Combination of eCD4-IgG1 Delivered by AAV9 and AZD5582 in SIV-infected, ART-suppressed infant RM

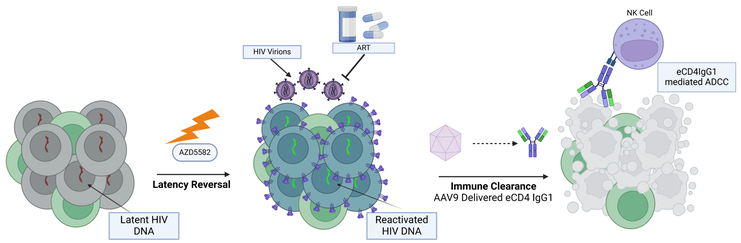
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BACKGROUND

- Children under 14 represent 25% of total HIV cases and 10% of new infections[1].
- While antiretroviral therapy (ART) suppresses HIV replication, it fails to eliminate latent reservoirs, necessitating curative strategies.
- Current latency-reversing agents (LRAs) reactivate the reservoir but do not effectively eliminate infected cells[2].
- **Latency Reversal and Clearance** strategies aim to overcome this limitation by combining LRAs with immunotherapy to clear reactivated cells.



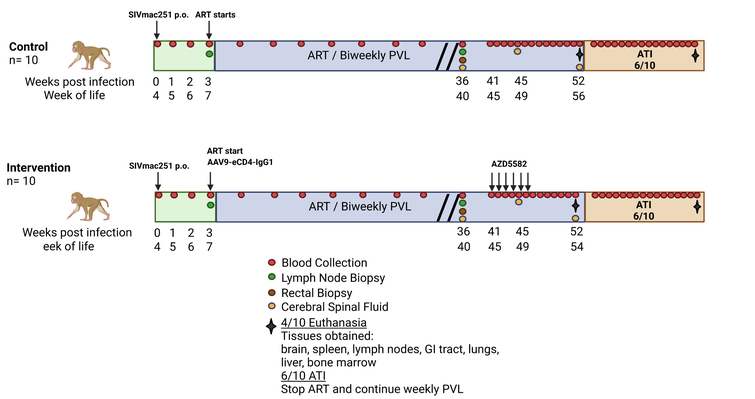
AZD5582 for Latency Reversal:

- Stimulates the non-canonical NF-κB pathway to reactivate latent HIV/SIV[3].
- Infant-specific considerations: Previous studies show reduced efficacy at doses effective in adults, aligning with evidence that pediatric HIV reservoirs are more resistant to stimulation[4,5].

AAV9-eCD4-IgG1 for Immune Clearance:

- eCD4-IgG1 is a fusion protein comprising the CD4 ectodomain, an IgG1-Fc, and the TYR-sulfated regions of CCR5 that bind to HIV/SIV Env on the cell surface, preventing viral escape due to the high conservation of its binding regions[6].
- More potent than broadly neutralizing antibodies (bnAbs) in neutralizing HIV/SIV and promoting antibody dependent cytotoxicity (ADCC).
- Engineered for long-term expression via AAV9, eliminating the need for repeated infusions.

STUDY DESIGN



CONCLUSIONS:

First proof-of-concept study combining an AAV9-delivered immunotherapeutic with a latency-reversing agent (LRA) in infants.

A single AAV9 dose led to persistent eCD4-IgG1 expression.

AZD5582 triggered robust on-ART viremia, exceeding levels previously observed in adult studies.

A subset of treated infants exhibited partial viral control following rebound, we are currently investigating the immune mechanisms driving suppression.

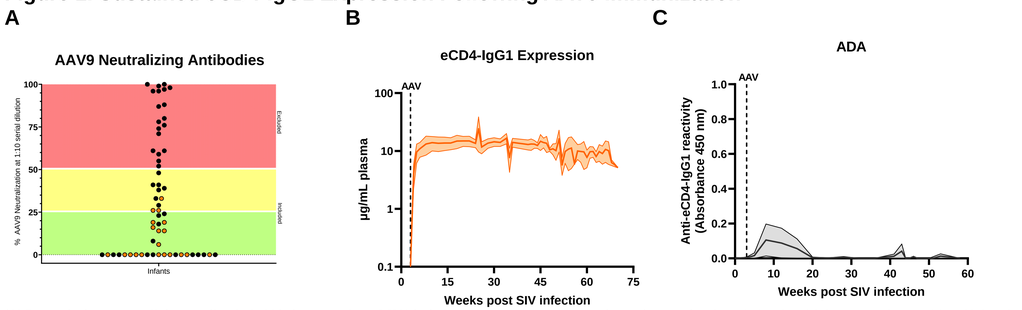
Partial correlation analyses revealed a strong negative association between peak AZD5582-induced on-ART viremia and the area under the curve of viremia post-ART interruption (r = –0.870, p = 0.055).

