various type of O:2-CRM conjugates were tested in Mice and found to be immunogenic
- Confirms the potential of O antigen as vaccine for S. paratyphi A

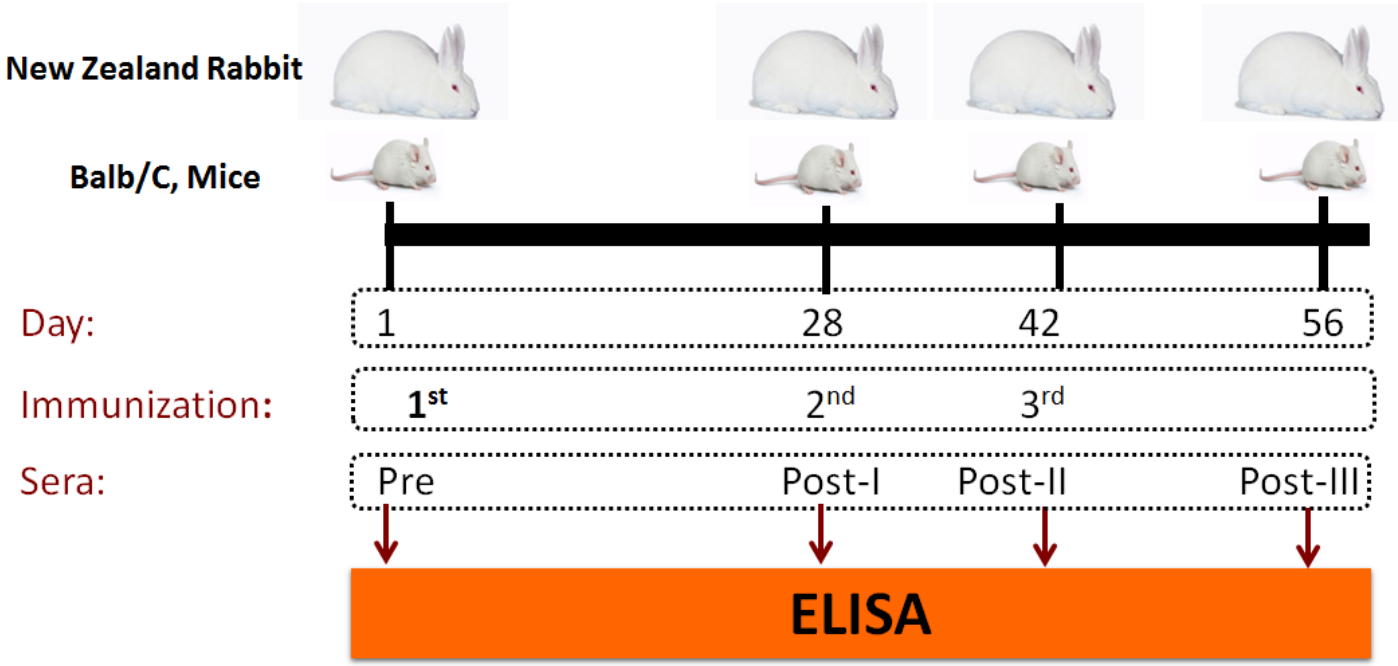
Biological E Approach to develop a Bivalent vaccine to combat Typhoid & Paratyphoid

Biological E. Limited



Mice and Rabbit model for Bivalent Pre-clinical Immunogenicity

Biological E. Limited



Route of immunization : Subcutaneous (SC)
Dose of Vaccine:
- **Mice: 1/10 SHD**
- **Rabbit: SHD**

