

Exploring *S. Typhi*-Specific HLA-E Restricted Immune Responses in Pediatric and Adult Ty21a Recipients

Mark Rudolph

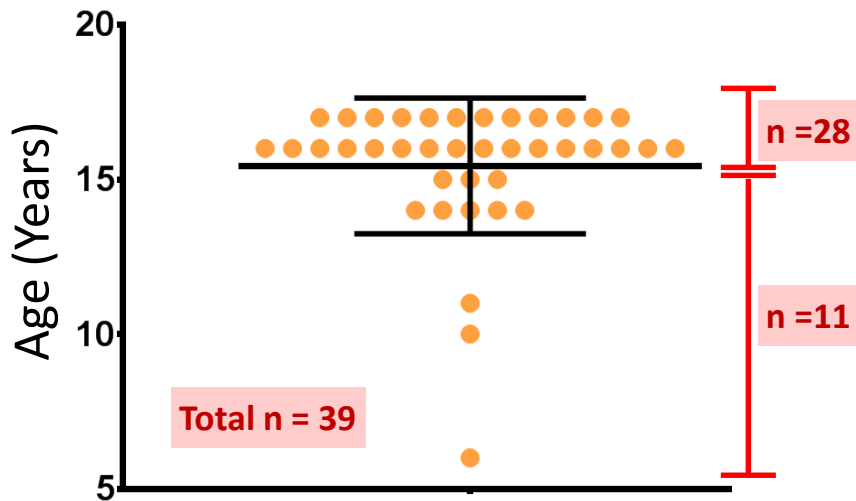
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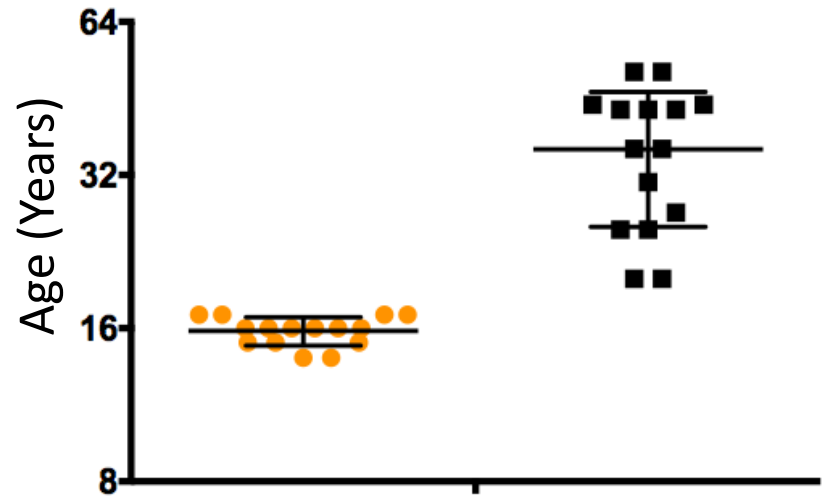
Center for Vaccine Development

Volunteer Information

Pediatric Volunteers



Age of Volunteers Tested



- Pediatric (total) $n = 15$
- Adult (total) $n = 15$ (9 individuals)

Pediatric volunteers receive Ty21a vaccine for medically indicated reasons (travel to endemic regions). Recruitment is ongoing.

