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INNOVATION AND ACCESS

# RTS,S Malaria Vaccine: *Toward an understanding of the immunologic basis of protection*

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PATH Malaria Vaccine Initiative  
March 21, 2018

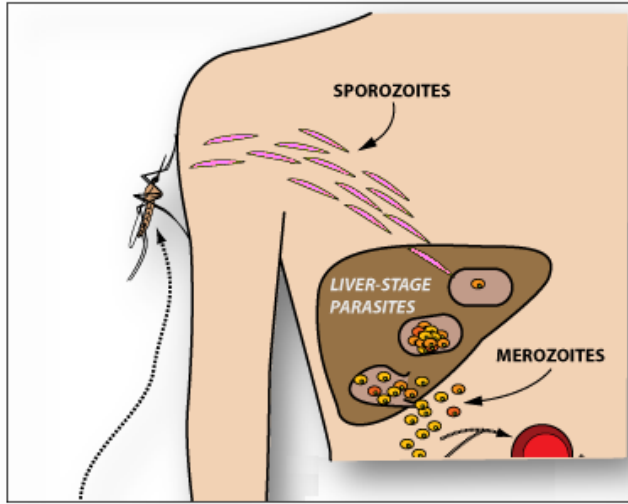


PATH/Doune Porter

# Outline

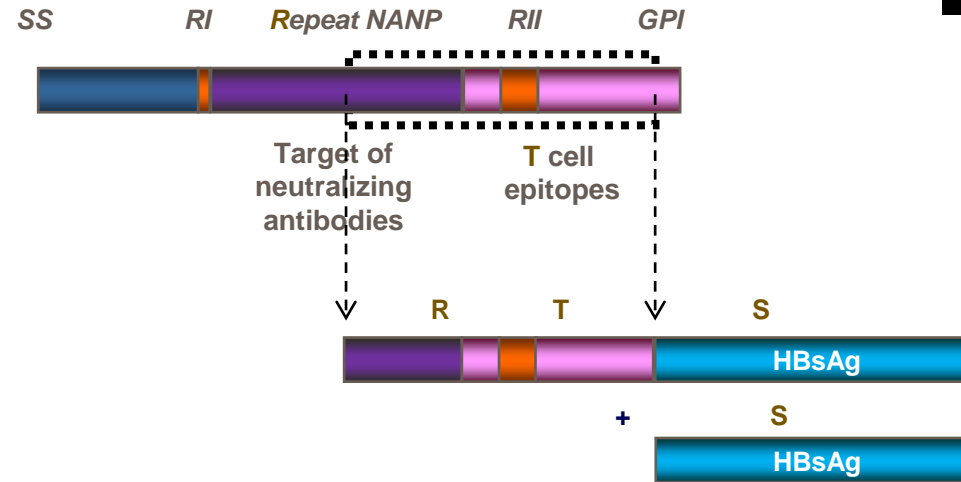
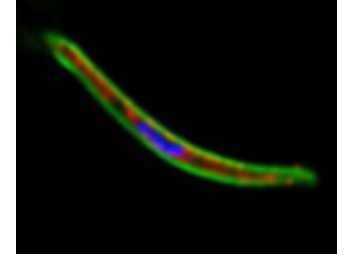
- 1) Biology of circumsporozoite protein in malaria infection
- 2) Immunologic correlates from Controlled Human Malaria Challenge Trials (CHMI)
- 3) Immunologic correlates from field trials under natural *P. falciparum* transmission
- 4) Opportunities for improvements and alternative strategies

# RTS,S antigen and Adjuvant Systems



## The Circumsporozoite Protein:

- ✓ is the major surface protein of the sporozoite, also expressed by early liver forms
- ✓ Liver entry function



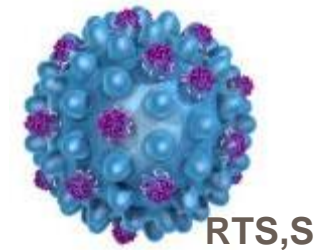
## The **RTS,S** vaccine particles:

- ✓ The **R** and **T** regions from CSP are fused to the Hepatitis B **S**urface protein (HBsAg)
- ✓ The fusion protein is co-expressed with HBsAg in yeast (*Saccharomyces cerevisiae*) where they spontaneously assemble into mixed particles

## The **AS01** Adjuvant System:

- ✓ Adjuvant System designed to induce strong antibody and Th-1 cell mediated immune responses
- ✓ Consists of MPL, QS21 and liposomes

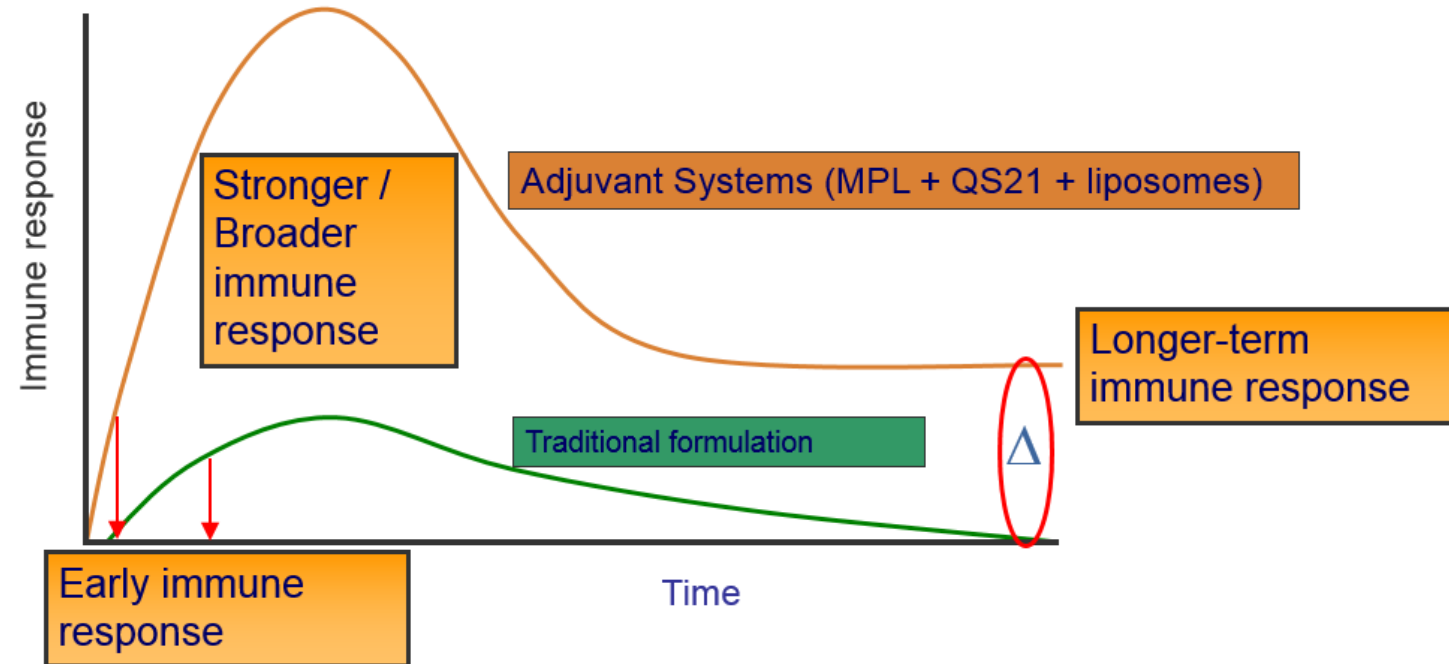
Gordon et al. *J Infect Dis* 1995;171:1576–85



