7TH GLOBAL FORUM ON TB VACCINES | 8-10 October 2024 Rio de Janeiro, Brazil

Driving innovation from discovery to access

Investigating Correlates of Protection after IV BCG Immunization

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Potential Uses for a TB Vaccine

Prevent:

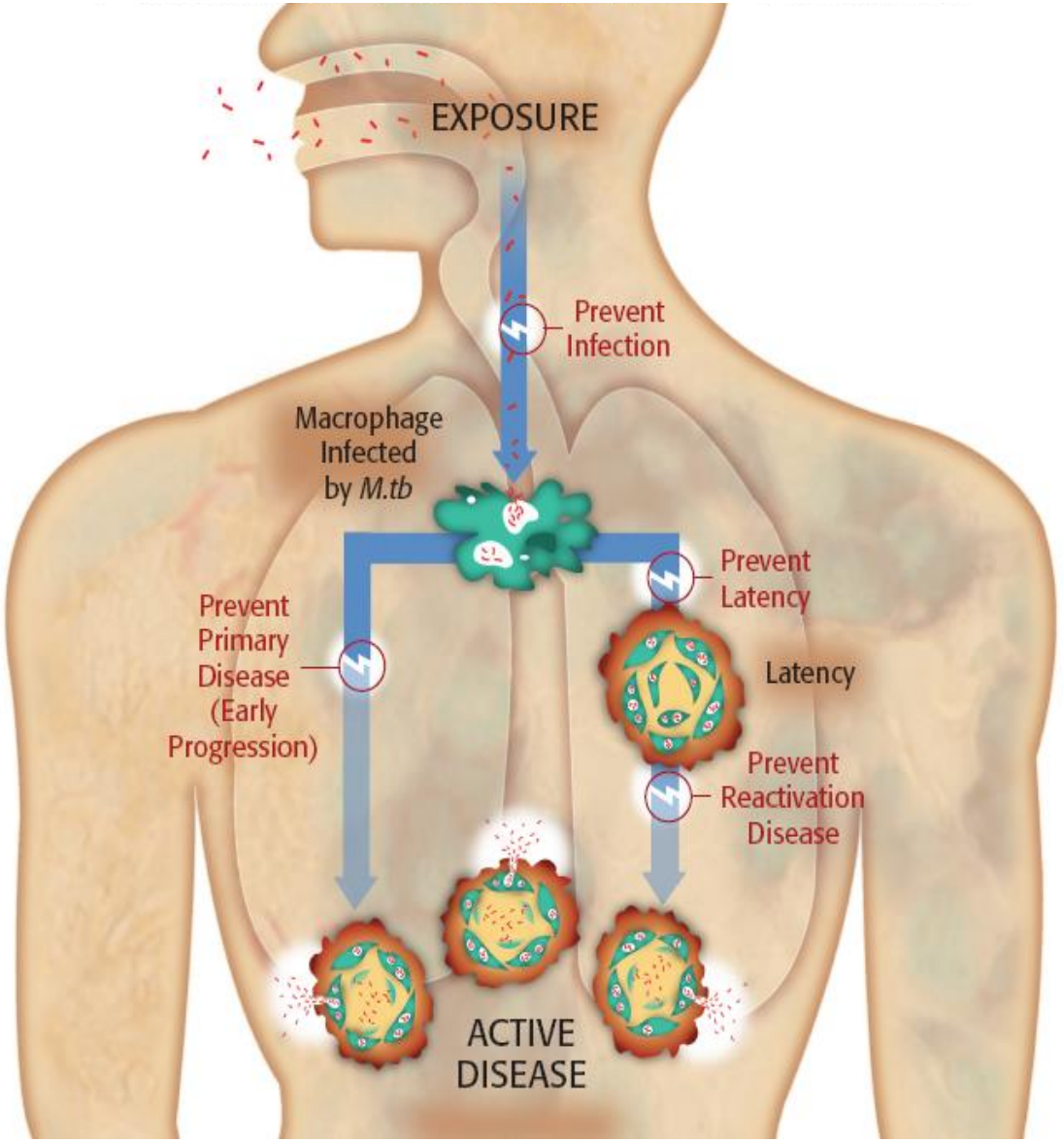
- Infection (Primary)
- Disease in LTBI
- Recurrence after antibiotics
- Re-infection

Which we are modeling by studying TB vaccines in rhesus macaques:

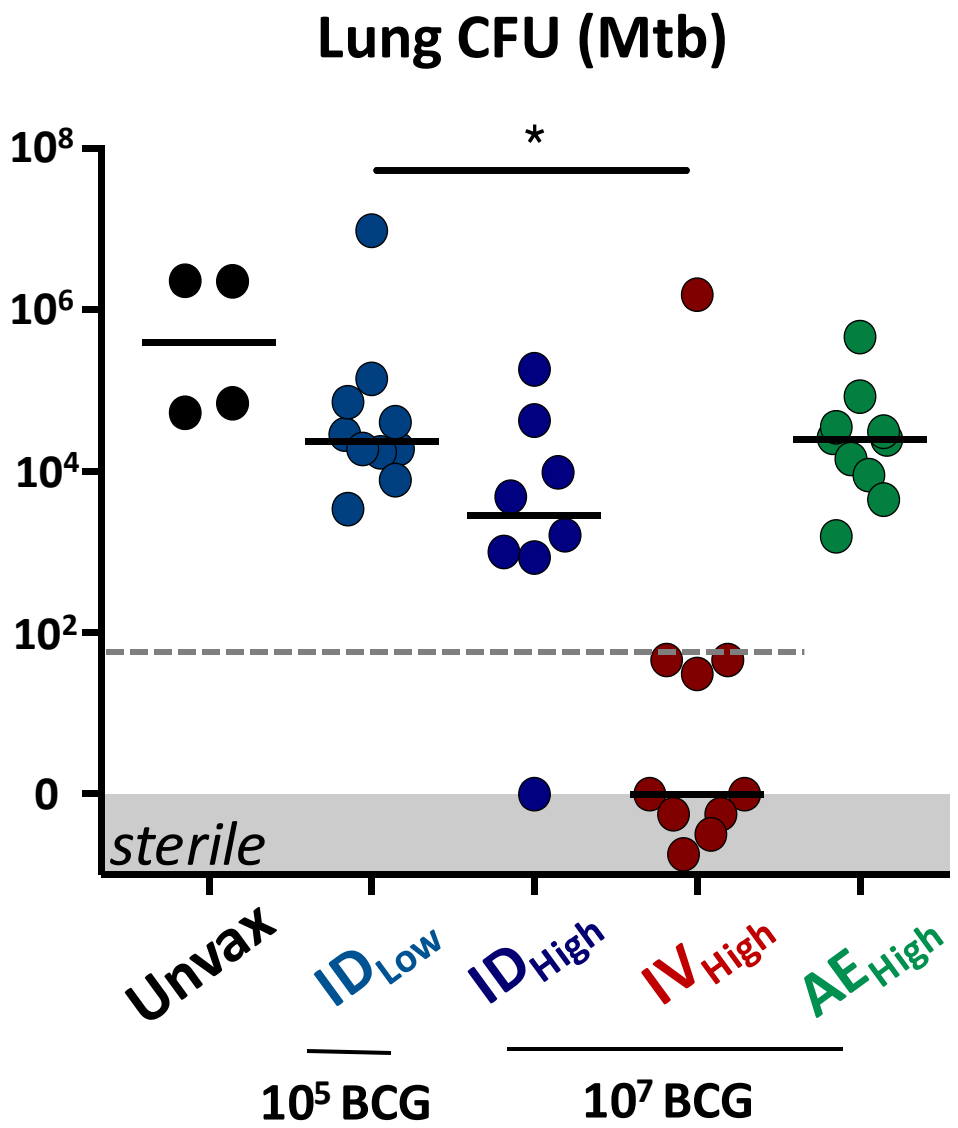
- Naïve (no BCG priming, IGRA-)
- Highly susceptible (no latency/Erdman)
- Prevent active disease from primary infection

Target population:

- IGRA neg adolescents
- Likely BCG-vaccinated at birth

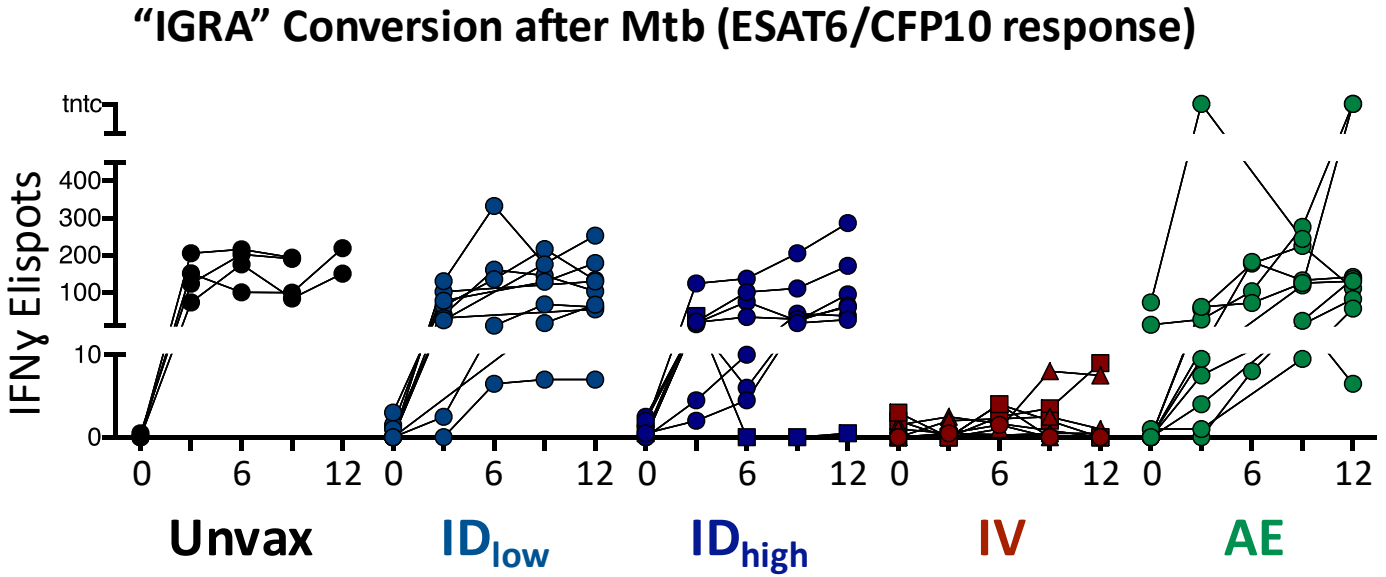


Protection against TB after IV BCG in Rhesus



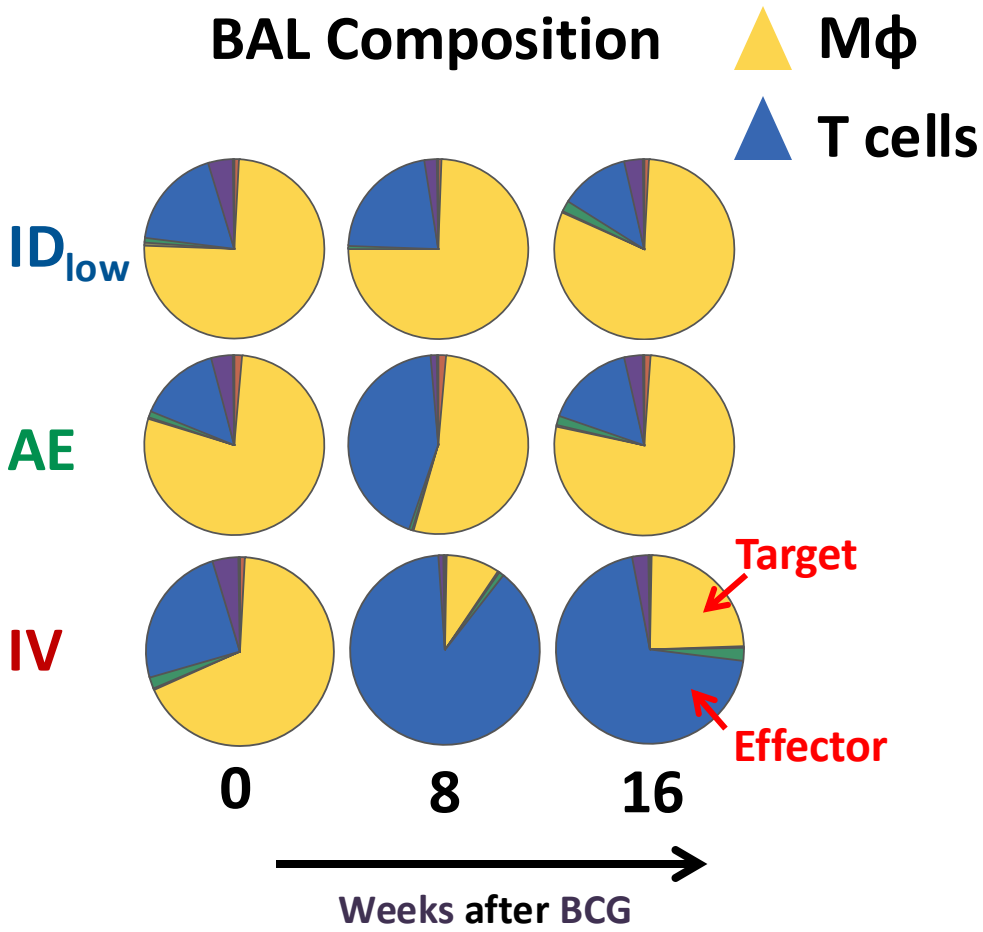
***Summary of protection after IV BCG*:**

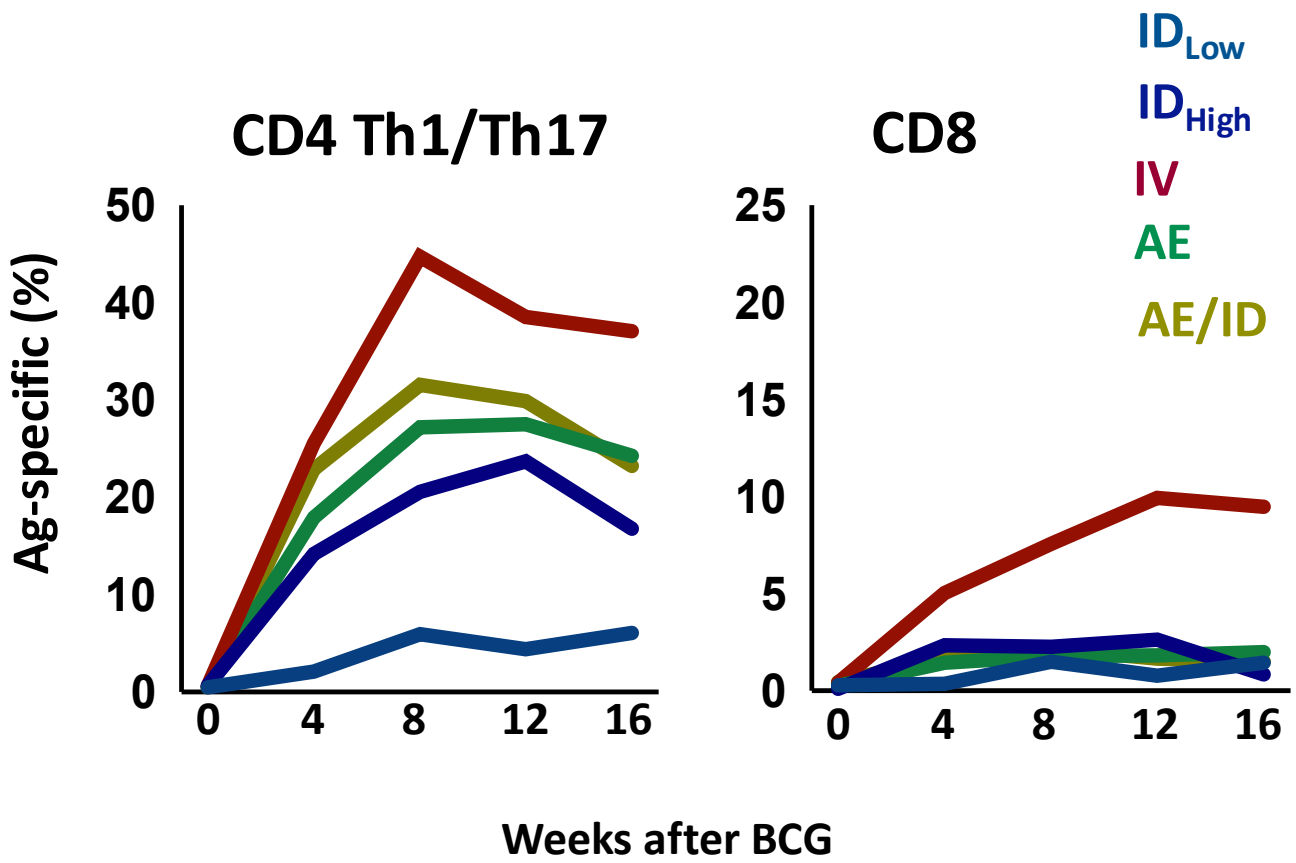
- **9/10 protected (<100 CFU)**
- **6/10 sterile (0 CFU)**



PA Darrah, et al. 2020. *Nature*. 577:95-102

IV BCG Vaccination Increases T Cells in the Airway

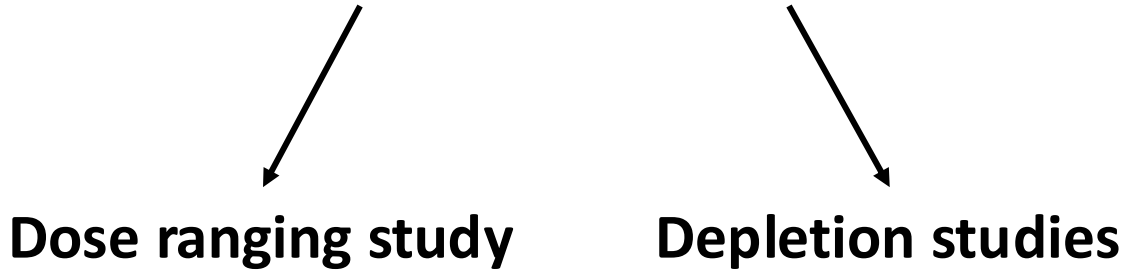




PA Darrah, et al. 2020. *Nature*. 577:95-102

Summary of BCG Route Study in NHP

- IV BCG conferred the highest level of protection in rhesus macaques
- Protection was associated with TB-specific CD4 and CD8 Trm in airway & lung
- **IV BCG provides a benchmark to study immune correlates and mechanisms of protection in a pre-clinical NHP model**



IV BCG Dose Ranging Study for Correlates Discovery

Goal:

