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SCHOOL OF MEDICINE
CENTER FOR VACCINE DEVELOPMENT
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Age-Associated Heterogeneity of Ty21a-induced T Cell Responses to HLA-E Restricted *Salmonella* Typhi Antigen Presentation

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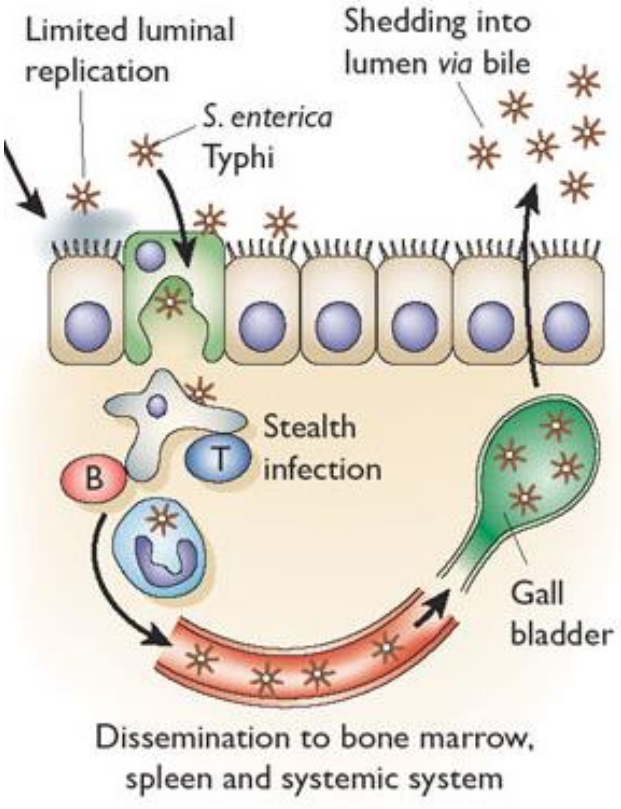
Salmonella Typhi

- *S.* Typhi is a gram negative human-restricted facultative intracellular pathogen
- Transmitted via fecal-oral route and invades through the gut epithelia
- Can disseminate into lymphatics and bloodstream by targeting DC and MΦ leading to systemic, sometimes chronic, infection

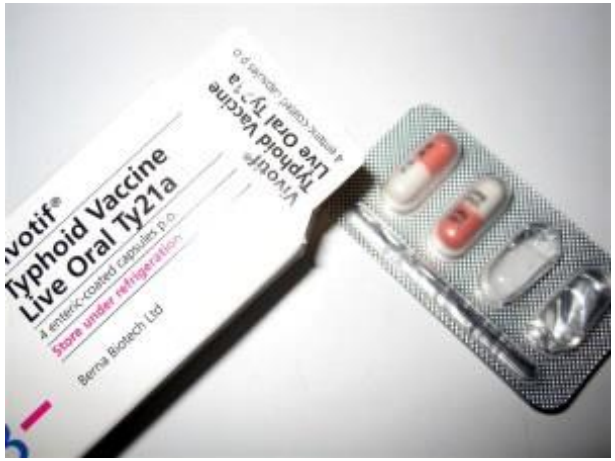


Rev Infect Dis. 1991 Jan-Feb;13(1):85-90

Nature Immunology **3**, 1026 - 1032 (2002)

Ty21a Vaccine

- Licensed Vi-neg oral live-attenuated vaccine derived from the wt Ty2 strain of *S.* Typhi



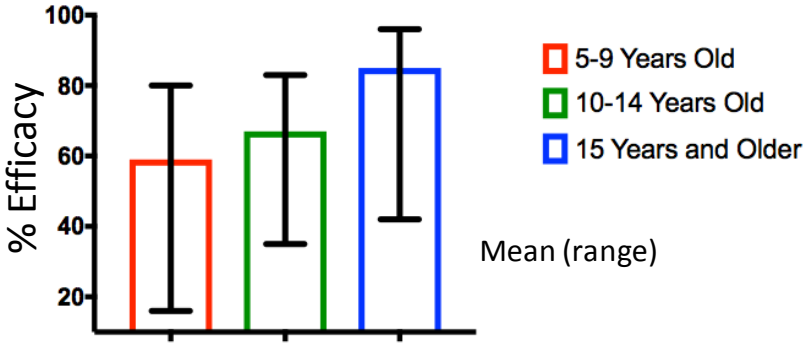
- Responses generated
  - Antibodies and memory B-cells to *S.* Typhi-specific O-LPS and H-flagella. However, these responses do not appear to correlate with protection in human wild-type challenge studies
  - *CD4+ and CD8+ Cell-mediated immune (T-CMI) responses*
    - *T_C1/T_C17 cytokine effectors*
    - *Contact-dependent cytotoxicity (CTL)*
    - *Helper T cell responses (e.g., Th1, Th17)*

Some of these T-CMI responses have been associated with protection in a human wild-type challenge study

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
(enteric-coated capsules)



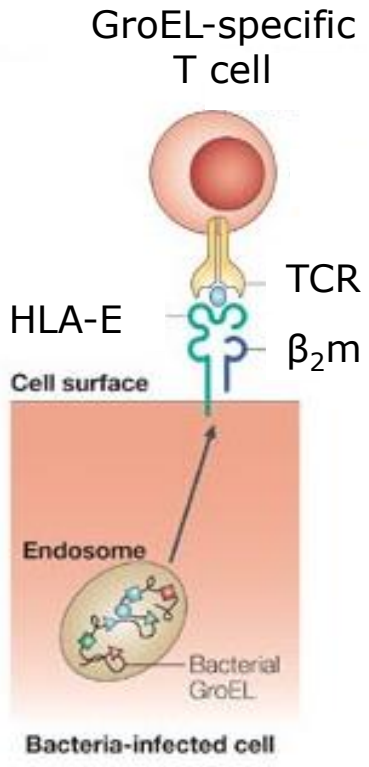
Adapted from: Levine et al., The Lancet 329: 1049-1052, 1987

- There is a poor understanding of human pediatric CMI immune responses
- Typically, younger children are less likely to develop robust, long-lasting CMI than adults and older children

Ty21a efficacy was previously observed to exhibit a trend to be lower at a younger age; however, these data showed large overlapping confidence intervals