# Immunologic principles of RTS,S-induced protection

## Preferred Performance Characteristics

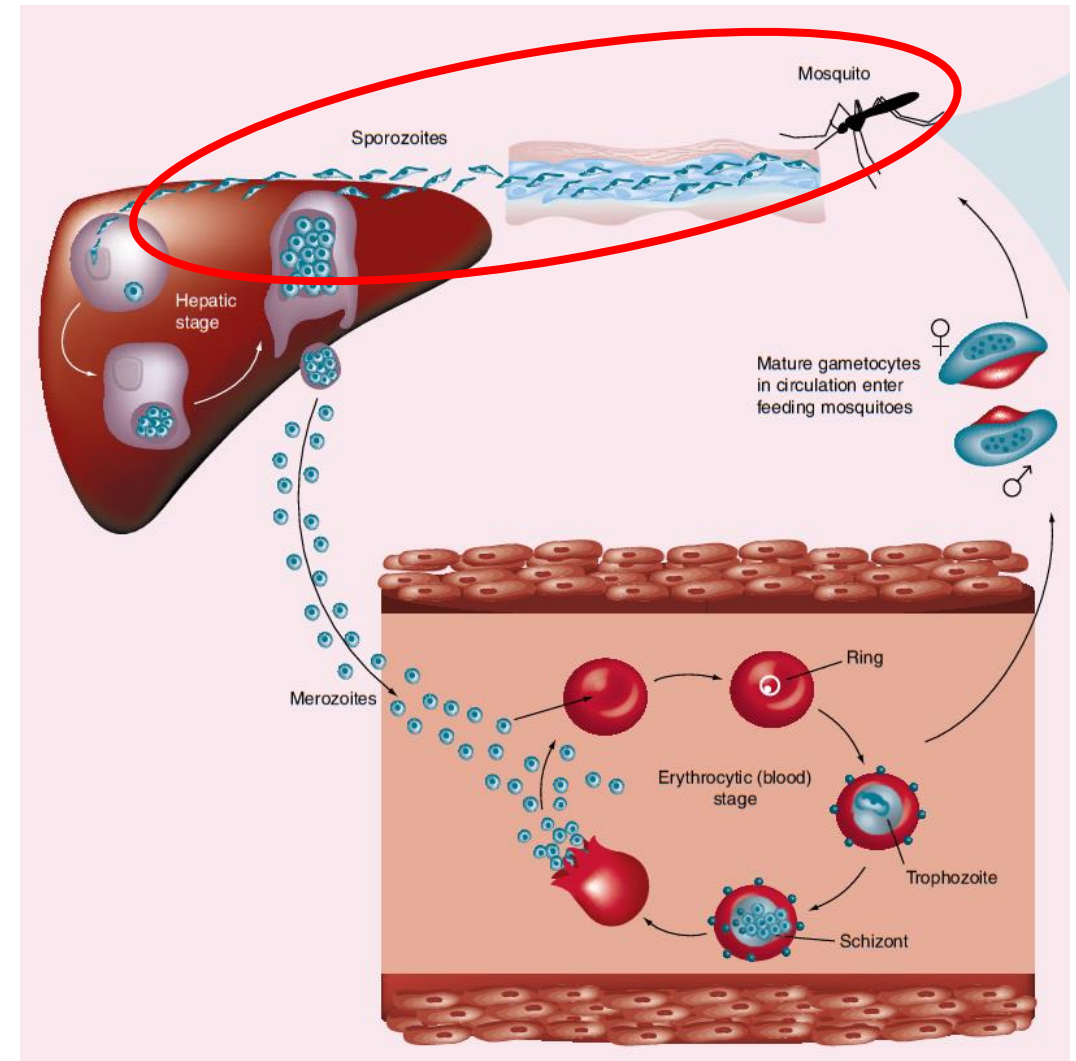
- Rapid onset and durable immunity
- Protection across age groups
- Protection across different epidemiologic settings
- Elicit protective humoral and cell-mediated immune responses
- Immunologic recall
  - ❖ *(Achilles heel of RTS,S)*



Pulendran B & Ahmed R. *Cell* 2006;124:849–863

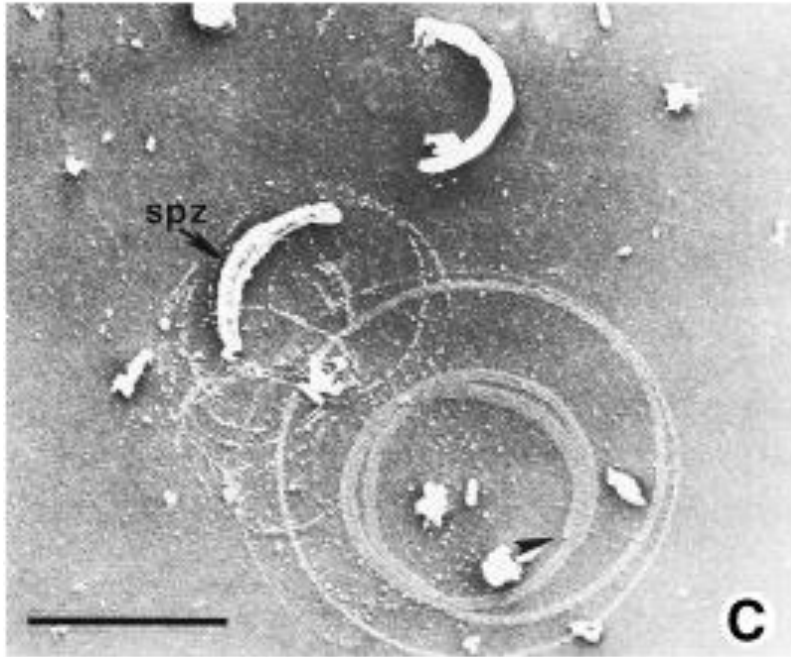
# Biology of the parasite informs the technical strategy

1. There is limited time to neutralize sporozoites before they invade liver hepatocytes
2. Effector cells and/or immunoglobulins need to be present at site of infection and at time of exposure to block infection
3. Insufficient time for anamnestic response



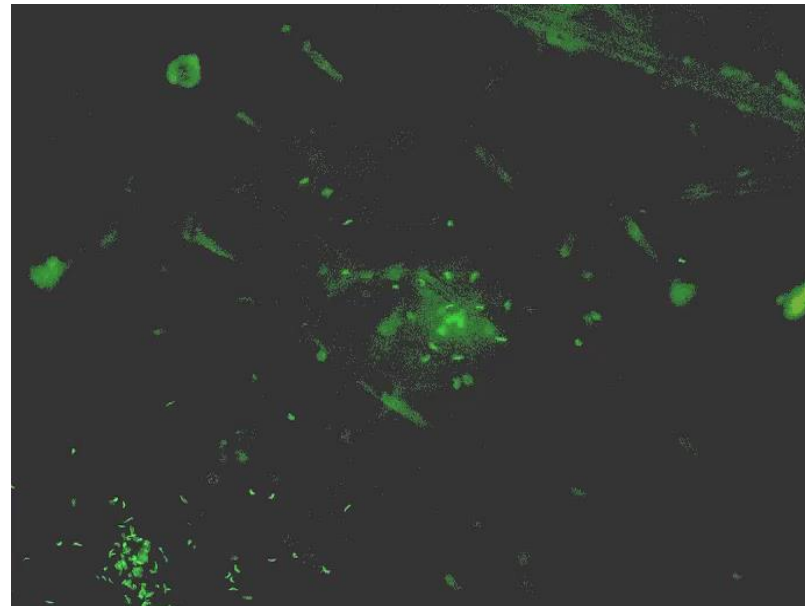
# Sporozoite (Spz) biology

- Spz are motile
- Motility essential for invasion
- Stopping motility prevents infection
- Spz are injected by the mosquito into the dermis by probing