- Force a 50% protection outcome by varying IV BCG dose

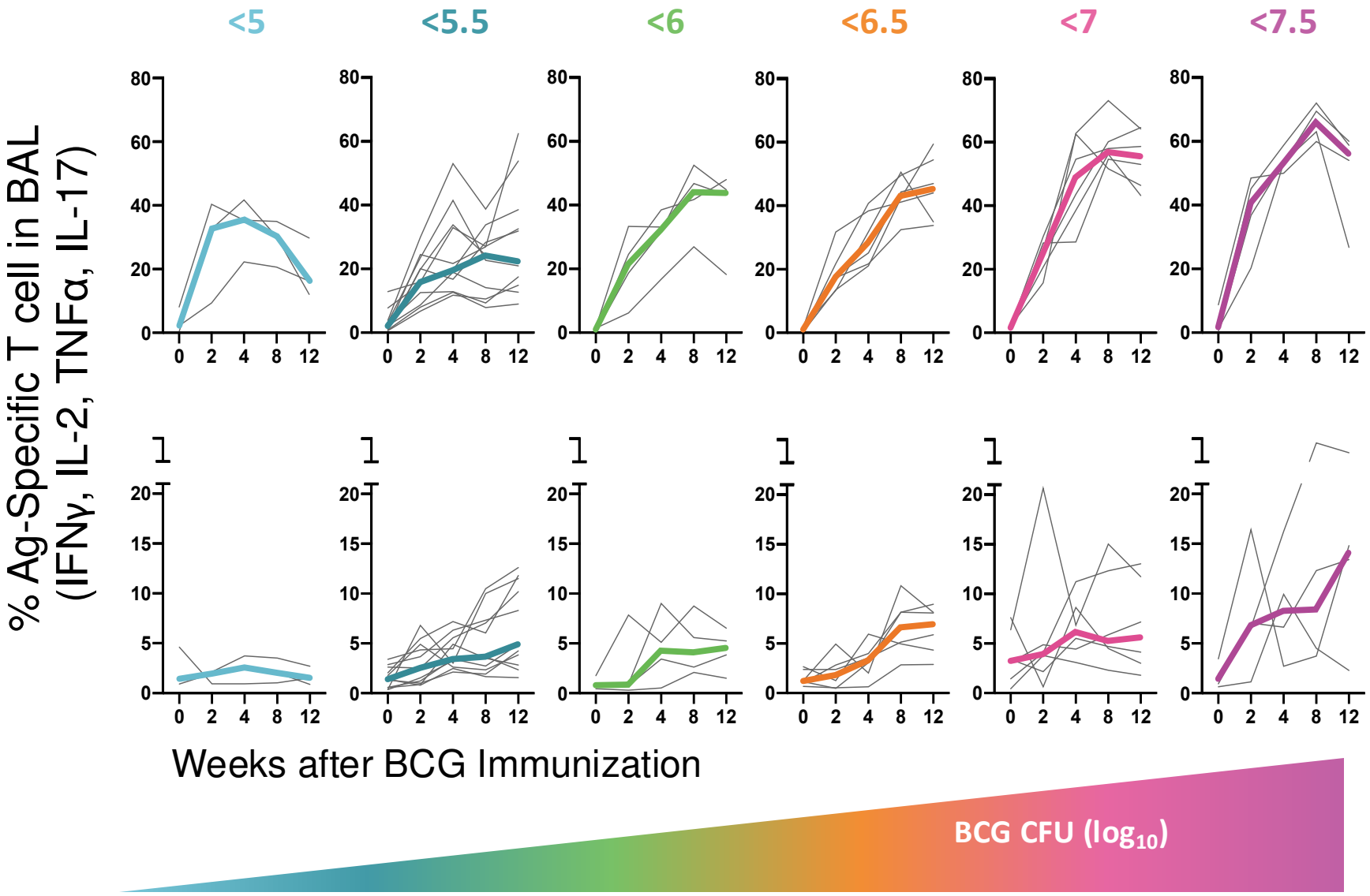
IV BCG dose (log_{10})	<5	<5.5	<6	<6.5	<7	<7.5	7.6
NHP cohort (n=34)	3	11	4	6	6	4	
Historical (n=10)							10

Challenge with Mtb Erdman (low dose) 6m later
Measure bacterial burden 12w post-challenge

PA Darrah, et al. Cell Host Microbe. 2023. 31(6):962

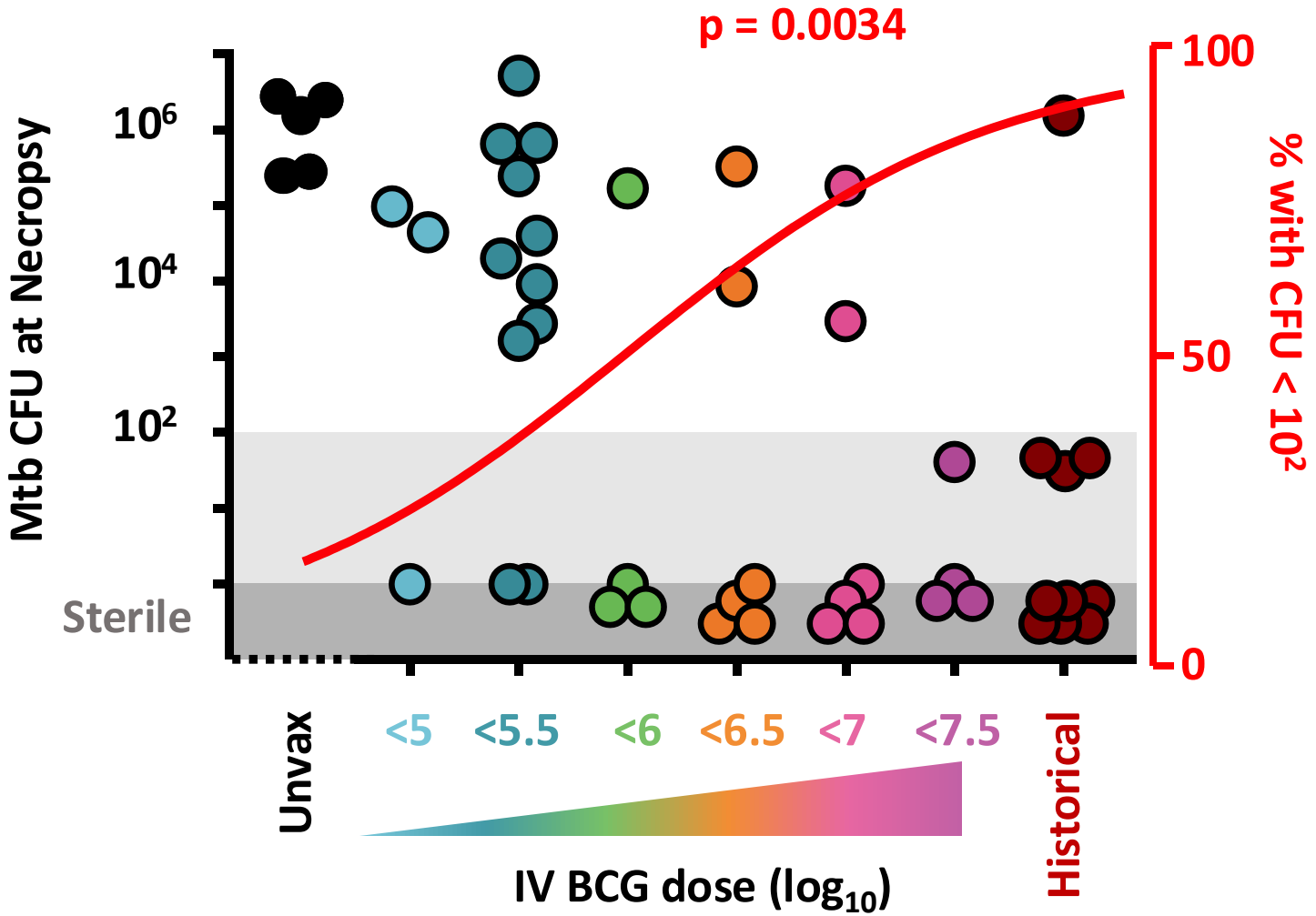
Cytokine Responses in the BAL and Blood Increase with Vaccine Dose

Cytokine Frequency in BAL



- PBMC responses
- Antibody titers

IV BCG Protects Across a Range of Doses



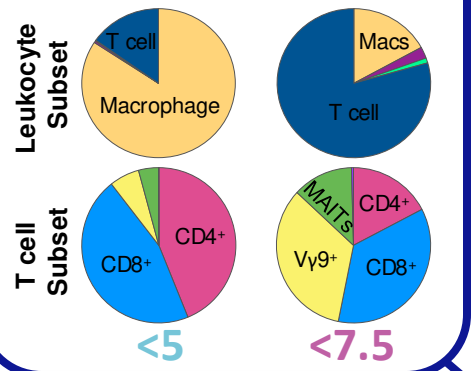
BCG Dose Study:
18/34 protected (<100 CFU)

PA Darrah, et al. Cell Host Microbe. 2023. 31(6):962

BAL and Blood Immune Parameters Fed into Systems Analysis

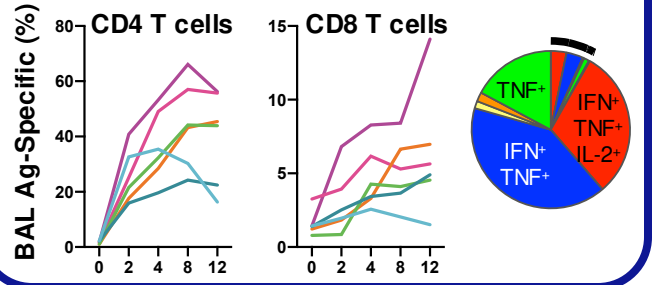
Cellular Composition (BAL & Blood)

Cell types/phenotypes



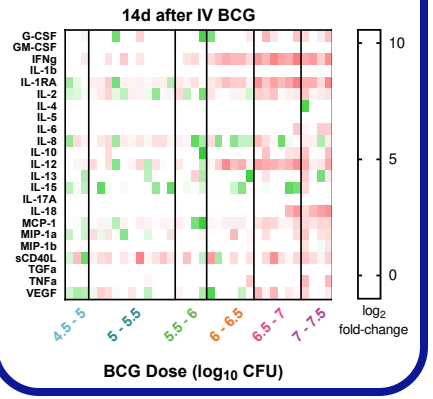
Ag-Specific T cell Responses (BAL & Blood)

CD4, CD8 Phenotype, function



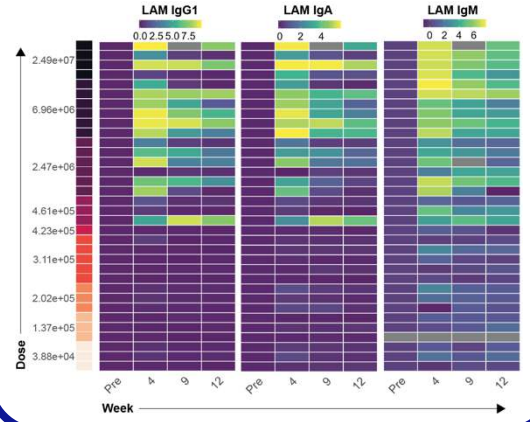
Plasma Cytokines

23-plex luminex

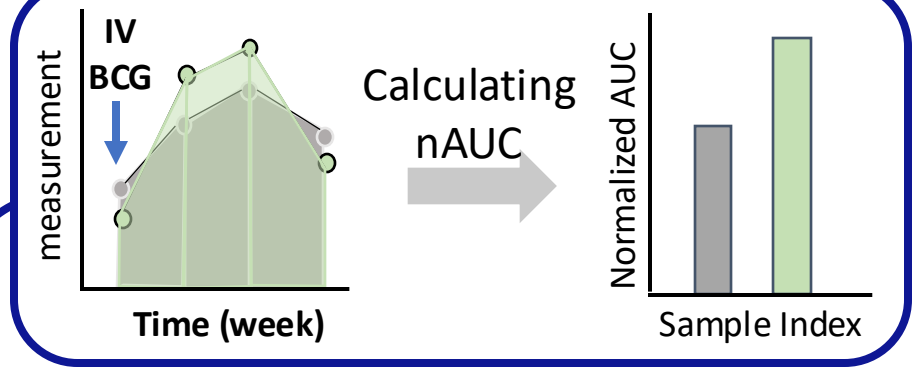


Ag-Specific Antibody Responses (BAL & Blood)