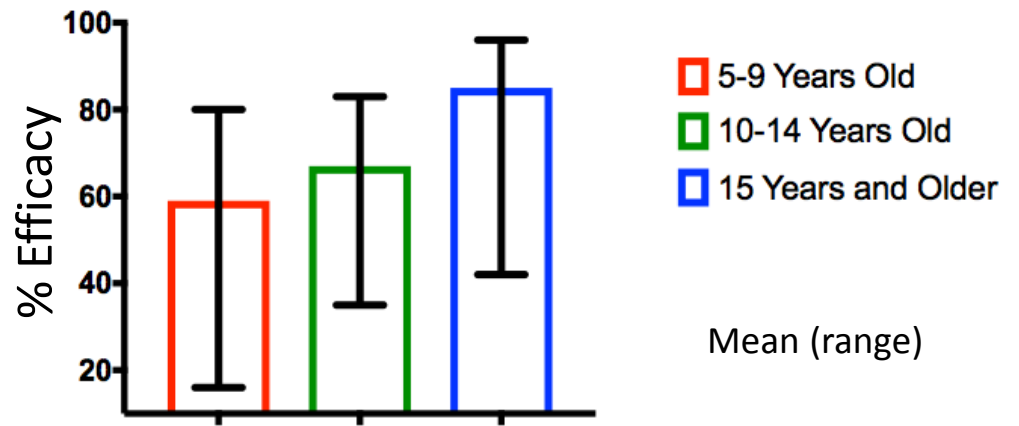
Pediatric Immune Responses

- Cell mediated immune responses (CMI) have been shown to change throughout childhood and into adulthood He et al., J Virol. 80: 11756-66, 2006
Pendergast et al., Nat Rev Immunol. 12: 636-48, 2012
- Thresholds of antigen exposure for inducing immunity also shift through increasing age Watson, B., J. Infect Disease. Suppl 2: S143-6, 2008
- Our lab has shown that children may have less multifunctional responses than adults in response to stimulation with superantigen Booth et al., Front Immunol. 5: 249, 2014
- We don't know how these differences may impact the pediatric response to Ty21a vaccination, or how could this affect Ty21a protection against the development of typhoid disease in different age groups

Ty21a Efficacy in Children - Field Studies

Age Group	Placebo	Vaccine
5-9 yr		
# of children	7193	7034
# of cases	25	10
Efficacy	-	59% (16-80%)
10-14 yr		
# of children	9710	9992
# of cases	32	11
Efficacy	-	67% (35-83%)
≥ 15 yr		
# of children	5001	5142
# of cases	13	2
Efficacy	-	85% (42-96%)

Ty21a Vaccine Recipients

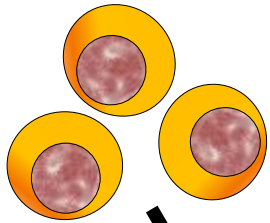


Adapted from: Levine et al., The Lancet 329: 1049-1052, 1987

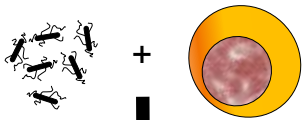
Ty21a efficacy was previously observed to be lower at a younger age; however, (1) these data showed large overlapping confidence intervals, and (2) responsiveness does not necessarily correlate with efficacy

Experimental Design

Target Cells
721.221.AEH



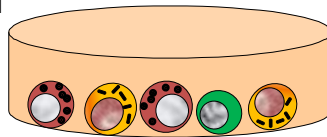
S. Typhi Infection



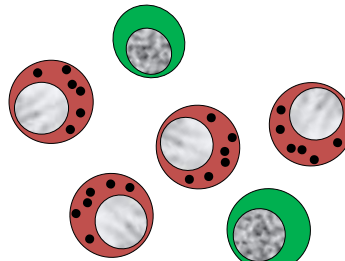
γ -irradiation



**Infected AEH
Co-culture**

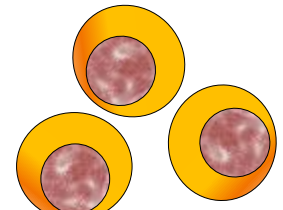


PBMC

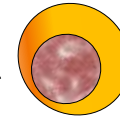


14-18 hr
incubation

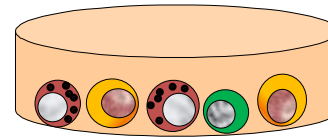
Target Cells
721.221.AEH



γ -irradiation



**Uninfected AEH Co-
Culture (Control)**



Mass Cytometry

(38 parameters)

