Testing TLR agonists and SIV mAbs in SIV-infected ART-suppressed macaques

Hannah King

December 16, 2022

Durable control of HIV

- Immunologic control of HIV to undetectable levels off antiretroviral therapy (ART)

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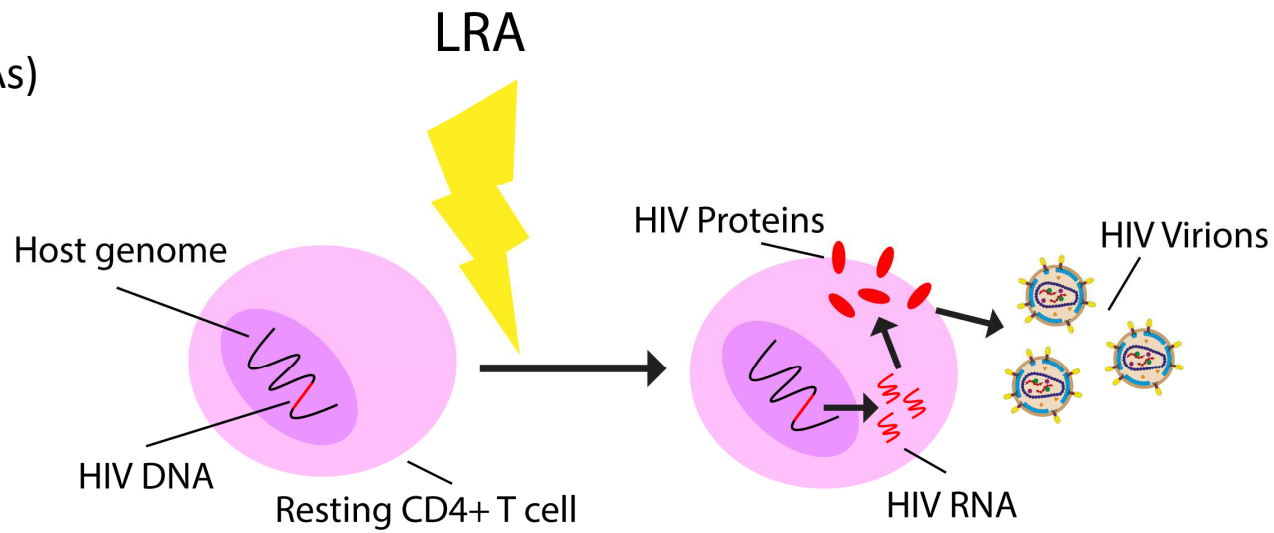
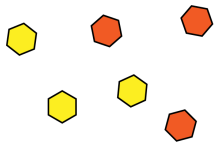
- Could be achieved by:
  - Shrinking the latent reservoir
    - i.e. Latency reversal agents (LRAs)



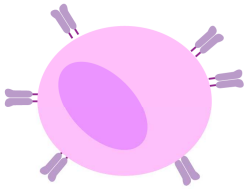
Durable control of HIV

- Immunologic control of HIV to undetectable levels off antiretroviral therapy (ART)

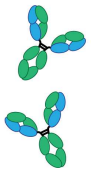
- Could be achieved by:
  - Shrinking the latent reservoir
  - Immune-mediated control



Innate factors (i.e. chemokines) block HIV entry

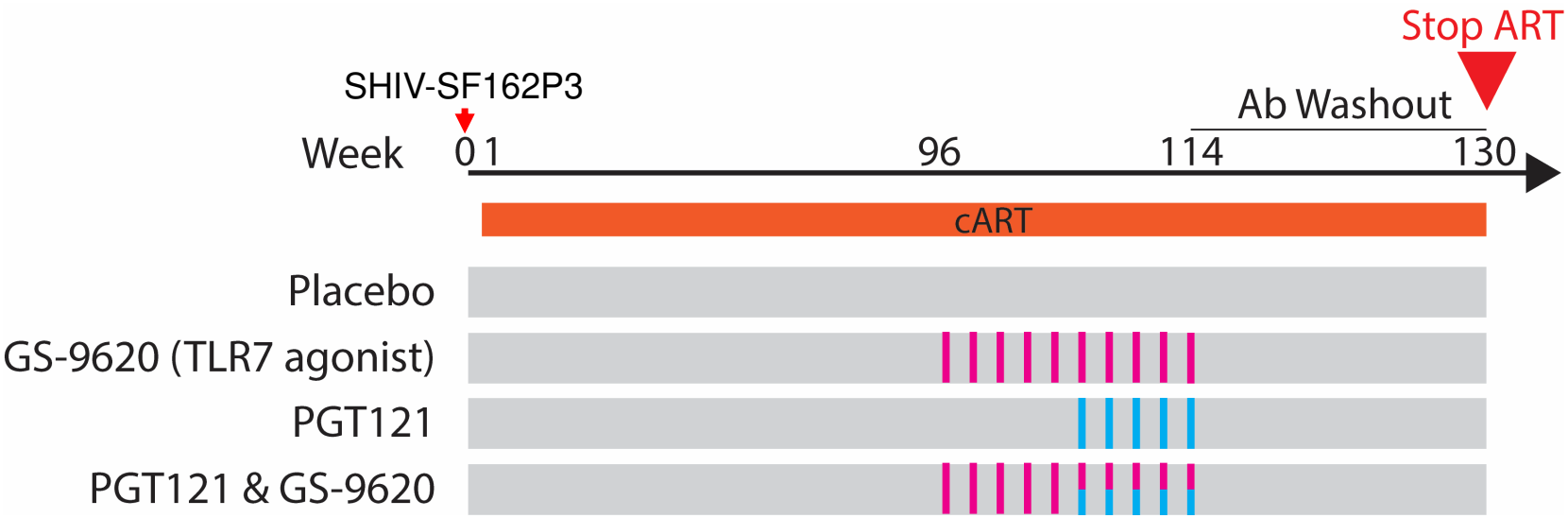


T cells kill infected cells, enhance antibody responses



Antibodies neutralize virus,
antibody-mediated killing of infected cells

Combining bNAbs and immune modulators



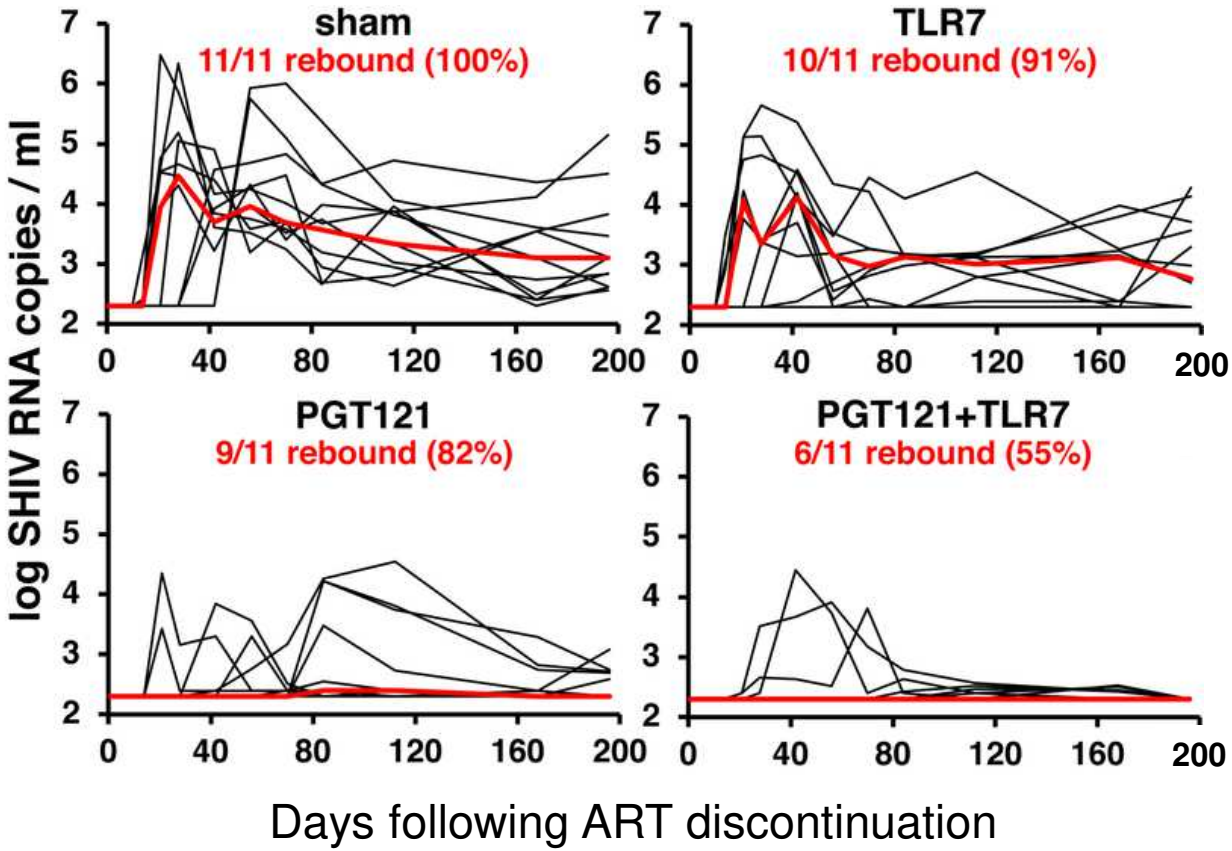
=TLR7 agonist administration

=mAb administration

Antibody and TLR7 agonist delay viral rebound in SHIV-infected monkeys

Erica N. Borducchi[1,6], Jinyan Liu[1,6], Joseph P. Nkolola[1,6], Anthony M. Cadena[1,6], Wen-Han Yu[2], Stephanie Fischinger[2], Thomas Broge[2], Peter Abbink[1], Noe B. Mercado[1], Abishek Chandrashekar[1], David Jetton[1], Lauren Peter[1], Katherine McMahan[1], Edward T. Moseley[1], Elena Bekerman[3], Joseph Hesselgesser[3], Wenjun Li[4], Mark G. Lewis[5], Galit Alter[2], Romas Geleziunas[3] & Dan H. Barouch[1,2]*

Immune modulator and bNAbs control infection



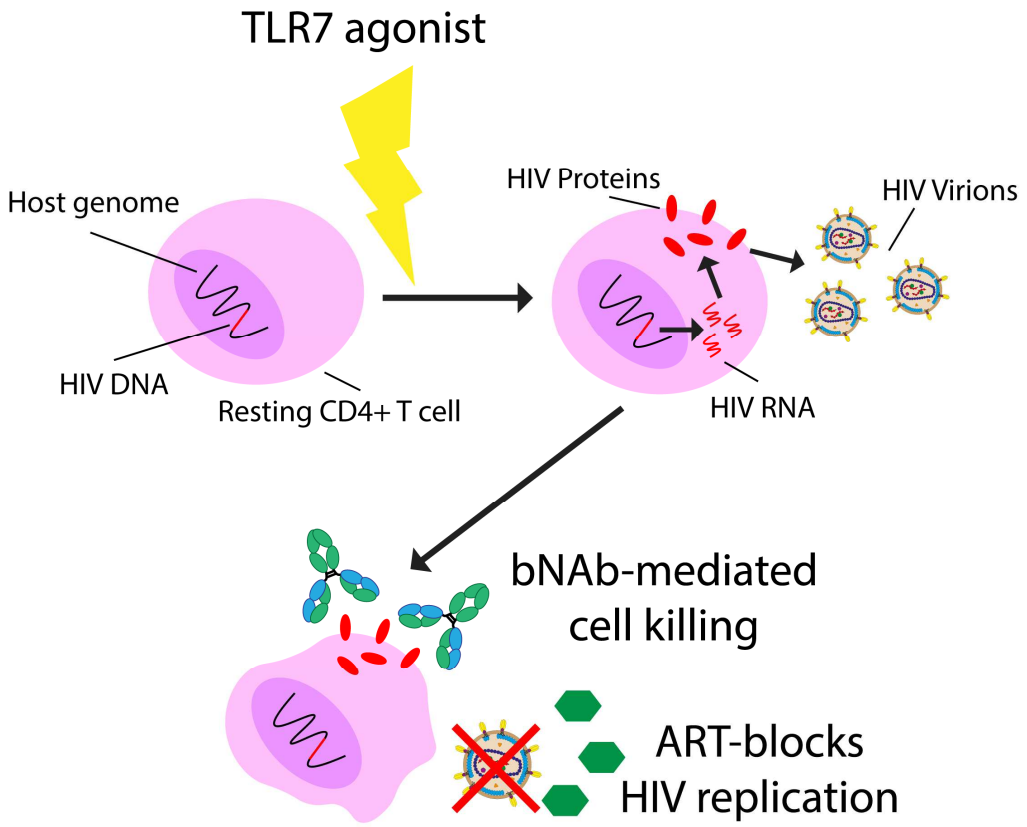
Borducchi et al, Nature, 2018

MHRP

Hypotheses for protection

- TLR7 agonist results in activation of CD4+ T cells
  - Leading to viral reactivation
    - Limited evidence of this occurring
  - Rendering them more susceptible to PGT121-mediated killing?