Isotypes (IgG, IgA, IgM)



~900 features



68 BAL and 83 blood immune features → protection

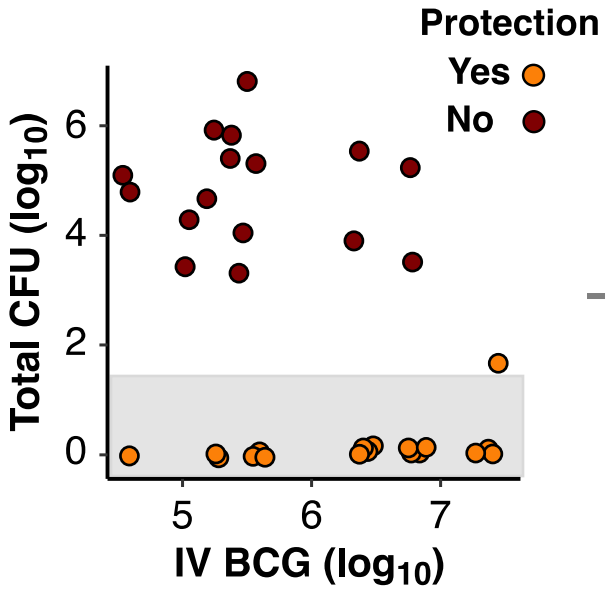
Multivariate systems analysis

Systems Serology
Eddie Irvine
Galit Alter (Ragon)

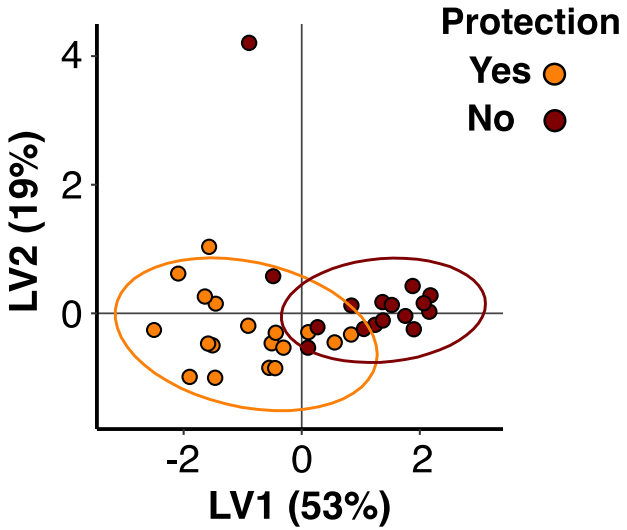
Chuangqi Wang
Doug Lauffenburger (MIT)

Multivariate Analysis to Distinguish Protection

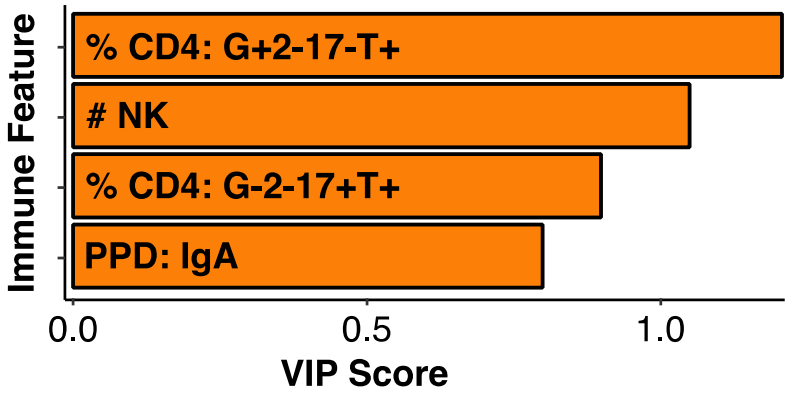
Define Protection



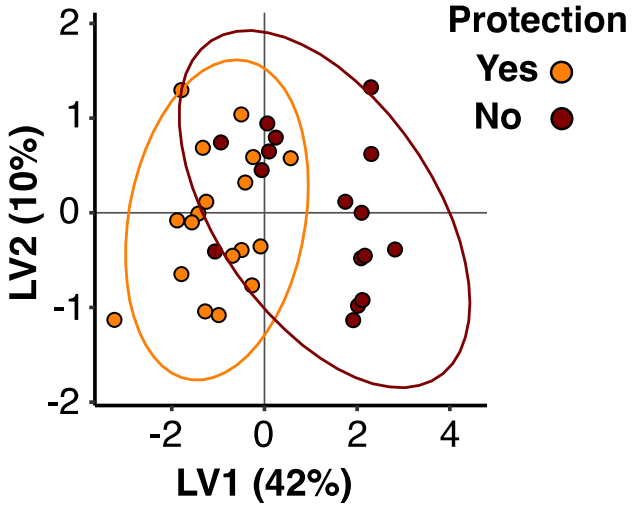
LASSO / PLS-DA

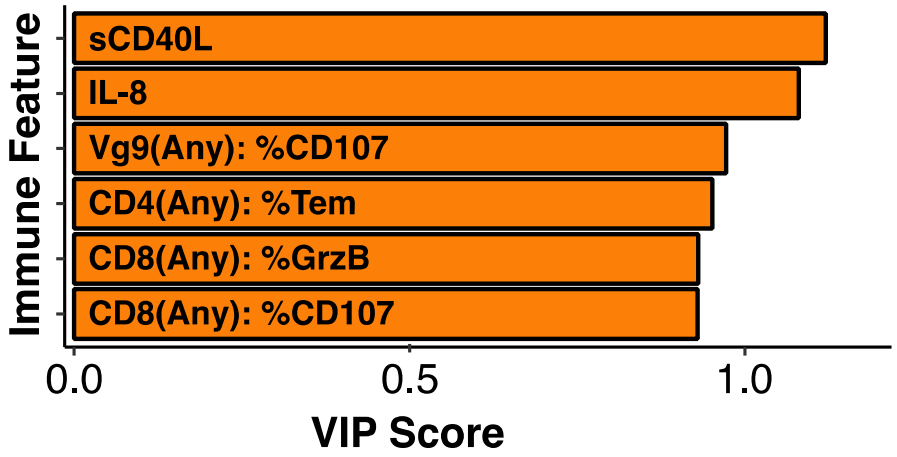


LASSO-selected features



BAL

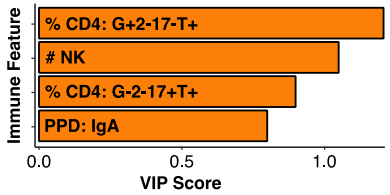




PBMC

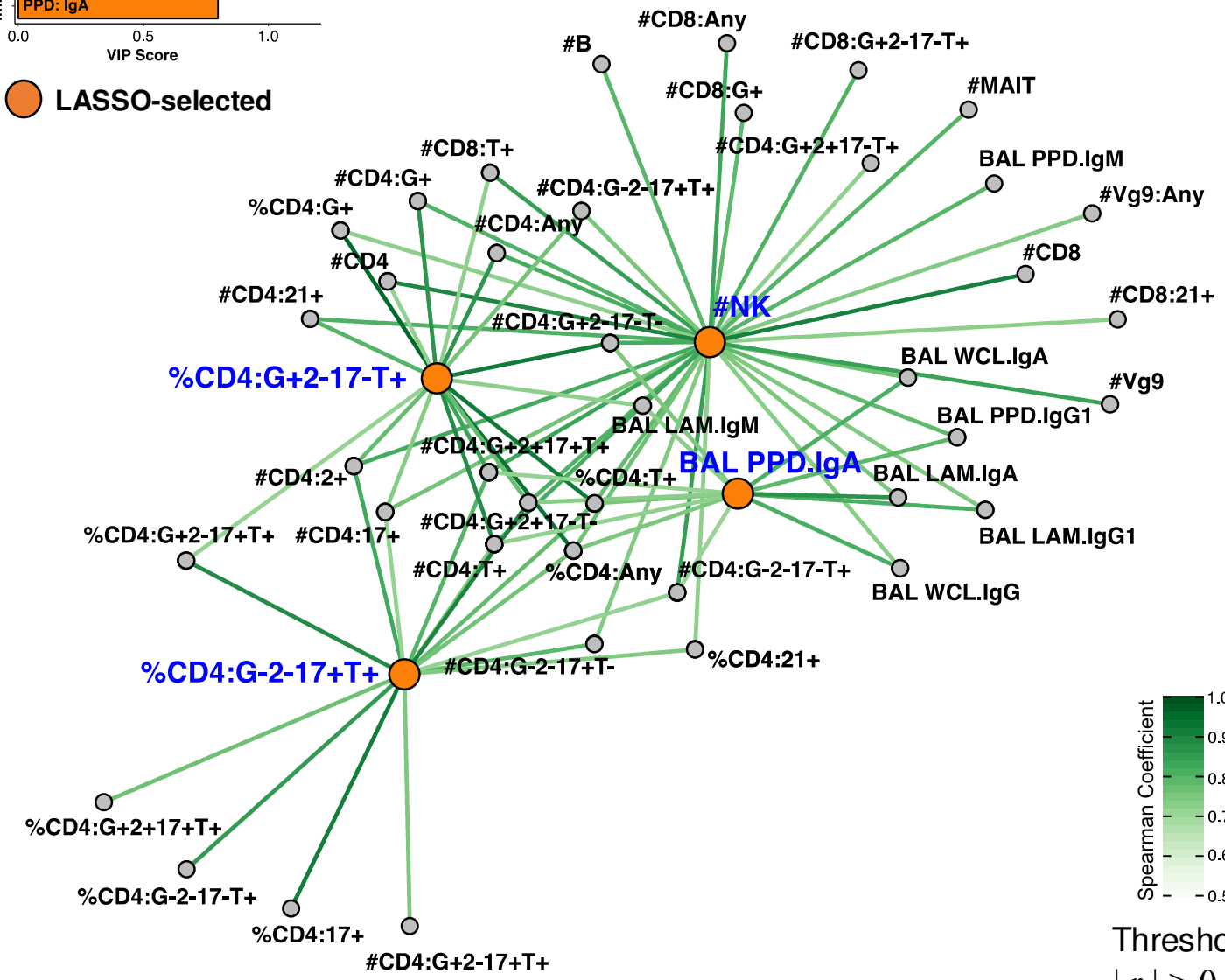
PA Darrah, et al.
Cell Host Microbe. 2023. 31(6):962