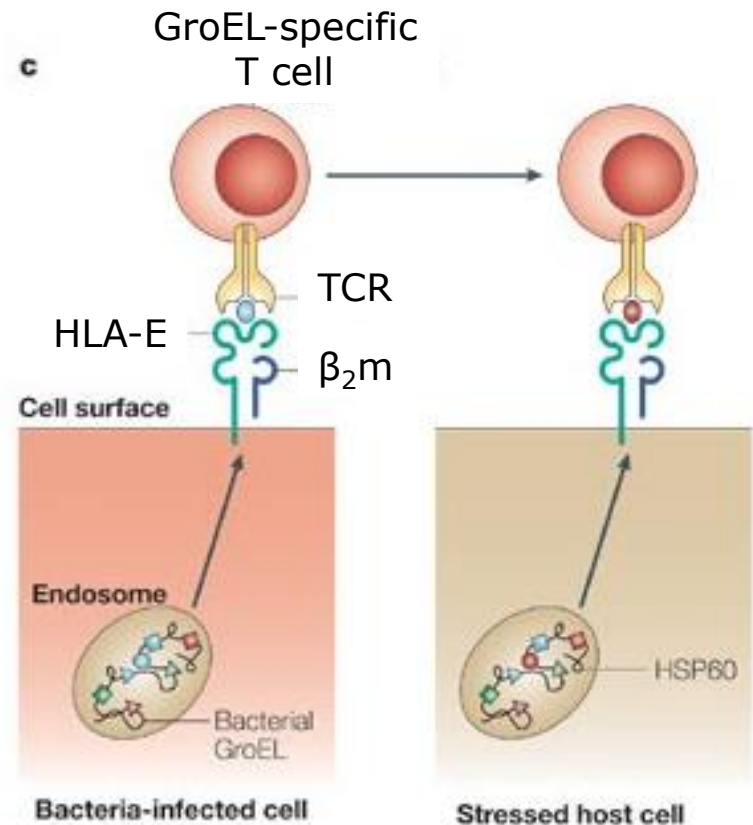
Memory subsets

Cytotoxic T cells

Helper T cells

HLA-E Restricted Antigen Presentation

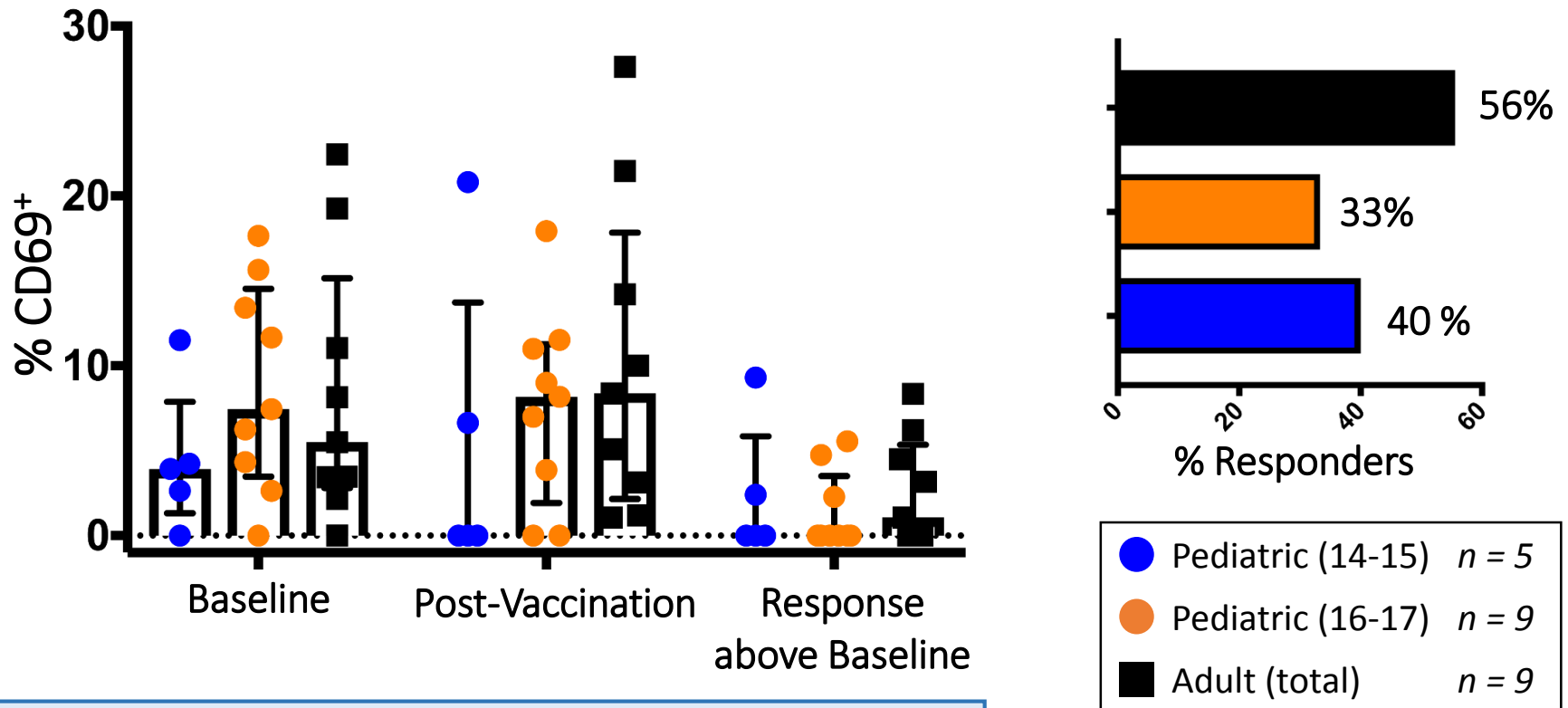
- Non-classical MHC class Ib
- Present a more conserved set of peptides:
 - HLA-A2 leader peptide (inhibitory)
 - *Bacterial chaperonins* (stimulatory)
 - *Heat-shock proteins* (stimulatory)
- We have shown in adult volunteers HLA-E restricted CD8⁺ T cell responses for up to 2 years following Ty21a vaccination