- Activated effector cells (NK cells and monocytes) mediating killing of infected CD4+ T cells



TLR agonist and mAb co-administration in SIV-infection model

➢Assess TLR agonist and mAb combination treatment in a rigorous SIV model of infection

- Less spontaneous viral control
- Species-matched antibodies compatible with simian Fc receptors
- Species-matched antibodies unlikely to generate anti-idiotype immune response

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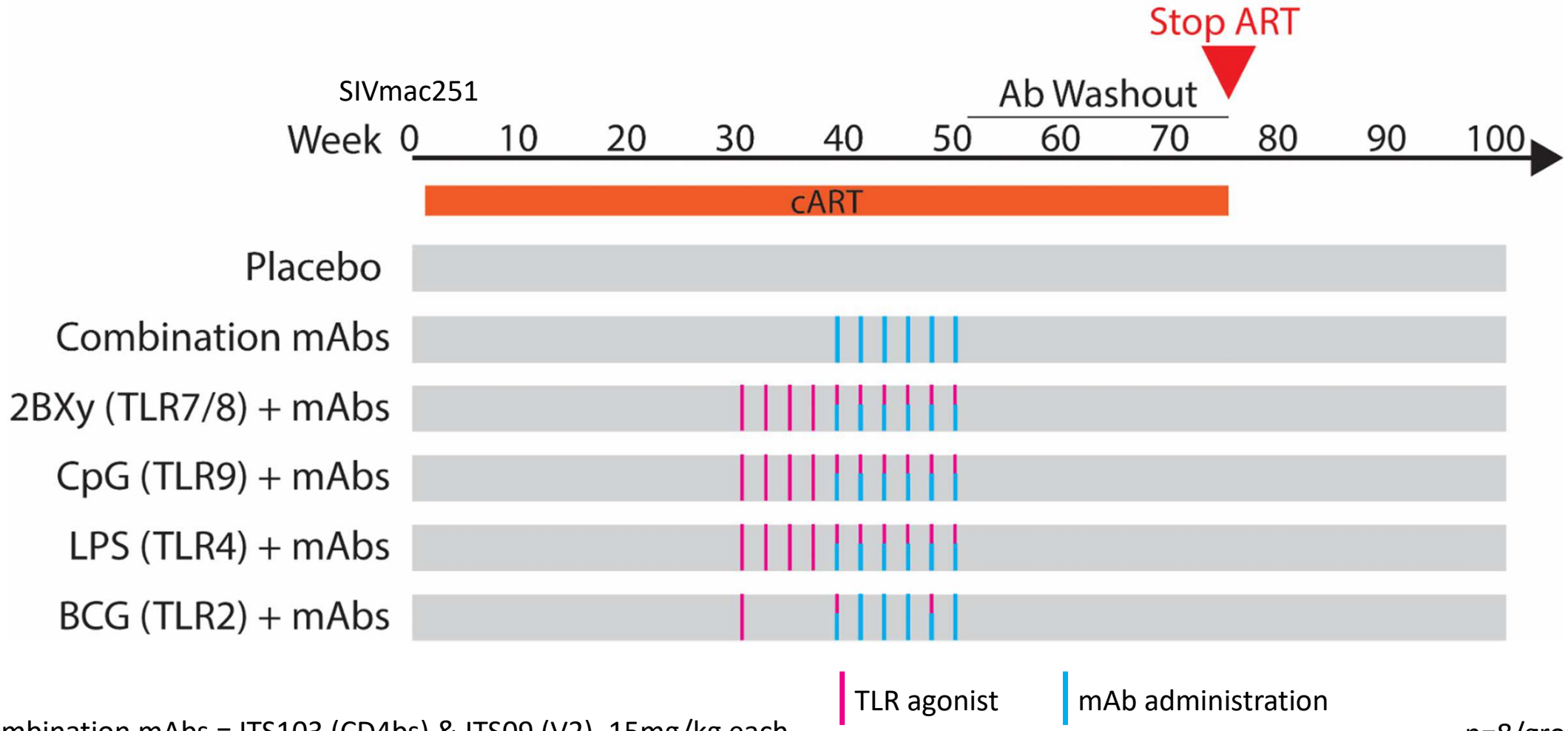
- Next generation anti-SIV mAbs enable evaluation in SIV model
  - ITS103 and ITS09 also bind to the surface of infected cells

Tier	Virus ID	ITS103.01[1]	ITS09.01[2]
1	SIVmac251.H9.1	0.027	0.024
	SIVmac251.6	0.009	0.344
2	SIVmac251.cs.41	0.015	0.028
	SIVmac251.30	0.015	>50
3	SIVmac239.cs.23	0.009	>50

[1]Welles, King…Mason et al, Plos Path, 2022; [2]Mason et al, Plos Path, 2016

Study design



Combination mAbs = ITS103 (CD4bs) & ITS09 (V2), 15mg/kg each

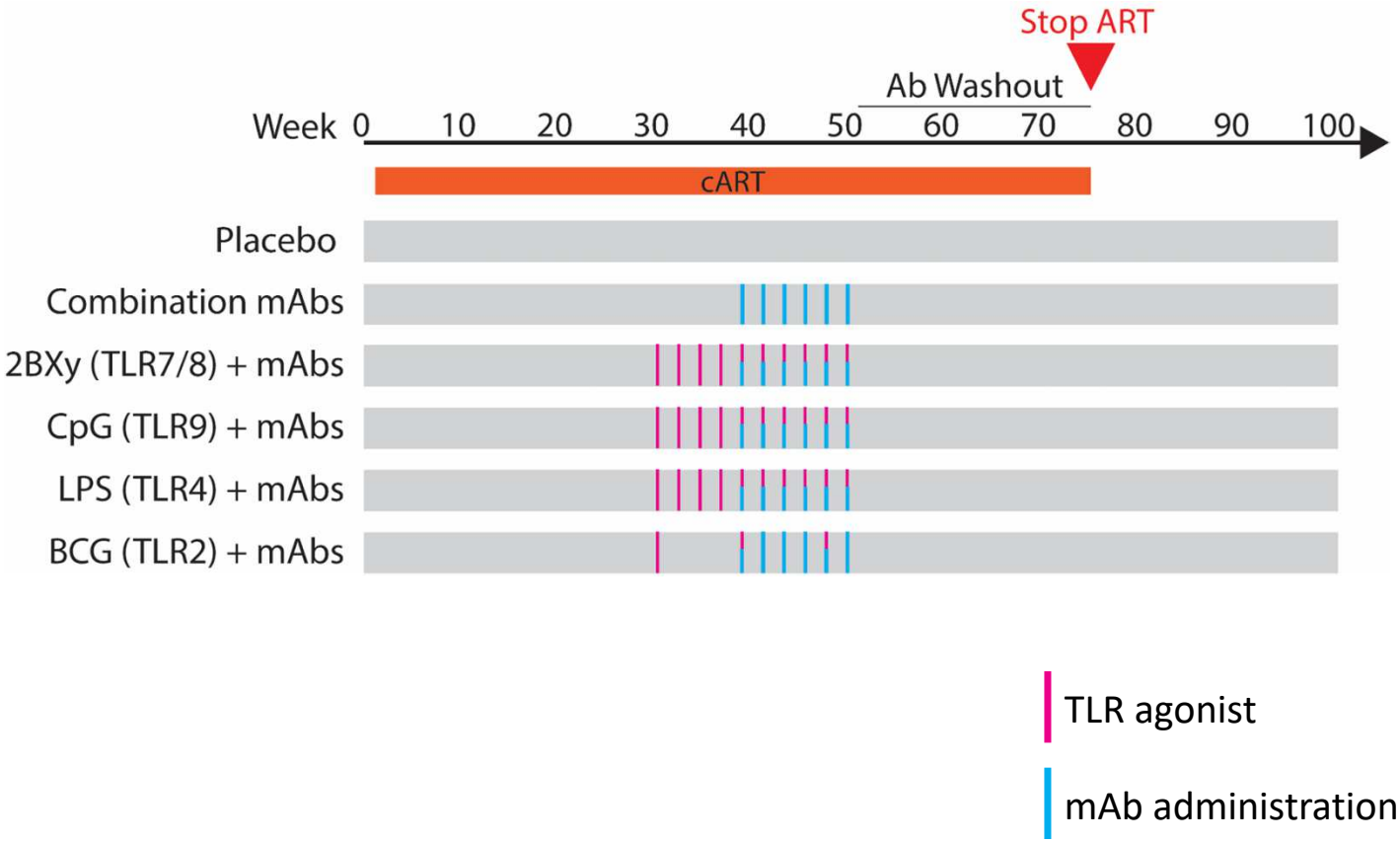
n=8/group

Immune agonists targeting different TLRs

Agonist	TLR	Rationale	Route	Dose
2BXy	TLR7/8	An oral TLR7 agonist + human bNAb achieved control against SHIV (Borducchi et al, 2018)	Intravenous	125µg/kg
CpG DNA	TLR9	Can protect against intracellular pathogens in animal models, trialed in cancer immunotherapy (Scheiermann et al, Vaccine, 2014)	Subcutaneous	250µg/animal
LPS (Lipopolysaccharide)	TLR4	Stimulates immune activation and viral reactivation in chronic SIV infection (Bao et al, PLOS One, 2014)	Intravenous	50µg/kg
BCG	TLR2 (& 4 & 9)	FDA approved for cancer immunotherapy, strong immune activator	Intravenous	$5x10^7$ CFU/animal

Measurement of immune activation

- Flow cytometry for innate activation markers
- Luminex for soluble cytokines
  - First administration of agent
- Antigen-specific T cell responses
- TLR agonists are expected to elicit strong immune activation