Network Analysis: Co-Correlated Immune Features



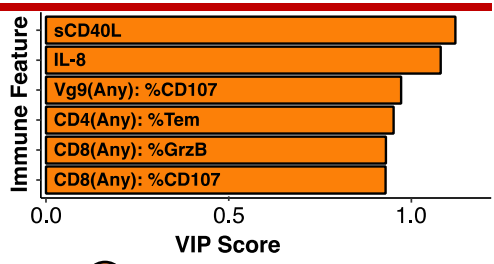
LASSO-selected

BAL

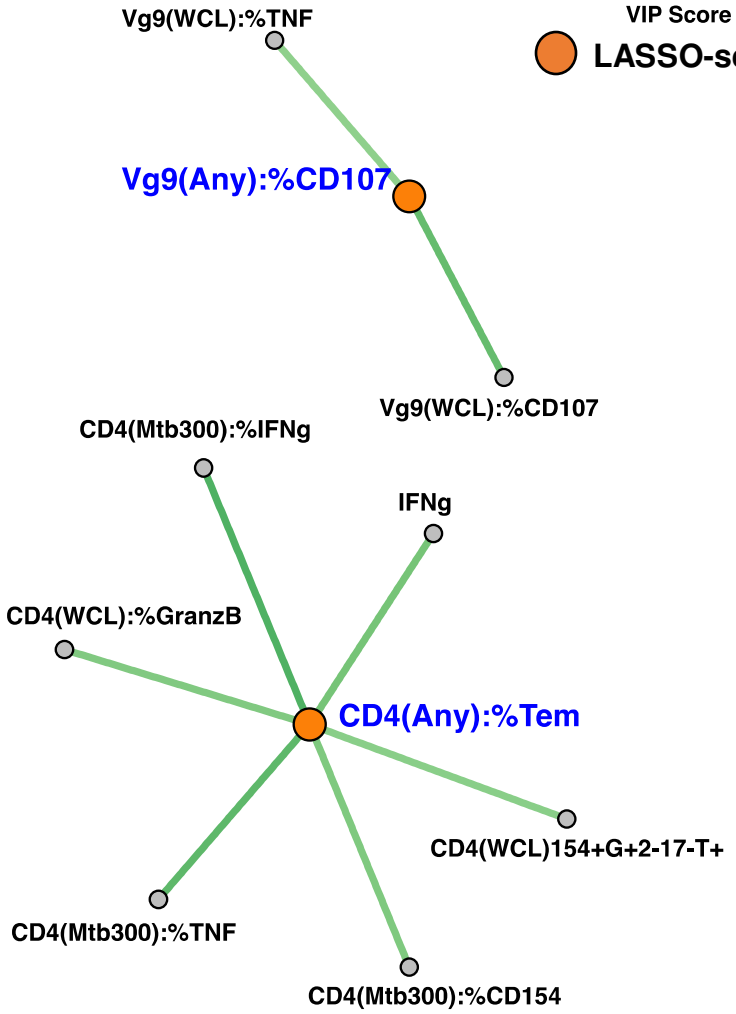


Threshold
$|r| > 0.7$

PBMC

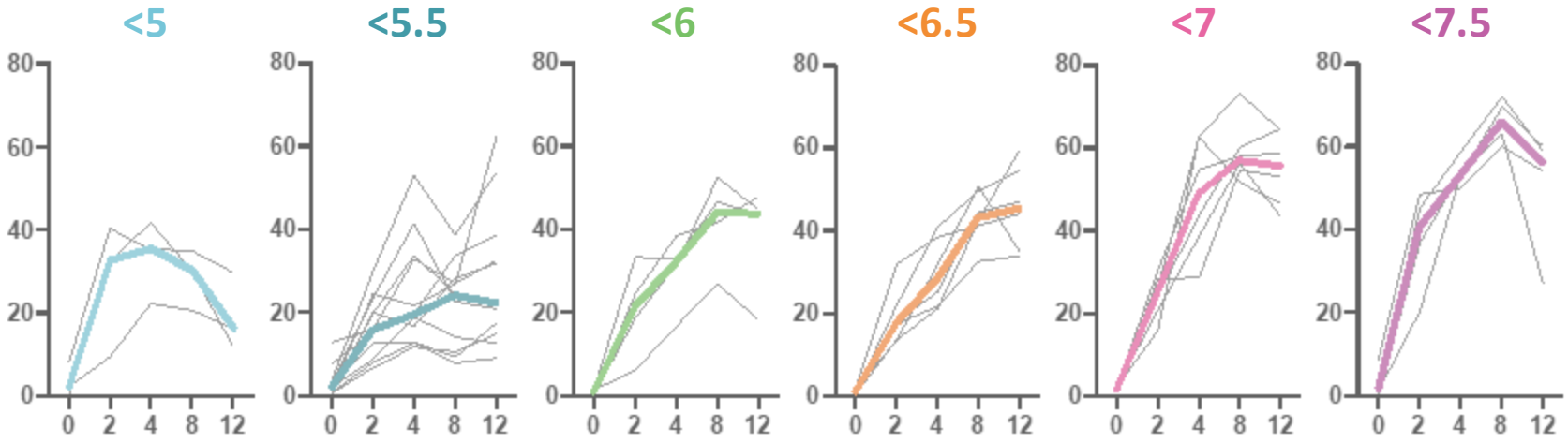


LASSO-selected



Identifying Dose-Independent Correlates

- Many immune features in BAL and blood correlate with IV BCG dose
  - *T cell responses, antibodies, plasma cytokines*