Human Leukocyte Antigen – E (HLA-E)

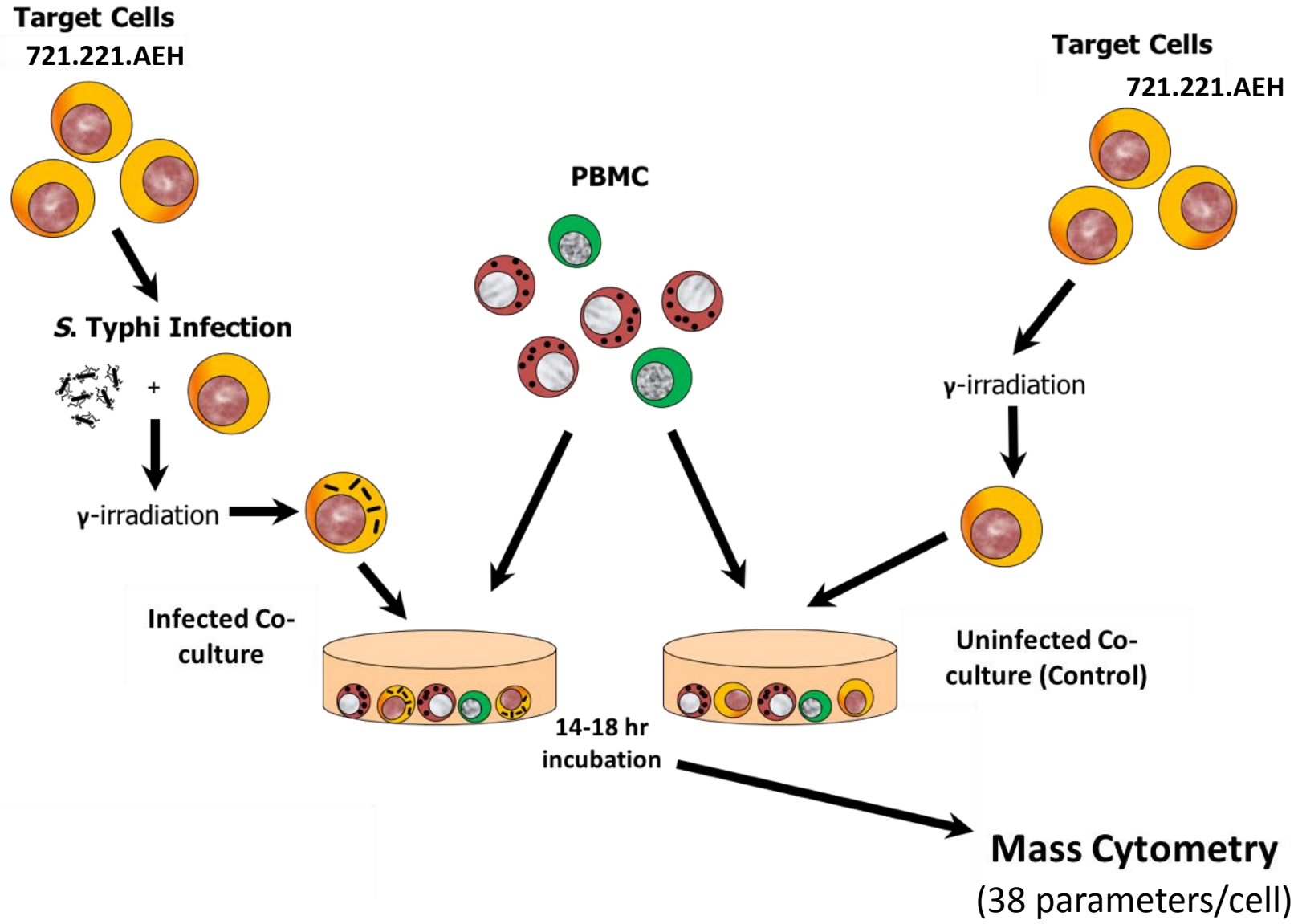
- Non-classical MHC class Ib
- HLA-E presents a conserved set of peptides
  - HLA-A2 leader peptide (inhibitory)
  - *Bacterial chaperonins (stimulatory)*
  - *Heat-shock proteins* (*stimulatory*)
- HLA-E restricted CD8+ T cell responses are present for up to 2 years following Ty21a vaccination in adult volunteers and are also induced upon wt *S.* Typhi challenge
- Differences in effector cytokine responses to autologous vs. HLA-E restricted *S.* Typhi-infected cells in Ty21a vaccinated adults



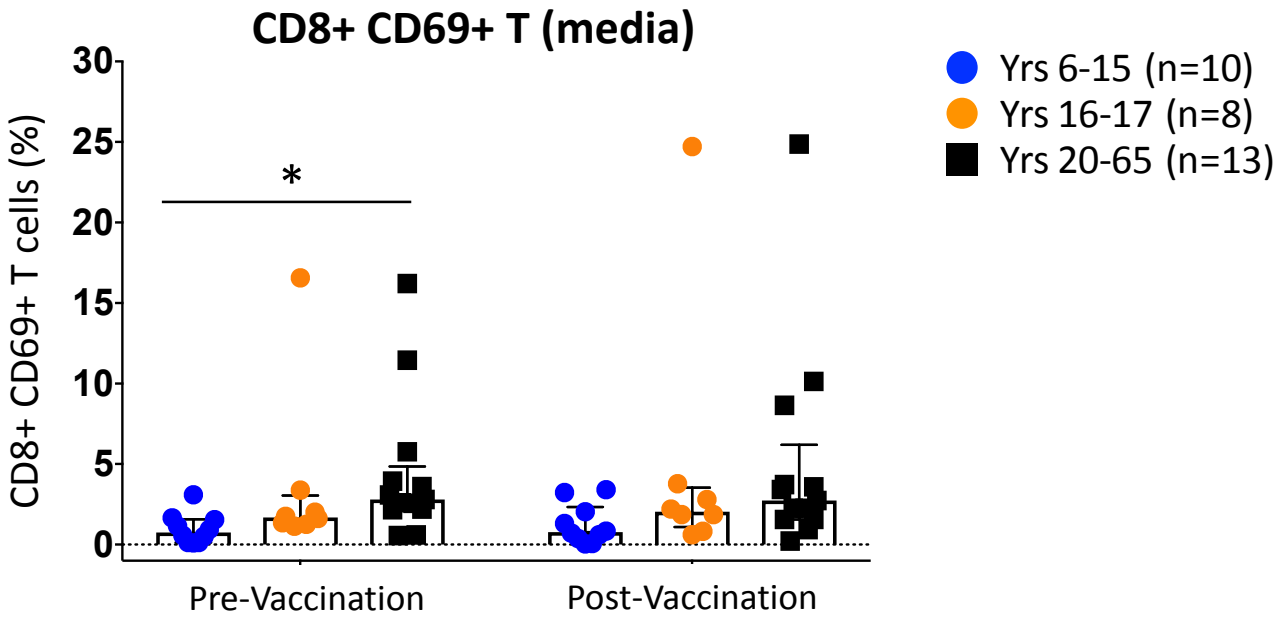
Salerno et al. J. Immunol. 173: 5852-5862, 2004
Salerno et al. Clin Vaccine Immunol 17: 1305-1314, 2010

