IMMUNE RESPONSE AND PROTECTION AGAINST ROTAVIRUS-LESSONS FROM BIRTH COHORT STUDIES

Gagandeep Kang

Christian Medical College, Vellore

Outline

- Description of birth cohorts so far
- Protection from disease in birth cohorts
- Immune responses in birth cohorts
- Implications for vaccine induced protection

Birth cohorts so far

Not including cohorts identified in nurseries, vaccination studies, followed up by required clinic visits

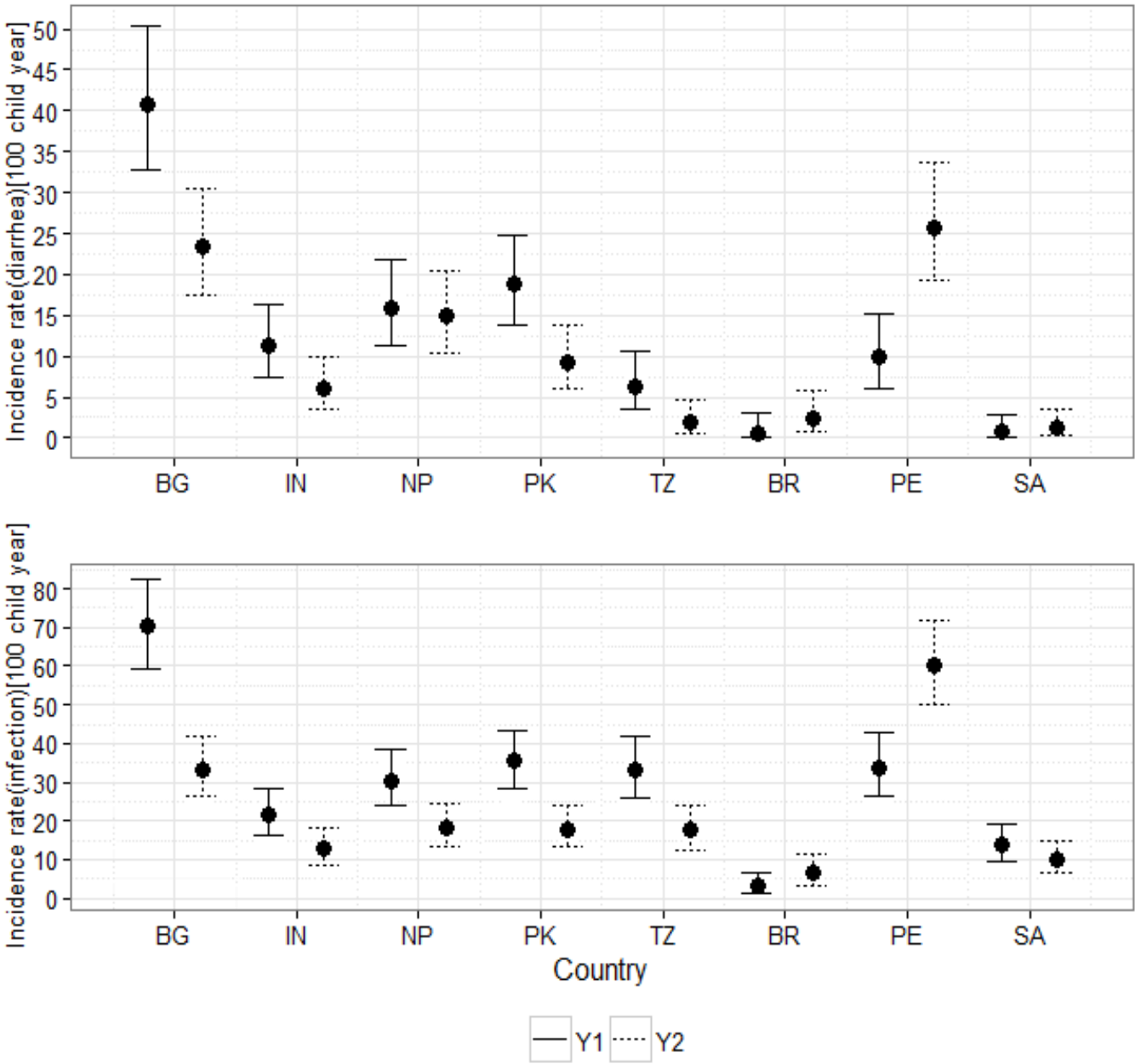
COMPARISON OF BIRTH COHORTS	Mexico City (2 years) Velazquez, NEJM 1996	Guinea-Bissau (2 years) Fischer JID 2002	Vellore, India (3 years) Gladstone NEJM 2011
No. of children	200, 77% follow up	200, 49% follow up	452, 373 with 99% follow up
Frequency of visits and stool	1/wk, 1/wk + diarr (15,503)	1/wk, 1/wk + diarr (11,406)	2/wk, 1 in 2 weeks + diarr (31,661)
Serum	Birth and 4 months (1080)	None	Birth and 6 months (2468)
Infections identified	316, 57% stool and 77% serology	116, by stool only	1103, 48% stool and 76% serology
Primary infections	52%	81%	30%
Proportion infected by 6 months	34%	26%	53%
Protection from infection	1 38% 2 60% 3 66%	1 52%	1 39% 2 52% 3 67%
Protection from disease	1 87% 2 100%	1 70%	1 18% 2 57%

