

Title: Extracellular condensates (ECs) are endogenous modulators of HIV transcription and latency reactivation

Wasifa Naushad¹, Lakmini S Premadasa², Vyshnavi Tallapaneni¹, Bryson C. Okeoma¹, Ashok Chaudhary^{3,4}, Jack T. Stapleton^{3,4}, Mahesh Mohan^{2*}, Chioma M. Okeoma^{1,5*},

¹Department of Pathology, Microbiology & Immunology, New York Medical College, Valhalla, NY, USA

²Host Pathogen Interaction Program, Southwest National Primate Research Center, Texas Biomedical Research Institute, San Antonio, TX 78227-5302 USA

³Department of Internal Medicine, Carver College of Medicine, University of Iowa, 200 Hawkins Drive, Iowa City, IA 52242-1109, USA.

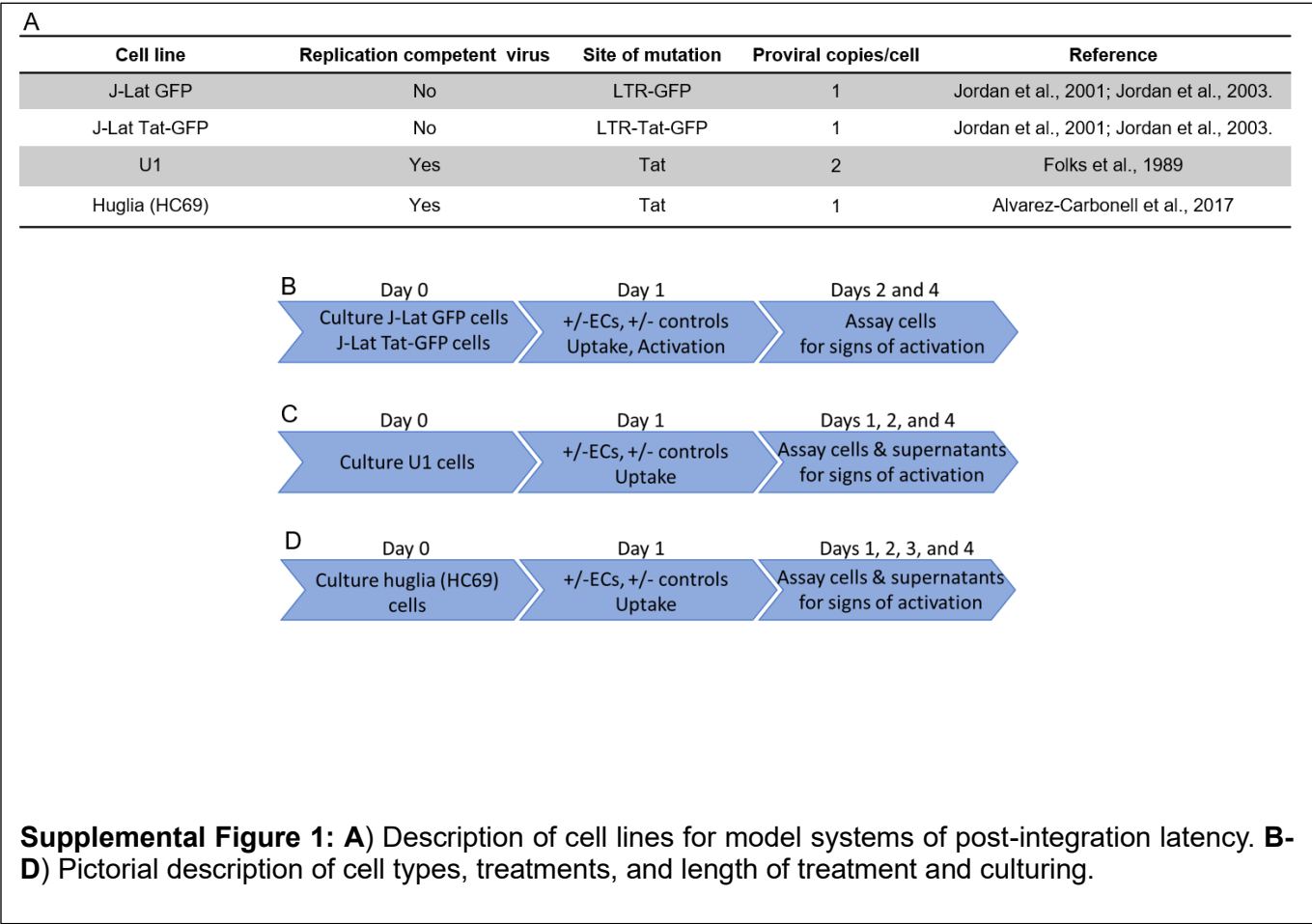
⁴Medical Service, Iowa City Veterans Affairs Medical Center, University of Iowa, 604 Highway 6, Iowa City, IA 52246-2208, US.