Adapted from: Rodgers et al. Nat Rev Immunol 5: 459-71, 2005

HLA-E Restriction Experimental Protocol



HLA-E Restricted Activation (CD8+ CD69+)



Younger children are less likely to have activated CD8+ T cells at baseline than older children or adults

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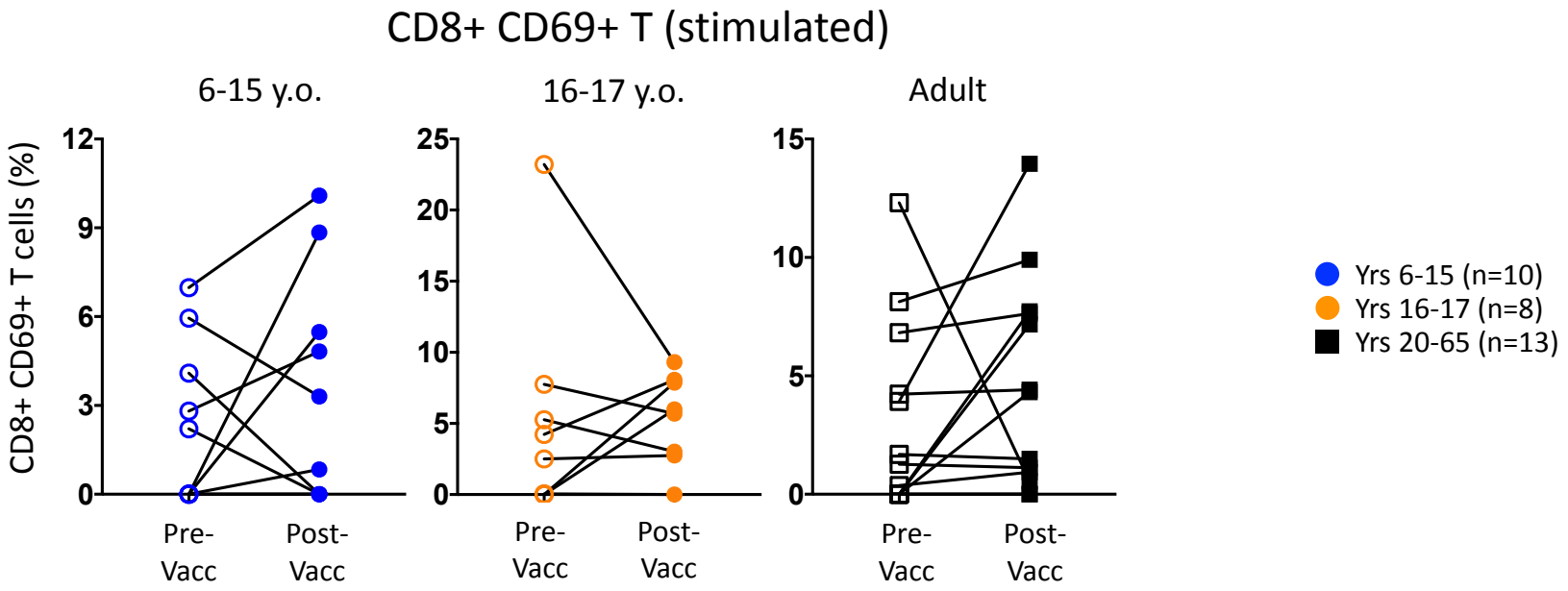
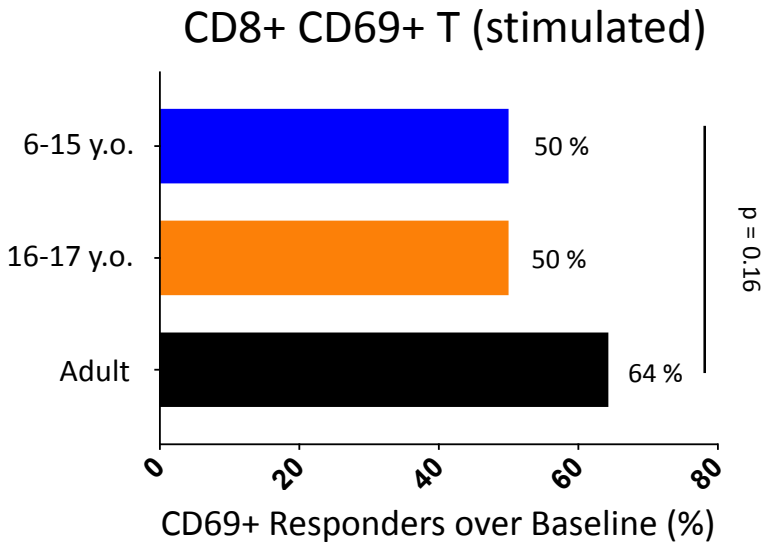
HLA-E Restricted Activation (CD8+ CD69+)





No significant differences were observed among age groups in the percentages of participants who exhibited increased activation to HLA-E-restricted *S.* Typhi-specific CD8+ T cell stimulation following Ty21a vaccination.