Vanderberg, JP. Parasitol Intl. 2014: 63:150-164

## Intravital microscopy



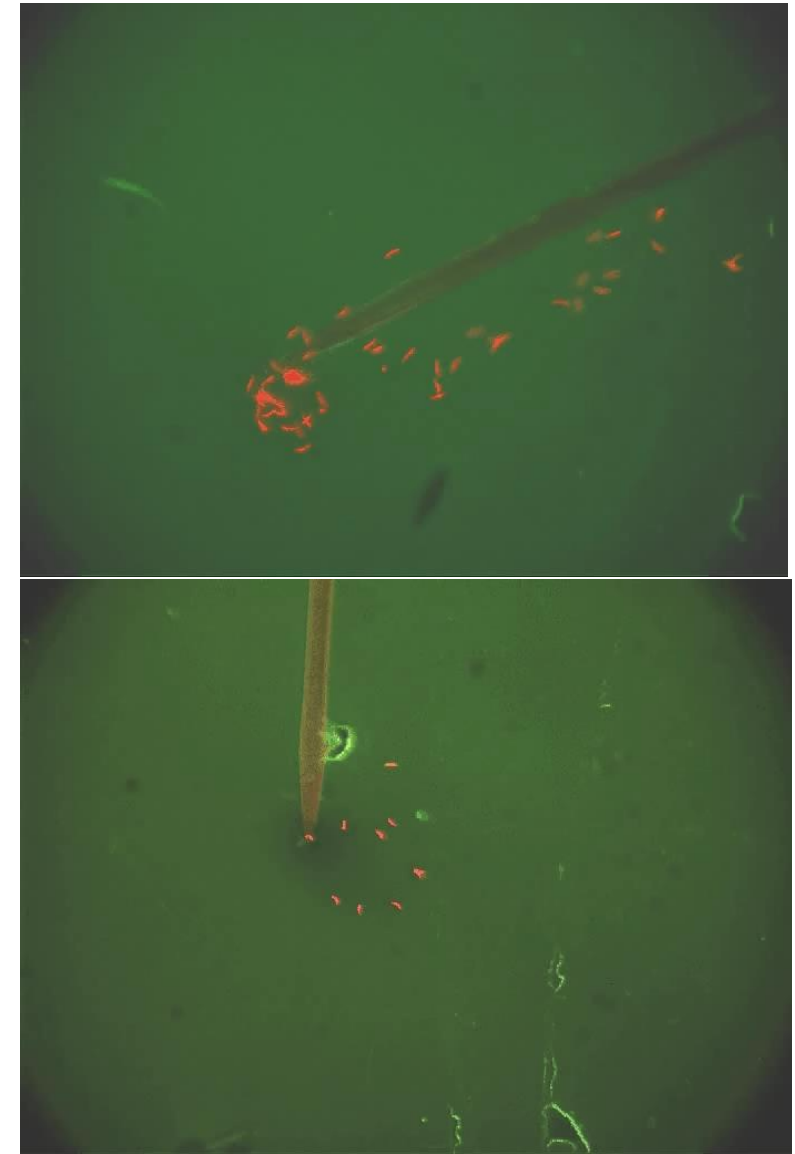
# Anti-CS immune sera in vitro

1. In vitro exposure of spz immune sera or anti-CSP sera result in “precipitin” called “CSP reaction”



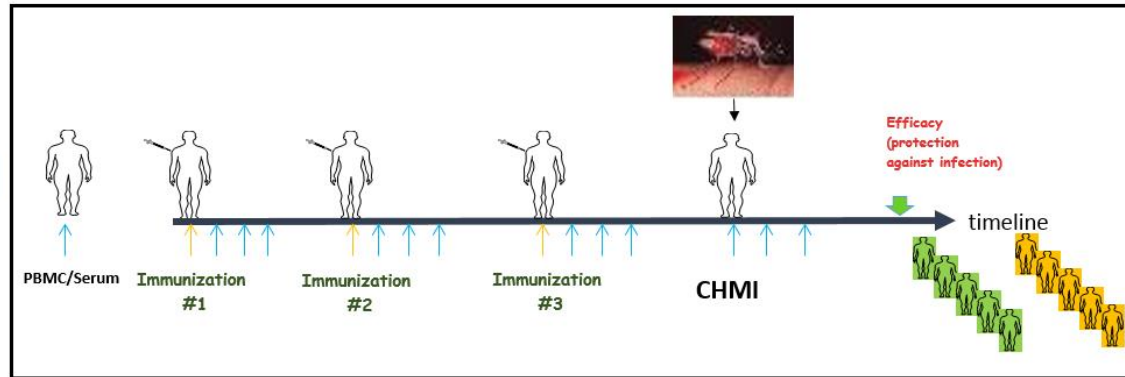
Vanderberg, JP. Parasitol Intl. 2014: 63:150-164

2. Blockage of Spz transmission in presence of immune sera



# Vaccine development: “the best correlate of protection is protection”

Controlled Human Malaria Infection (CHMI) & Field testing; an iterative process



## Upside

- ✓ De-risk clinical development

## Downside

- ✓ Making wrong assumptions
  - Age groups differences
  - Epidemiologic differences
  - Force of infection
  - Host genetic diversity
  - Parasite genetic diversity
  - Dose and schedule variation



# Immune correlates of protection/risk

Knowledge  $\Rightarrow$  Understanding (mechanistic)

Knowledge  $\Rightarrow$  Understanding  $\Rightarrow$  Action (predictive biomarker)

Ab titer  
CMI-cytokine  
Tfh cell analysis  
CMI –ELISPOT  
Ab function – phagocytosis  
Ab repertoire  
CMI-proliferation  
Immunofluorescence  
Systems biology (microarray/RNAseq)  
Systems immunology ((Fc receptor+ immune cells  
Ab gliding motility  
CMI-cytotoxicity  
Ab isotype/subclass  
Complement fixation  
B cell analysis  
Ab function – ILSDA/traversal  
Non-CS surrogates of protection  
Ab in vivo small animals  
Ab avidity/affinity  
CMI-Cytof  
CMI-ICS

X

NANP repeat domain  
C-terminus (Th2R&Th3R)

Computationally  
complex requiring  
integrated analysis

Reproducible

Robust

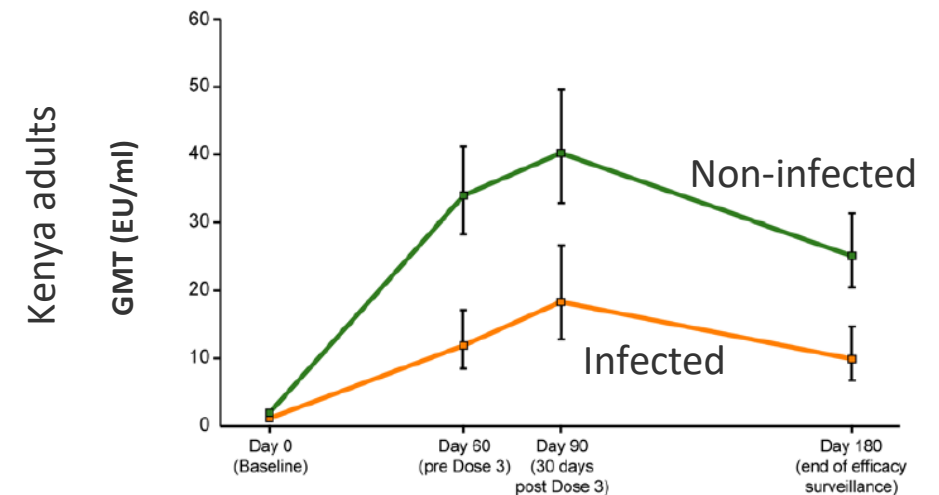
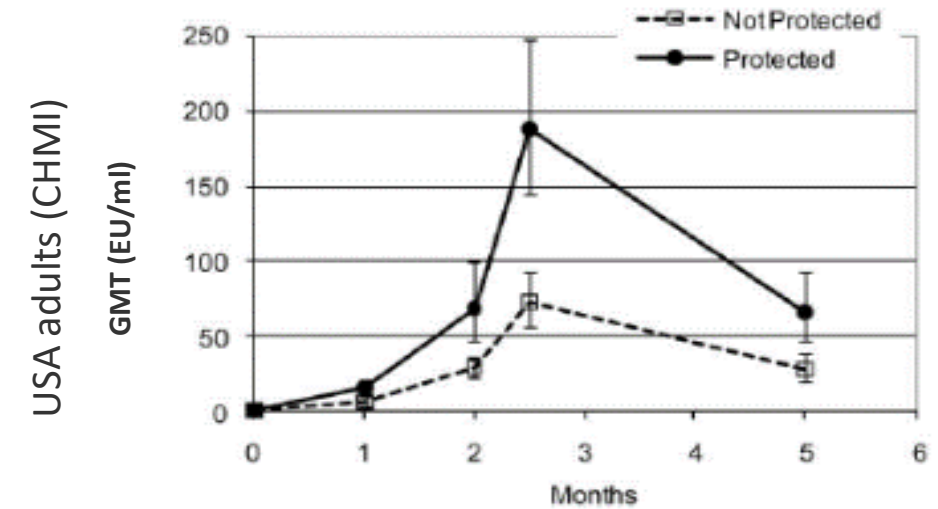
Feasible

# Anti-CS GMT to NANP repeat domain correlates with protection

*“Because that’s where the money is”*

Slick Willie Sutton, the Bank Robber

- Magnitude of anti-CS repeat matters – Higher levels of anti-NANP antibodies are associated with more protection
- Reproducibility across several CHMI studies
- Dependent upon specific vaccine regimen
- **There is no antibody level that reliably predicts protection above a defined threshold**