All cohorts are not equal

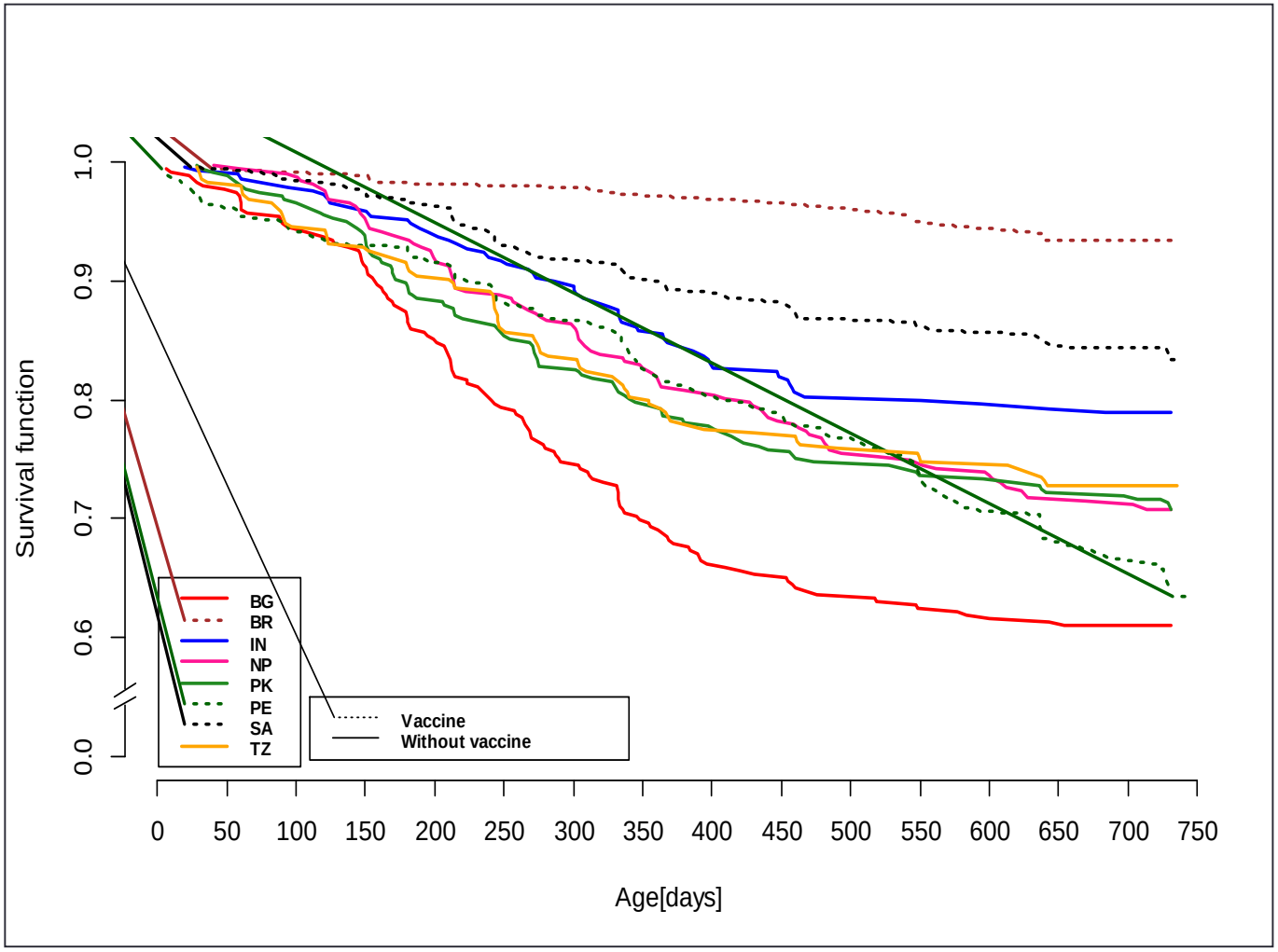
- Study designs
  - Follow up
  - Sampling
  - Laboratory methods
  - Analysis

BUT WHAT IF THEY WERE?

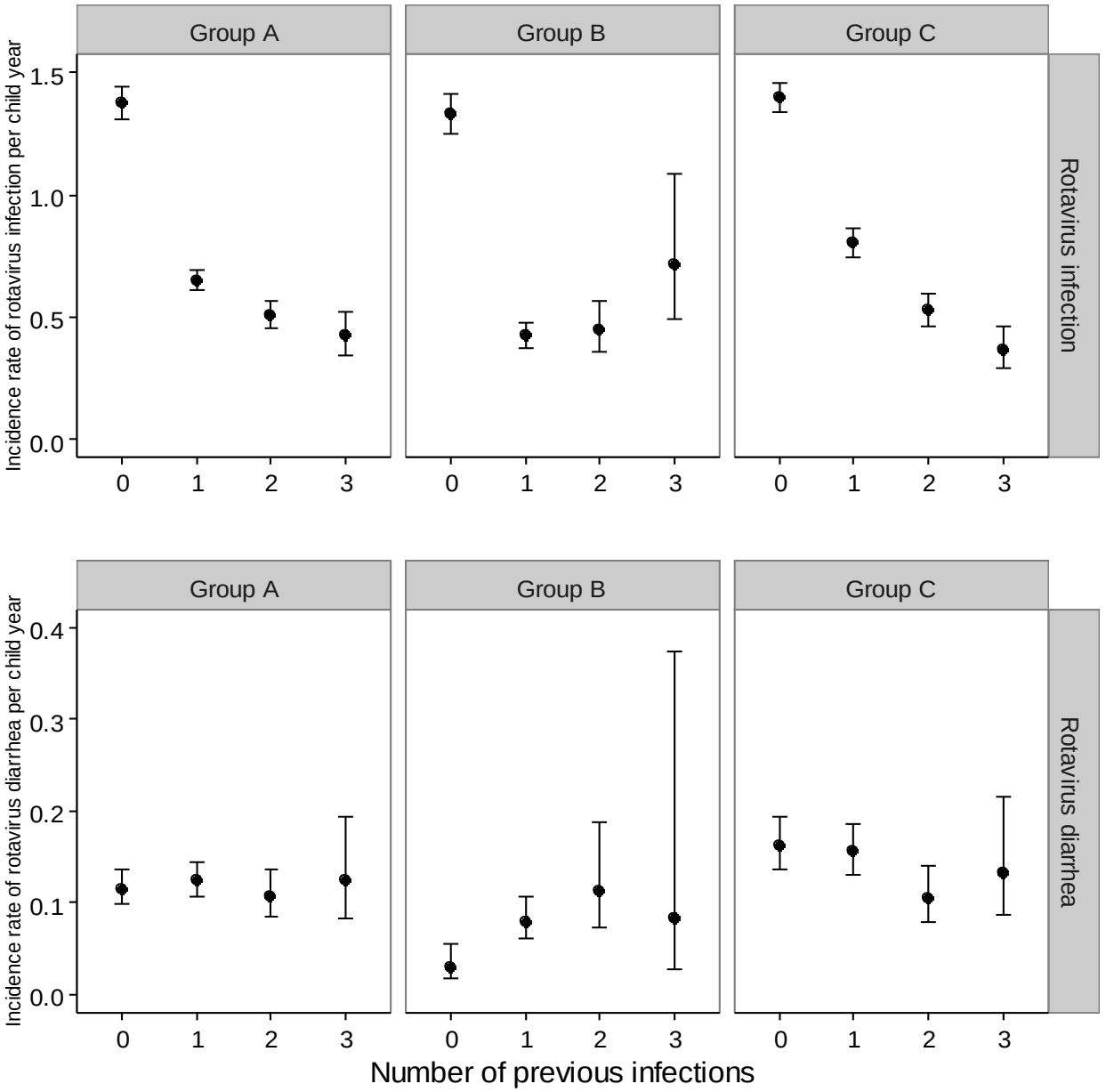
MAL-ED rotavirus infection and diarrhoea in 8 countries in the first two years of life



