



Evaluation of Different Treg Depletion Approaches as Strategies for Improved SIV Reactivation and Clearance

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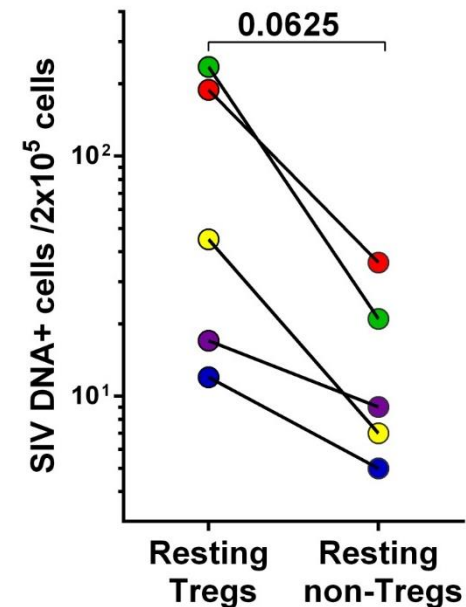


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BACKGROUND

- **T regulatory cells (Tregs) can be latently infected with HIV and may represent a potentially important HIV reservoir**
- They expand in blood and tissues in chronically HIV-infected patients and SIV-infected macaques¹
- HIV/SIV DNA positive cells are more abundant in Tregs than in non-Tregs in patients on ART and in rhesus macaques (RMs)^{2,3}
- Tregs are less susceptible to cell death than conventional T cells



1. Moreno-Fernandez, M. E., Presicce, P. & Chougnet, C. A.. *J Virol* **86**, 10262–10269 (2012).
2. Tran, T.-A. *et al. PLOS ONE* **3**, e3305 (2008).
3. Allers, K. *et al. J. Virol.* **84**, 3259–3269 (2010).



BACKGROUND



- **T regulatory cells (Tregs) have a critical contribution to the shape of the viral reservoir**
- During acute infection, Tregs decisively contribute to the establishment of HIV reservoir by reversing CD4⁺ T cell immune activation status
- During chronic infection, Tregs contribute to the impairment of CTL responses, as their expansion correlates with loss of CTL function and their *ex vivo* depletion enhances T cell responses to HIV/SIV antigens.
- The HIV-specific CD8⁺ T cells from elite controllers evade Treg suppression



BACKGROUND



- **These observations support a major involvement of Tregs in suppressing protective effector immune responses against HIV**
- **This Treg effect may be critical for the “shock and kill” strategies, which require increased virus killing of the reactivated virus**



HYPOTHESIS



Treg depletion is a valid HIV cure approach, as through a single intervention we can

- **reduce the size of the reservoir**
- **reactivate the virus**
- **boost cell-mediated immune responses**



RATIONALES



for the different therapeutic interventions aimed at Treg depletion

Anti-CCR4 DT

High concentrations of CCR4 are present on the surface of Tregs, which make CCR4 a plausible target for Treg depletion.¹

IL2 DT

The IL-2 receptor is CD25, which is a surface marker of Tregs. As such, CD25 is a plausible target for Treg depletion.²

Combination DT

A combination of the anti-CCR4 DT and the IL2 DT could improve the efficacy of Treg depletion (which is needed as Fox-P3 cannot be directly targeted being an intracellular marker).

Cyclophosphamide

Low doses of Cyclophosphamide have been shown to selectively deplete Tregs, although the mechanism is poorly understood.³

1. Wang, Z. *et al. Mol Oncol* **10**, 553–565 (2016)
2. He, T. *et al. J. Immunol.* **197**, 4535–4539 (2016).
3. Zhao, J. *et al. Cancer Res.* **70**, 4850–4858 (2010)