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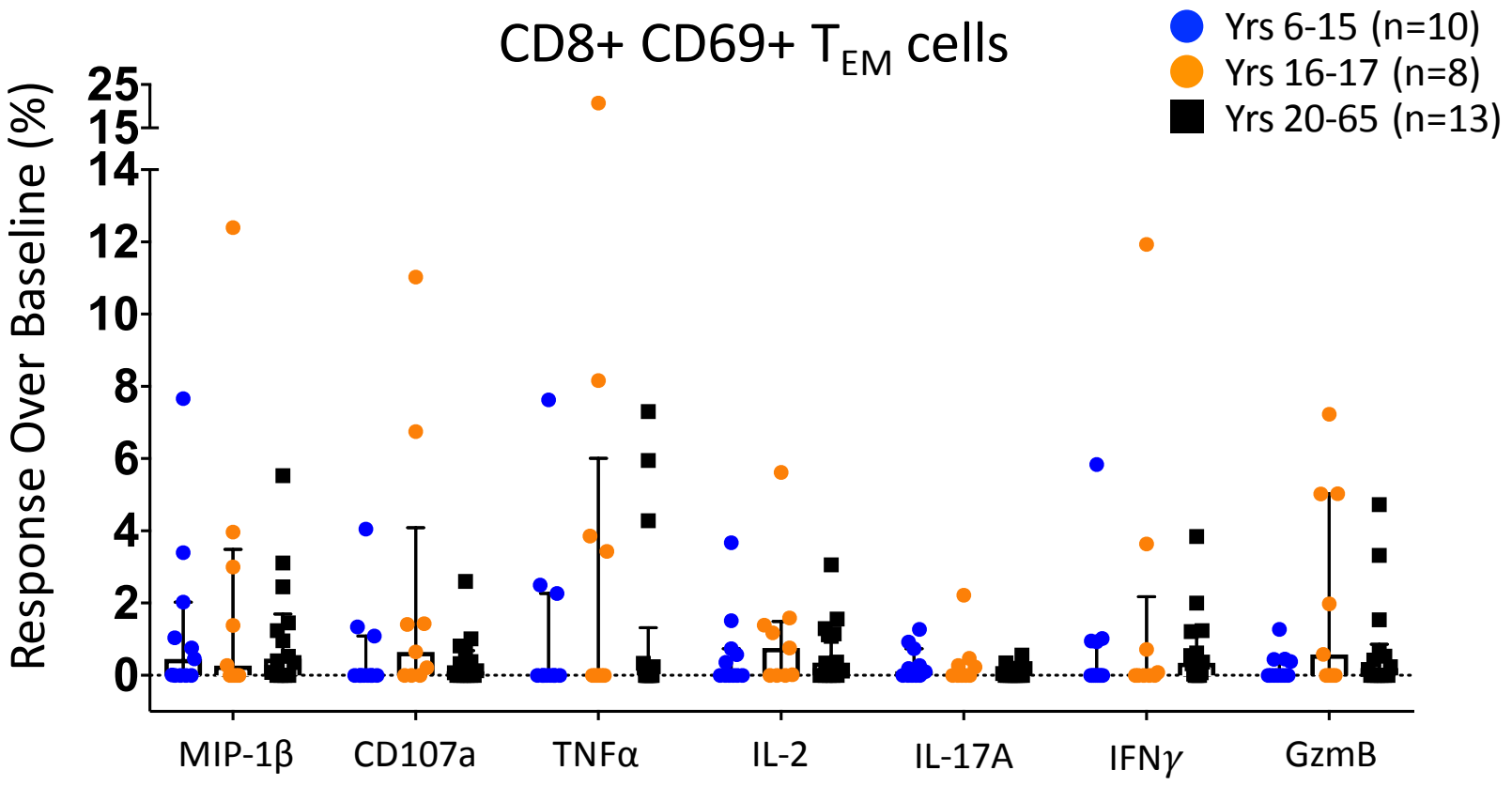
HLA-E Restricted CD8+ T_{EM} Responses



No significant age-associated differences were observed in CD8+ T_{EM} effector responses following vaccination

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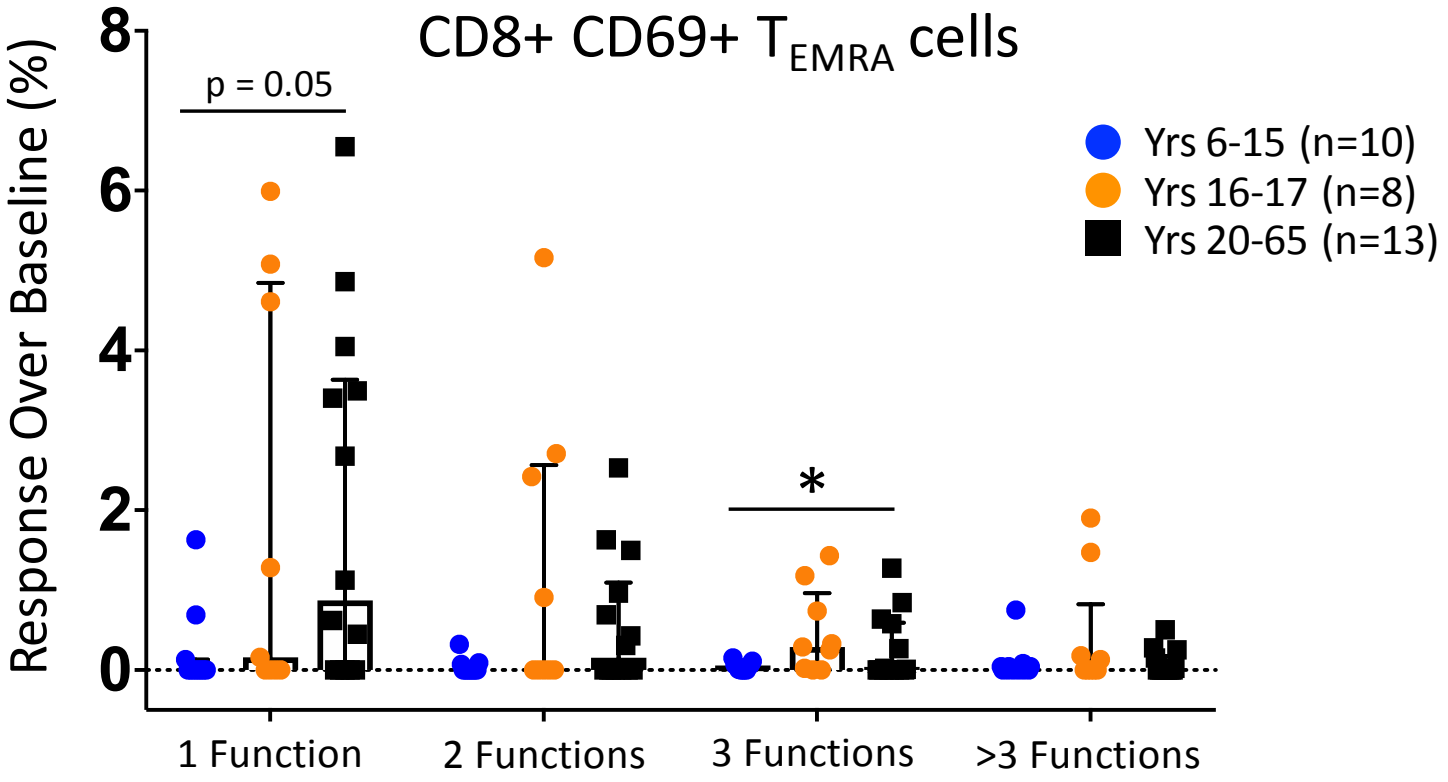
HLA-E Restricted CD8+ T_{EMRA} Responses