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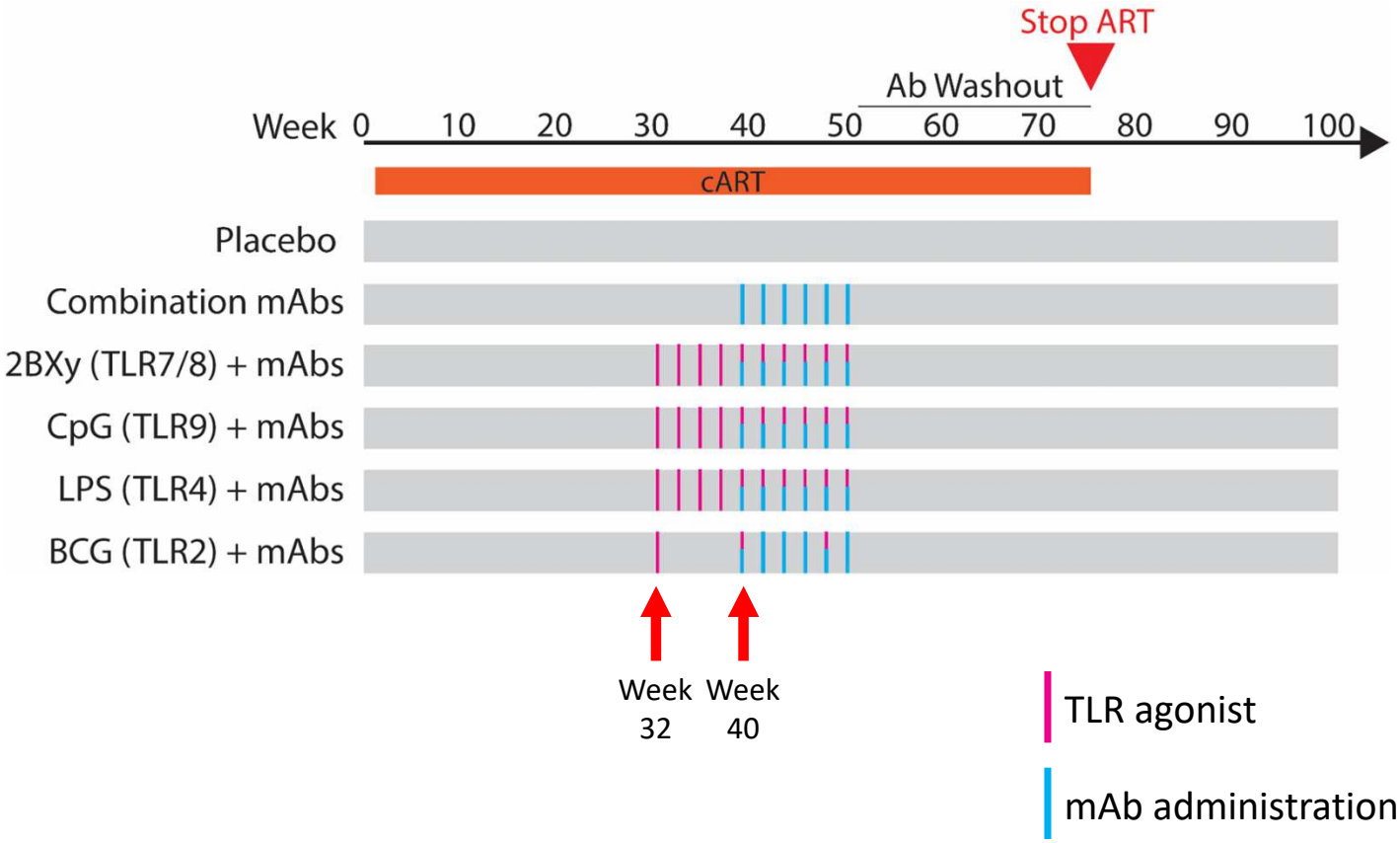
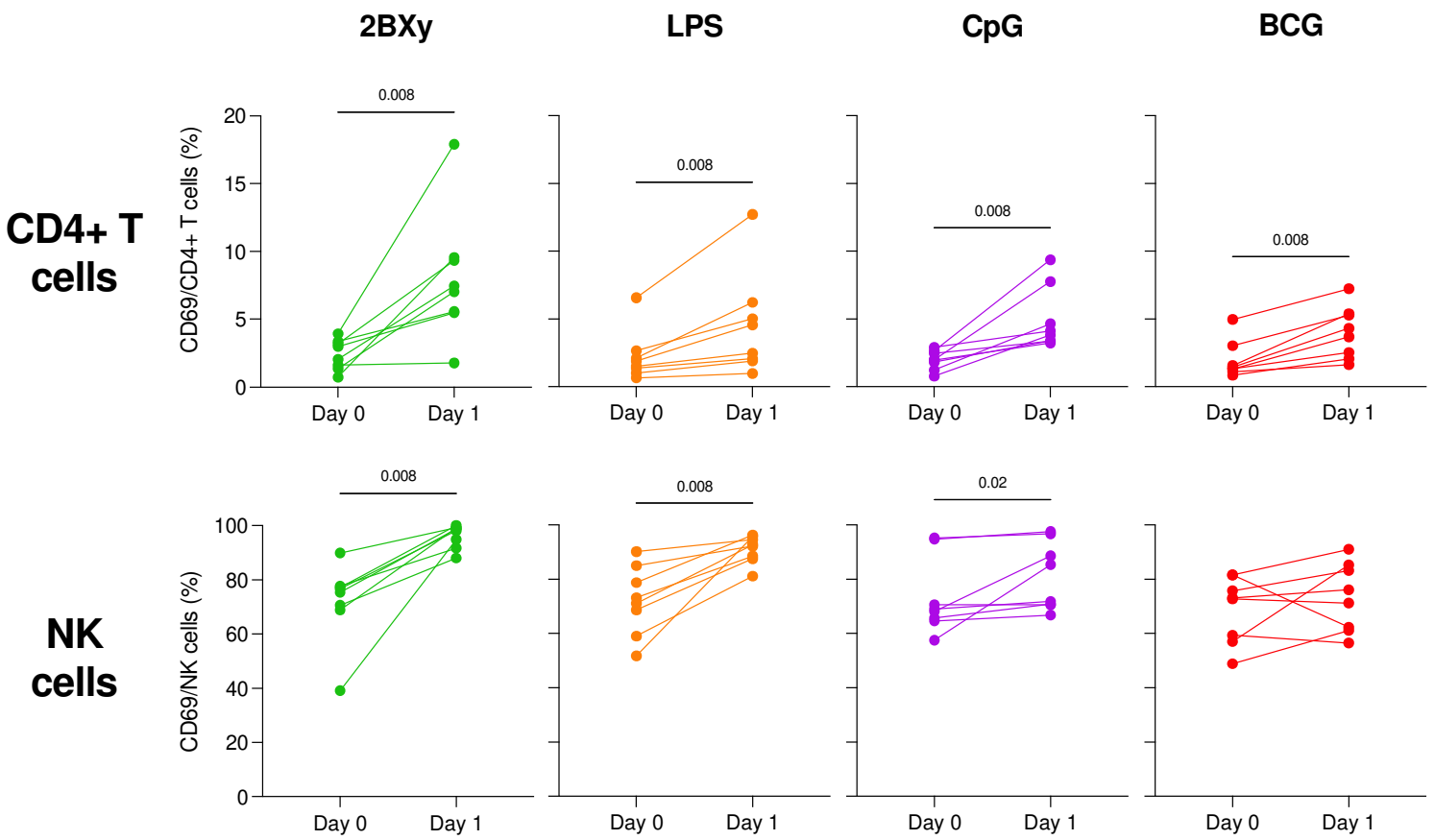
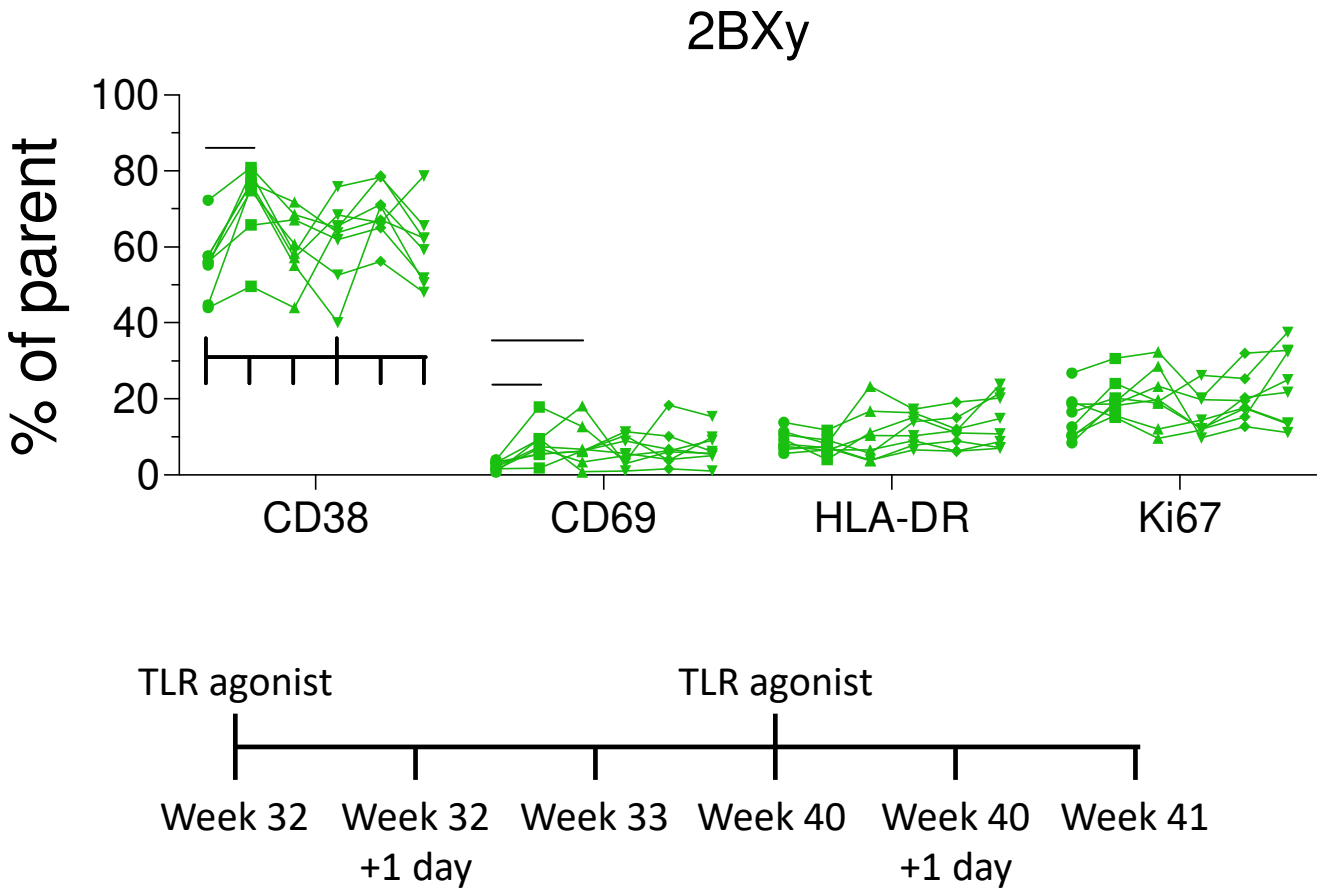