Treg Depletion

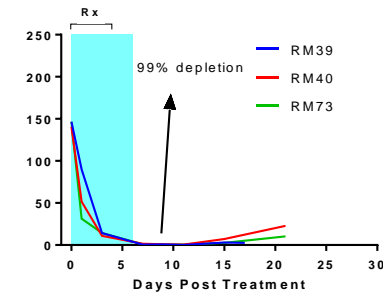
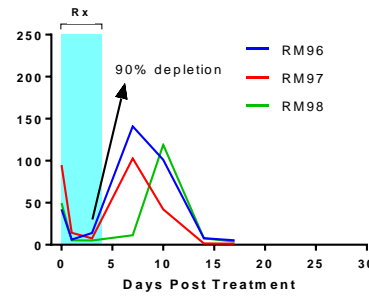
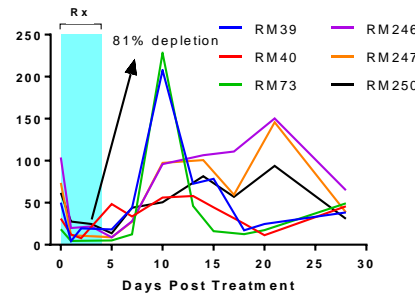
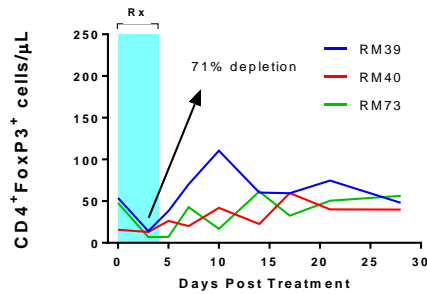
Anti-CCR4 DT

IL2-DT

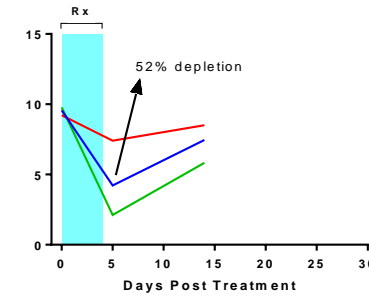
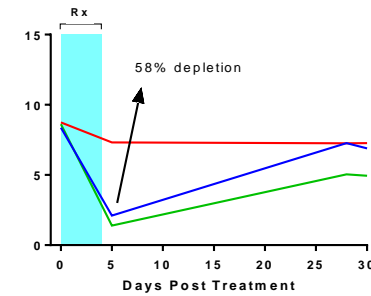
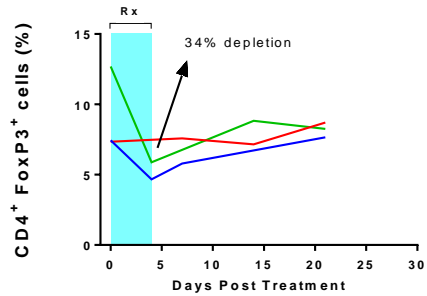
Combination DT

Cyclophosphamide

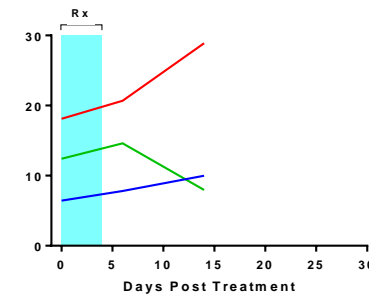
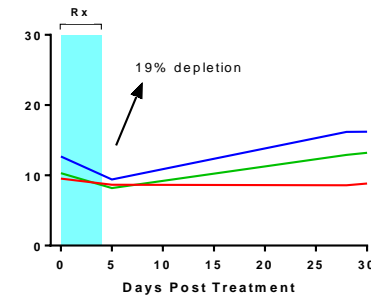
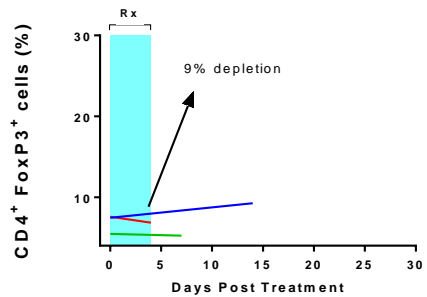
Blood