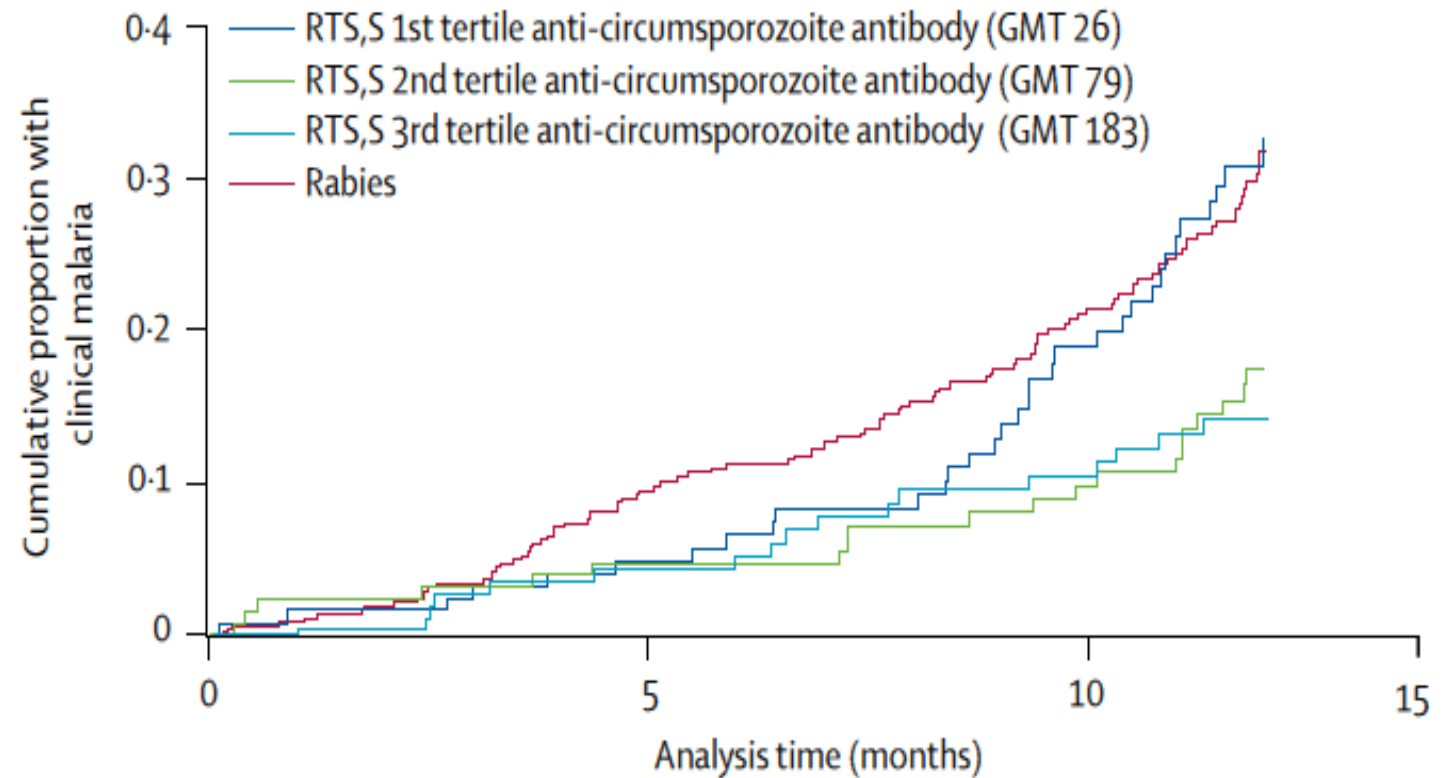


Kester. et al. JID 2009; 200: 337-46 Polhemus. et al. PLoS One 2009; 4: e6465

# Antibody titer to CS NANP repeat correlate with protection in children

- Reproducibility across studies (Phase 2, Phase 3)
- Reproducible across different epidemiologic settings

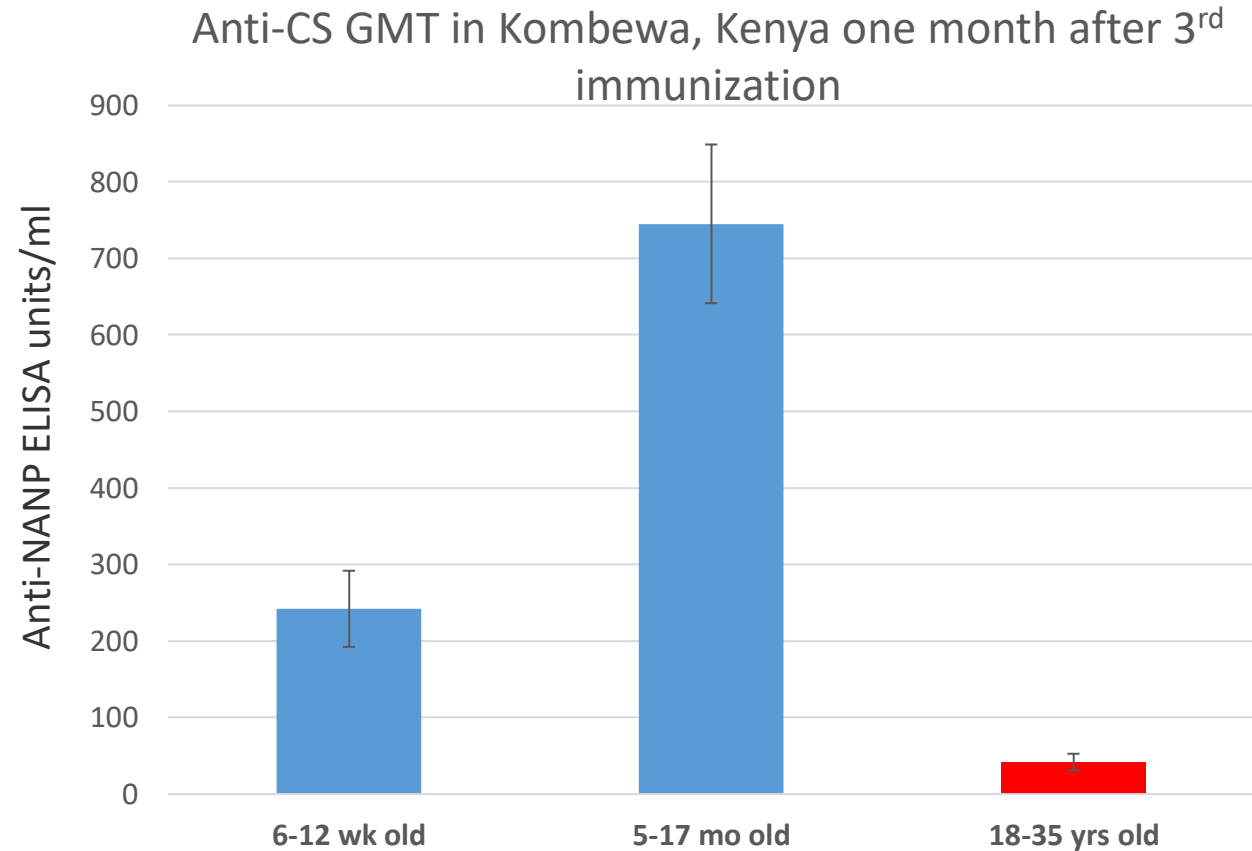
Phase 2 study in African children 6-17 mo old (Kenya & Tanzania) Stratification by titer



Olutu et al Lancet Inf Dis. 2011; 11:102-109

# Antibody GMT in infants and children compared to adults in Sub-Saharan Africa

- Age-specific differences in antibody titer
- Low antibody levels in adults with prior exposure to *P. falciparum* infection
- Immune hyporesponsiveness due to concurrent subclinical blood-stage infection (?)