CD69 is upregulated on T and NK cells following TLR agonist administration

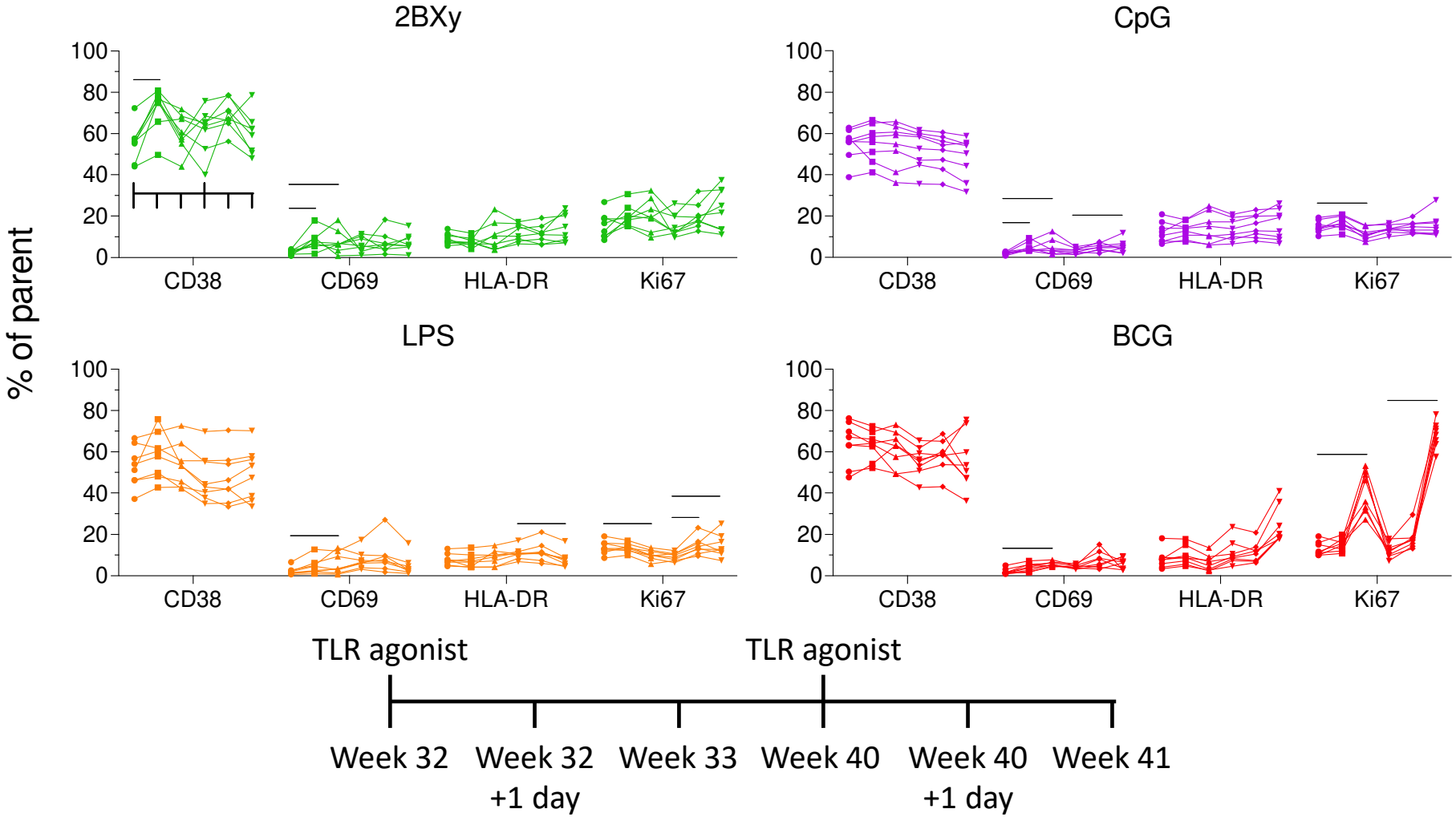


CD4+ T cell activation



Friedman test with Dunn's post-test, comparing to time of infusion

CD4+ T cell activation



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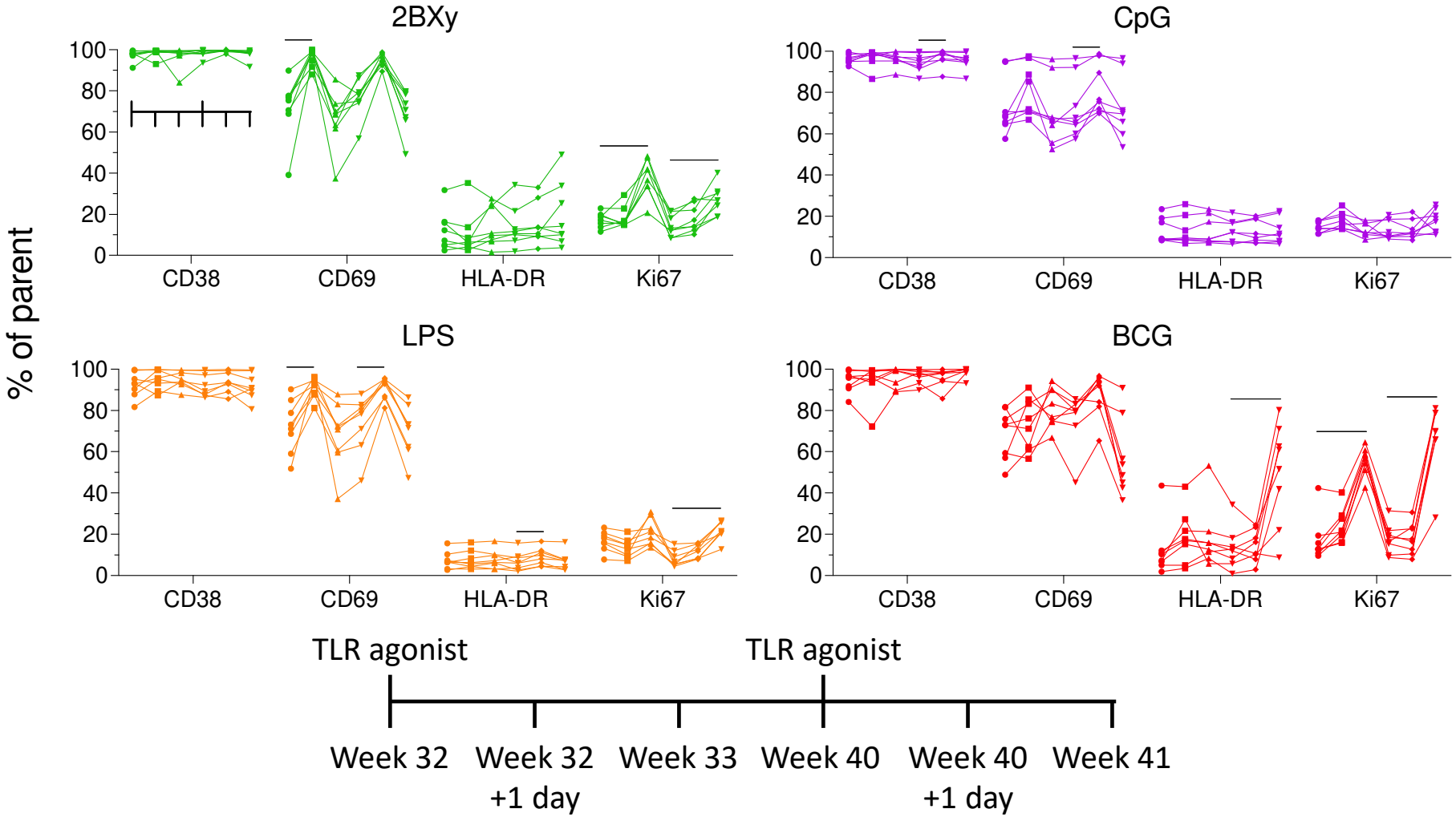
NK cell activation



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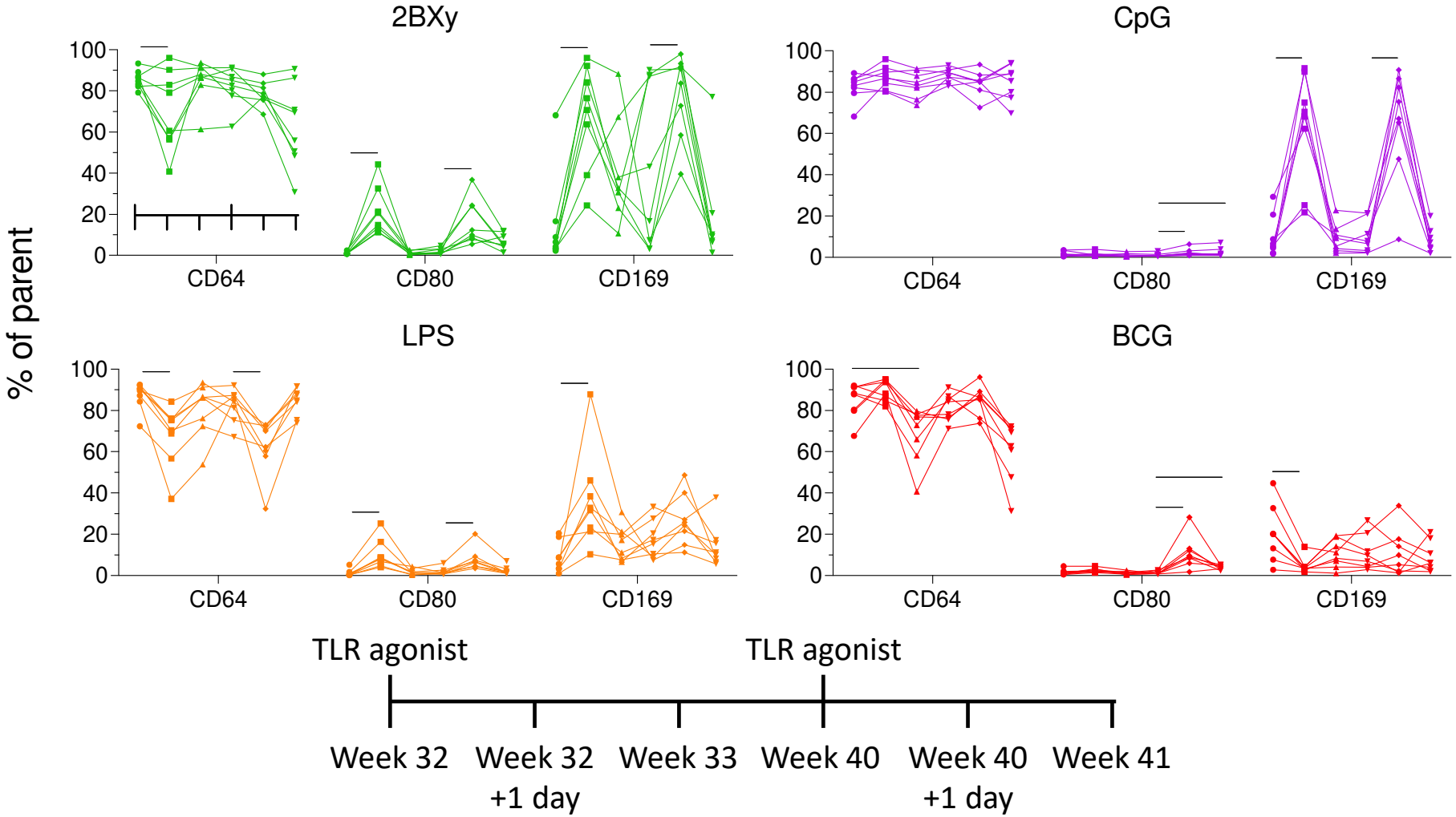
Monocyte activation



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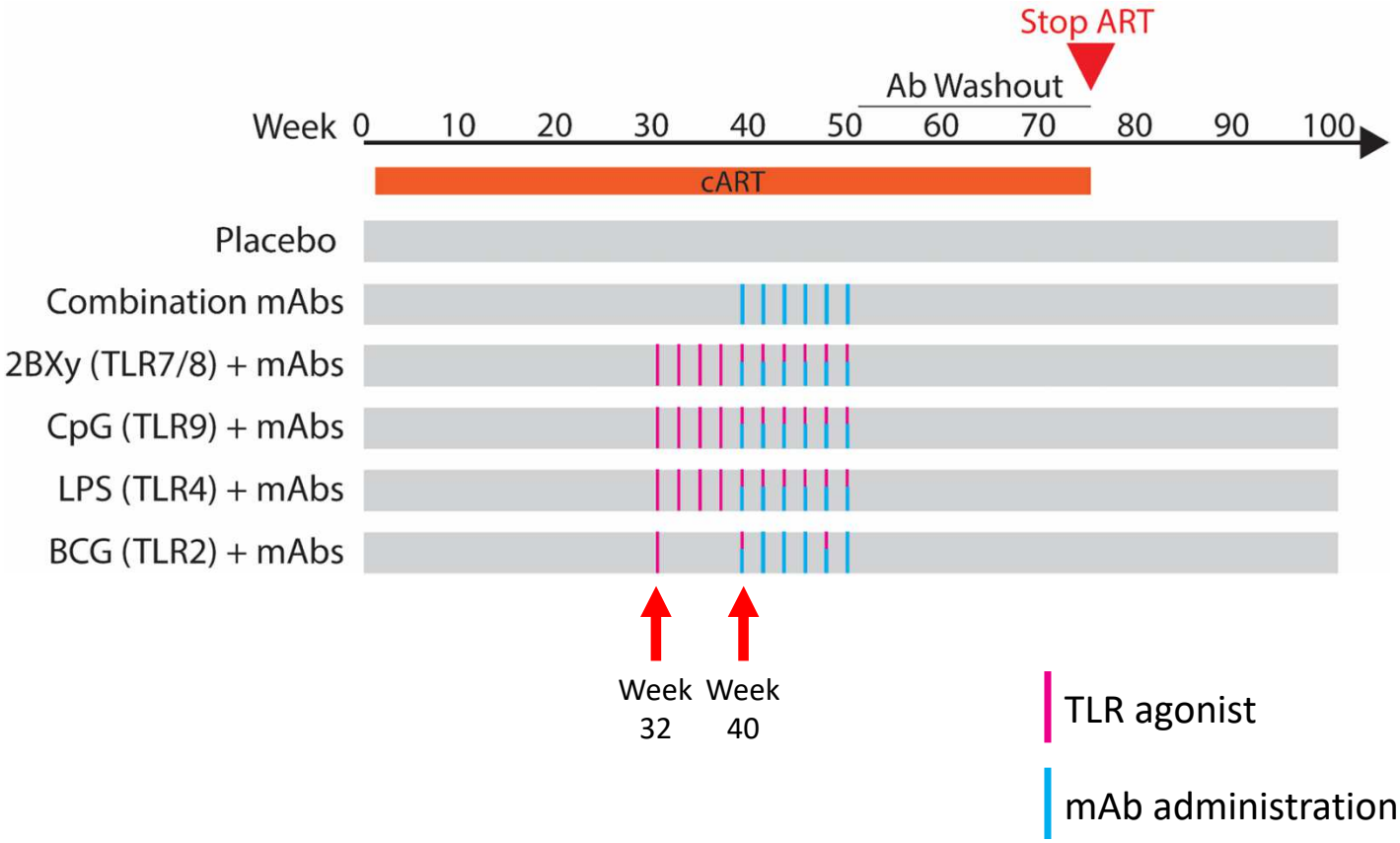
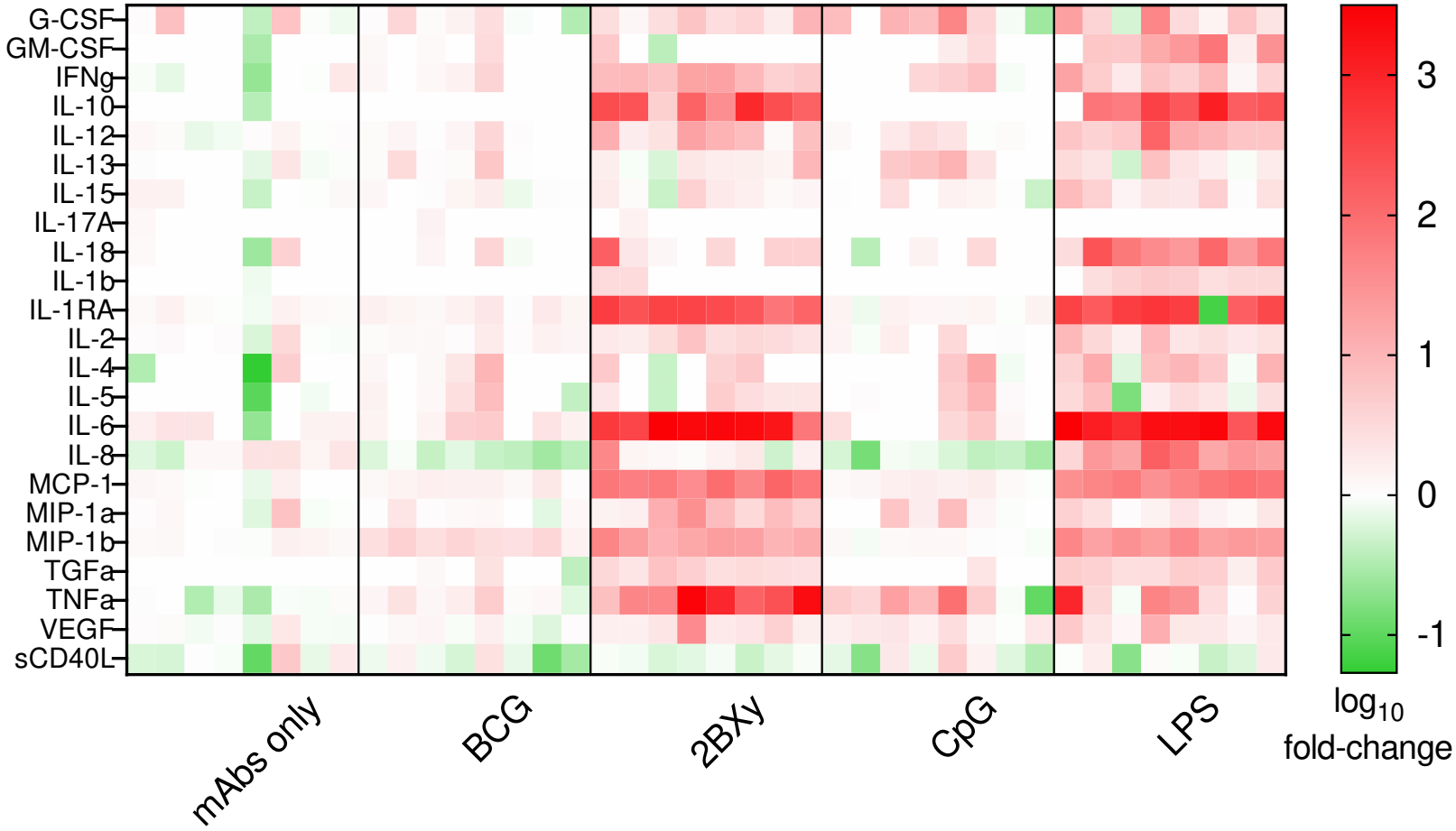
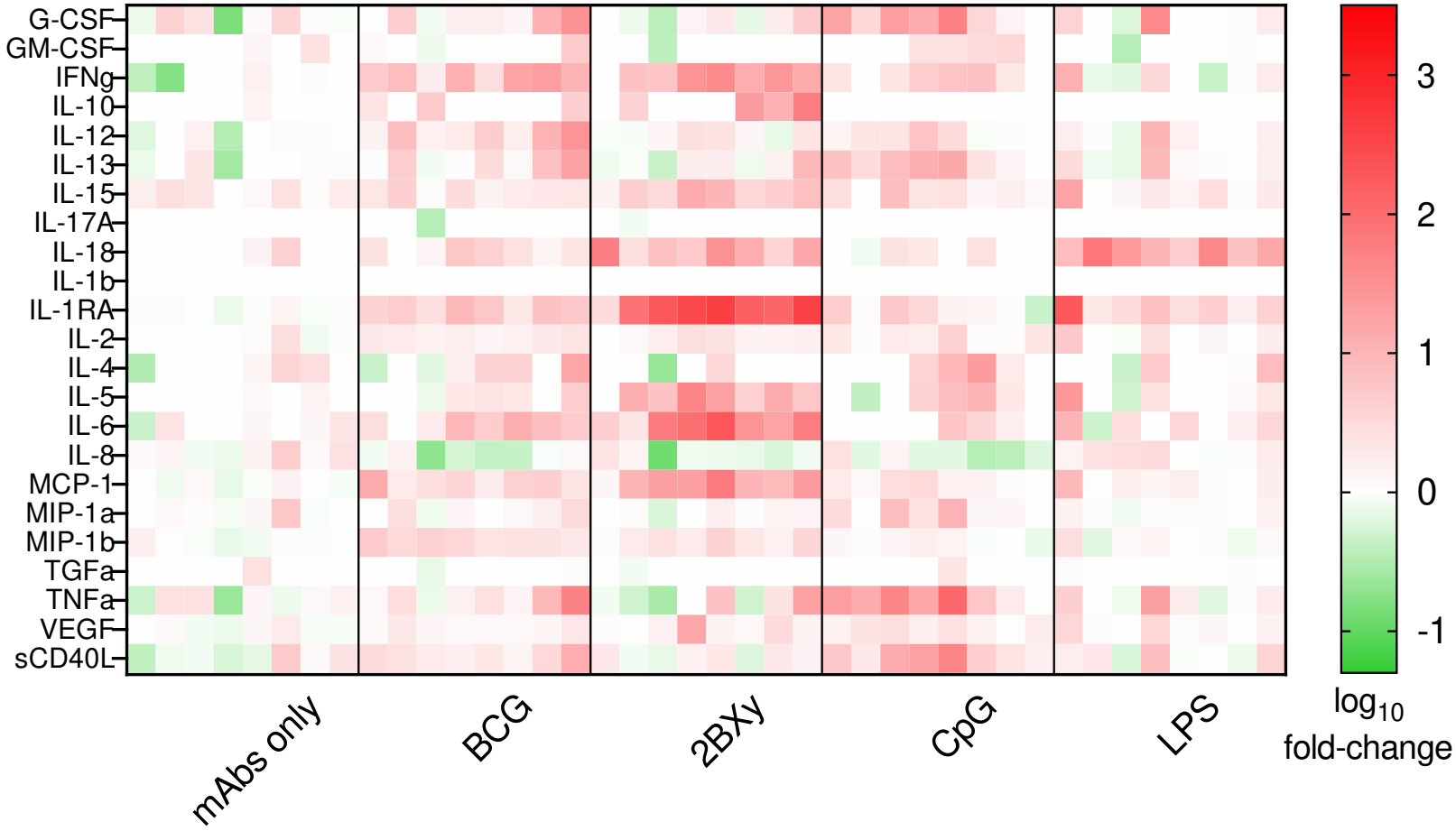