CD8 Activation (CD69+)

HLA-E restricted *S. Typhi* responses

Gated on CD8⁺ T effector memory (CD62L-/CD45RA-) Cells

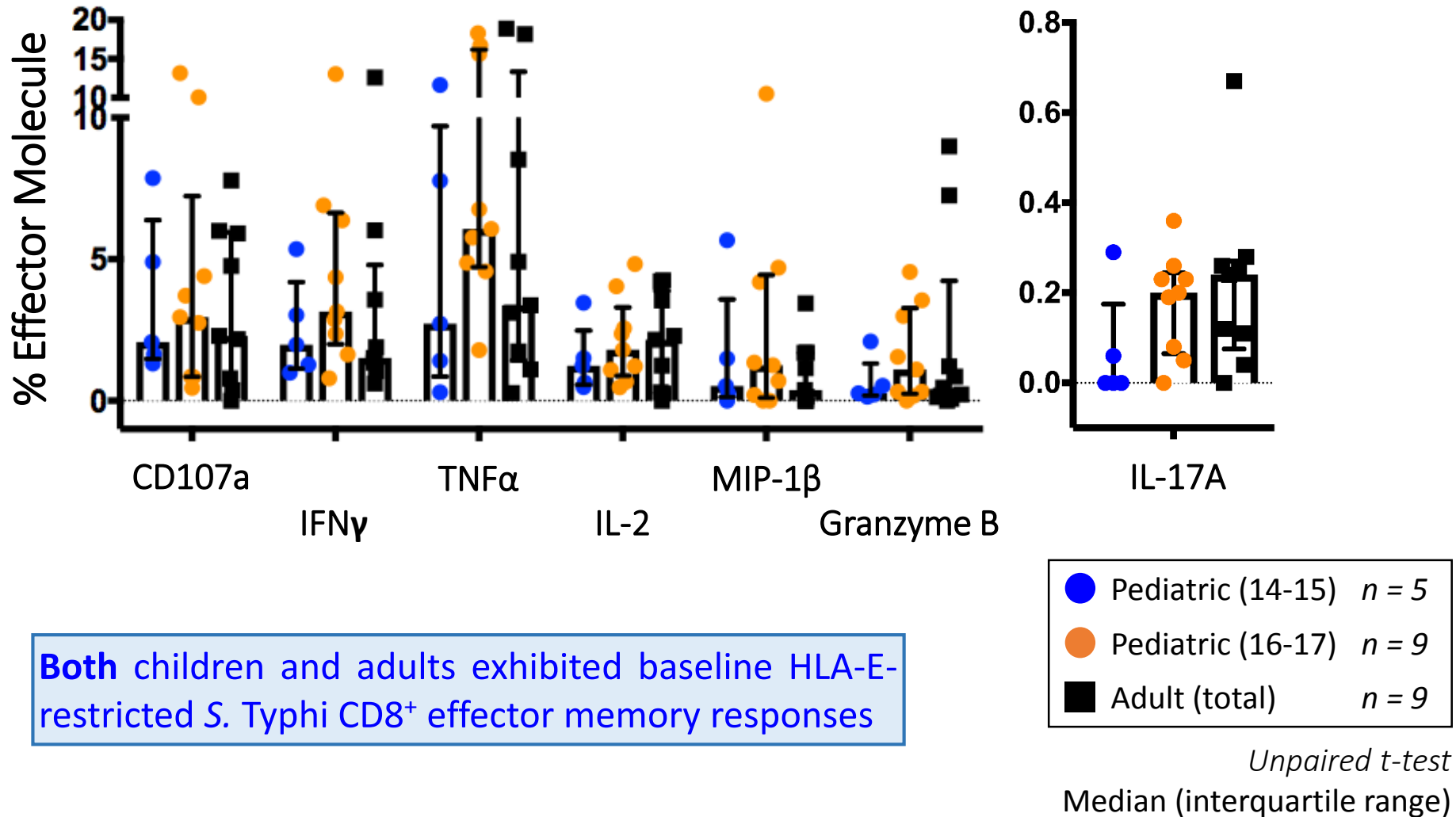


Expression of the activation marker CD69 on CD8⁺ T_{EM} cells provides preliminary data that HLA-E-restricted *S. Typhi* responses are observed in a higher proportion of adults than children

Unpaired t-test
Median (interquartile range)