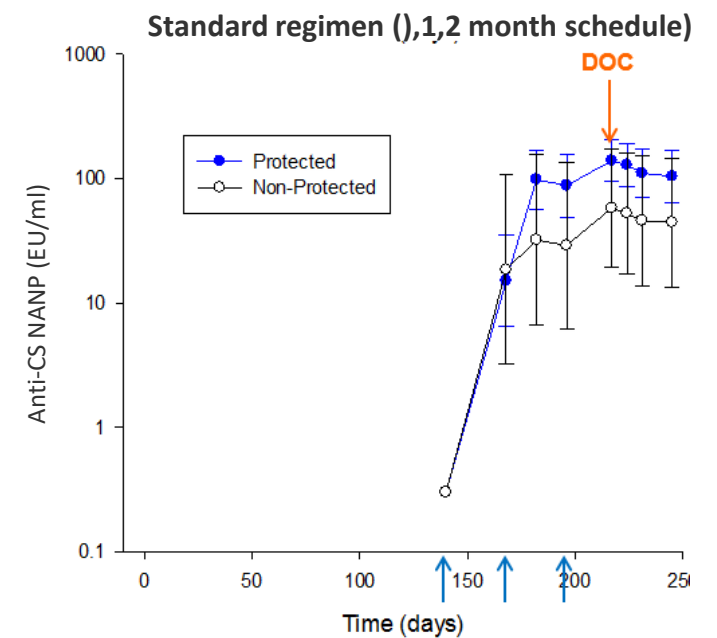


# Strategies to improve vaccine efficacy:

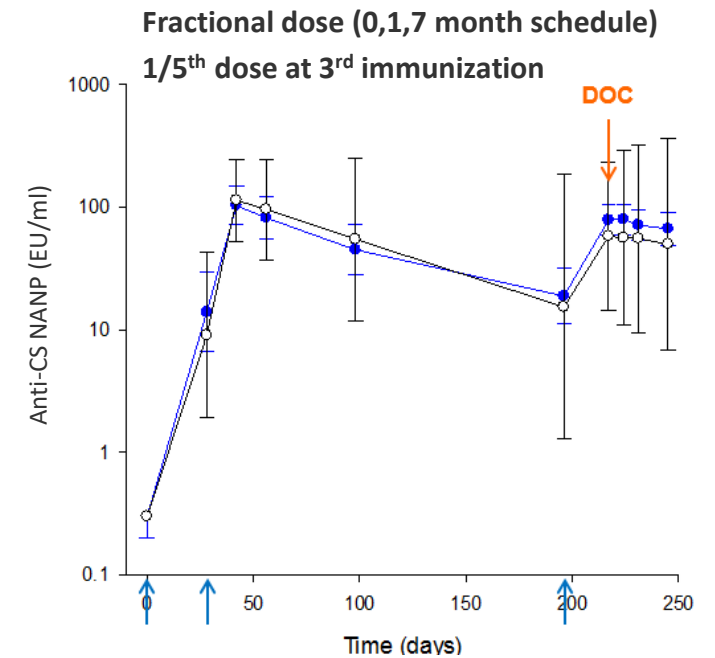
*Adjustments to dose & schedule can have profound effects on outcome and immune correlates*

- Immune correlate(s) of protection may change depending on changes in dose and schedule, prime-boost strategies
- Magnitude of antibody is essential but not sufficient for protection

62.5 % protection  
95% CI (29.4-80.1)



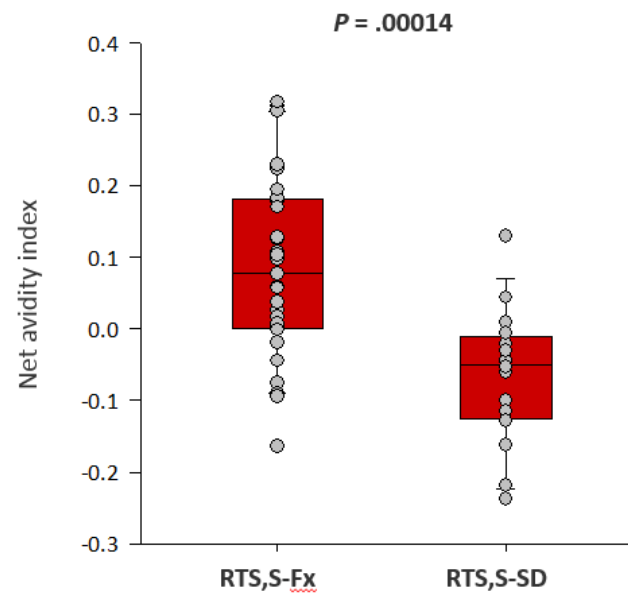
86.7 % protection  
95% CI (66.8-94.6)



# Antibody avidity associated with improved vaccine regimens

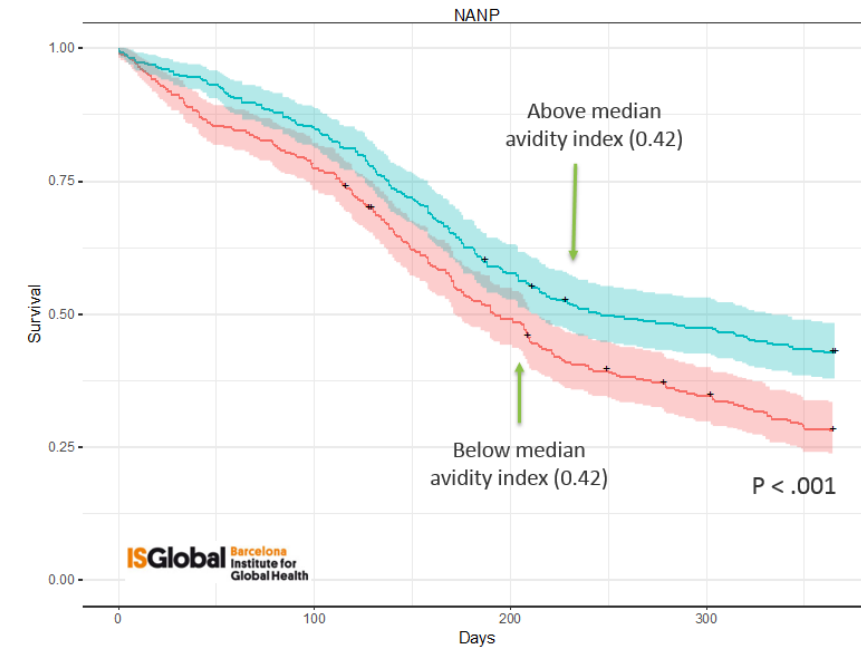
- Ab avidity increasingly observed as contributing independently to protection in both CHMI challenge studies and in clinical trials of RTS,S in Phase 2 and Phase 3 trials
- Increases in antibody avidity with each subsequent immunization seen as important observation that requires further investigation

Fractional dose versus Standard dose  
RTS,S/AS01 (CHMI)



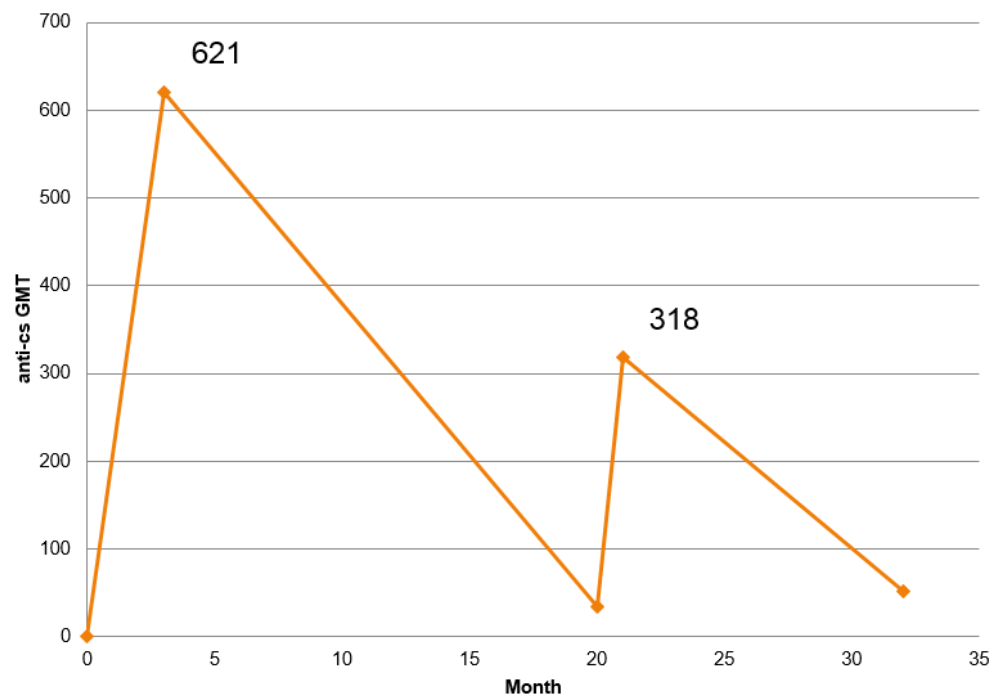
Regules et al., *J Infect Dis.* 2016 214(5):762-71

Higher avidity anti-NANP responses associated with increased protection in Burkinabé and Ghanian children

