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## BACKGROUND

Intact proviral DNA (IPD) assay is a measure of the replication competent HIV reservoir. Little is known about decay patterns of IPD in people with HIV (PWH) during long-term (15-20 yr) suppressive ART.

## METHODS

Participants in ACTG A5321 with chronic HIV and documented suppression of viremia (<50 cp/mL) for >15 yr had measurements of intact, 5' or 3' defective, and total proviral DNA from blood samples. A biexponential model for IPD was estimated using non-linear regression.

## RESULTS

- Fourteen participants (5 assigned female sex at birth) evaluated longitudinally from ART yr 1 to ART yr 17-23 (median 20 yr; 8-10 timepoints). (Table 1)
- Median pre-ART plasma HIV RNA 4.2 log<sub>10</sub> cp/mL; median pre-ART CD4 count 377/mm<sup>3</sup>.
- At yr 1 of ART, median IPD: 204 cp/million CD4+ T-cells; median intact provirus percentage (intact divided by total proviruses): 66%
- At last time point (median 20 yr on ART), median IPD fell to 16 cp/million CD4+ T-cells; intact provirus percentage fell to 7%
- Decay of intact proviruses but not defective proviruses over the course of time on ART: intact proviruses declined by 13-fold from first to last time point, whereas total proviruses (which includes intact provirus) declined by only 3-fold and defective provirus levels were generally stable (median 0.7-fold reduction) (Table 2, Fig 2).
- Five participants had biphasic IPD decay, 3 had biphasic decay with a second phase plateau (slope effectively zero), and 2 showed evidence of increased IPD levels during the second decade (Figure 1)
- Inflection or transition of decay occurred at a median of 5 yr after ART initiation (range 2-13 yr).
- Median first phase IPD half-life was 1 yr (n=10), whereas median second phase IPD half-life was >25 yr (n=8).
- In 4 participants, there was a variable pattern of IPD decay, or substantial censored DNA levels, perhaps due to fewer cells assayed or lower IPD levels. Not included in modeling

## CONCLUSIONS

- In PWH receiving two decades of suppressive ART, selective decay of intact proviruses with three patterns of intact provirus decay revealed: 1) initial rapid decline followed by markedly slower second phase decline; 2) initial rapid decline followed by zero-slope plateau; 3) initial rapid decline followed by late increases in intact proviruses.
- Slowing or reversal of intact provirus decay suggests mechanisms of infected cell clearance and persistence are different during first phase decay compared with second phase.
- Findings may reflect 1) increasing inability to clear cells with intact but transcriptionally silent proviruses; 2) clonal expansion of cells with intact proviruses.