Vaccines used for immunization

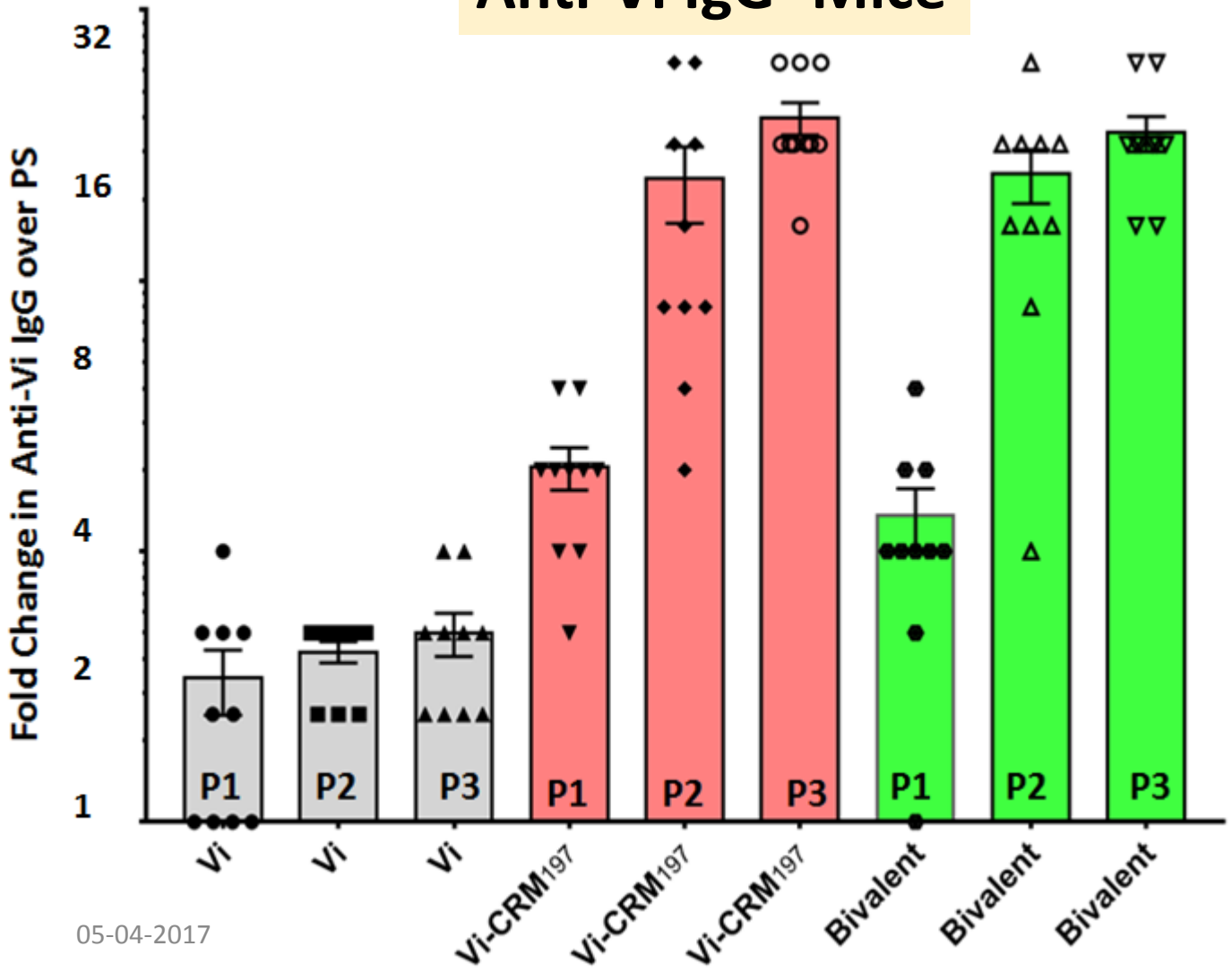
Groups	Vaccine
1	Bivalent (Vi-CRM_{197}+O:2-CRM_{197})
2	Vi-CRM_{197}
3	O:2-CRM_{197}
4	Vi (unconjugated)
5	O:2 (Unconjugated)
6	PBS

Anti-Vi IgG was measured in the groups with Vi and Anti-O:2 IgG response was evaluated in O:2 containing groups

Immunogenicity of Bivalent (Vi-CRM$_{197}$ + O:2-CRM$_{197}$) vaccine in Balb/C Mice: Anti-Vi IgG response