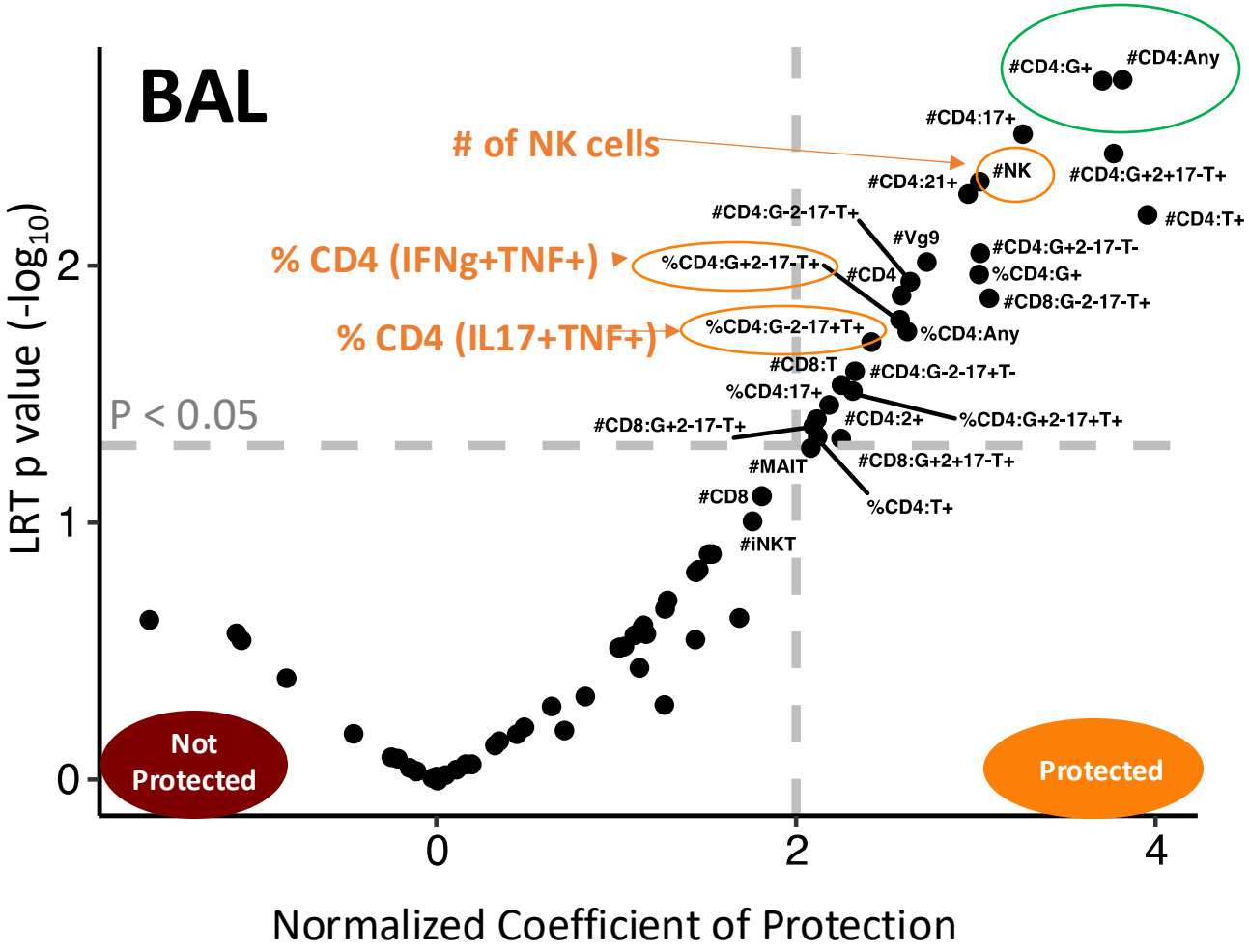


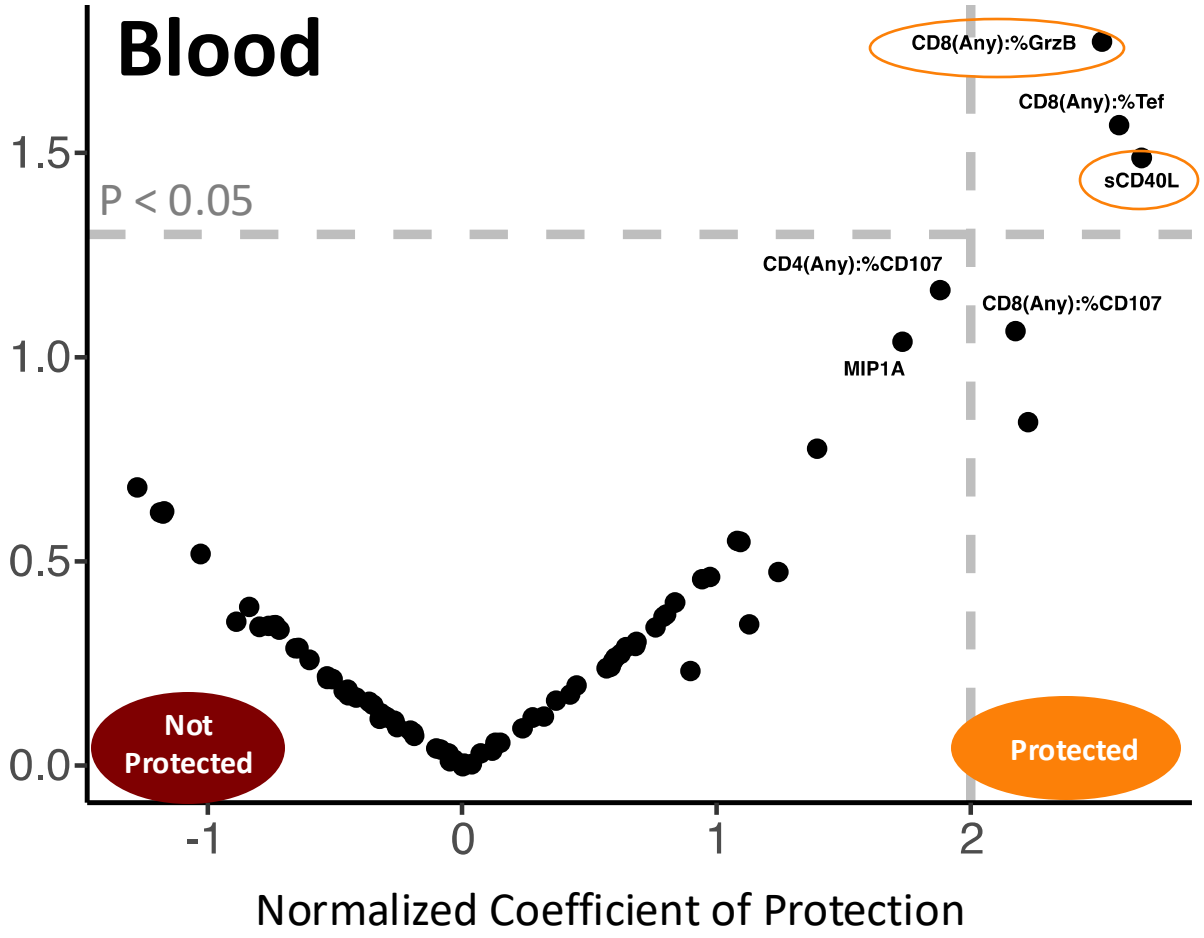
CD4 T cell responses

- Can we identify dose-independent correlates?

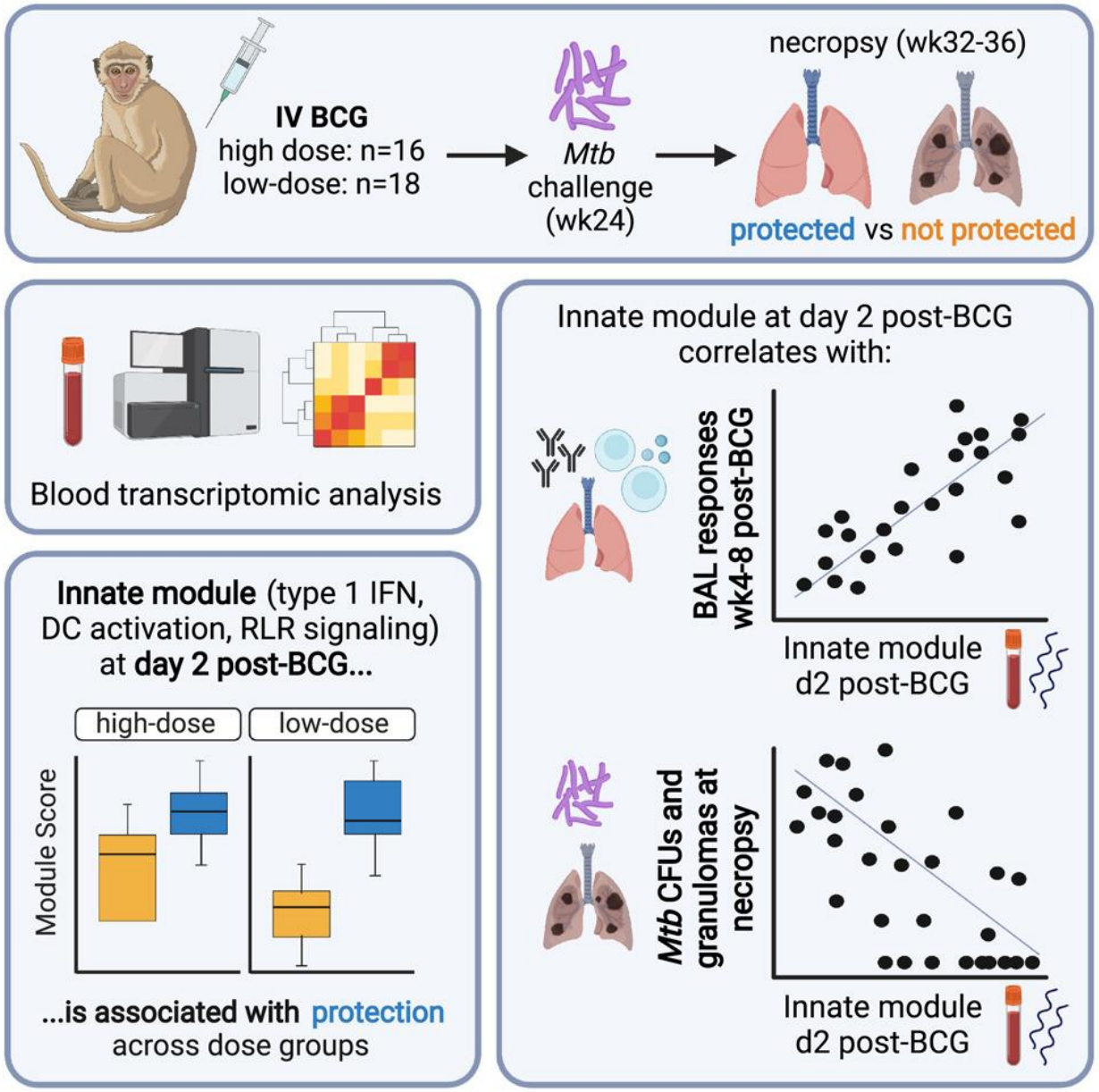
Nested Mixed Linear Model

❖ ***Corrects for IV BCG vaccine dose and animal cohort batch effects***





Blood transcriptional correlates of BCG-induced protection



YE Liu… P. Khatri.
Cell Reports Med. 2023. 4(7):101096

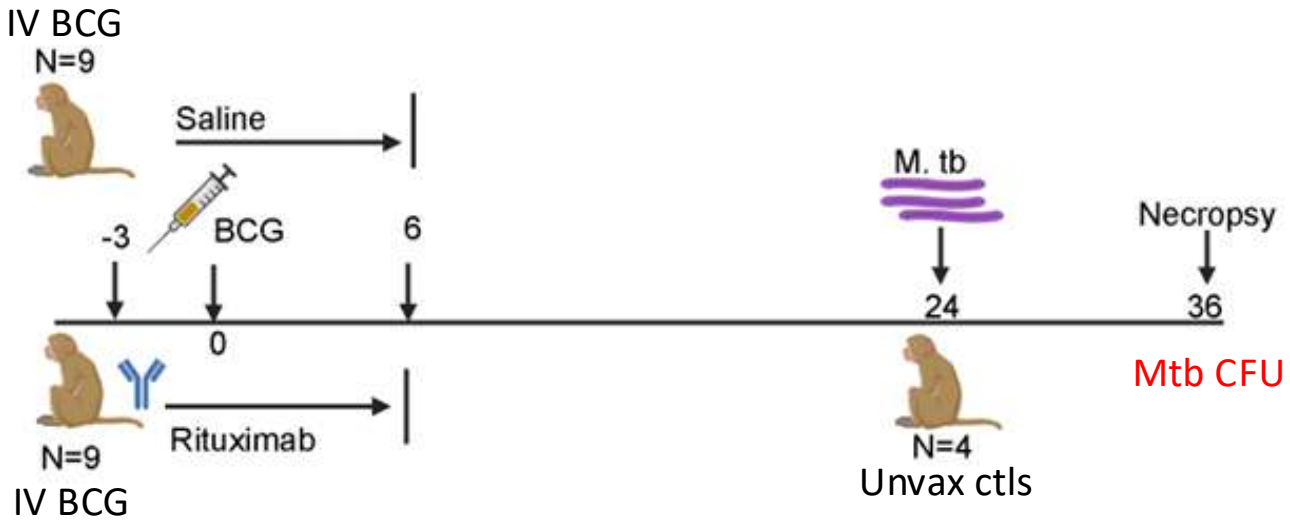
Correlates are not *necessarily* mechanisms