SLN



Gut





Total Lymphocytes CD4⁺ T-Lymphocytes

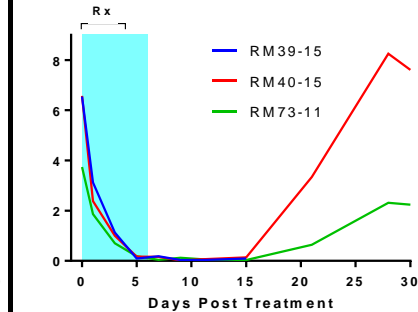
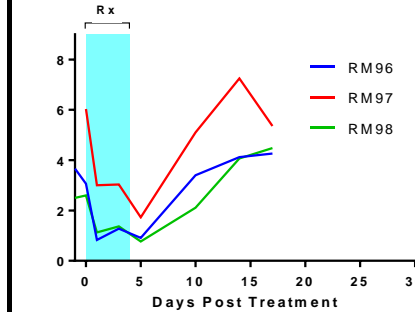
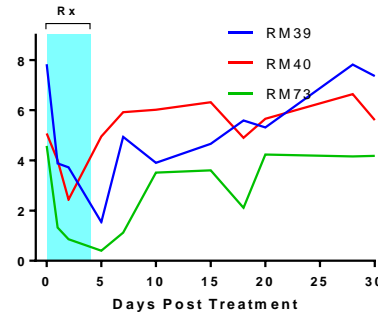
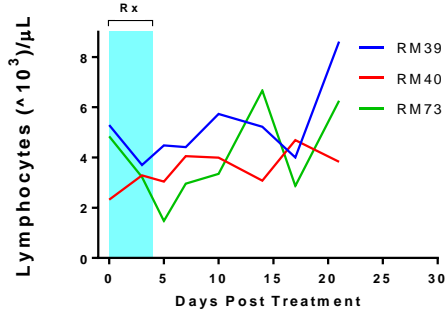
Anti-CCR4DT

IL2-DT

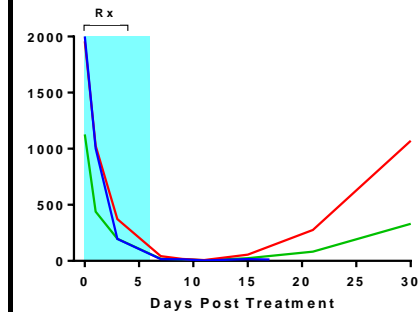
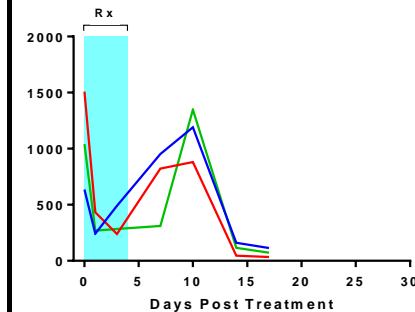
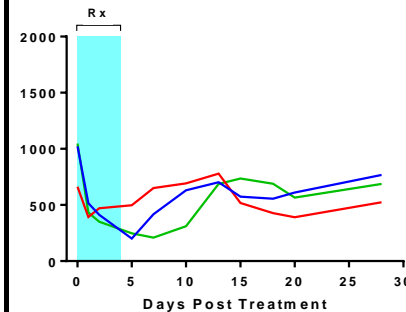
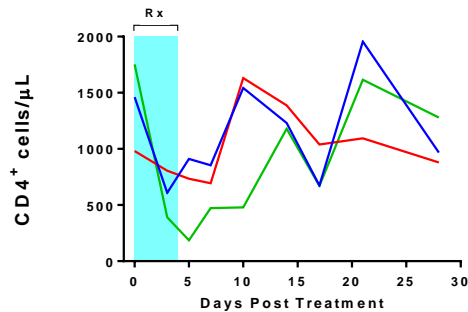
Combination DT