Significant age-associated differences in CD8+ T_{EMRA} effector mono and trifunctional responses following Ty21a vaccination

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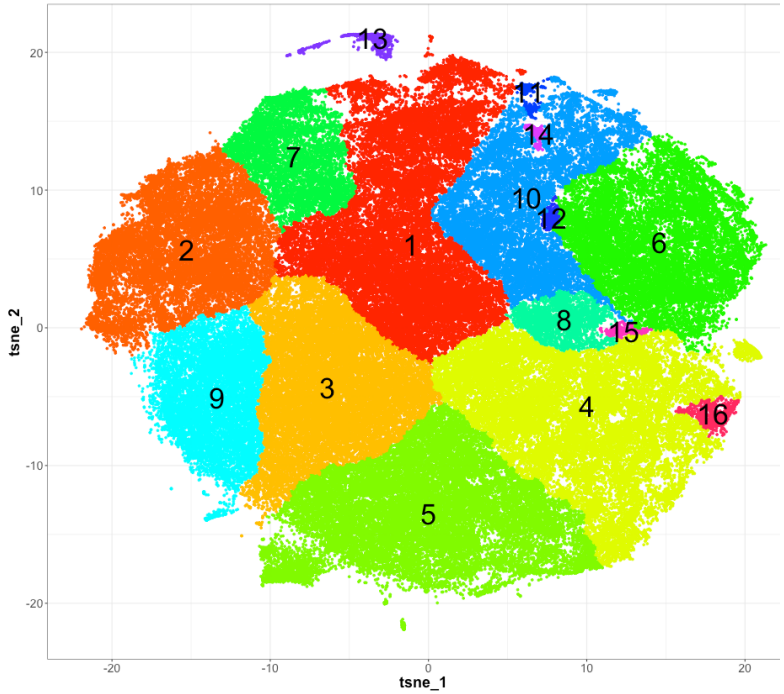
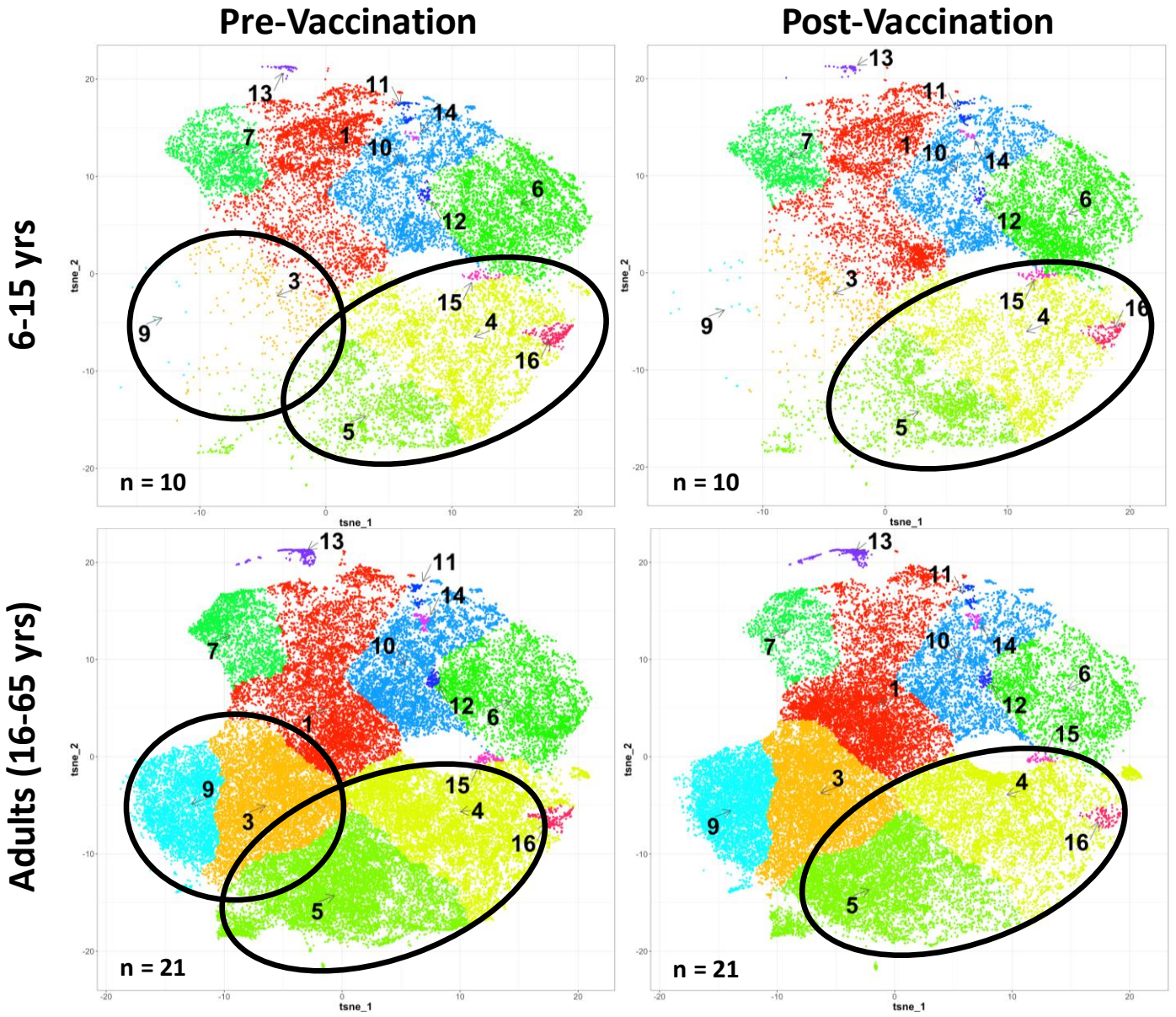
t-Distributed Stochastic Neighbor Embedding (tSNE)