MAL-ED first infections



MAL-ED infection and diarrhoea following prior infections



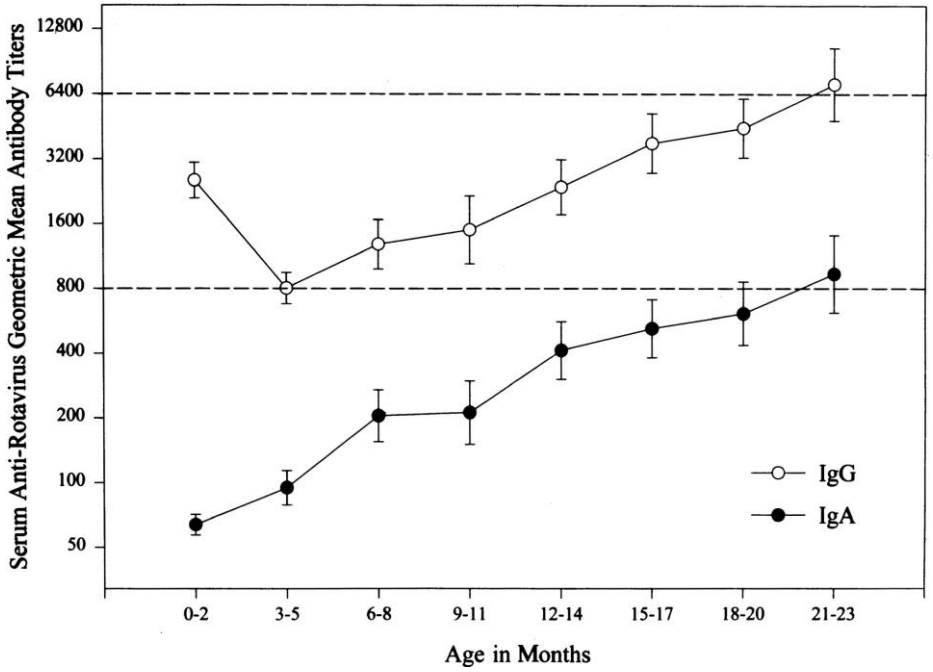
A-all countries
B-countries with vaccine
C-countries without vaccine

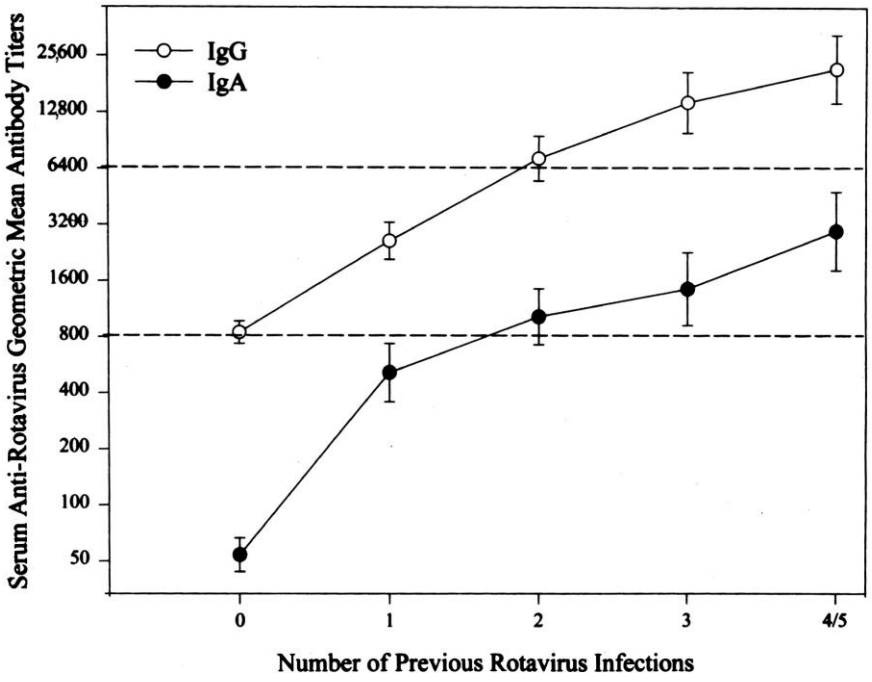
	Mexico City (2 years)	Guinea-Bissau (2 years)	Vellore, India (3 years)	MAL-ED (2 years)
Frequency of visits and stool	1/wk, 1/wk + diarr (15,503)	1/wk, 1/wk + diarr (11,406)	2/wk, 1 in 2 weeks + diarr (31,661)	2/wk, 1/month +diarr (30,086)
Serum	Birth and 4 months (1080)	None	Birth and 6 months (2468)	7 and 15 months
Infections identified	316, 57% stool and 77% serology	116, by stool only	1103, 48% stool and 76% serology	892 by stool, 91% infected by serology at 7 months
Primary infections	52%	81%	30%	80%
Proportion infected by 6 months	34%	26%	53%	12.5%
Protection from infection	1 38% 2 60% 3 66%	1 52%	1 39% 2 52% 3 67%	1 43% 2 67% 3 74%
Protection from disease	1 87% 2 100%	1 70%	1 18% 2 57% 3 79%	1 4% 2 36% 3 18%

Immune responses in birth cohorts

- Mexico
- India

Antibody estimations in birth cohorts-Mexico





IgA titer >1:800

79% against any rotavirus infection, 81% against asymptomatic infections, and 84% against any rotavirus-associated diarrhoea (82% against mild diarrhoea and 100% against moderate-to-severe diarrhoea)

IgG titer >1:6400

49% against any rotavirus infection, 40% against asymptomatic infection, 46% against rotavirus-associated diarrohea, and 71% against moderate-to-severe diarrhoea