Cyclophosphamide

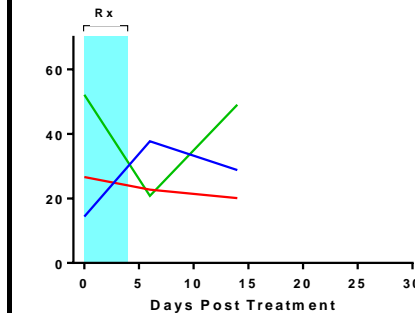
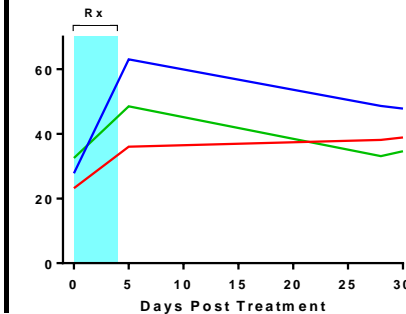
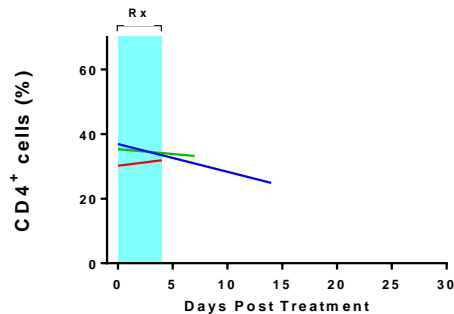
Blood