- Non-linear variation of principal component analysis
- Groups all variables of interest into a multiple one-dimensional representations containing every cell event
- Combines one-dimensional representations to generate the most descriptive two-dimensional plot
- Clustering algorithms divide the resulting plot into groups based on each cells' similarity to its neighbors
- Unbiased means of identifying key population features that would be impossible to discern using conventional gating methods

CD8+ CD69+ T_{EM} cells



HLA-E Restricted CD8+ T_{EM} tSNE Data



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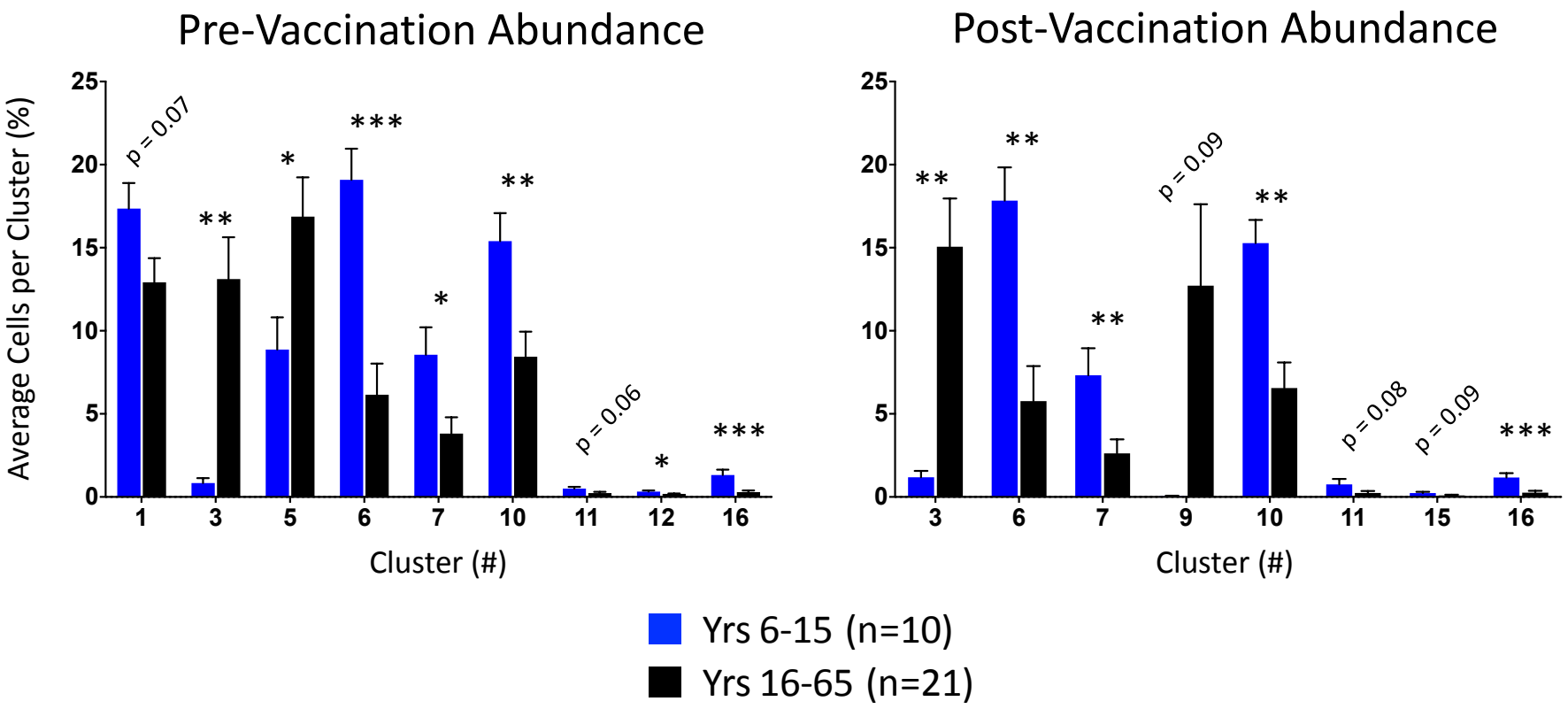
HLA-E Restricted CD8+ T_{EM} tSNE Data