Biological E. Limited

Anti-Vi IgG -Mice

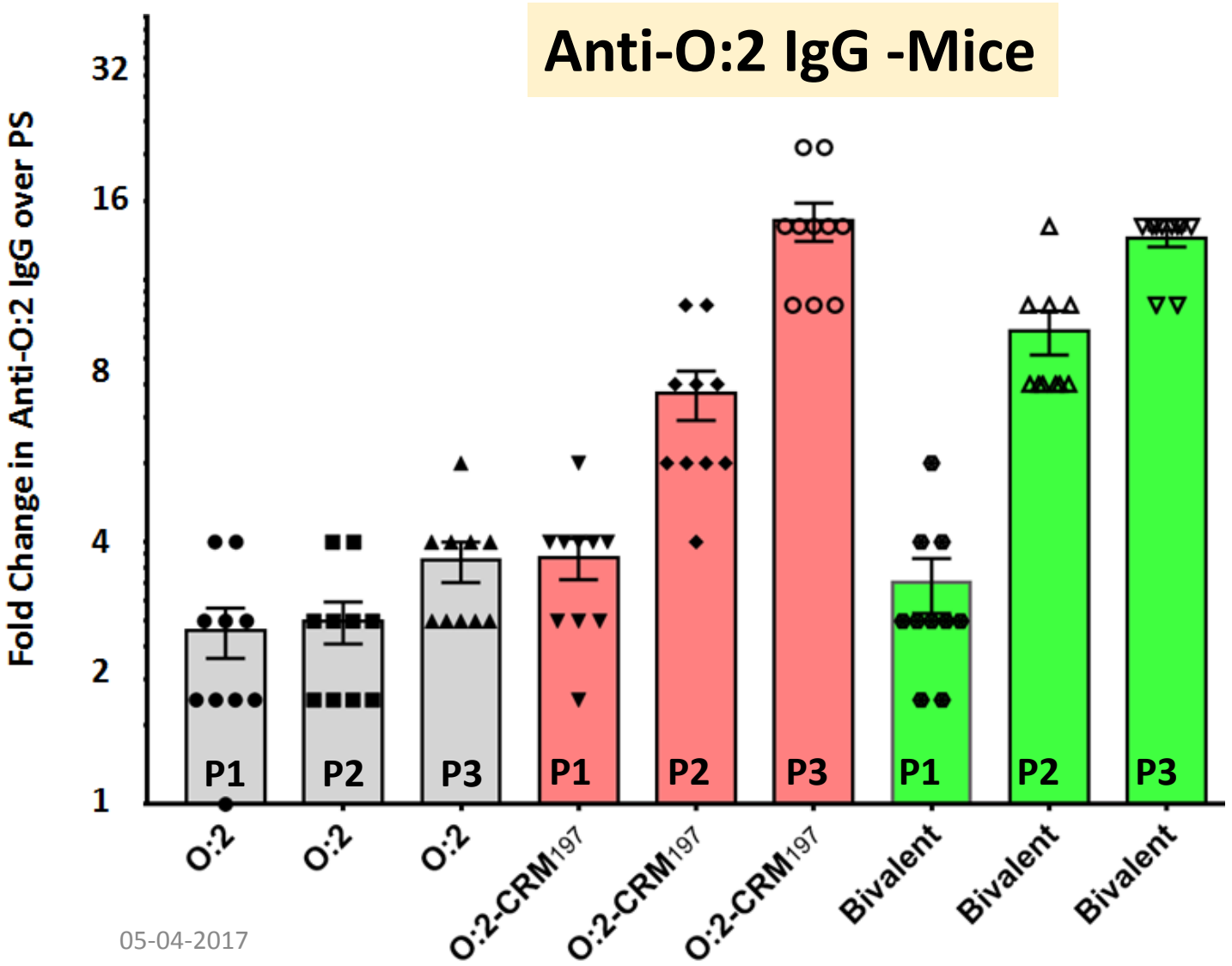


P1: Post Dose 1 (D28)
P2: Post Dose 2 (D42)
P3: Post Dose 3 (D56)

- A significant increase in the Anti-Vi antibody observed in monovalent conjugate (Vi-CRM$_{197}$) and Bivalent group when compared with group immunized with only Vi.
- Following the second injection, a significant secondary antibody response has been observed in Both Monovalent and Bivalent vaccines

Immunogenicity of Bivalent (Vi-CRM$_{197}$ + O:2-CRM$_{197}$) vaccine in Balb/C Mice: Anti-O:2 IgG response