Blood



Gut





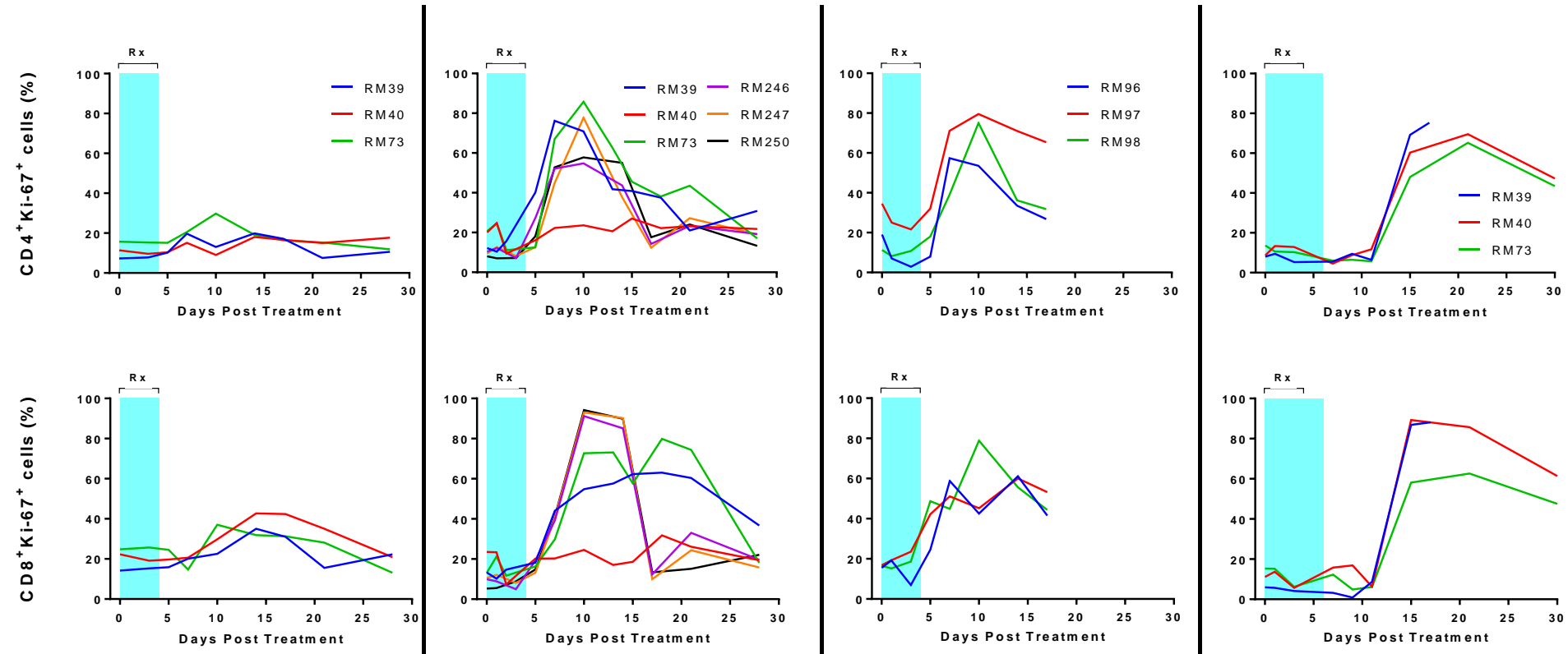
Immune Activation

Anti-CCR4DT

IL2-DT

Combination DT

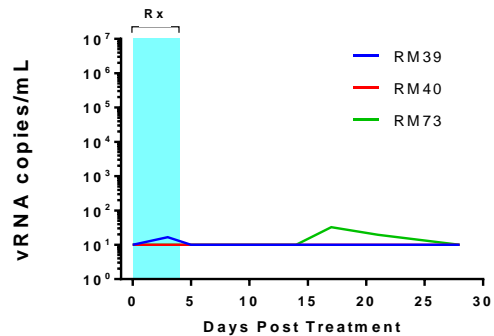
Cyclophosphamide



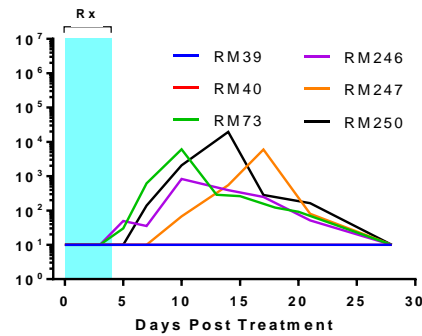


Plasma Viral Loads

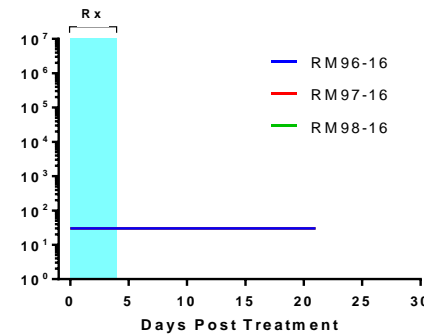
Anti-CCR4DT



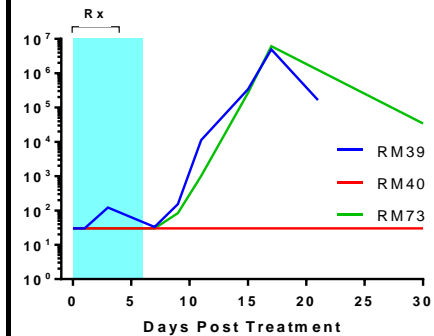
IL2-DT



Combination DT



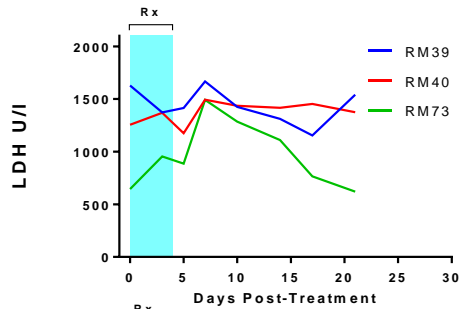
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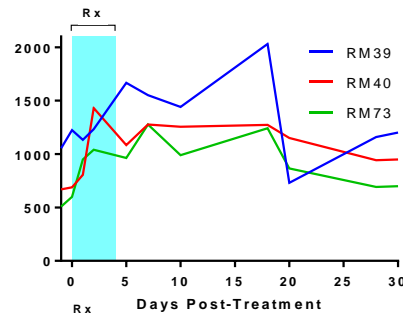