F. Raul Velazquez et al. J Infect Dis. 2000;182:1602-1609

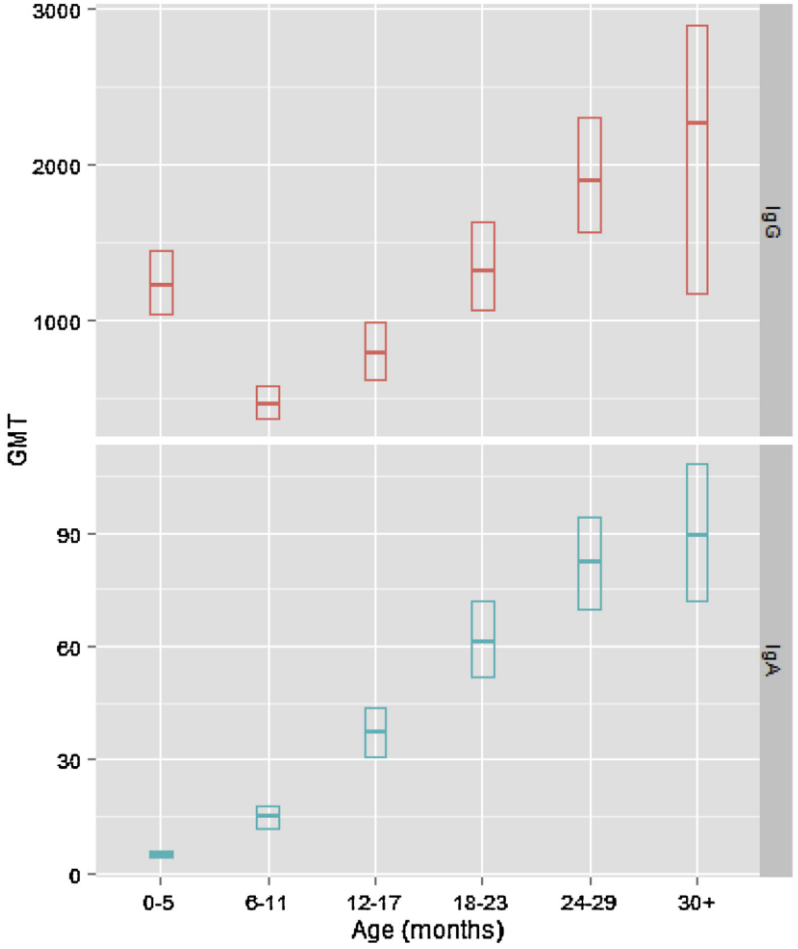
Rotavirus infection and IgA estimation in natural infection in India

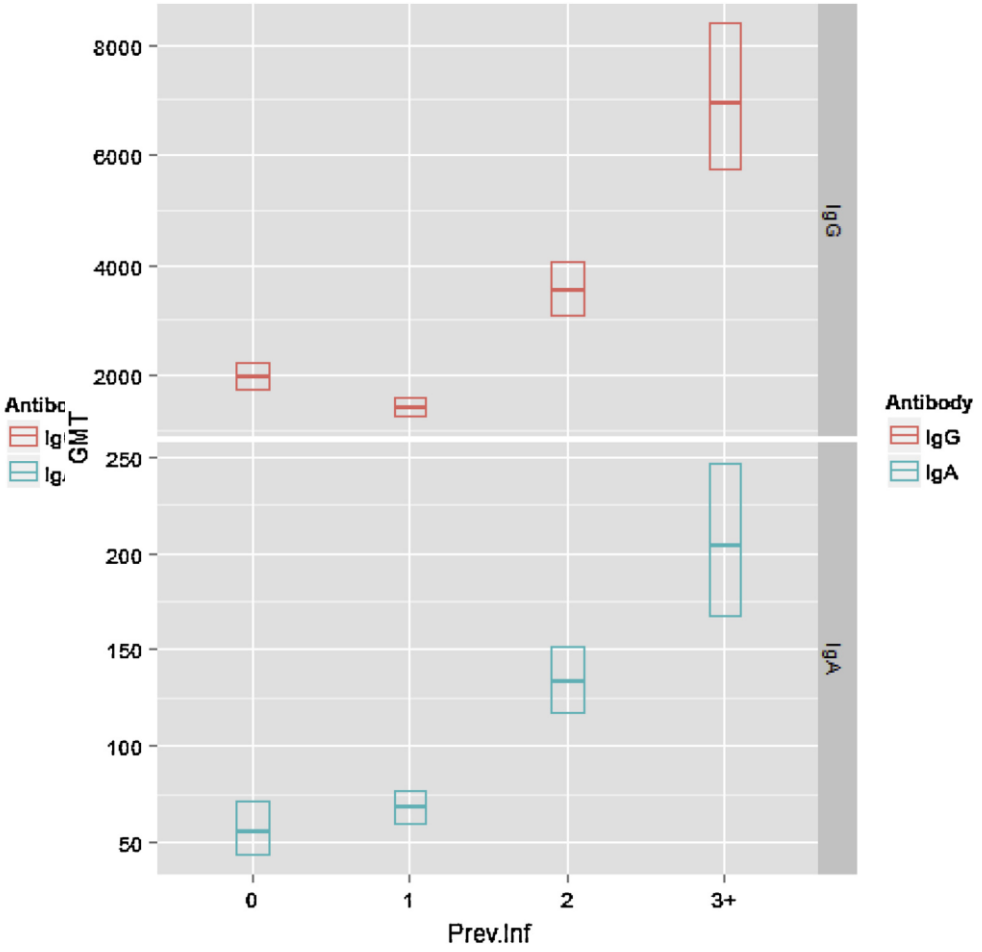
Natural Infection	
No. of children who completed follow up	373
No. of children who ever seroconverted	356 (95.4%)
No. of children with four fold change > 20 units	342 (91.7%)
No. episodes with four fold conversion	1580
No. episodes with four fold conversion > 20 units	1337 (84.6%)

IgA responses in natural infection

Four fold change	**1st increase (n=342)**	**2nd Increase (n =152)**	**3rd Increase (n =24)**	**4th Increase (n =2)**
Median (IQR) Pre-infection IgA	**0** (0 - 0)	**52.9** (20.8 - 137.3)	**120.1** (54 - 199)	**530.3** (299 .6 - 761.1)
Median (IQR) Post-infection IgA	**105** (48.6 -409)	**607** (211.7 - 2012.2)	**1295.8** (718.6 - 2987.3)	**2883** (1673 - 4093.2)
	P < 0.0001	P < 0.0001	P < 0.0001	p = 0.17

Antibody estimations in birth cohorts-India





Premkumar et al, Vaccine 2014 32S:A55-61

Antibodies and protection in India

- Classified by IgG and IgA deciles
  - Rotavirus infection was reduced by 72% (95% CI: 58–81%) among children with IgG values > 20,818 compared with those with values ≤100
  - Rotavirus infection was reduced by 68% (95% CI:54-77%) among children with IgA values > 619 compared with those with values of 0
- No significant relationships between IgA and IgG antibody titers with rotavirus disease

