

**Upregulation of IL-32 isoforms in virologically-suppressed HIV-infected individuals:
Potential role in persistent inflammation and transcription from stable HIV-1 reservoirs**

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Supplemental materials

Variable	HIV+ on cART Viremia <50 copies/ml	HIV+ ON/Off ART Viremia > 50 copies/ml	HIV ^{neg}
Gender	Female N=51 (13.5%)	Female N= 9 (19.5%)	Female N= 22 (20.5%)
	Male N= 328 (86.5%)	Male N= 37 (80.5%)	Male N= 85 (79.5%)
Age (Years)	54.9±9.19	53.98±12.84	55.03±9.88
Viral load (Log ₁₀ Copies/ml)	1,57±0,21	2.93±1.26	NA
Nadir CD4 count (Cells/mm ³)	216±167	288±211	NA
CD4 counts (Cells/mm ³)	647±866	480±323	Unknown
Duration of infection (Years, Average ± SD)	17.22±7.94	18.96±9.62	NA
Duration of cART (Years, Average ± SD)	12.7±6.7	ON ART: 8.95±9 (n=20) OFF ART: 13±8 (n=17) ART unknown (n=9)	NA

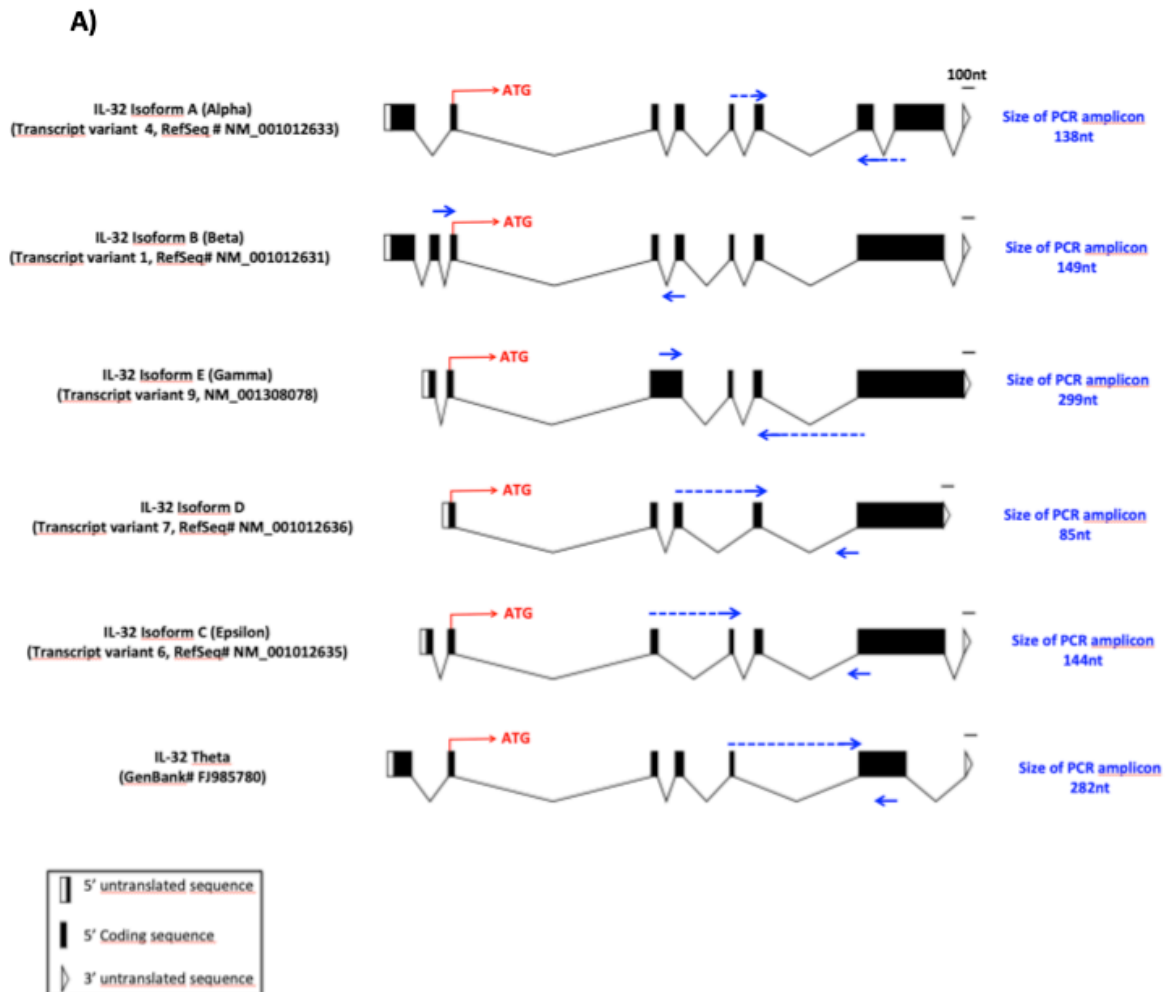
Supplemental Table 1: Demographic and clinical parameters of study participants at the time of study entry. NA: non-applicable.

Gene/Isoform	NCBI Reference number	Forward Primer	Reverse Primer
β-Glucuronidase	NM_000181.4	TCTGTCAAGGGCAGTAACCTG	GCCACGACTTTGTTTTCTG
IL-32α	NM_001012633	CACCCAGAGCTCACTCCTCT	GGCTCCGTAGGACTTGTCAC
IL-32β	NM_001012631	GAAGACTGCGTGCAGAAGGT	CTTTCTATGGCCTGGTGCAT
IL-32γ	NM_001308078	GGTAATGCTCCTCCCTAC	GAGGAGTGAGCTCTGGGTGC
IL-32D	NM_001012636	AGGACGTGGACAGGACGACT	AGGAGTGAGCTCTGGGTGCT
IL-32ε	NM_001012635	AGGCCCGAATGGTGATGT	GGCACCGTAATCCATCTCTT
IL-32θ	FJ985780	GAGCTGGAGGAGCTCACTCC	GAGCTCTGACAGAGAGCAGC

Supplemental Table 2: Primer sequences for qRT-PCR assays to amplify IL-32 isoforms and the reference house-keeping gene.

PCR reaction	Primer sequence
Reverse transcription and 1st PCR	
Forward	5'-ATG CCA CGT AAG CGA AAC TCT GGG TCT CTC TDG TTA GAC-3'
Reverse	5'-CCA TCT CTC TCC TTC TAG C-3'
Nested Real-time PCR	
Forward	5'-ATG CCA CGT AAG CGA AAC T-3'
Reverse	5'-CTG AGG GAT CTC TAG TTA CC-3'
Probes	5'-/FAM/CA CTC AAG G/ZEN/C AAG CTT TAT TGA GGC /3IABkFQ/-3'

Supplemental Table 3: Primer sequences for Real-time RT-PCR assays of HIV RNAs.



Supplemental Figure 1: Primer positions for specific RT-PCR amplification of the IL-32 isoforms α , β , γ , D, ϵ , and θ . Schematic representation for the IL-32 exons within each of the individual IL-32 isoforms detected and quantified using specific primers sets. Sequence blast was done using individual IL-32 transcript variants, with the NCBI RefSeq numbers noted to the left of each isoform and IL-32 gene on chromosome 16 (RefSeq# NG_029254.1). Forward and Reverse Primer positions are indicated with blue arrows (dashed arrows indicate that the primer sequence was designed to overlap the extremities of two adjacent exons). Expected amplicon's size is shown to the right of each isoform.