2BXy and LPS show strong soluble cytokine induction in plasma 2 hours after administration



Fold-change in signal 2 hours over baseline

All TLR agonists elicit cytokine expression in plasma 1 day post-administration



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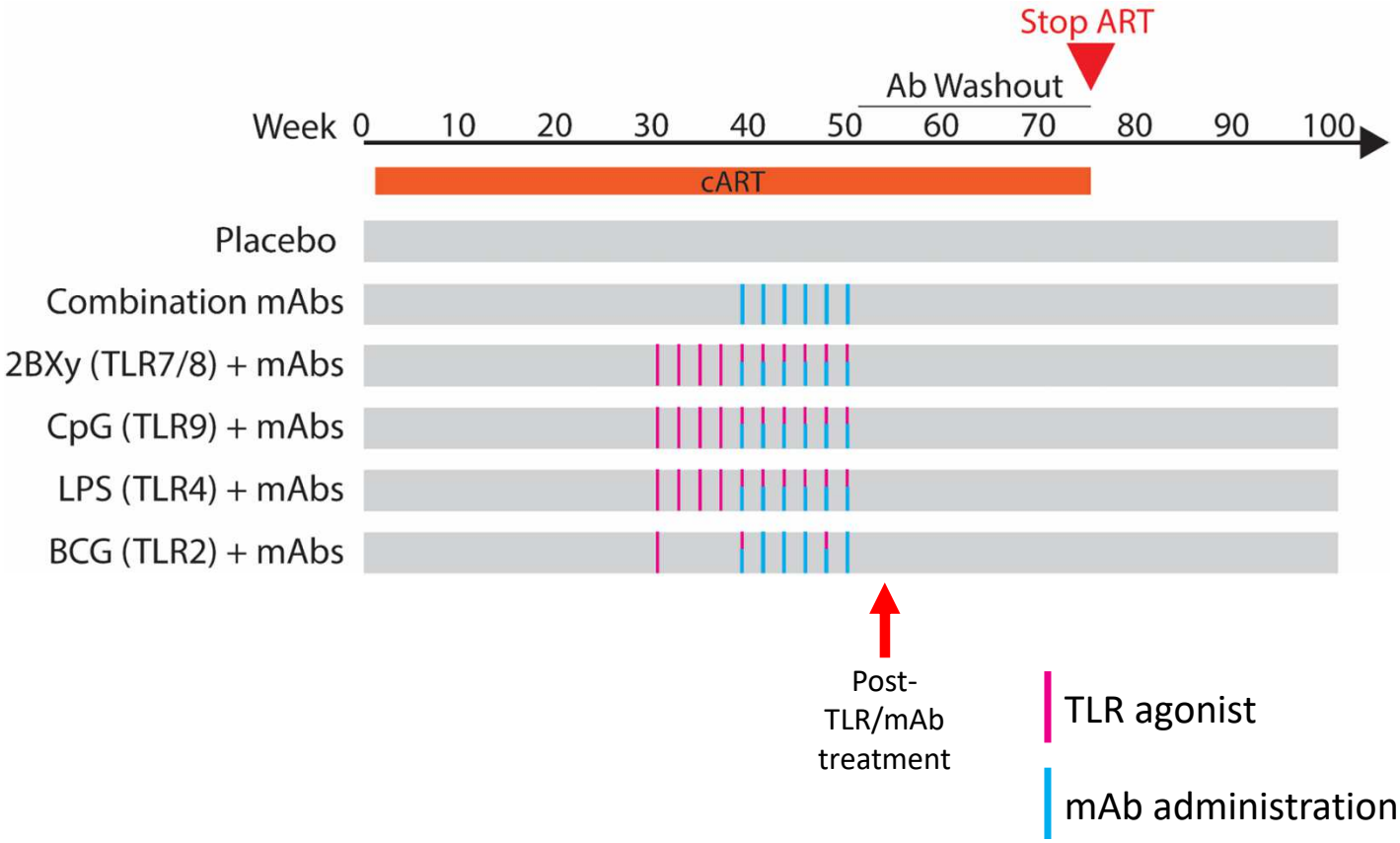
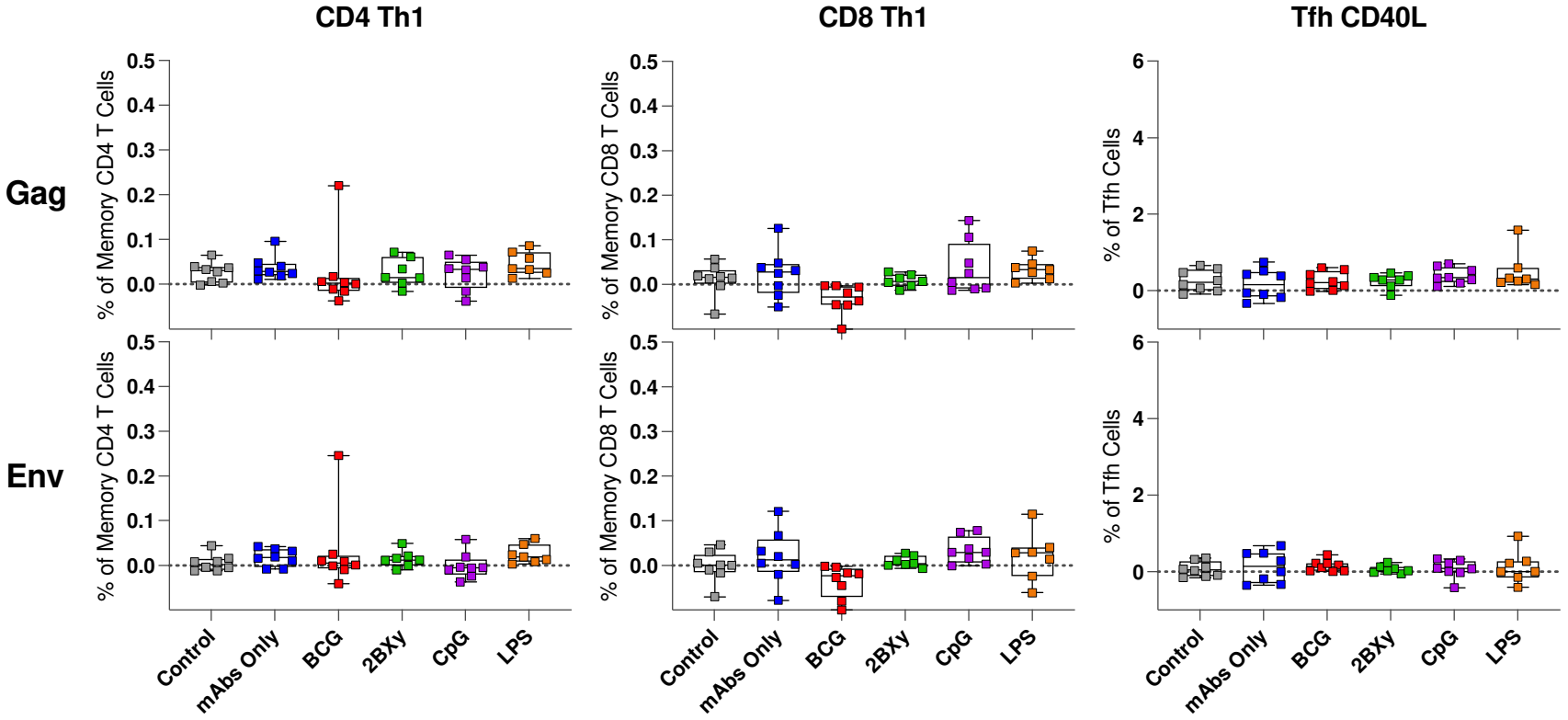