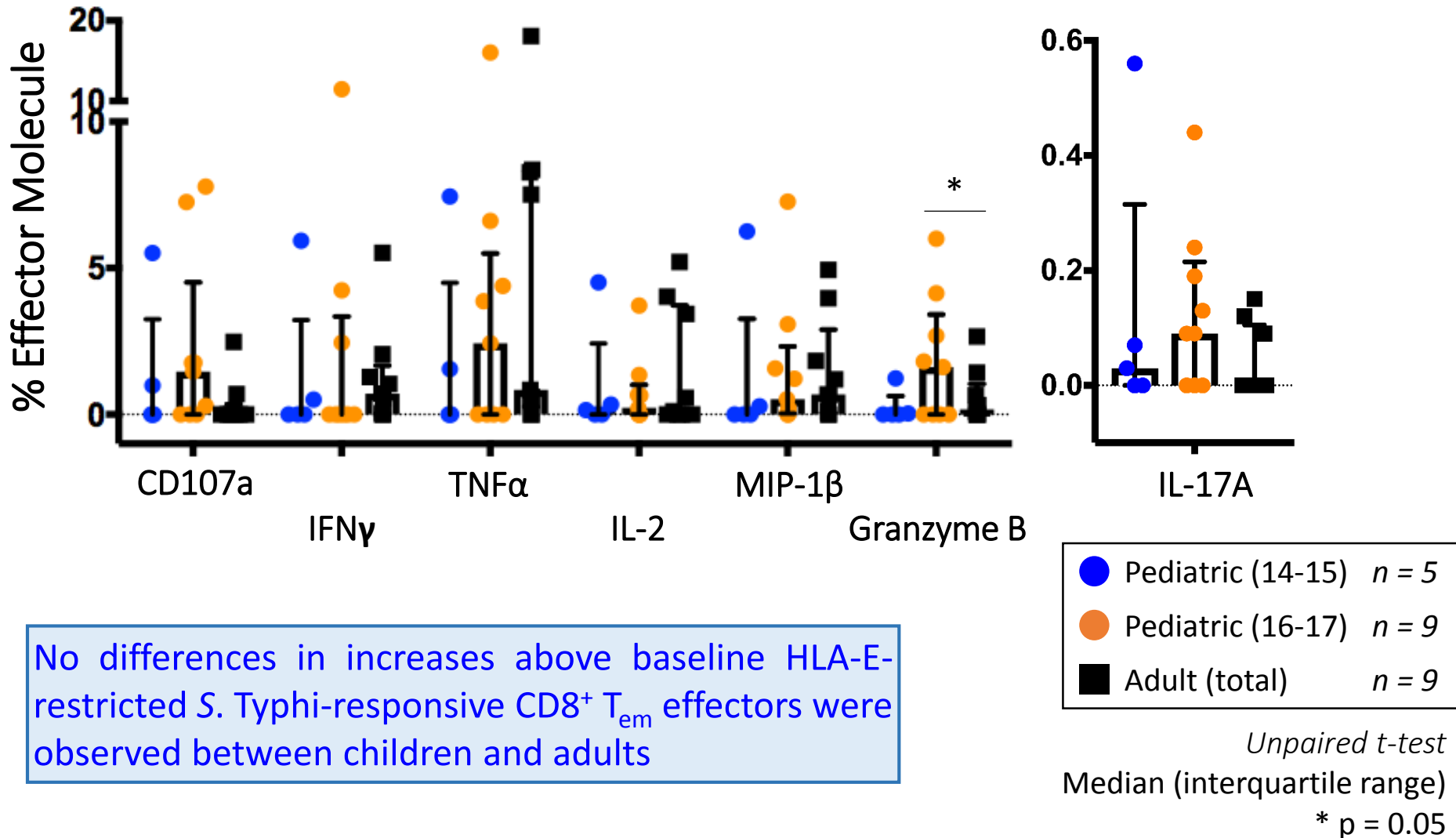
Baseline HLA-E restricted *S. Typhi*-responses

Gated on CD8⁺ T effector memory (CD62L⁻/CD45RA⁻) Cells