RV genes chosen for protein expression based on identification of prevalent strains

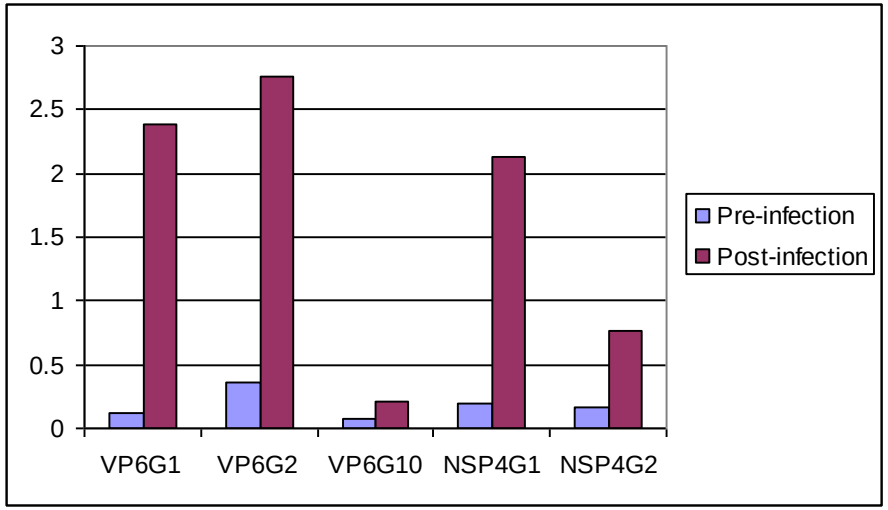
	G1P[8]	G2P[4]	G9P[8]	G10P[11]
VP4				
VP6				
VP7				
NSP4				

Total -12 genes chosen for protein expression

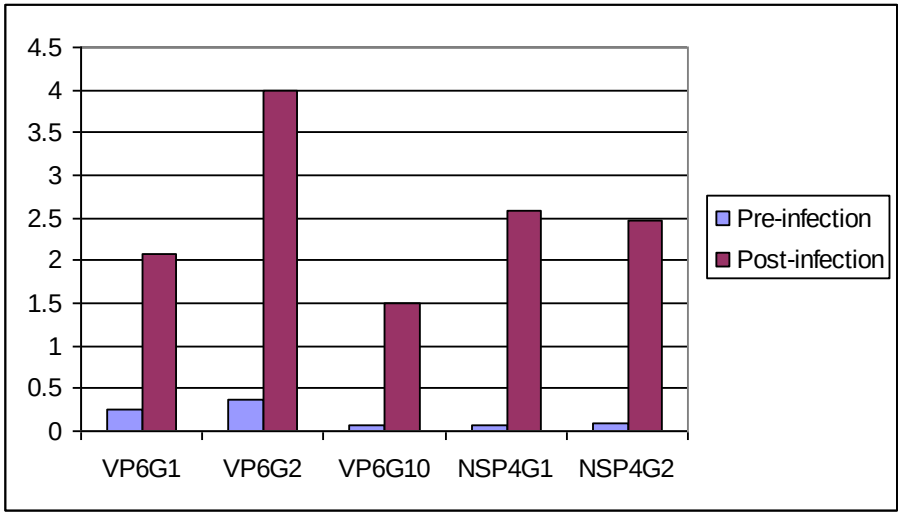
Proteins expressed using a baculovirus expression system in insect cells, purified and characterized

Immunoassays established in EIA formats

Results from Pre- and Post- RV infection sera
VP6 & NSP4 IgG Assay



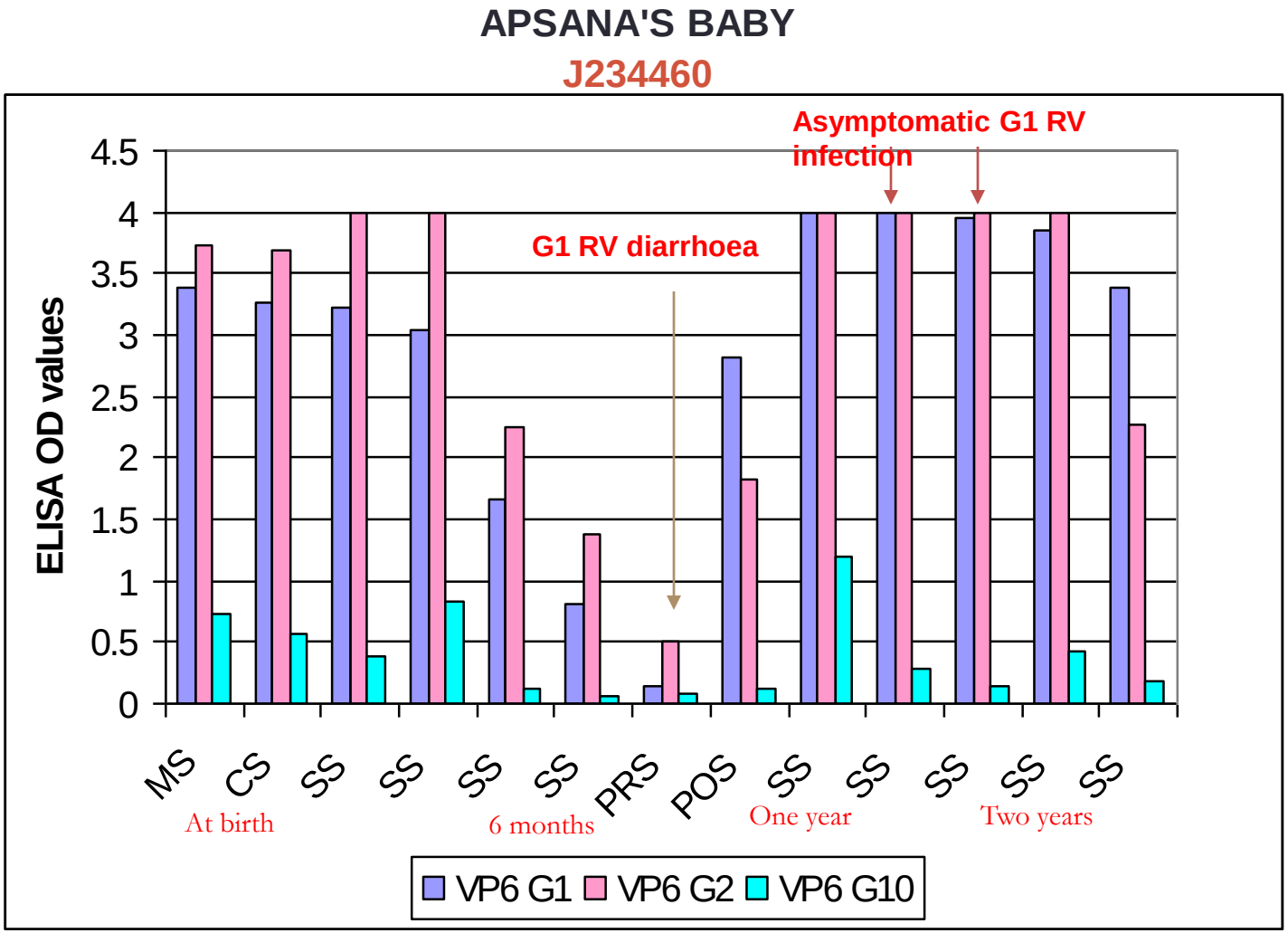
Child 1: G1P[8]