No enhancement of T cell responses post-TLR treatment

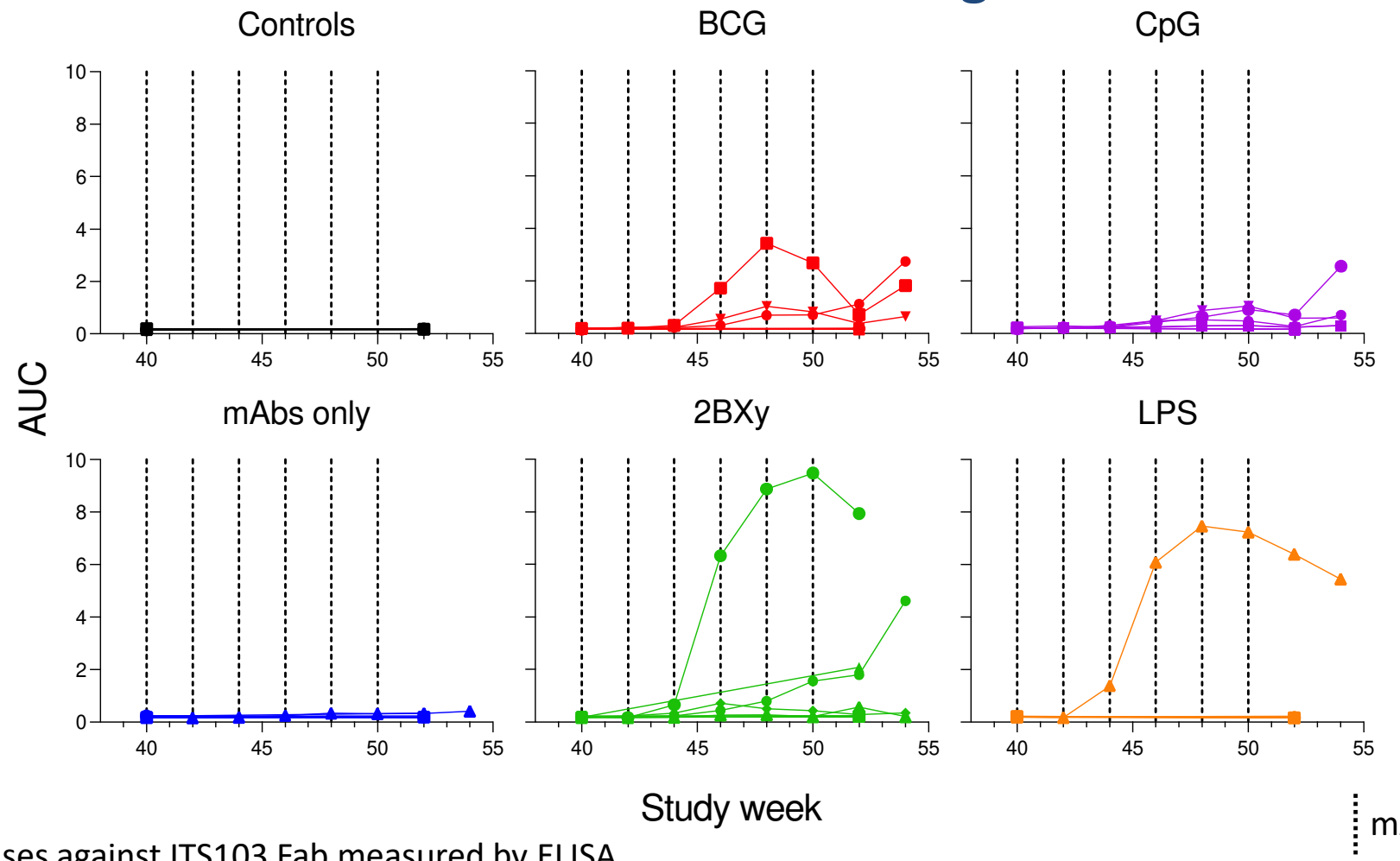


- T cell responses measured 4 weeks post-TLR/mAb treatment
- Also measured at baseline, post-ART initiation, peak VL post-ATI and setpoint VL post-ATI
  - No significant differences observed

Some animals displayed adverse reactions to the TLR agonists

- Likely kidney failure in one animal following 8th LPS administration
- Acute reaction to 10th 2BXy administration
- 3 animals lost weight and poor appetite after 3rd BCG

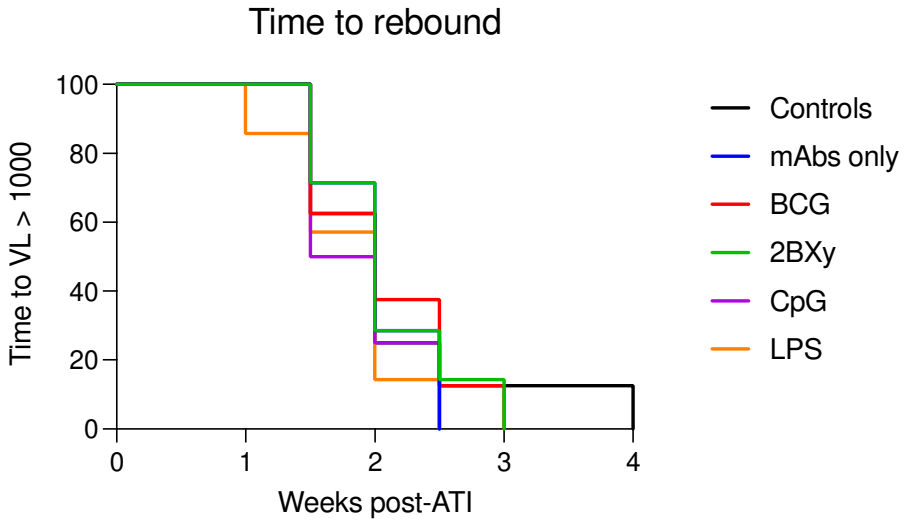
Anti-drug antibodies (ADA) elicited against mAbs when co-administered with TLR agonist

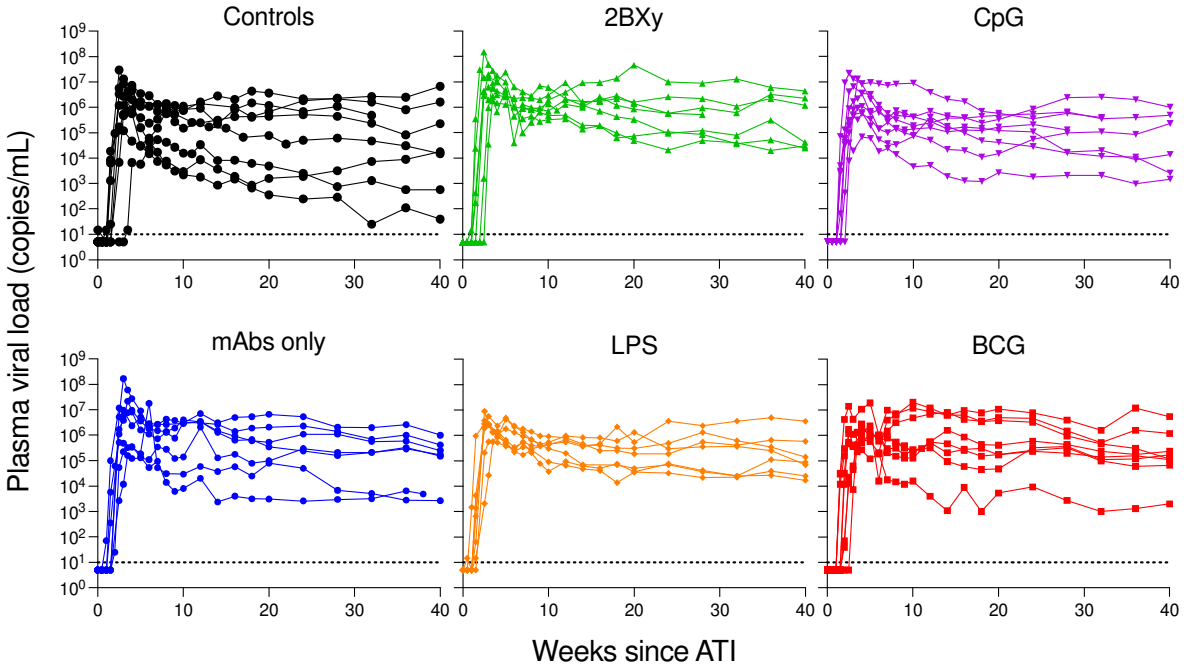


Binding responses against ITS103 Fab measured by ELISA

Did combination mAb and TLR agonist treatment delay rebound in SIV infected monkeys?

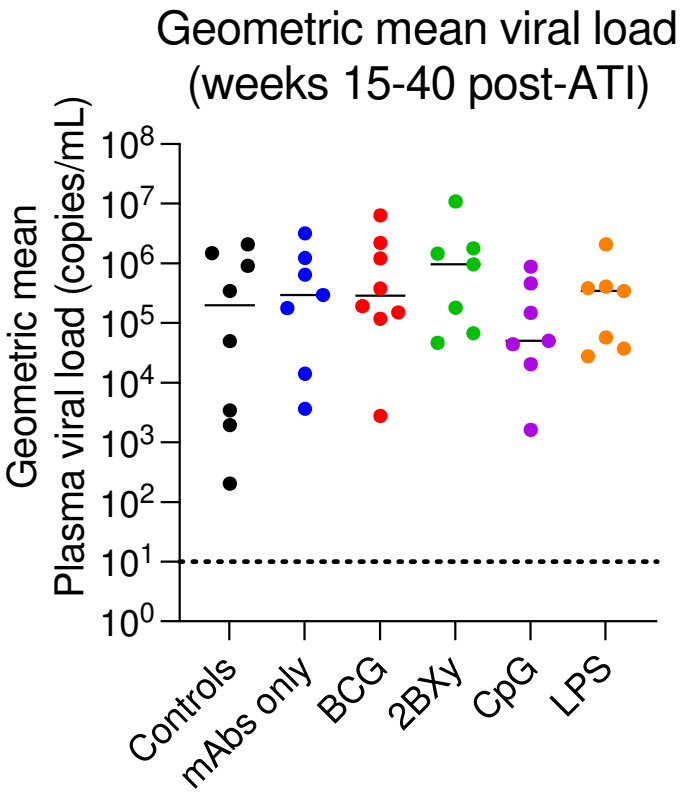
No evidence of delayed viral rebound following treatment

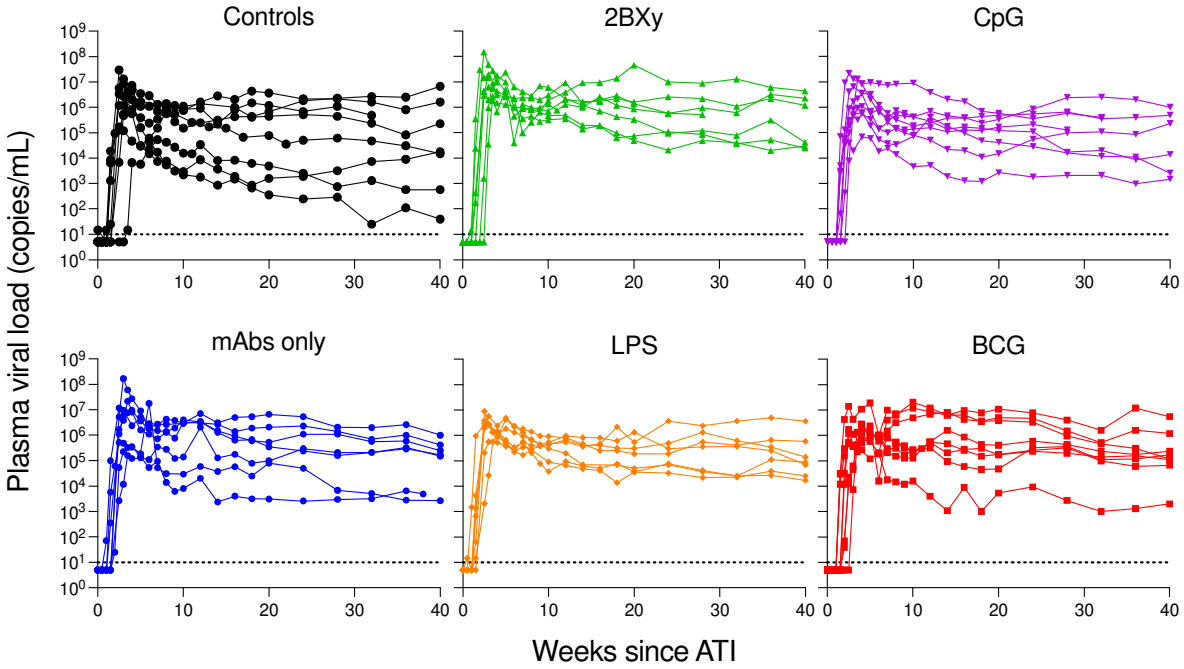




- Consistent early rebound observed in control animals
  - Timing consistent with rebound in PLWH suppressed during Fiebig I at ATI (Colby et al, 2018)
- All animals rebound within 4 weeks of ATI

No evidence of viral control post-rebound





Conclusions

- IV and SC administered TLR agonists used in this study are strongly immunostimulatory
  - Induced antibodies to simian anti-SIV mAbs that are normally not immunogenic
    - Did TLR agonists break the immune tolerance against infused rhesus mAbs?
  - Can elicit adverse events after repeat administrations

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  - Can elicit adverse events after repeat administrations

- In the SIV model with the agonists used here, TLR agonist + bNAb combination treatment administered during ART does not result in control of viremia
  - SIV model too stringent?

- **Key question:** Is TLR agonist and bNAb treatment an effective cure strategy in the SIV model of infection?