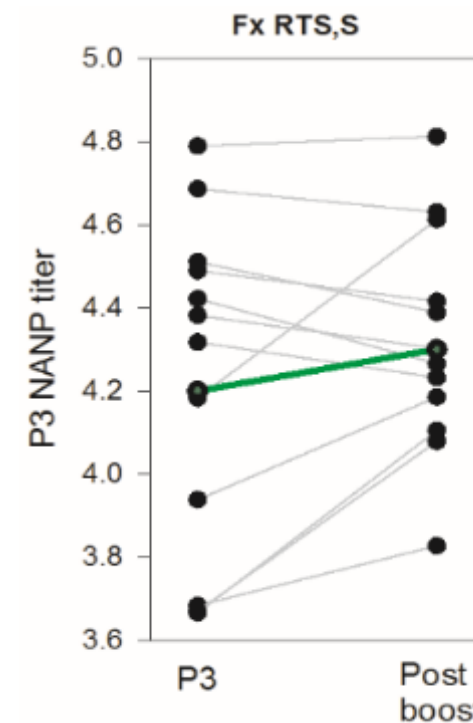
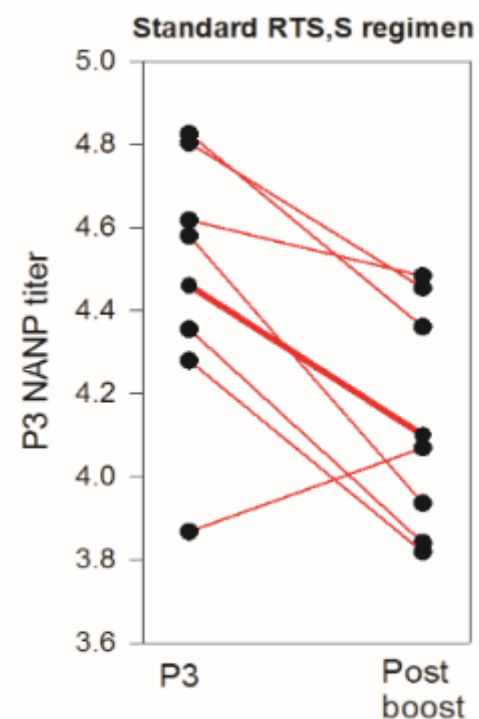


# How CHMI trials may inform reasons around hyporesponsiveness following 4<sup>th</sup> dose RTS,S

## Young children RTS,S Phase 3 Clinical trial



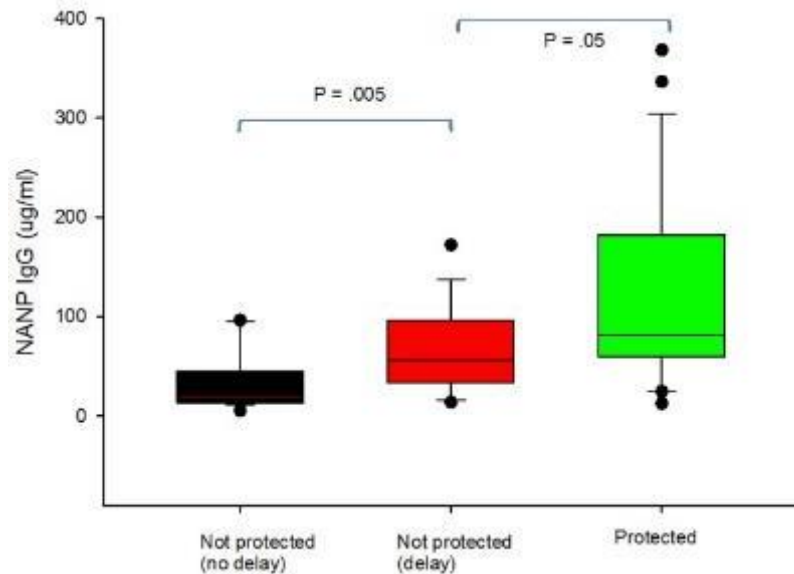
## CHMI MAL071



# Antibody functionality

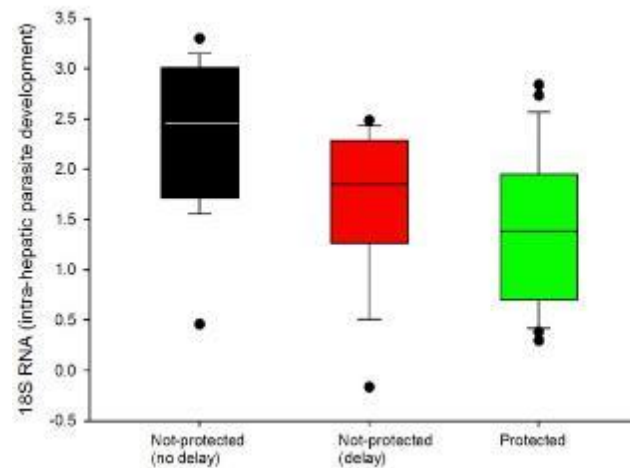
- Functional assay needs to add value over more traditional assays
- Example below – no added information from assay that measures antibody inhibition of sporozoite invasion/development over a single ELISA titer