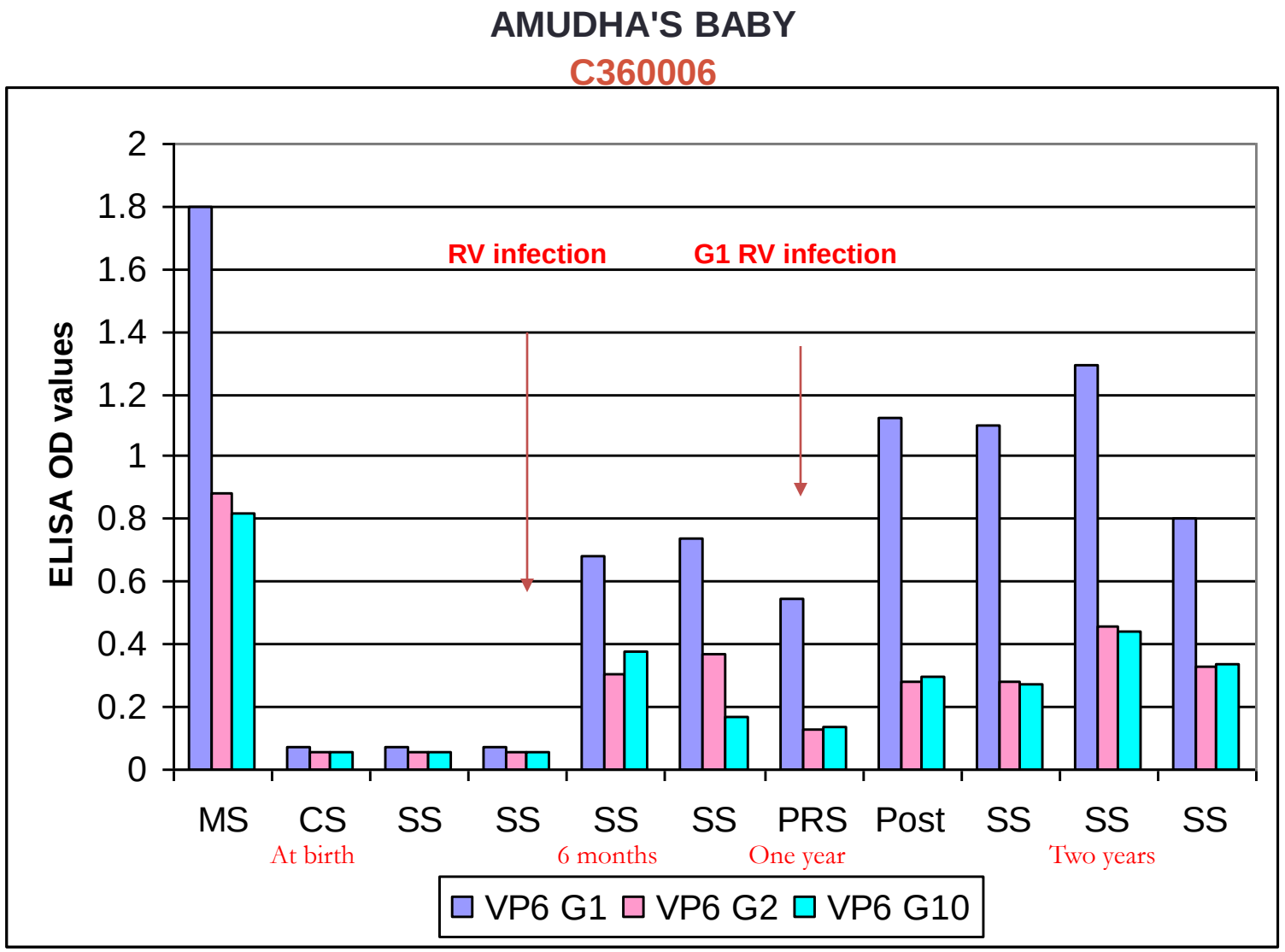
Child 2: G1+G2+G9P[8]