**Table 1: Participant Characteristics**

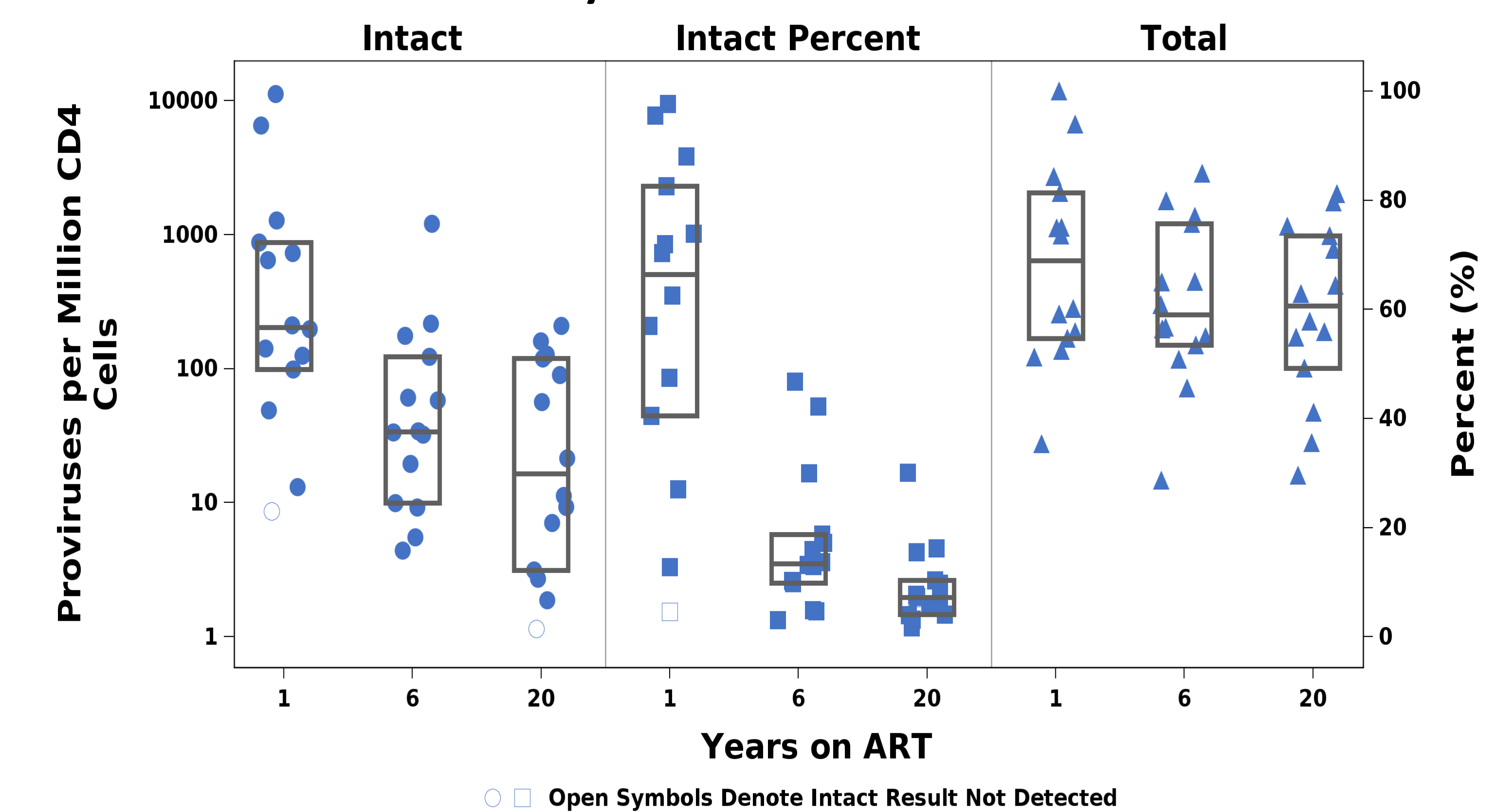
	Sex	Age (yr) Last Timepoint	Years on ART	Pre-ART CD4 count (cells/mm <sup>3</sup> )	IPDA Grouping
1	F	50	18	524	A) Rapid Decline/Slower 2 <sup>nd</sup> Phase
2	M	53	21	216	A) Rapid Decline/Slower 2 <sup>nd</sup> Phase
3	M	57	22	316	A) Rapid Decline/Slower 2 <sup>nd</sup> Phase
4	M	60	17	49	A) Rapid Decline/Slower 2 <sup>nd</sup> Phase
5	M	68	23	347	A) Rapid Decline/Slower 2 <sup>nd</sup> Phase
6	F	57	19	353	B) Rapid Decline/Plateau
7	M	63	20	553	B) Rapid Decline/Plateau
8	F	74	19	762	B) Rapid Decline/Plateau
9	F	66	22	47	C) Rapid Decline/Late Increase
10	F	75	19	746	C) Rapid Decline/Late Increase
11	M	45	22	401	Variable
12	M	54	21	443	Variable
13	M	67	20	505	Variable
14	M	72	22	27	Variable

**Table 2: Decay of intact proviruses but not defective proviruses**

Fold-reduction from first to last timepoint		
	Median (Q1, Q3)	(Minimum, maximum)
Intact Proviral DNA	13.4 (4.6, 46.0)	(1.8, 91)
Defective Proviral DNA	0.7 (0.3, 1.7)	(0.1, 4.5)
Total Proviral DNA	2.7 (1.2, 4.9)	(0.3, 6.7)

Decreases in intact, defective, and total proviral DNA from first to last timepoint. First timepoint is year one on ART. Last timepoint is year 17-23 on ART. Q1: 25<sup>th</sup> percentile. Q3: 75<sup>th</sup> percentile.

**Figure 2: Intact Proviruses, Intact Percent (intact/total), Total Proviruses Over 20 years of ART**



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**Figure 1: Intact Provirus Decay over Two Decades of Suppressive ART:**

- 5 participants: initial rapid decline followed by markedly slower second phase decline (Pattern A).
- 3 participants: initial rapid decline followed by zero-slope plateau (Pattern B)
- 2 participants (both female): initial rapid decline followed by late increases (nadir to final, 2.9-fold, 6.3-fold) (Pattern C).