RESULTS

Table 1: Group Characteristics Before ART Initiation

Group	Control	Intervention	P
Male to Female Ratio	1:1	1:1	>0.9999
Number of Viral Challenges for Infection (Mean)	2	1.6	0.6255
Age of Infection in Weeks (Mean, SD)	6.3 (2.55)	5.4 (2.70)	0.4147
A01 Allele	6/10	4/10	0.3711
Peak Plasma Viral Load (Mean)	5.93E+07	5.48E+07	0.8401
Plasma Viral Load at week 2 (Mean)	5.56E+07	4.05E+07	0.54
Plasma Viral Load at week 3 (Mean)	1.94E+07	2.44E+07	0.5897

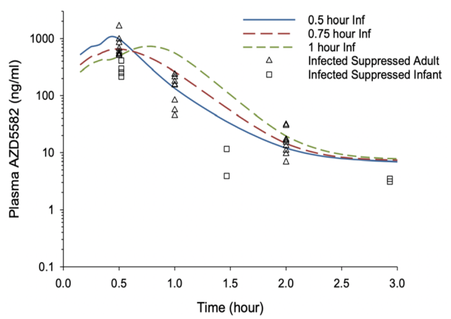
Figure 1. Sustained eCD4-IgG1 Expression Following AAV9 Immunization



(A) Pre-existing neutralizing antibody (nAb) titers against AAV9 in infant rhesus macaques prior to immunization. Animals with low or undetectable nAb levels were selected for inclusion. **(B) Longitudinal plasma eCD4-IgG concentrations following AAV9-eCD4IgG immunization**. eCD4-IgG levels were measured up to 70 weeks post-immunization, demonstrating stable transgene expression. **(C) Anti-drug antibody (ADA) responses against eCD4-IgG over time in immunized infant macaques.** One animal developed transient ADA responses, which did not significantly impact eCD4-IgG expression.

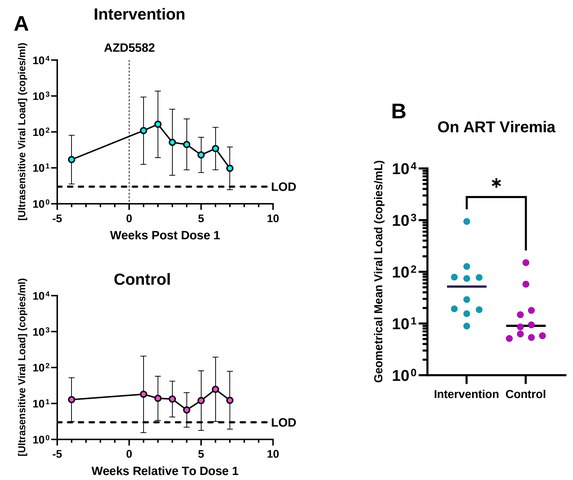
Figure 2: A 0.2 mg/kg Dose of AZD5582 Can Replicate Adult Macaque's Pharmacokinetics in Infants

Pharmacokinetic (PK) simulations of a 0.2 mg/kg dose of AZD5582 across varying infusion durations. The simulations are based on previous studies conducted by our laboratory, where plasma concentrations of AZD5582 were examined in infant macaques (n=15) across doses ranging from 0.1 to 0.25 mg/kg. These simulations are tailored for an ideal monkey of average weight.
The colored lines represent AZD5582 simulations for different infusion durations. To contextualize these simulations, actual AZD5582 levels observed in adult and infant macaques from two different studies, both administered with the previously effective dose of 0.1 mg/kg, are included for comparison.



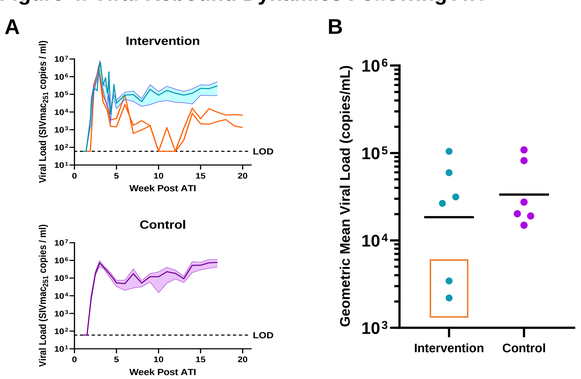
RESULTS

Figure 3. Latency Reversal Following AZD5582 Administration



(A) On-ART viremia following AZD5582 administration in intervention and control animals. Intervention animals received a loading dose of 0.25 mg/kg, followed by five weekly doses of 0.2 mg/kg. Viremia was measured weekly for 7 weeks post-loading dose using an ultrasensitive assay. During treatment, 8/10 infant RM had on-ART viremia >60 copies/mL, peaking at 5,600 copies/mL. Of 67 viral load measurements, 47 (65.7%) exceeded baseline.
(B) Geometric mean of SIV viremia over time post-AZD5582. Intervention animals had significantly higher viremia than controls (p=0.0147, Mann-Whitney test).
Viremia was quantified using an ultrasensitive assay (LOD: 3 SIV copies/mL).

Figure 4. Viral Rebound Dynamics Following ATI



(A) Mean plasma viremia (± SEM) over time following ATI. Six macaques per group were monitored for 20 weeks post-ATI. Despite sustained eCD4-IgG expression, all intervention animals rebounded within 6–11 days post-ART cessation.
(B) Geometric mean viremia per animal over time post-ATI. While there were no significant differences between groups, two intervention animals exhibited a geometric mean viremia 1.2076 log-units lower than the control group (~16-fold reduction), suggesting potential post-rebound viral control in a subset of treated animals.

References

1. UNAIDS. 2023.
2. Singh V. Curr HIV/AIDS Rep. 2021;18(2):117-27.
3. Nixon CC, Mavigner M, Nature. 2020;578(7793):160-5.
4. Bricker KM. J Virol. 2022;96(7):e0169921.
5. Dhummakupt A. JCI Insight. 2020;5(4).
6. Davis-Gardner ME. PLoS Pathog. 2017;13(12):e1006786.

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