TOXICITY

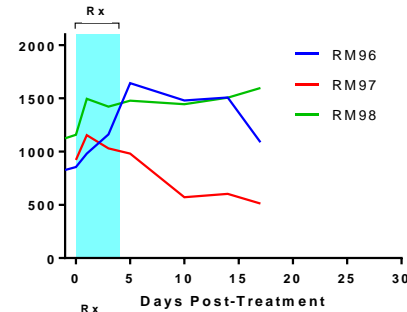
Anti-CCR4DT



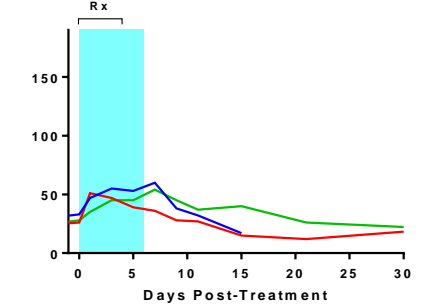
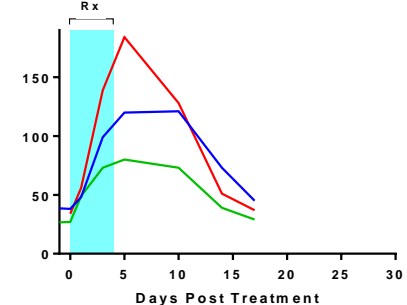
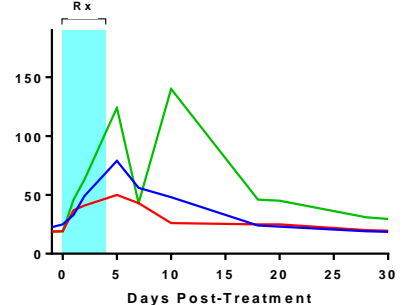
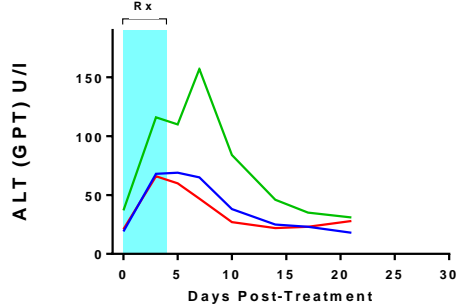
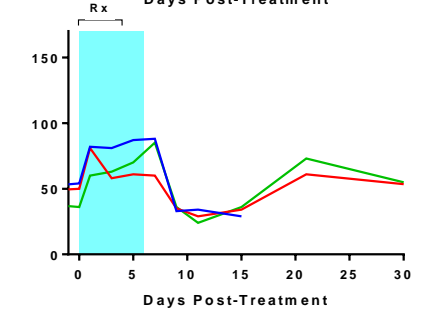
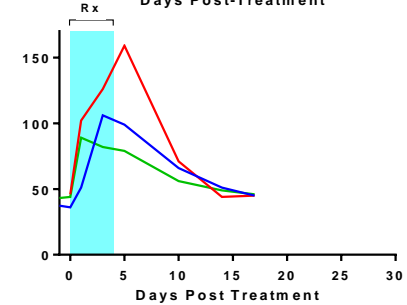
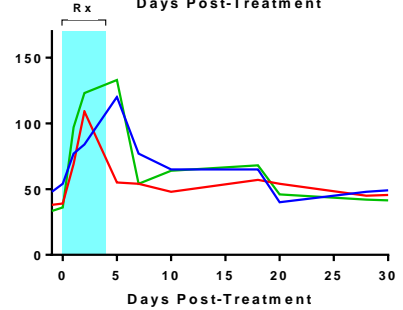
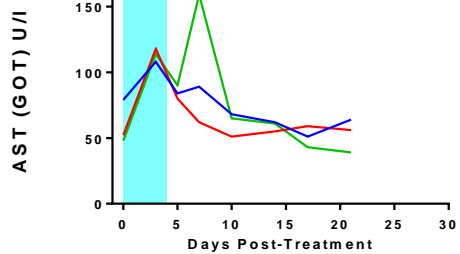
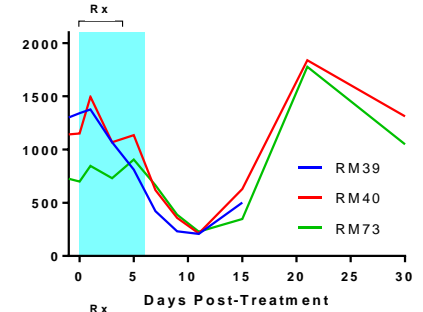
IL2-DT