- **Key finding(s):**
  - IV and SC TLR agonists used in this study stimulate large immune activation
    - Different agonists elicit distinct activation profiles
  - TLR agonists used in this study and bNAb treatment ineffective as a cure strategy in SIV model
- **Next steps**: Test SIV mAbs in SIV infection model with previously successful Gilead TLR7 agonist (vesatolimod) which is administered orally

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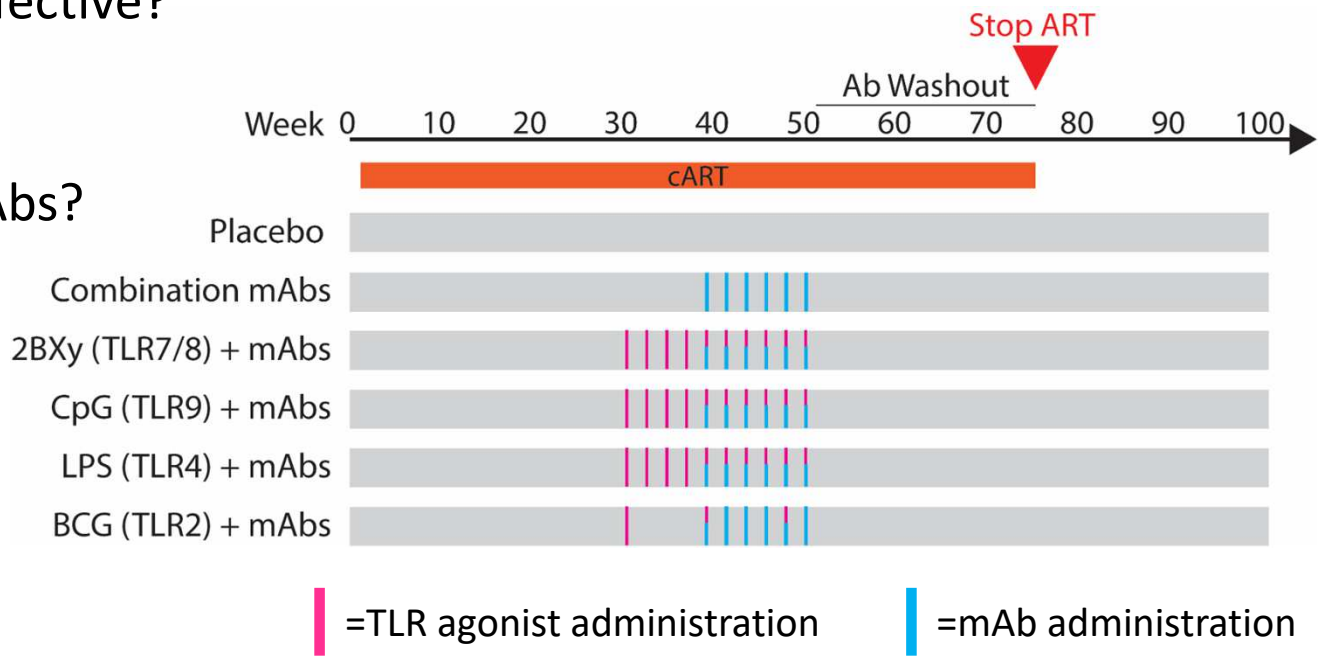
Dale and Betty Bumpers Vaccine Research Center

MHRP U.S. MILITARY HIV RESEARCH PROGRAM

GILEAD

Why was the combination treatment not effective?

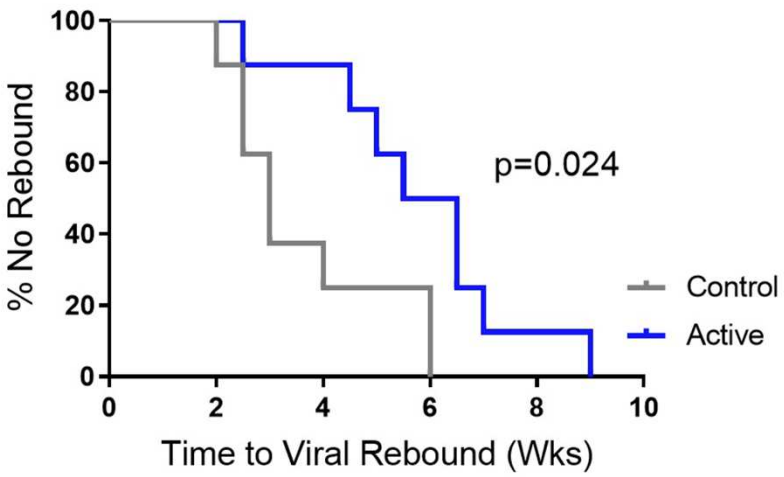
- SIV model?
  - Control may be difficult to achieve
  - IV infection route leads to establishment of a large viral reservoir
- Was the TLR7 agonist used ineffective?
  - IV route
- ADA elicited against infused mAbs?

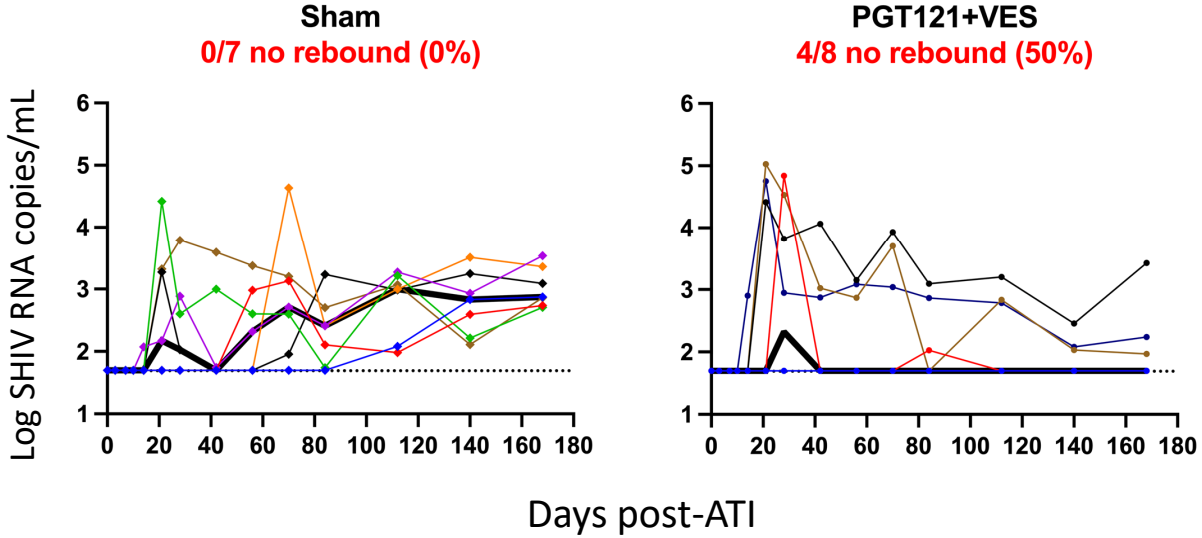


Additional experiments

- Cell-associated viral load in PBMC and lymph node
- PK analysis for antibodies
- Endogenous humoral response measurements

Other studies have (at least partially) replicated the original study





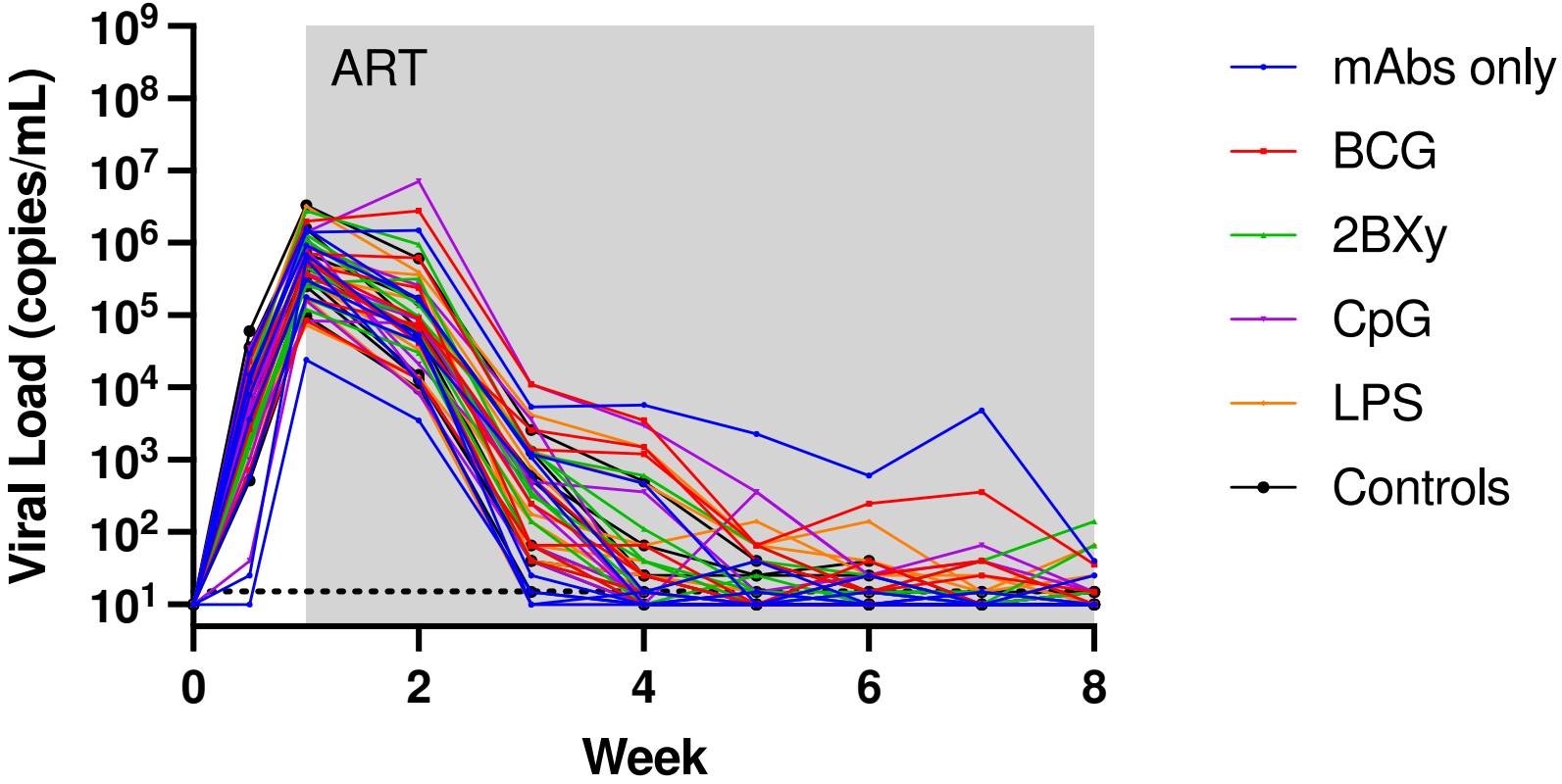
Hsu et al, PLoS Path, 2021

- SHIV 1157ipd3N4
- IR infection route
- HIV bNAbs PGT121 & N6
- TLR7 agonist GS-986 via oral gavage
- ART day 14

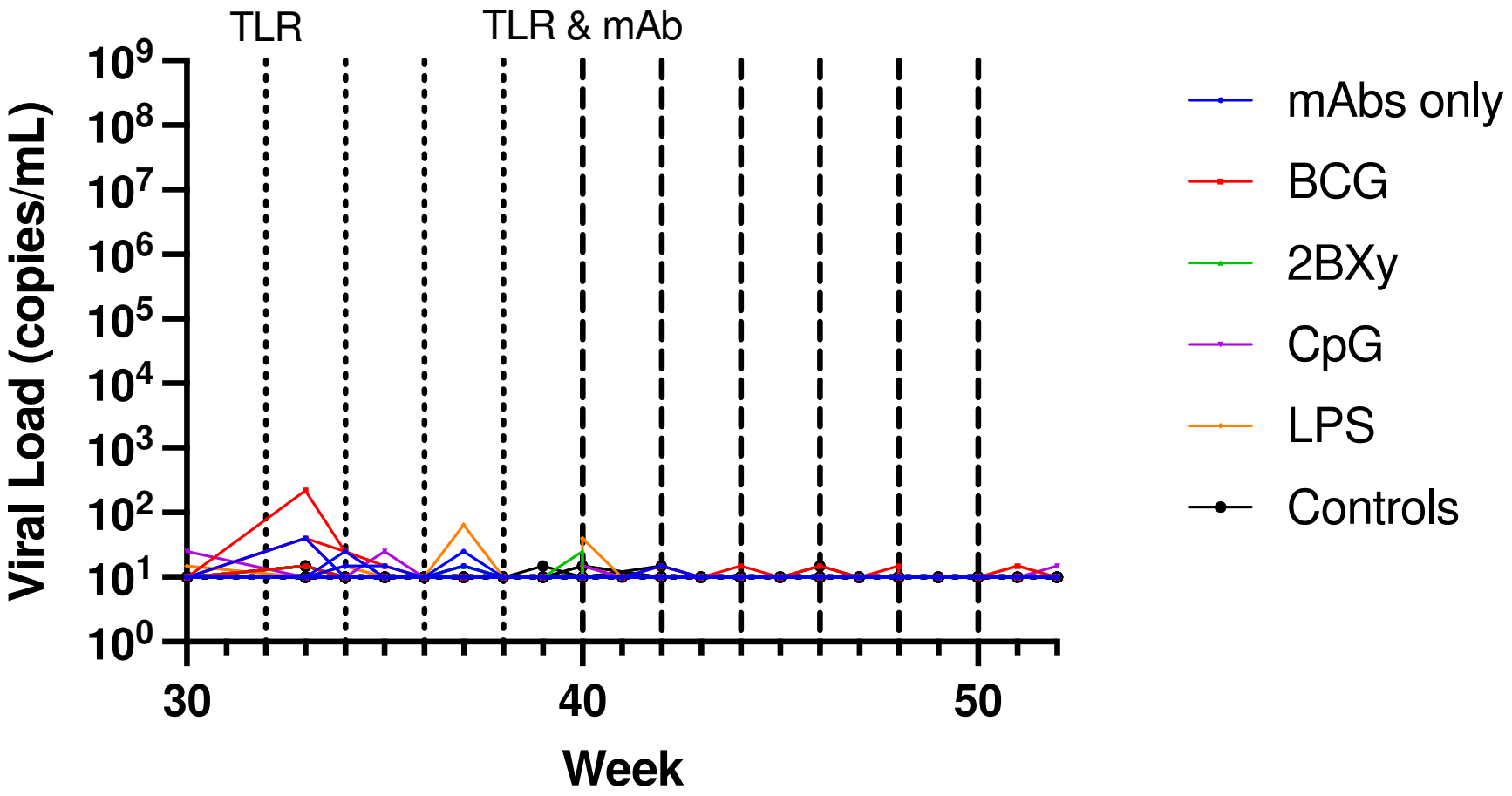
Moldt et al, PLoS Path, 2022

- SHIV SF162P3
- IR infection route
- HIV bNAb PGT121
- TLR7 agonist GS-9620 (vesatolimod) via oral gavage
- ART at 1 year