Biological E. Limited

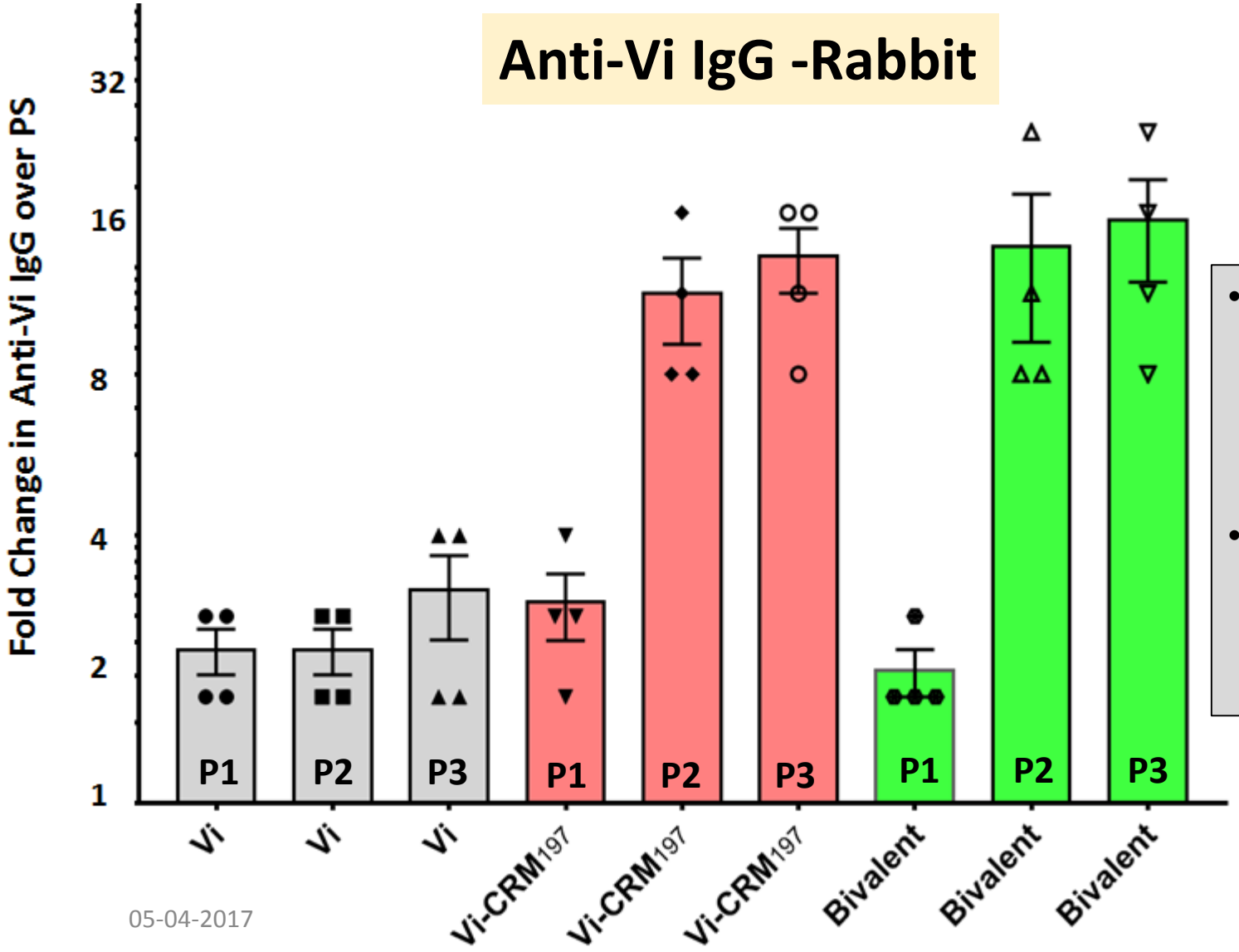


P1: Post Dose 1 (D28)
P2: Post Dose 2 (D42)
P3: Post Dose 3 (D56)

- A significant increase in the Anti-O:2 antibody observed in monovalent conjugate (O:2-CRM$_{197}$) and Bivalent group when compared with group immunized with only O:2.
- Following the second injection, a significant secondary Antibody response has been observed in Both Monovalent and Bivalent vaccines

Immunogenicity of Bivalent (Vi-CRM_{197} + O:2-CRM_{197}) vaccine in Rabbits: Anti-Vi IgG response