Longitudinal follow up of study subject J234460

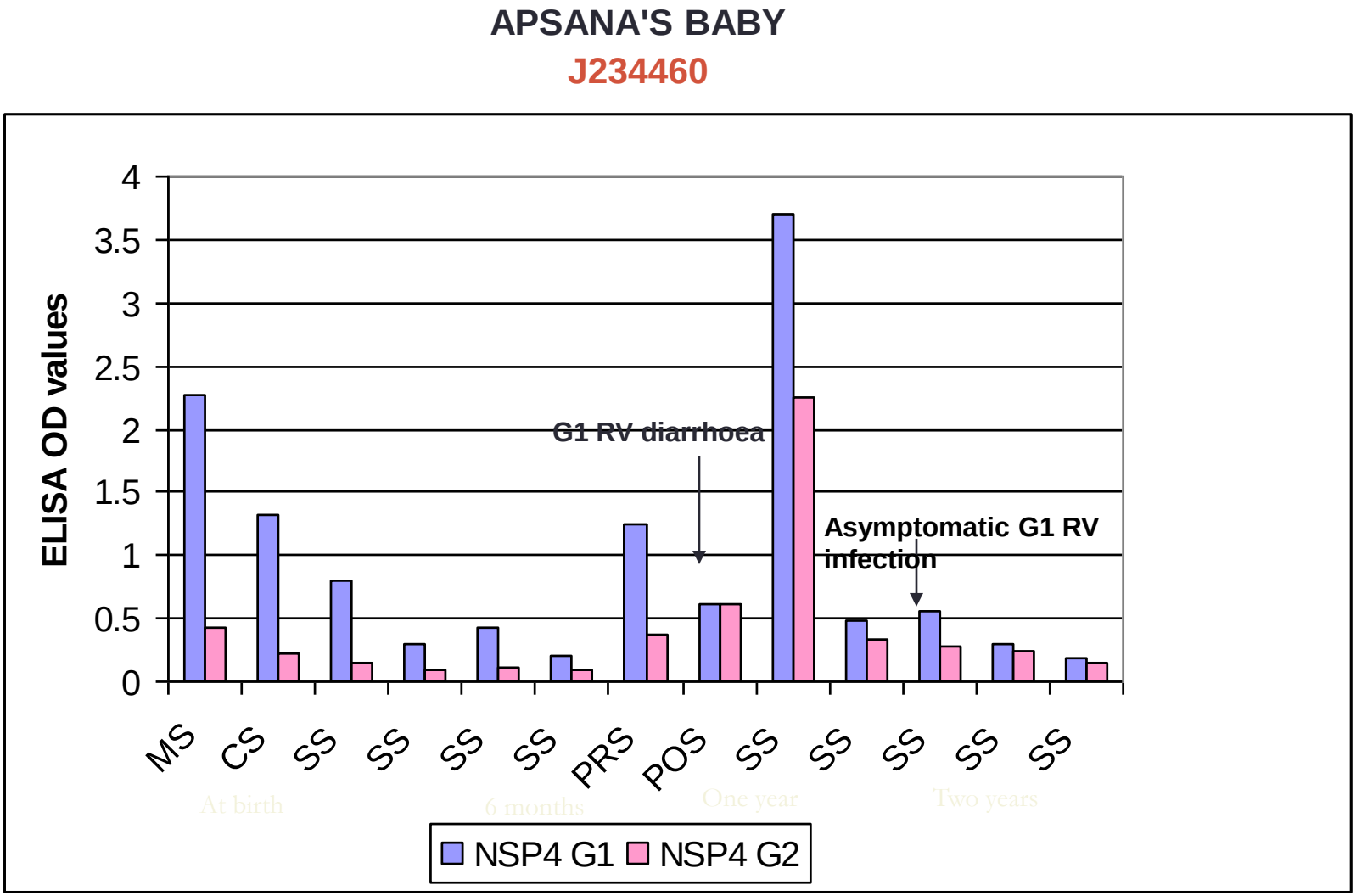
rVP6 IgG



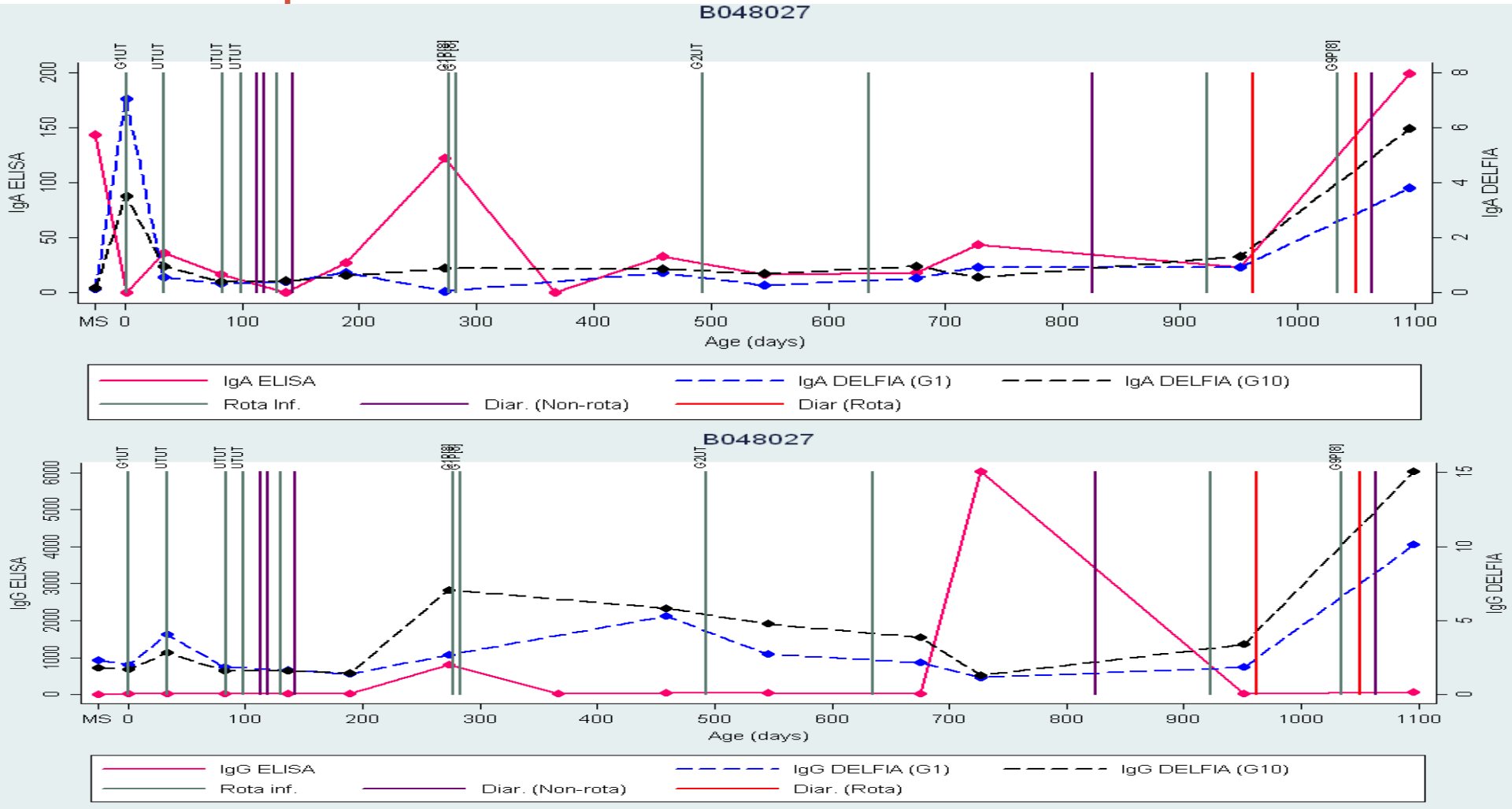
Longitudinal follow up of study subject C360006 rVP6 IgA