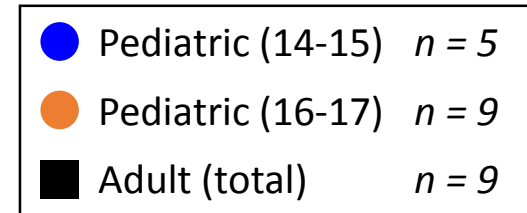
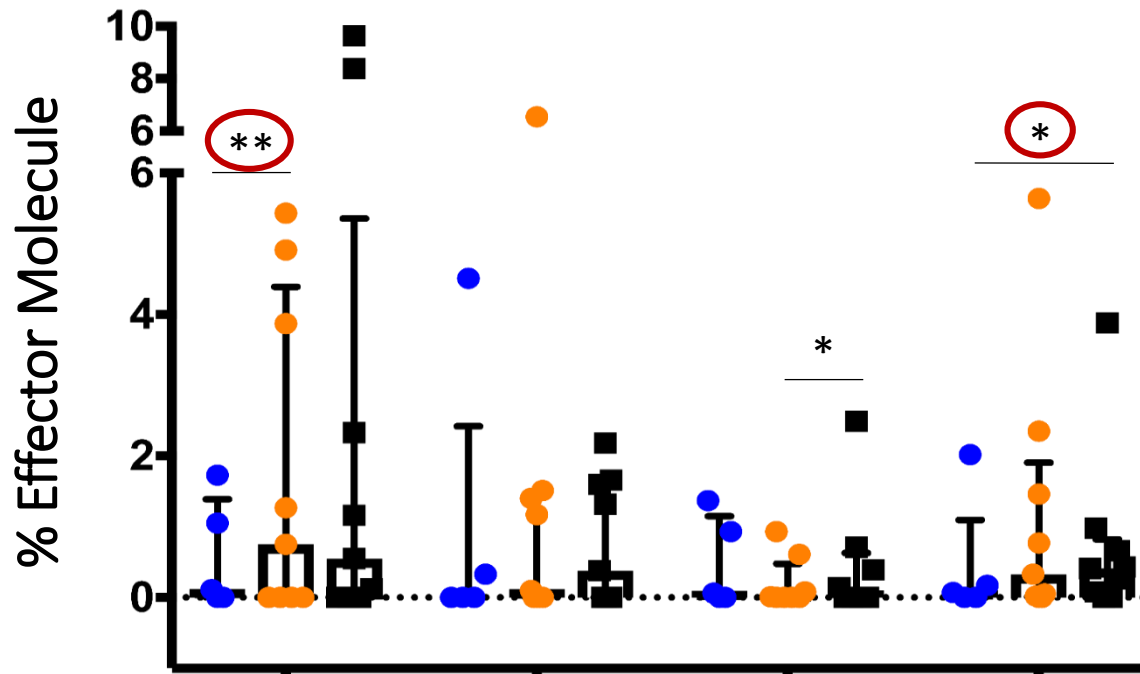
Ty21a vaccination elicits increases in HLA-E restricted *S. Typhi* responses