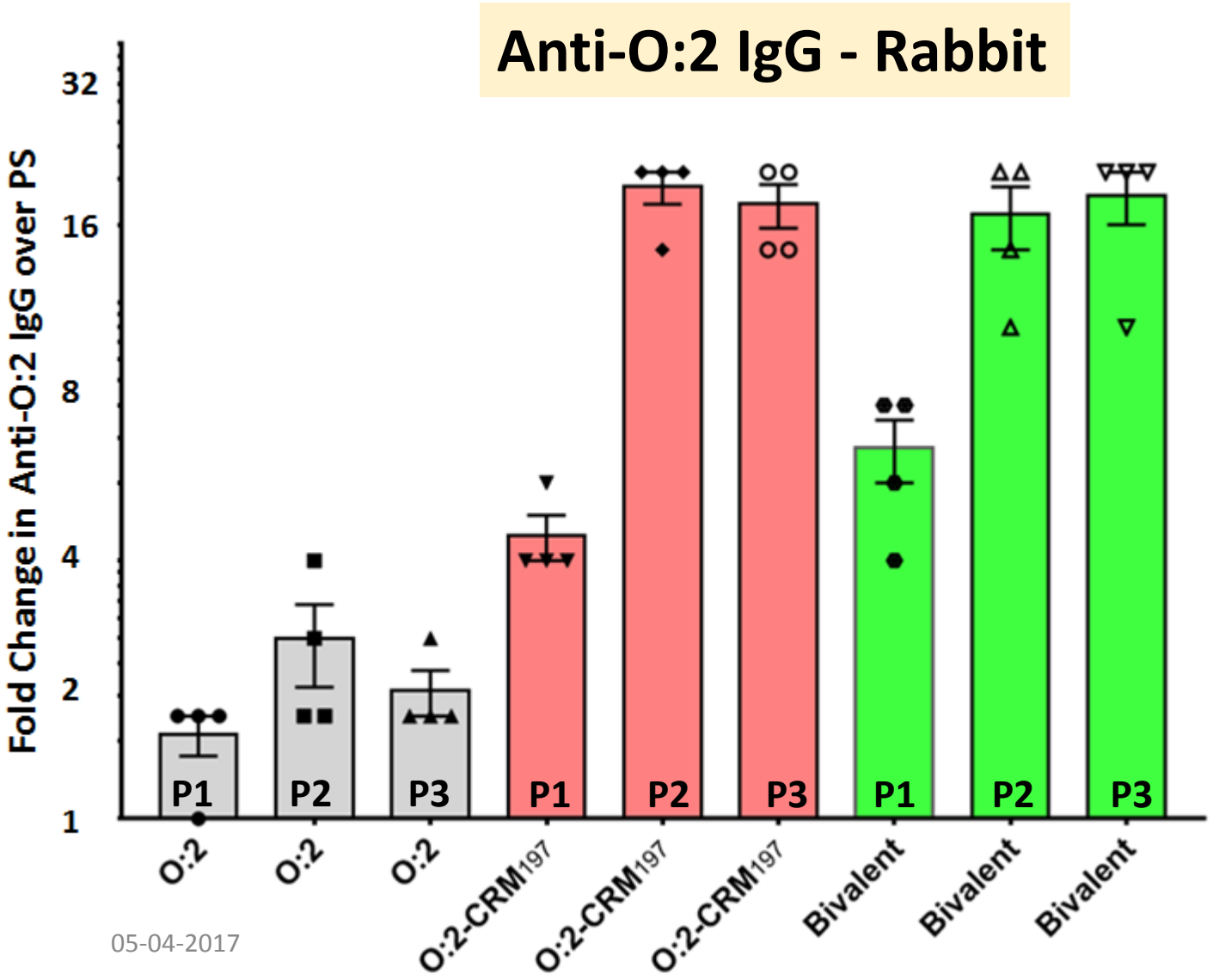
Biological E. Limited



P1: Post Dose 1 (D28)
P2: Post Dose 2 (D42)
P3: Post Dose 3 (D56)

- A significant increase in the Anti-Vi antibody observed in monovalent conjugate (Vi-CRM_{197}) and Bivalent group when compared with group immunized with only Vi.
- Following the second injection, a significant secondary Antibody response has been observed in Both Monovalent and Bivalent vaccines

Immunogenicity of Bivalent (Vi-CRM$_{197}$ + O:2-CRM$_{197}$) vaccine in Rabbits: Anti-O:2 IgG response



P1: Post Dose 1 (D28)
P2: Post Dose 2 (D42)
P3: Post Dose 3 (D56)

- A significant increase in the Anti-O:2 antibody observed in monovalent conjugate (O:2-CRM$_{197}$) and Bivalent group when compared with group immunized with only O:2.
- Following the second injection, a significant secondary Antibody response has been observed in Both Monovalent and Bivalent vaccines

Conclusion and Future Perspectives

Biological E. Limited

- A Novel Bivalent conjugate vaccine containing Vi and O:2 polysaccharide was developed and found to be immunogenic in Mice and Rabbits
- No interference between Anti-Vi and Anti-O:2 antibody was observed when Vi-CRM and O:2-CRM conjugate vaccine administered together in Bivalent vaccine
- The Pre-Clinical Toxicological (PCT) study is planned and to be executed to evaluate the safety parameters of Bivalent vaccine

Acknowledgements:

- The Vaccine Technical Development Team, Biological E.

- Biological E. management for their support

- Biological E. Bivalent vaccine project is a part of technology transfer from GSK Vaccines Institute for Global Health (GVGH)

- Sabin Institute and Coalition against Typhoid (CaT) for Travel Award to attend the conference