Longitudinal follow up of study subject J234460 rNSP4 IgG



We tried to develop more sensitive assays and better follow up

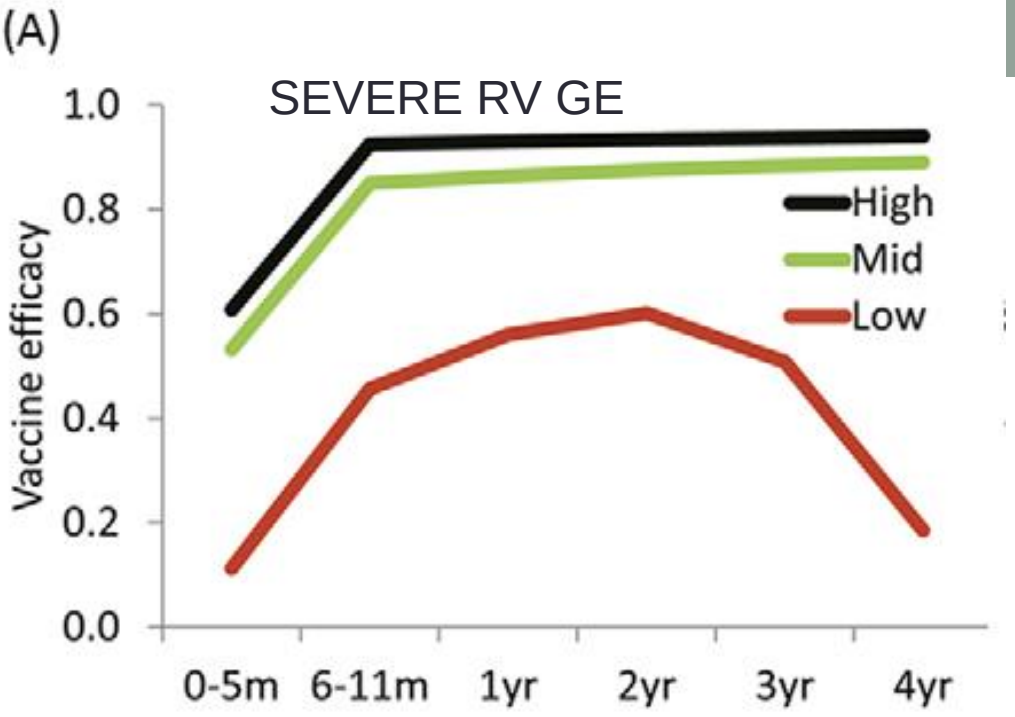


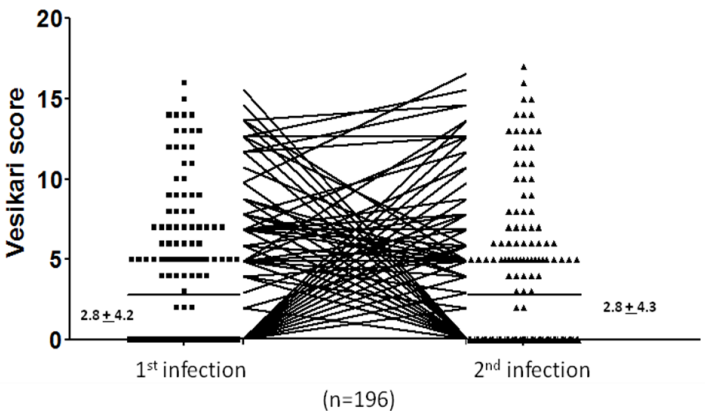
- Cohort data is difficult to interpret

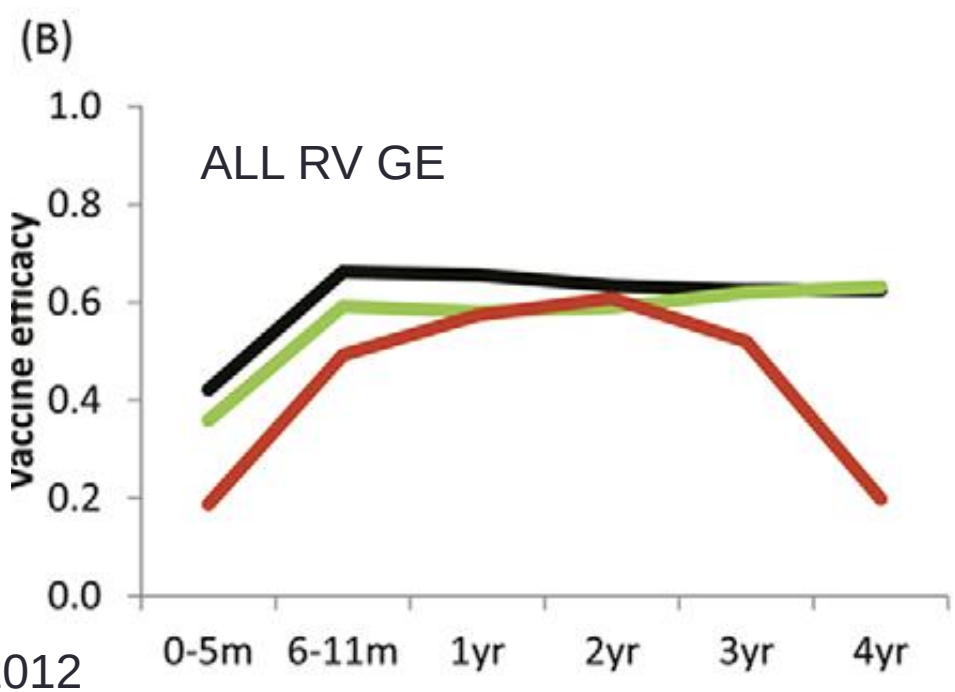
- This was the point at which I gave up and moved to clinical trials
  - Defined time of exposure
  - Defined dose

Implications of data from birth cohorts for vaccination

- Predicted 50% efficacy
- Difference in protection from all and severe GE by setting







Lopman PLoSOne 2012

Key take aways

- Age and prior infection are the best predictors of protection from infection and disease with rotavirus
  - This holds true in multiple settings
- Serum IgA and IgG antibody responses increase with infection, but there is no cut-off that predicts protection
- Faecal IgA and salivary IgA measured in a subset were variable
- Recombinant protein based assays may be useful, but a cohort study may not be the best design for their evaluation