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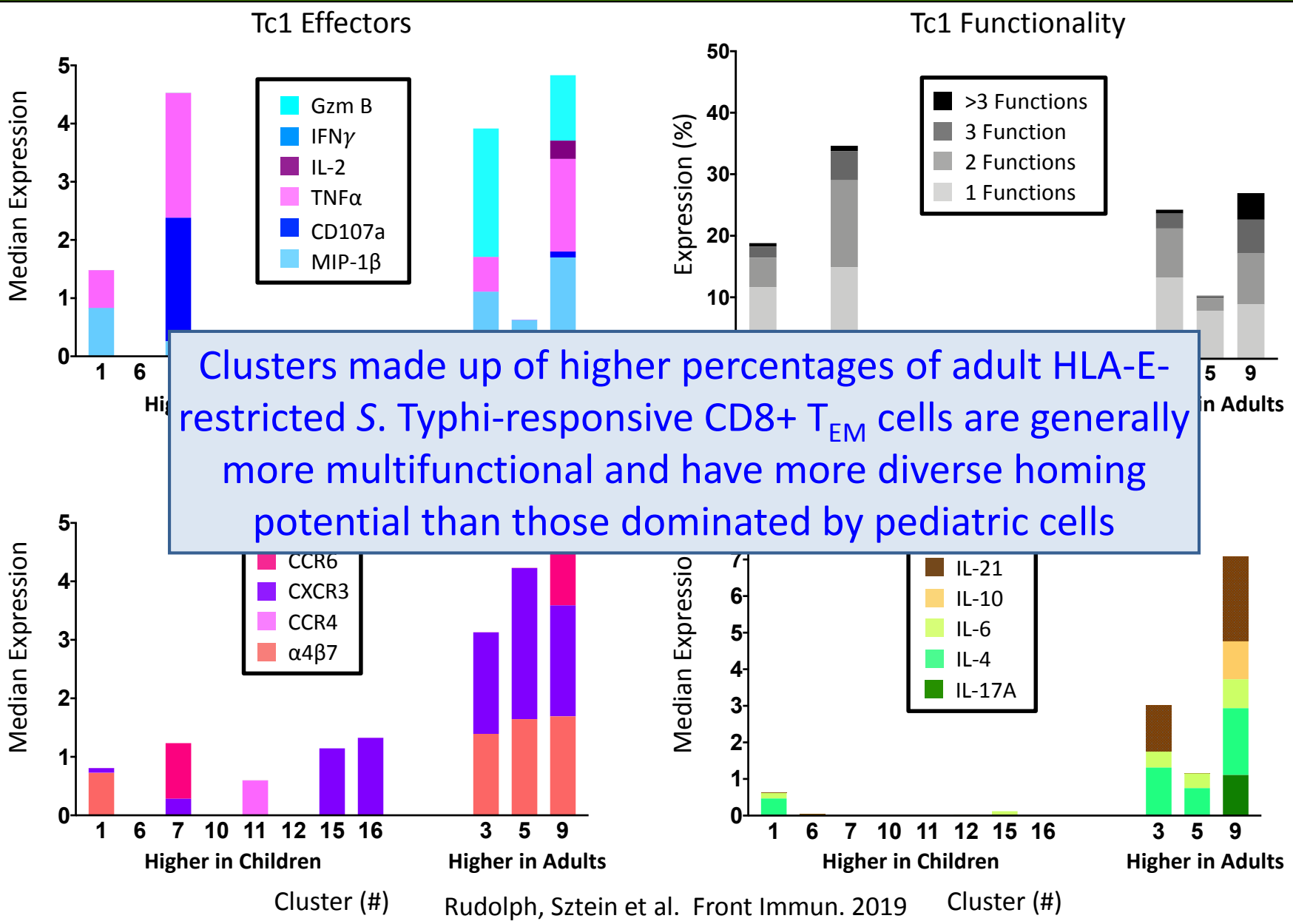
HLA-E Restricted CD8+ T_{EM} tSNE Data



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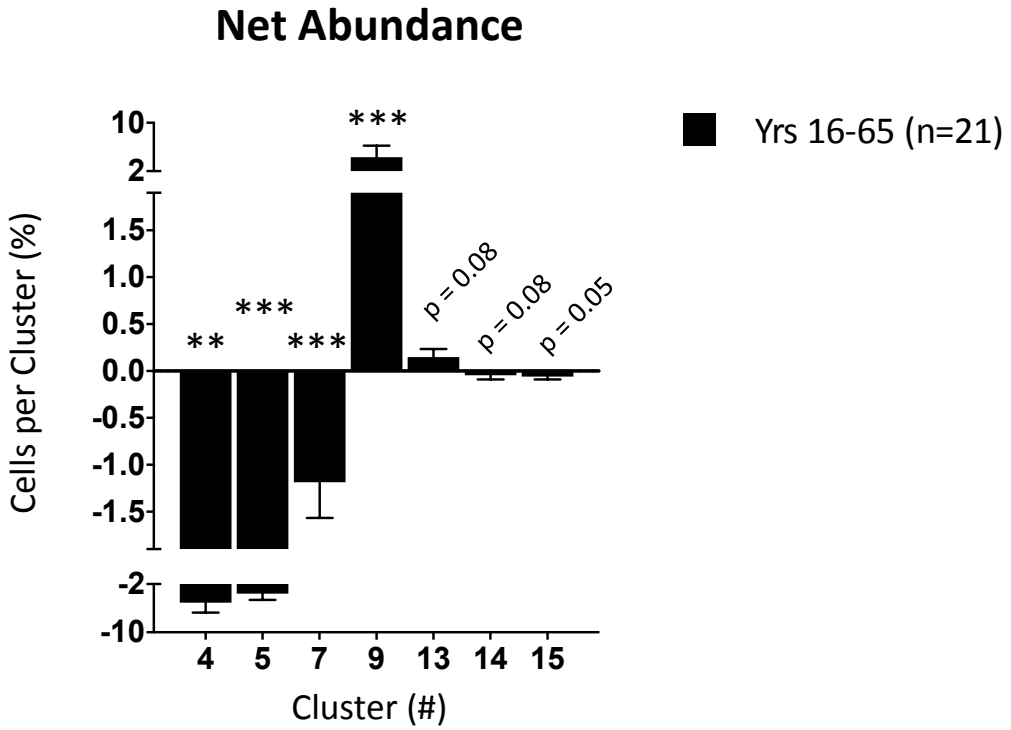
HLA-E Restricted CD8+ T_{EM} tSNE data in adults following Ty21a vaccination



Ty21a vaccination only significantly changed percentages of adult HLA-E-restricted *S.* Typhi-responsive cells within select tSNE clusters