### NANP IgG titers

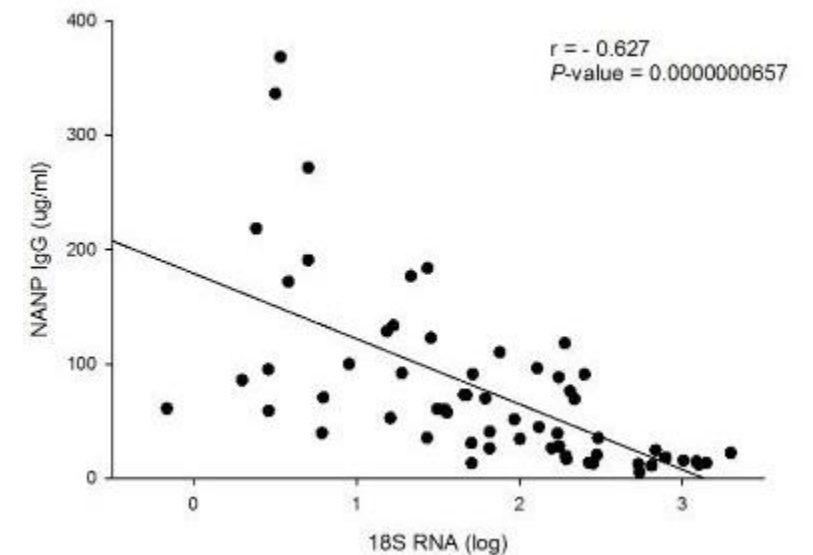


### ILSDA

### Inhibition of liver stage development assay

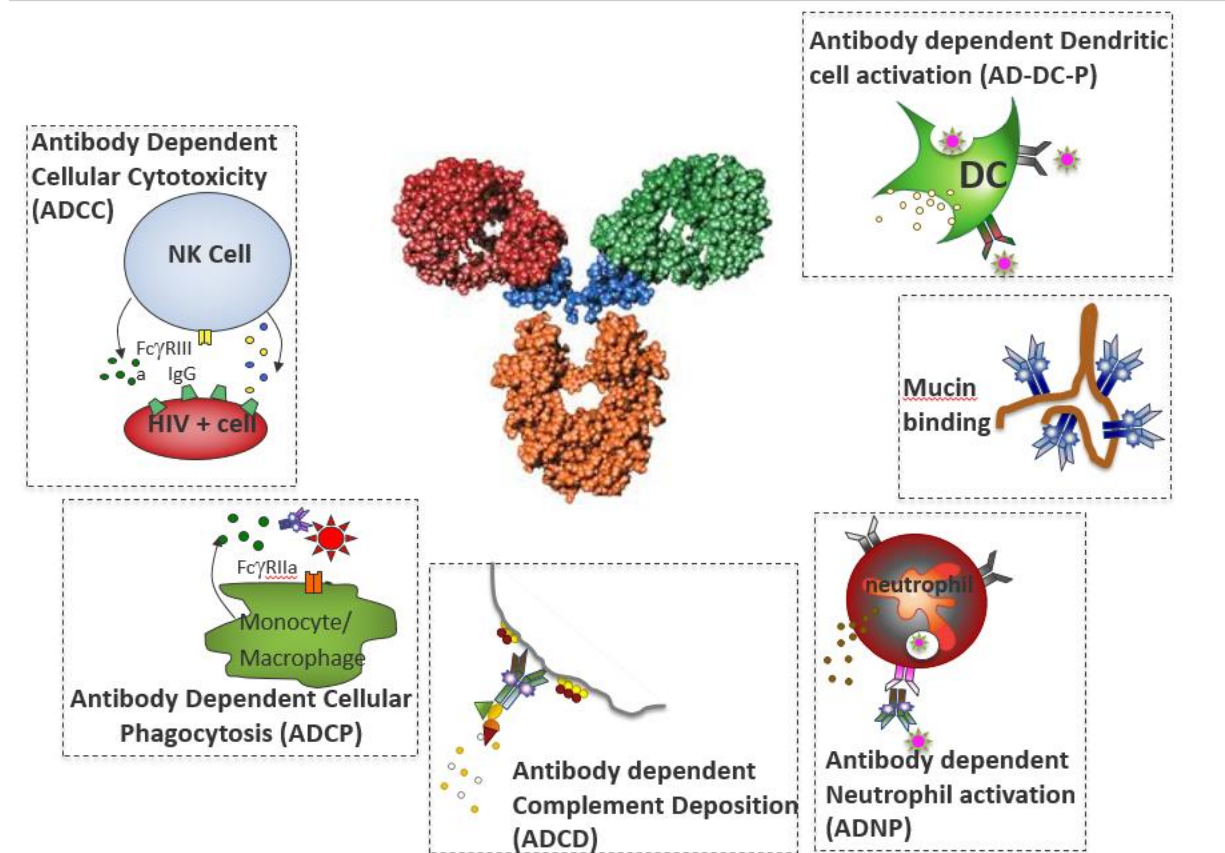
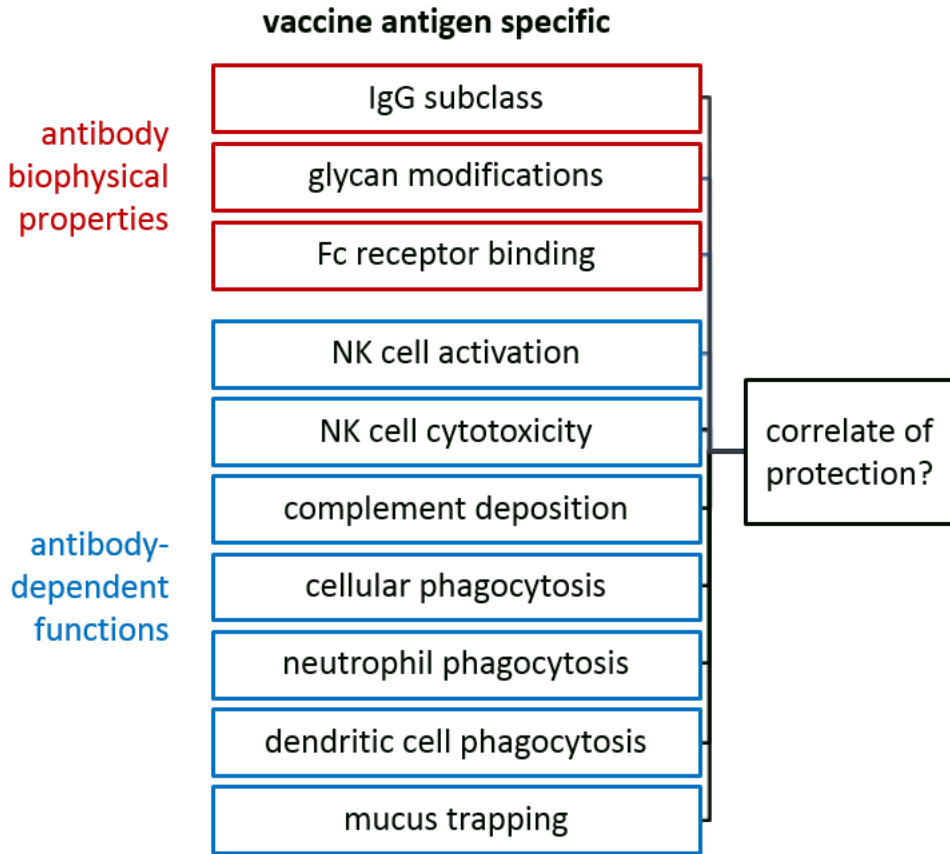


### Inverse correlation





# Systems immunology



Courtesy G. Alter, Ragon Institute and MGH

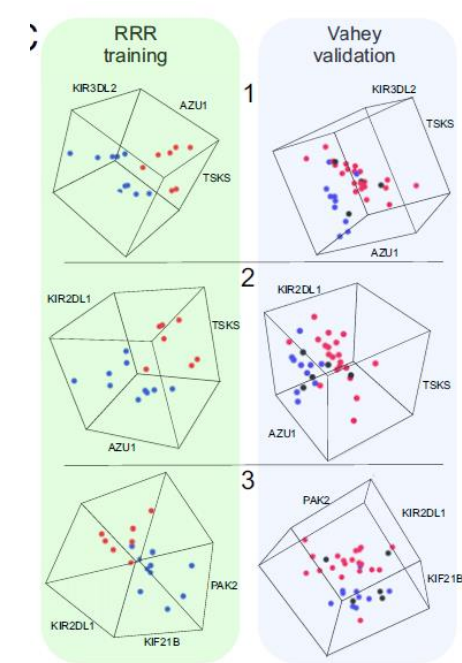
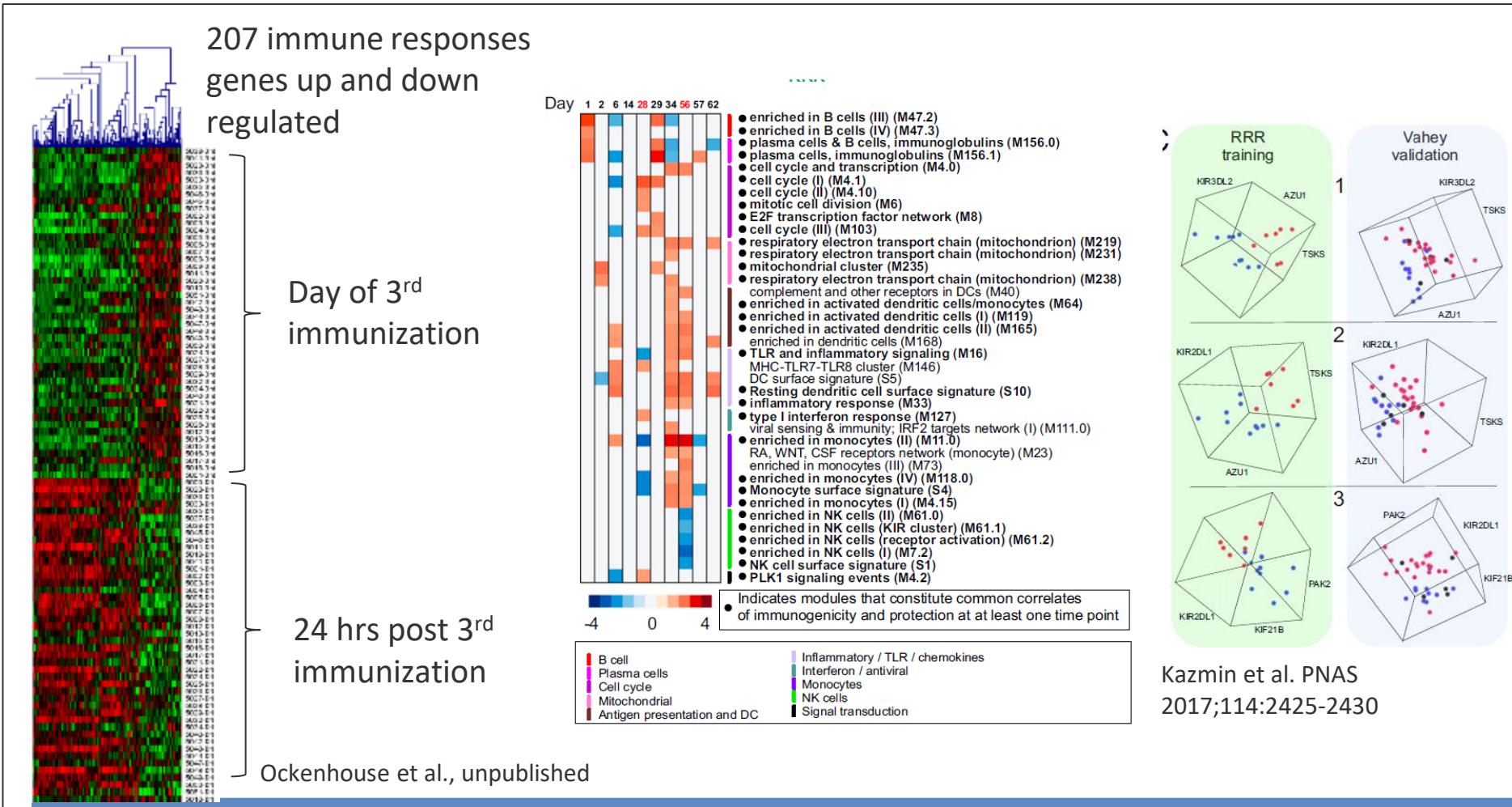
# Systems biology – Molecular signatures of protection