Combination DT



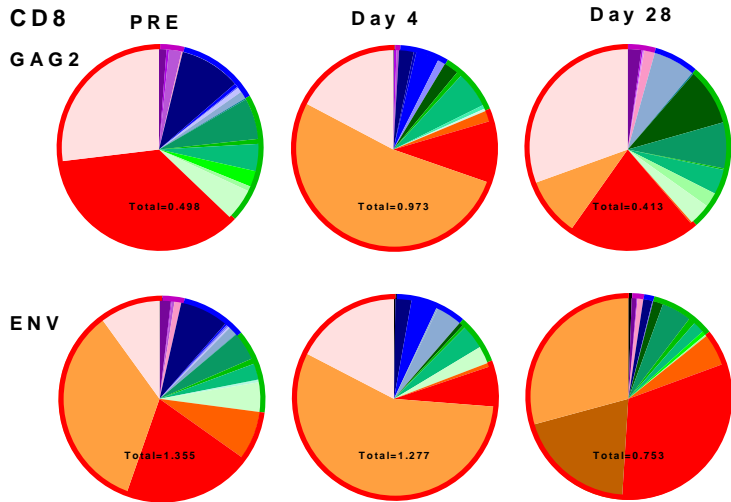
Cyclophosphamide



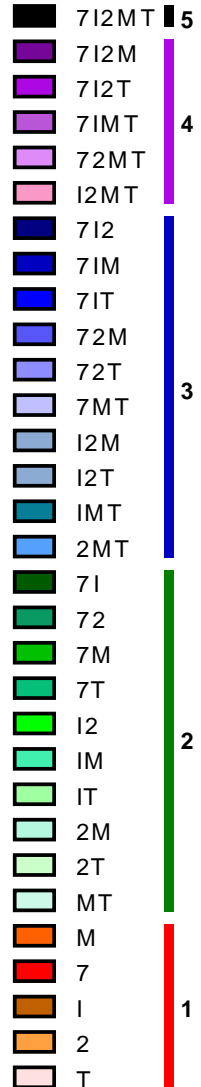
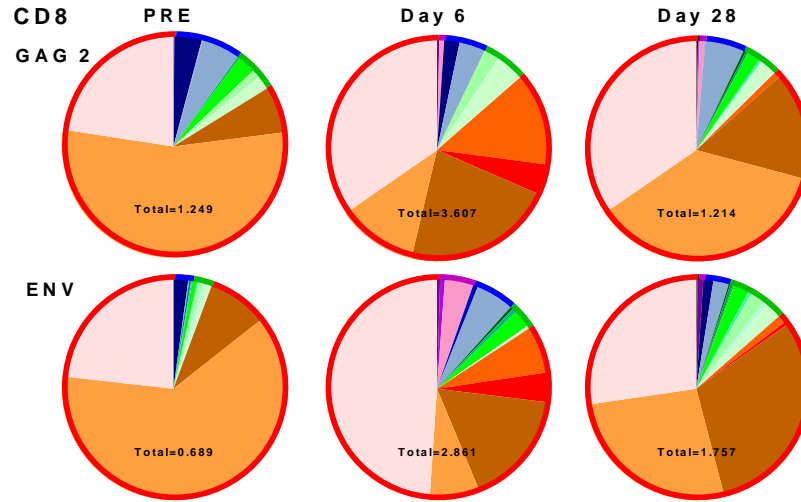


Cytotoxic T Lymphocytes

Anti-CCR4DT



IL2-DT





CONCLUSIONS



- Cy proved to be an effective cyto-reductive agent and its impact on the reservoir has to be investigated.
- Immunotoxin therapies were relatively similarly effective in depleting Tregs, yet virus reactivation and the boost of SIV-specific immune responses were more prominent after IL2-DT, probably due to the IL-2-induced activation or to the loss of suppressing abilities of residual Tregs after IL2-DT-treatment.
- Treg depletion boosted CTLs, clearing the reactivated virus.



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