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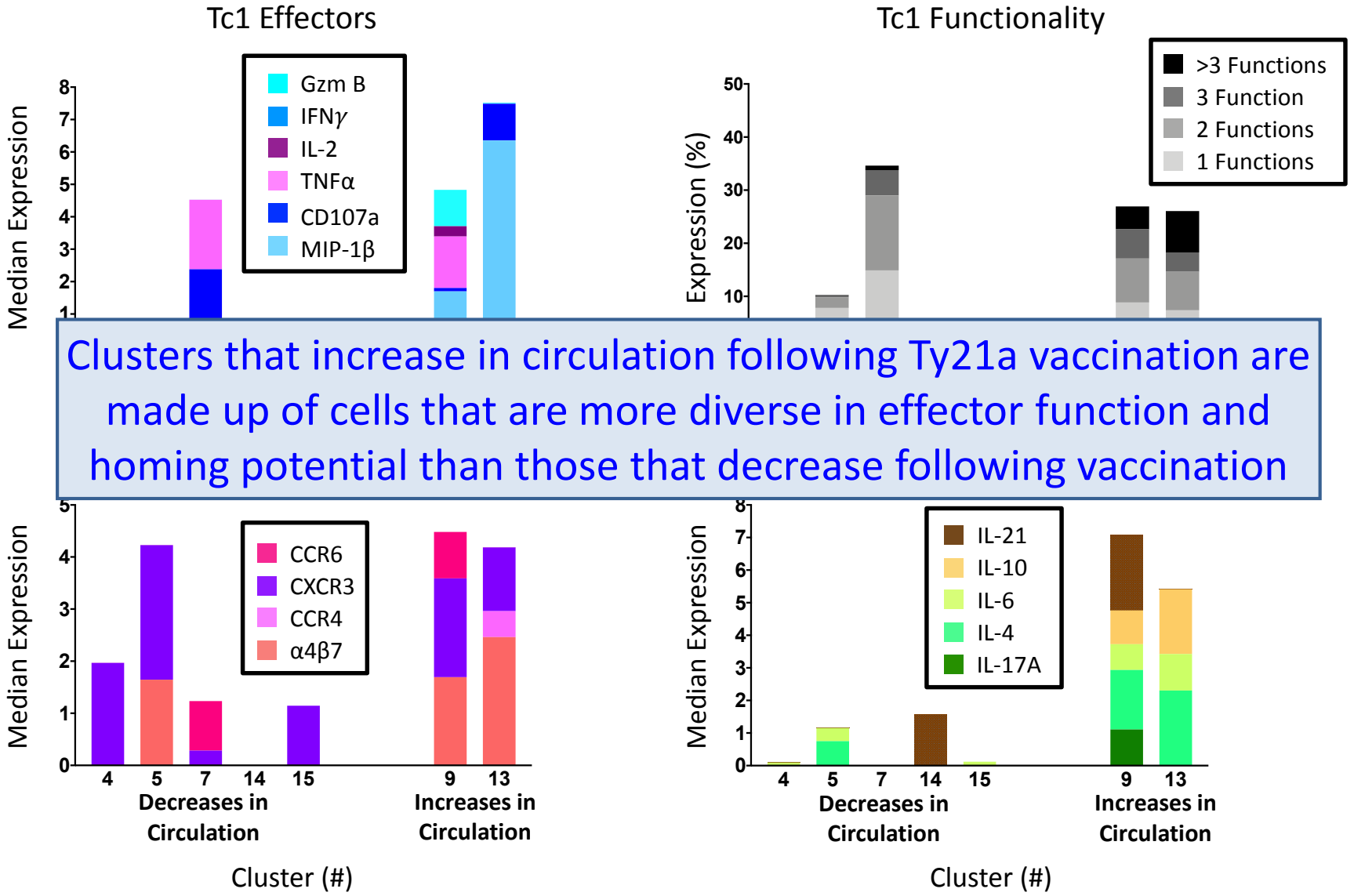
HLA-E Restricted CD8+ T_{EM} tSNE Data in adults



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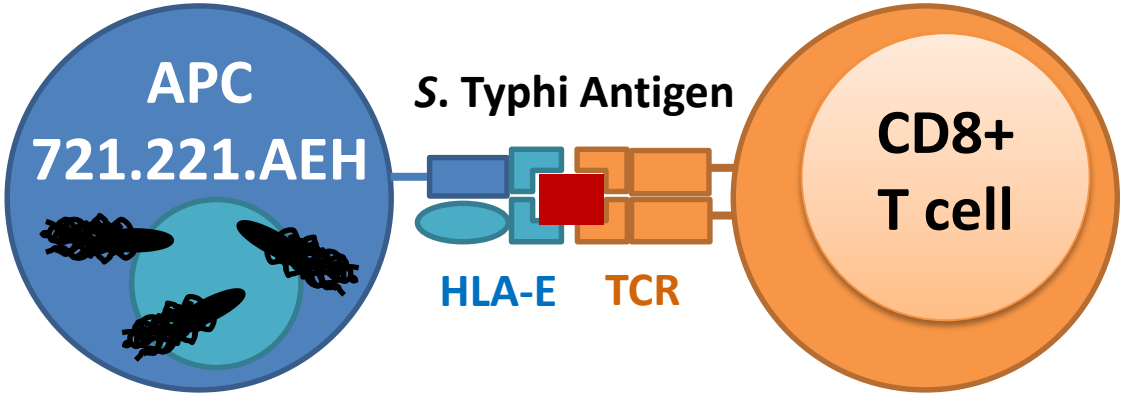
CD8+ HLA-E Restricted Responses: Summary



T Cell Subtype	Children	Adults*
CD8+ T Cells	↓ Baseline activation (CD69+) Heterogeneous T_{EM} post-Ty21a ↓ T_{EMRA} MIP1β, TNFα Fewer cells in T_{EM} and T_{EMRA} tSNE clusters dominated by gut/inflammation homing MF populations	↑ Baseline activation (CD69+) Heterogeneous T_{EM} post-Ty21a ↑ T_{EMRA} MIP1β, TNFα More cells in T_{EM} and T_{EMRA} tSNE clusters dominated by gut/inflammation homing MF populations

*** 16-17 year old T cell activation and functions are more adult-like**

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CD8+ HLA-E Restricted Responses to Ty21a Immunization in Children: Overall Conclusions

- These data demonstrate the presence of major differences in the "fine granularity" of HLA-E-restricted CD8+ responses between children and adults before and after Ty21a immunization
- These include an increased multifunctionality of the responses in adults and marked differences in the cytokines being produced and the homing potential of the responding cells
- These data emphasizes the importance of in depth studies of the CMI immune response in children during the development of future generations of typhoid vaccines, as well as other vaccines targeted for children

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