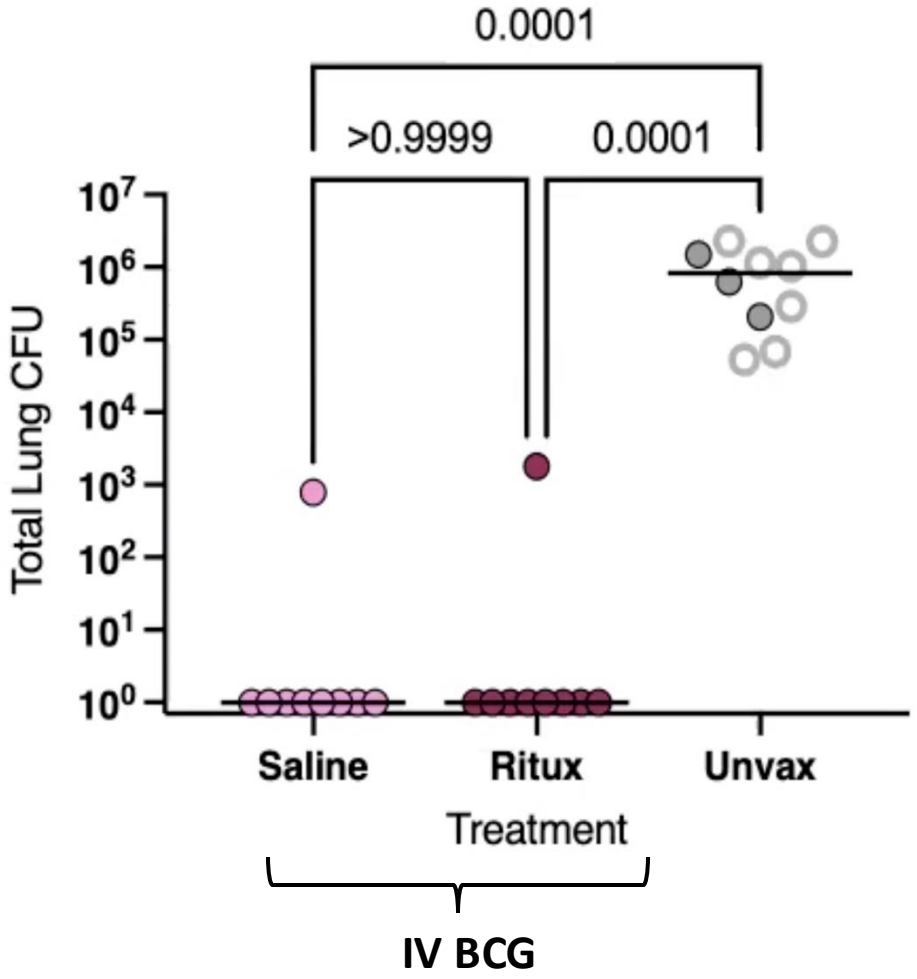
- Are antibodies a mechanistic correlate of IV BCG-mediated protection?
  Rituximab study

- Are T cells a mechanistic correlate of IV BCG-mediated protection?
  T cell depletion study

B Cell Depletion in Rhesus: Experimental Design

Depleting B cells using Rituximab (anti-CD20) during IV BCG immunization-- limits antibody response



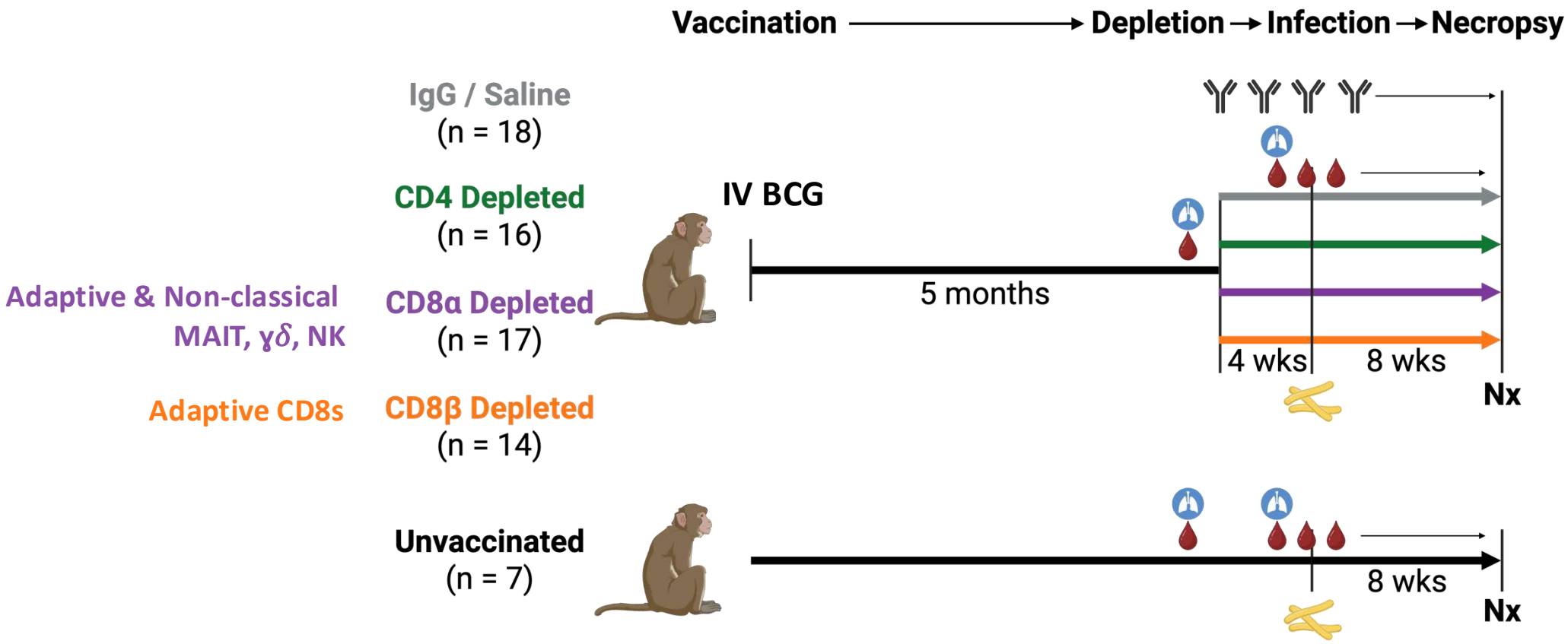


Flynn Lab (Pitt) & Lauffenburger Lab (MIT)
Wang, Myers, Irvine, et al. BioRxiv. 4/16/2024

❖**B cell depletion did not impair protection conferred by BCG IV**

T cell Depletion in Rhesus: Experimental Design

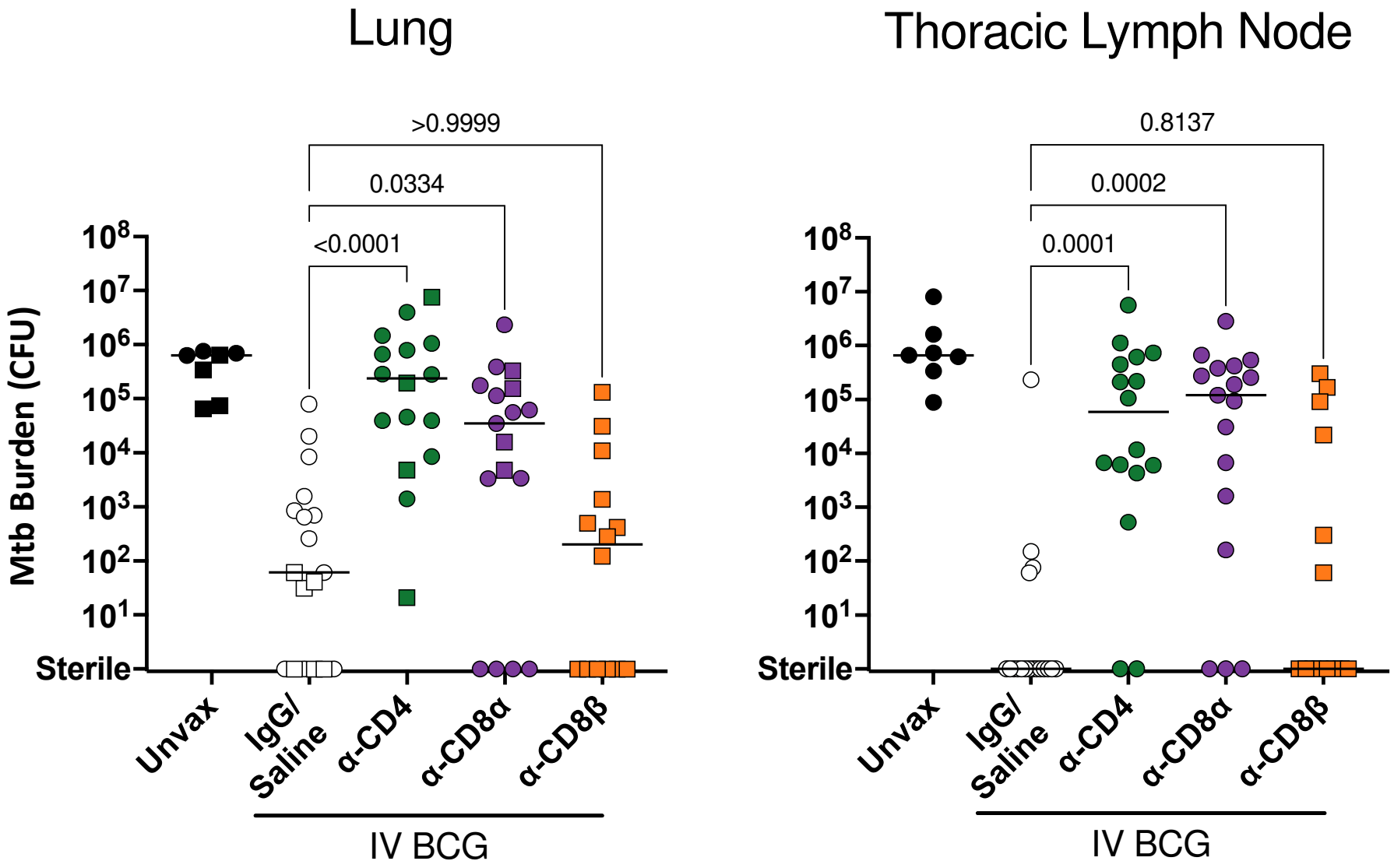
Depleting T cells AFTER IV BCG immunization but BEFORE Mtb challenge;
Limits T cell response, preserves Ab response