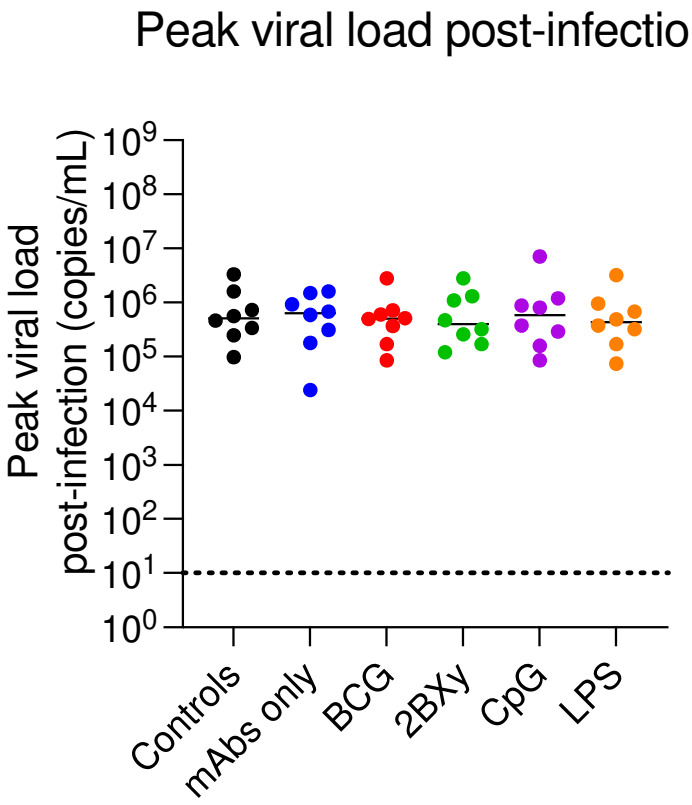
Viral loads were equivalent prior to treatment

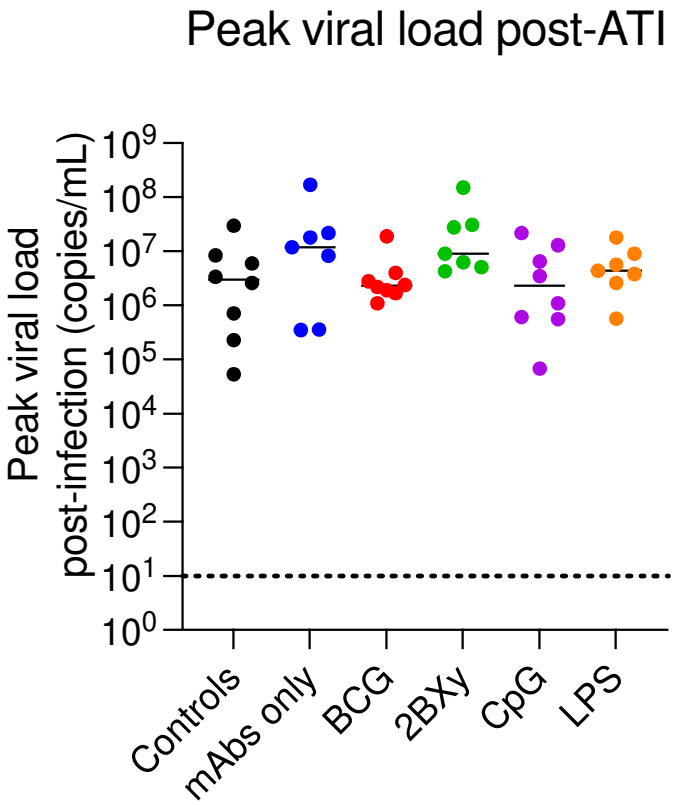


No change in viral loads during TLR agonist treatment

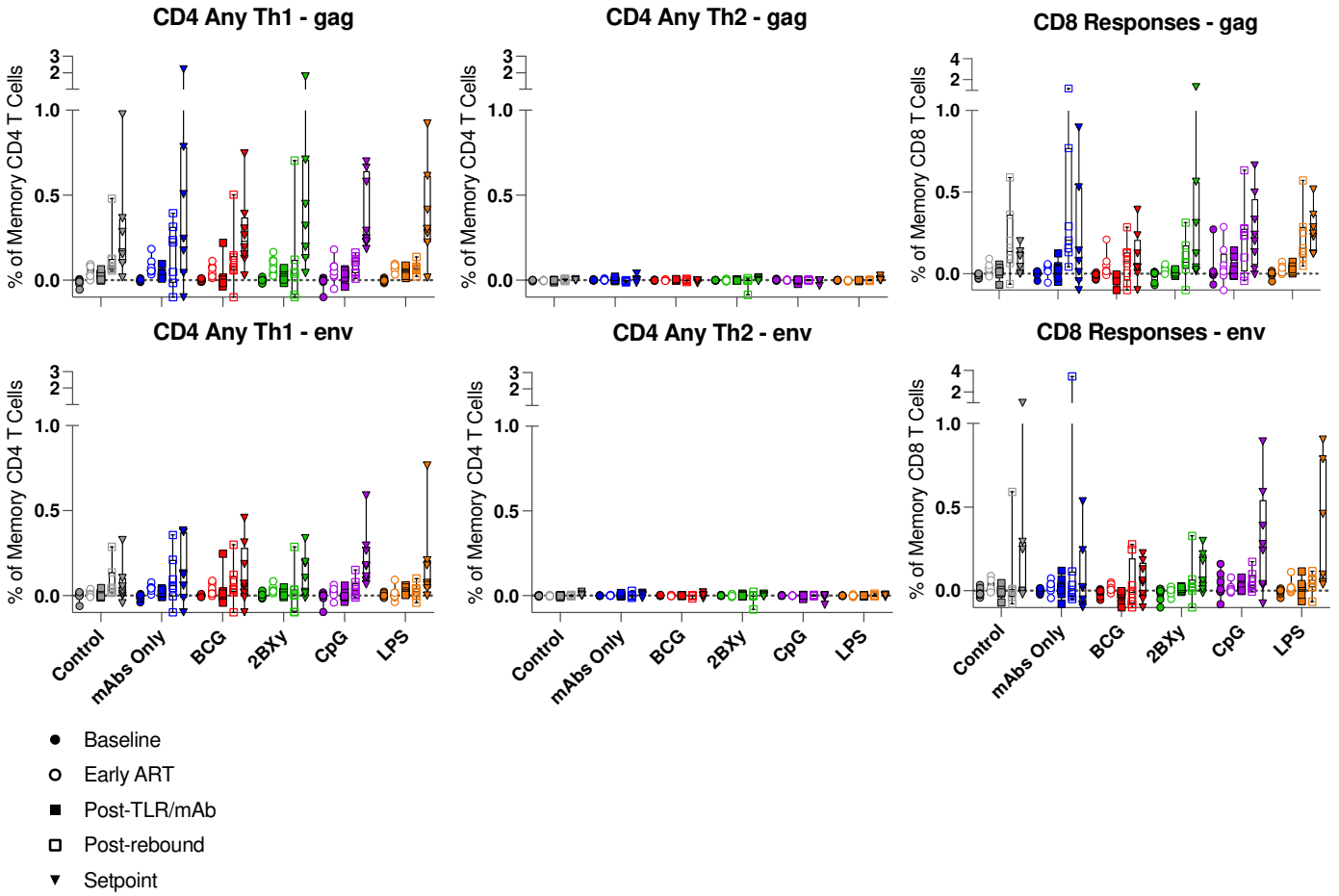


No difference in peak viral loads in early infection or post-rebound





SIV-specific T cell responses equivalent at all timepoints measured



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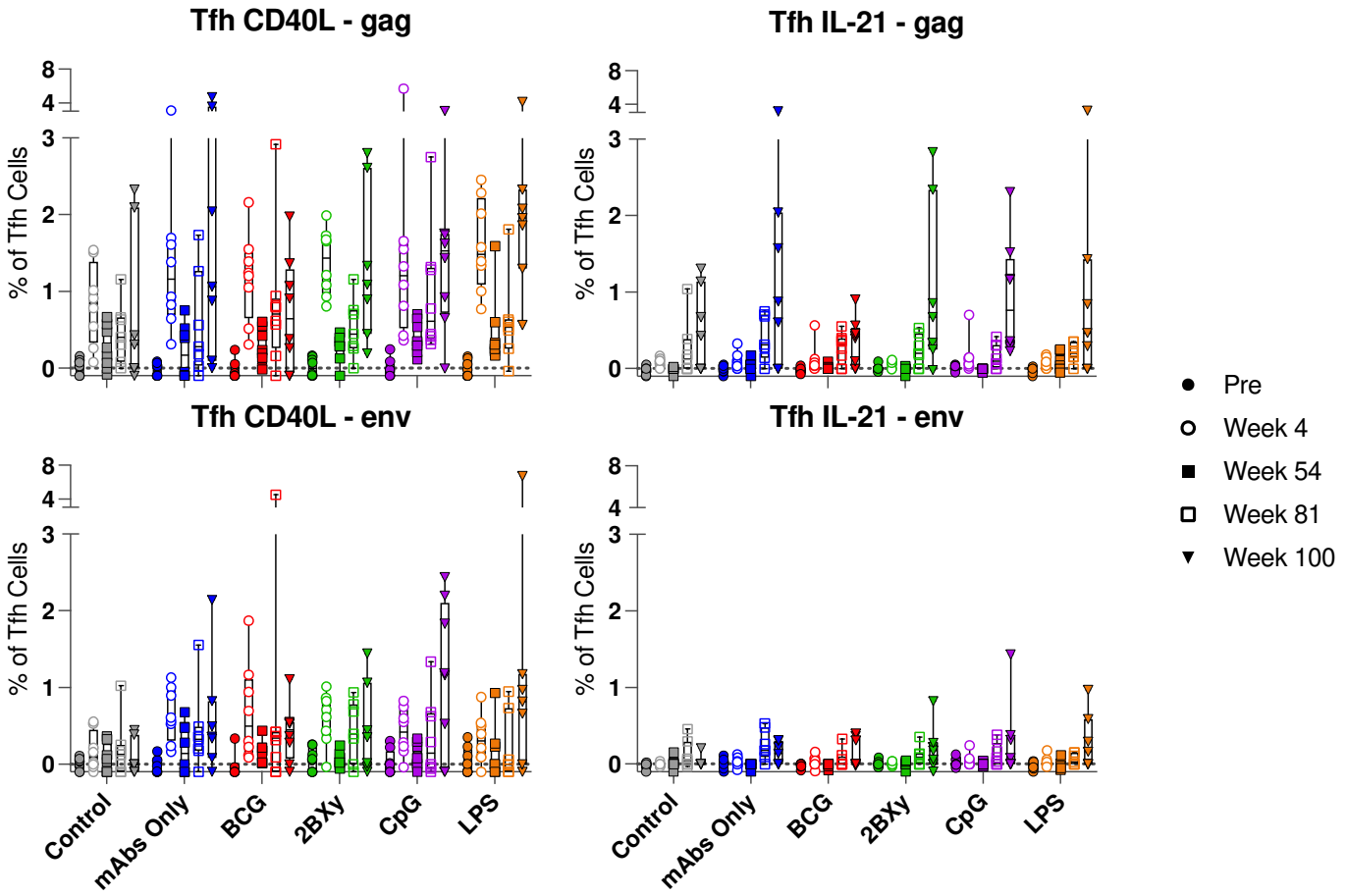

Some animals developed immune responses to 2BXy but not LPS