## Microarray and RNAseq transcriptomics

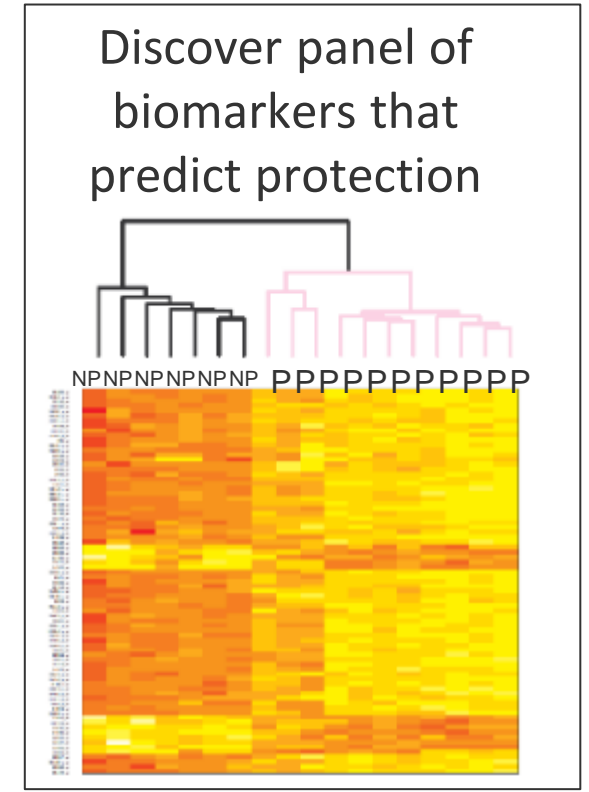
Where we are now – knowledge & understanding



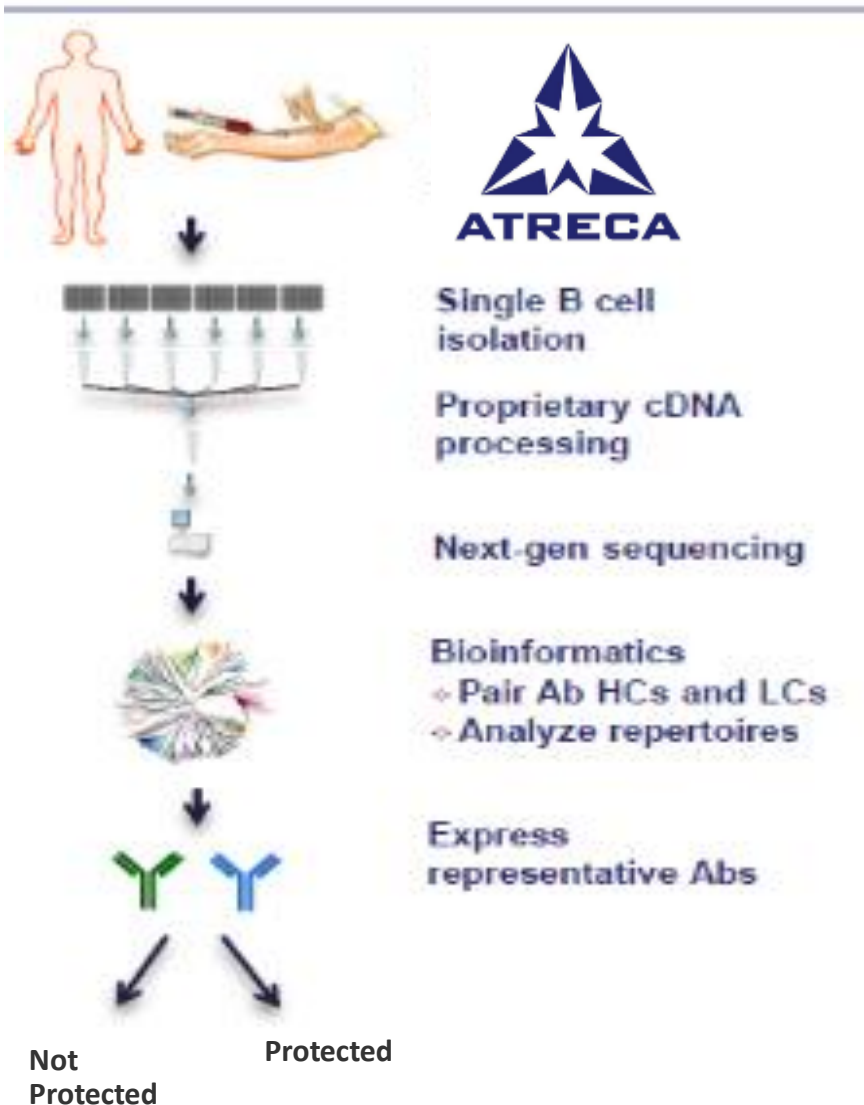
Where one hopes to be



Kazmin et al. PNAS 2017;114:2425-2430



# Opportunities and promising technologies – *Antibody repertoires*



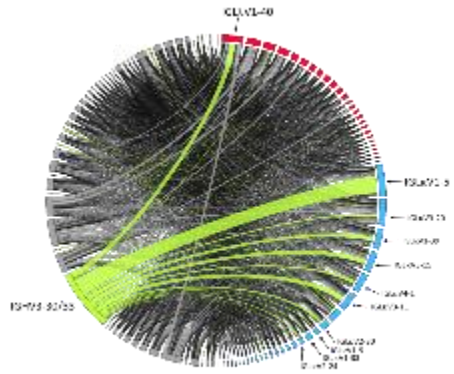
## Knowledge → Understanding

- Understand how somatic hypermutation leads to increased antibody affinity for target antigen in polyclonal serum
- Understand how different vaccine regimens induce repertoire signatures of protection

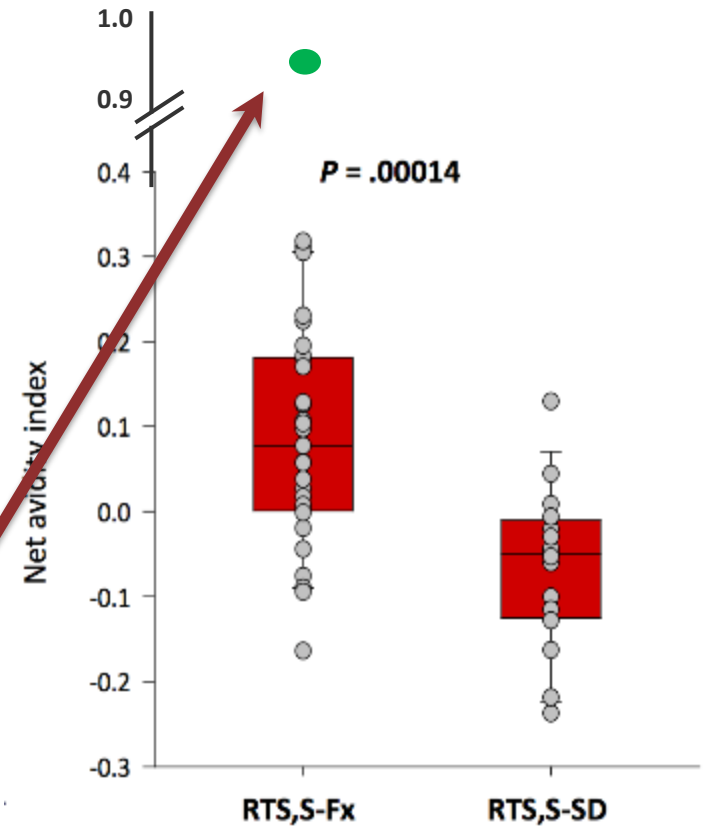
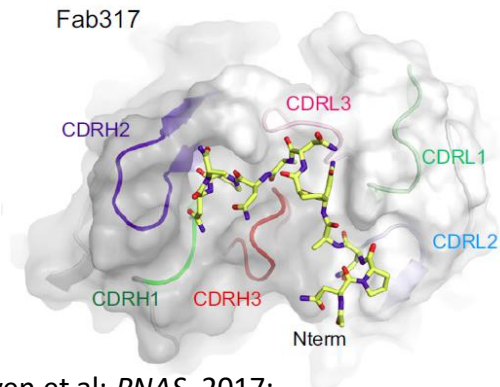
## Understanding → Action

- Passive immunization as viable alternatives to active immunization

# Clinical lead CSP mAb (AB-000317)



- **Characteristics of AB-000317**
  - Produced as IgG1
  - Binds to NPNANPNANPNA peptide
  - Protection
    - Chimeric Pb/PfCSP: 100%
    - PfCSP (hu liver mice): 99.3%
  - Affinity constant: 0.21 nM
  - Net avidity index (NANP<sub>6</sub>) = 0.95



Regules et al., *J Infect Dis.* 2016 Sep 1;214(5):762-71

Oyen et al; *PNAS.* 2017; 114(48):E10438-E10445

## • Critical questions

- Can a single CSP mAb, targeting the central repeat region, protect humans?
- Can a 'high avidity mAb' result in protection at low serum concentrations?

# Acknowledgements



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Ulli Wille-Reece, PhD



BILL & MELINDA GATES foundation



Thank you

[malaria vaccine.org](http://malaria vaccine.org) | [path.org](http://path.org)