NIH VRC, U.Pitt (Flynn) & MIT (Fortune)
Simonson, Zeppa, Bucsan, et al. BioRxiv, 5/17/2024

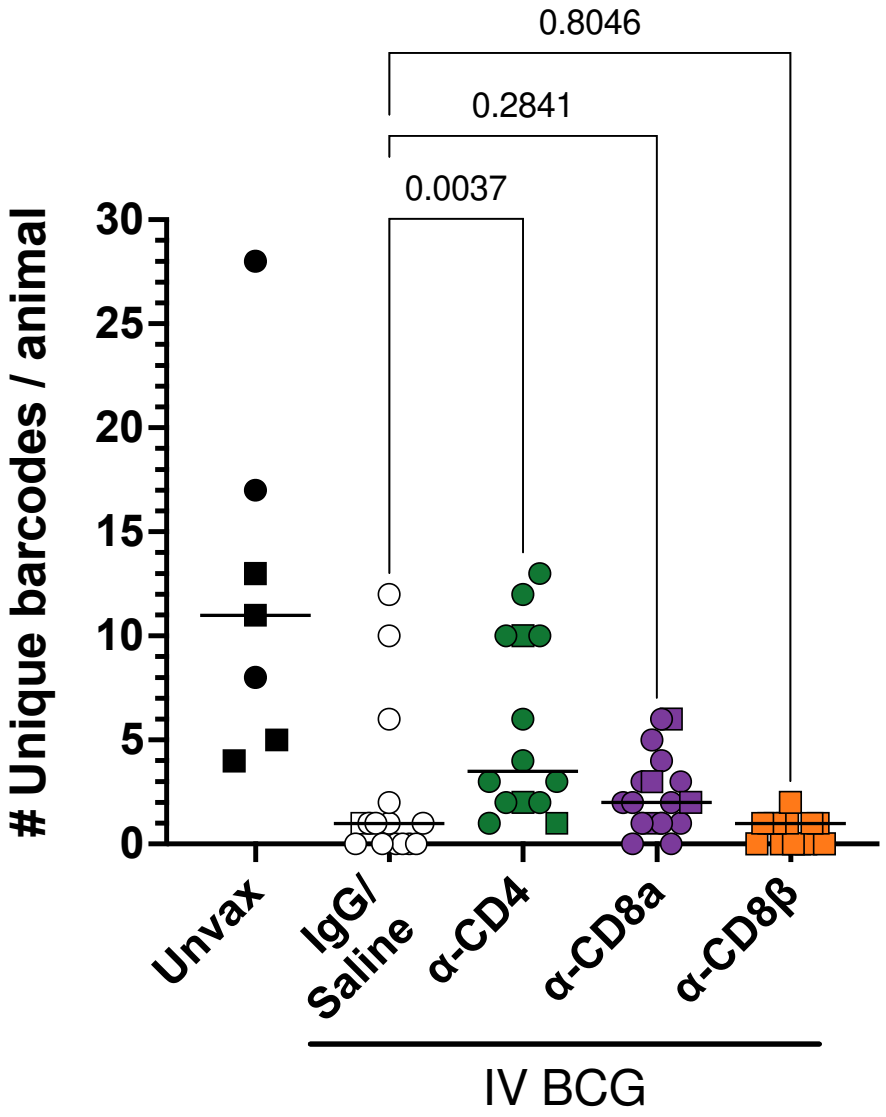
Andrew Simonson (Pitt)
Allison Bucsan (VRC)

T cell depletion: Mtb Burdens



❖ **CD4 or CD8a depletion after IV BCG abrogates protection**

T cell depletion: Mtb Barcode Data

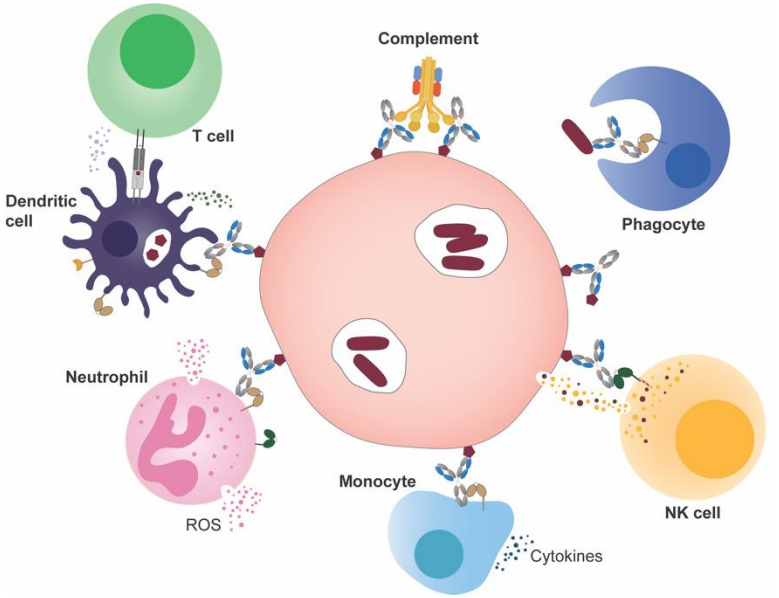


Michael Chao
Sarah Fortune
(Harvard)

❖ **Restriction of Mtb establishment is maintained in absence of T cells**

Mechanisms of IV BCG-Mediated Protection

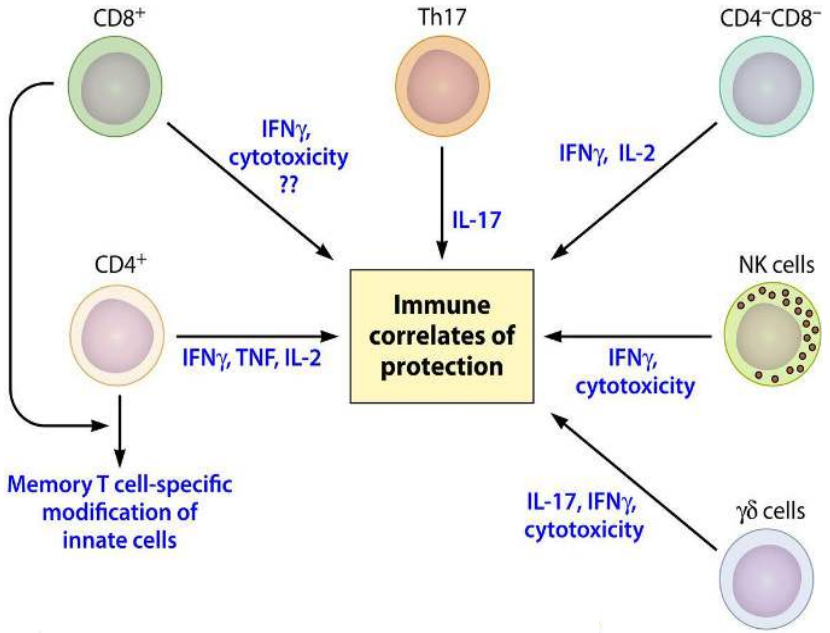
Antibodies



B cell depletion
No effect on IV BCG protection

Wang, Myers, Irvine, et al. BioRxiv. 4/16/2024

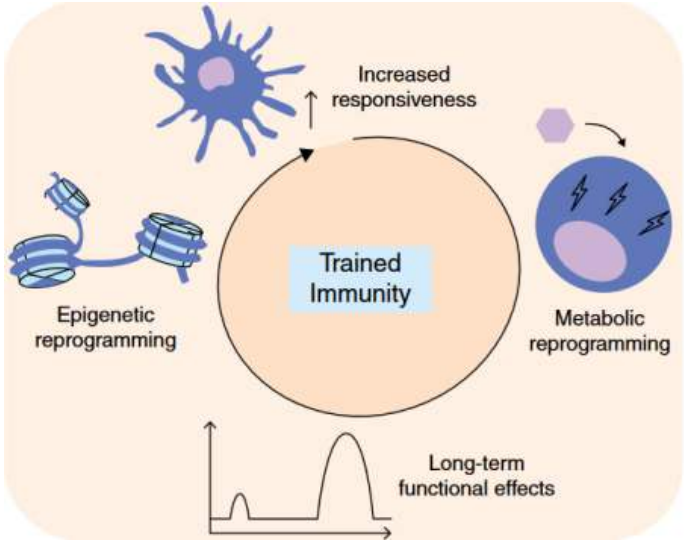
T Cells



CD4 depletion = no protection
CD8a depletion = less protection

Simonson, Zeppa, Bucsan, et al. BioRxiv, 5/17/2024

Trained Immunity



Mechanism (?)
Evidence in mouse model
Signatures in rhesus

Vierboom, et al. Cell Rep Med. 2021

Modified from Bhatt et al. 2015. Clin. Vaccine Immunol

Does IV BCG Inform Future Vaccine Strategies?

IV BCG

- Systemic immunity
- Self adjuvanted (innate immunity)
- Diverse immune responses (cellular, humoral, trained)
- Broad antigen repertoire (~4000 proteins + lipids)
- Durability: persistence maintains memory

How can studying IV BCG inform future vaccine design?

- Delivery: achieve robust lung immunity with better AE delivery
- Safety: use more attenuated strains of BCG (rBCG, auxo, irradiated, kill-switch)
- Antigens: map responses from IV BCG animals to discover new T cell antigens
- Mimic training signals to combine with subunit vaccines (mRNA, viral vectors)

Acknowledgments

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Alex Smith
Philana Ling Lin
Pauline Maiello
Joe Zeppa
Jake Borish
Mark Rodgers

VRC TRP
JP Todd
Ruth Woodward

VRC Flow Core
David Ambrozak

VRC Seq Core
Amy Ransier
Farida Laboune

Harvard/MIT
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Chuangqi Wang

Sarah Fortune
Michael Chao

Galit Alter
Edward Irvine

Stanford Bio-X
Purvesh Khatri
Yiran Liu

BMGF
Alison Kraigsley
Ann Ginsberg
Karen Makar