Gated on CD8⁺ T effector memory (CD62L⁻/CD45RA⁻) Cells



Ty21a vaccination elicits increases in HLA-E restricted multifunctional *S. Typhi*-responses

Gated on CD8⁺ T effector memory (CD62L⁻/CD45RA⁻) Cells



Unpaired t-test
 Median (interquartile range)
 * *p* = 0.05
 ** *p* = 0.01

TNFα	+	+	+	+
IFNγ	-	+	-	+
IL-2	-	-	+	+

Preliminary analyses showed some differences in increases above baseline HLA-E-restricted *S. Typhi*-responsive MF CD8⁺ T_{em} effectors between children and adults

Conclusions

- Expression of the activation marker CD69 on CD8⁺ T_{EM} cells provides preliminary data that HLA-E-restricted *S. Typhi* responses are observed in a higher proportion of adults than children
- Children exhibited baseline HLA-E-restricted CD8⁺ effector memory responses to *S. Typhi* antigens, suggesting previous exposure to HLA-E-restricted antigens
- It appears that for some functions the proportion of responders following vaccination in adults are higher than in children (analyses ongoing)
- Preliminary analyses showed some differences in increases above baseline HLA-E-restricted *S. Typhi*-responsive MF CD8⁺ T_{EM} effectors between children and adults. These differences, however, appear to be less than those observed following SEB stimulation.

Future Directions

- Expand our HLA-E restriction experiments to include our youngest volunteers (6-14 years, n=8)
- Continue our analysis of soluble antigen (GroEL, OmpC, FliC) responses in pediatric and adult helper T cells
- Use Epstein-Barr Virus transformed autologous B-lymphoblastoid cell lines (already generated) to measure responses to classical HLA-restricted (HLA-A,B,C) *S. Typhi* antigen presentation in pediatric and adult T cells
- Assess Ty21a vaccination induced correlations between *S. Typhi* immune responsiveness and functional characteristics of the gut microbiota (in cooperation with the Institute for Genome Sciences)

Acknowledgements

Center Vaccine Development, UMB

Immunology Group

Marcelo B. Sztein

Monica McArthur

Stephanie Fresnay

Rosangela Mezghanni

Franklin Toapanta

Jay Booth

Rekha Rapaka

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Marcelo Sztein (advisor)

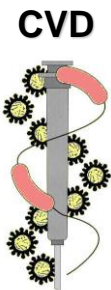
Monica McArthur

Larry Magder

Claire Fraser

Eileen Barry

George Lewis



Supported by grants U19 AI082655 (UM-CCHI) &
R01 AI-36525 from the NIH