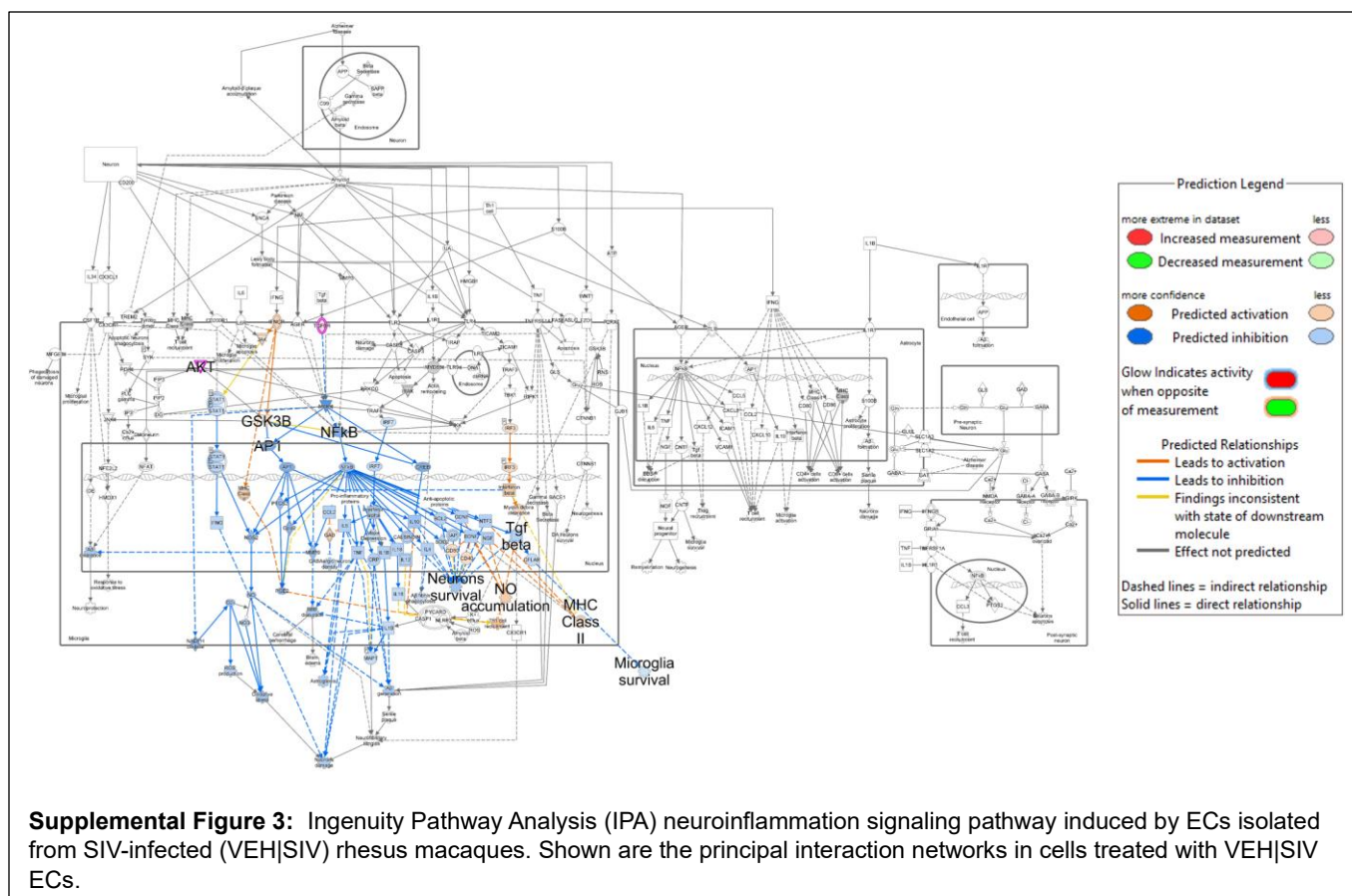
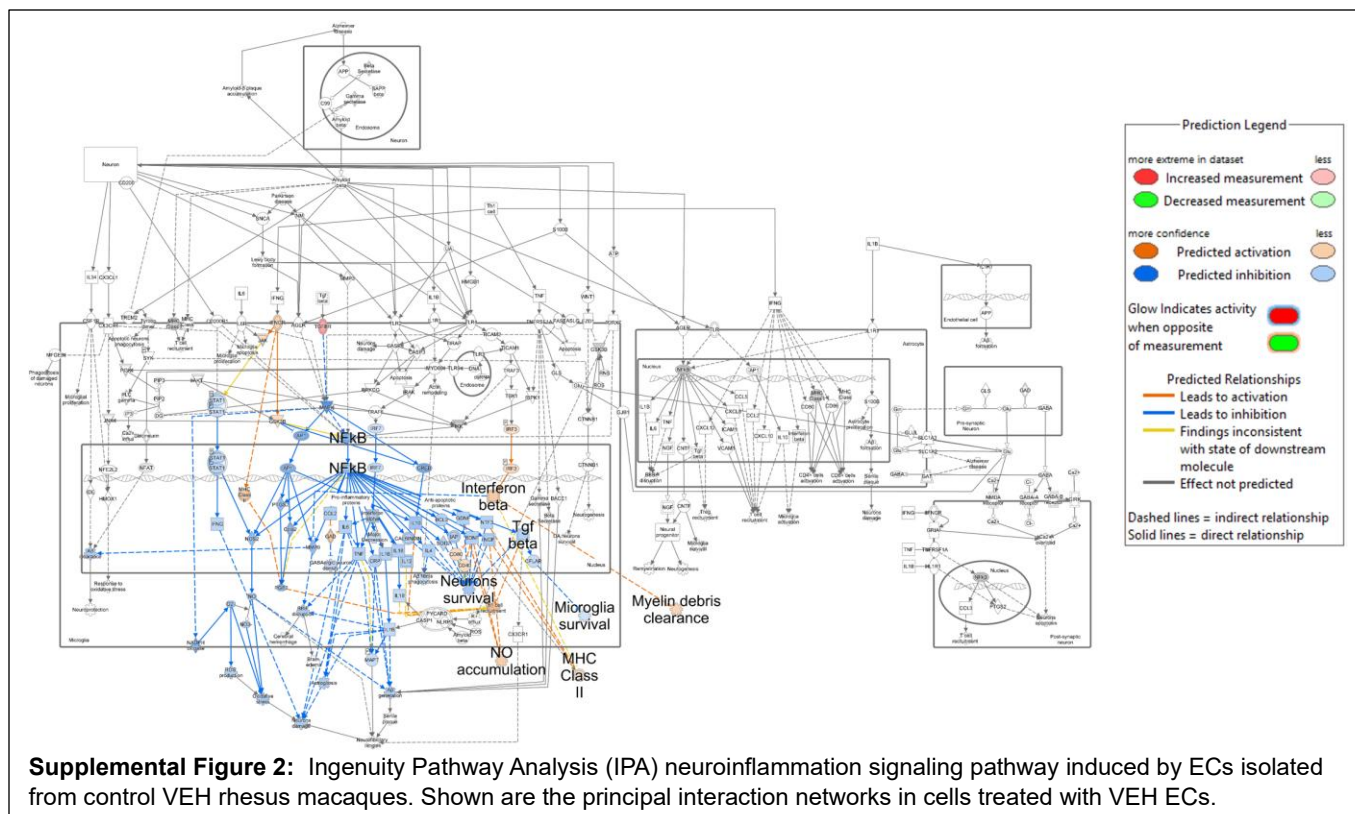
⁵Lovelace Biomedical Institute, Albuquerque, NM 87108-5127, USA.

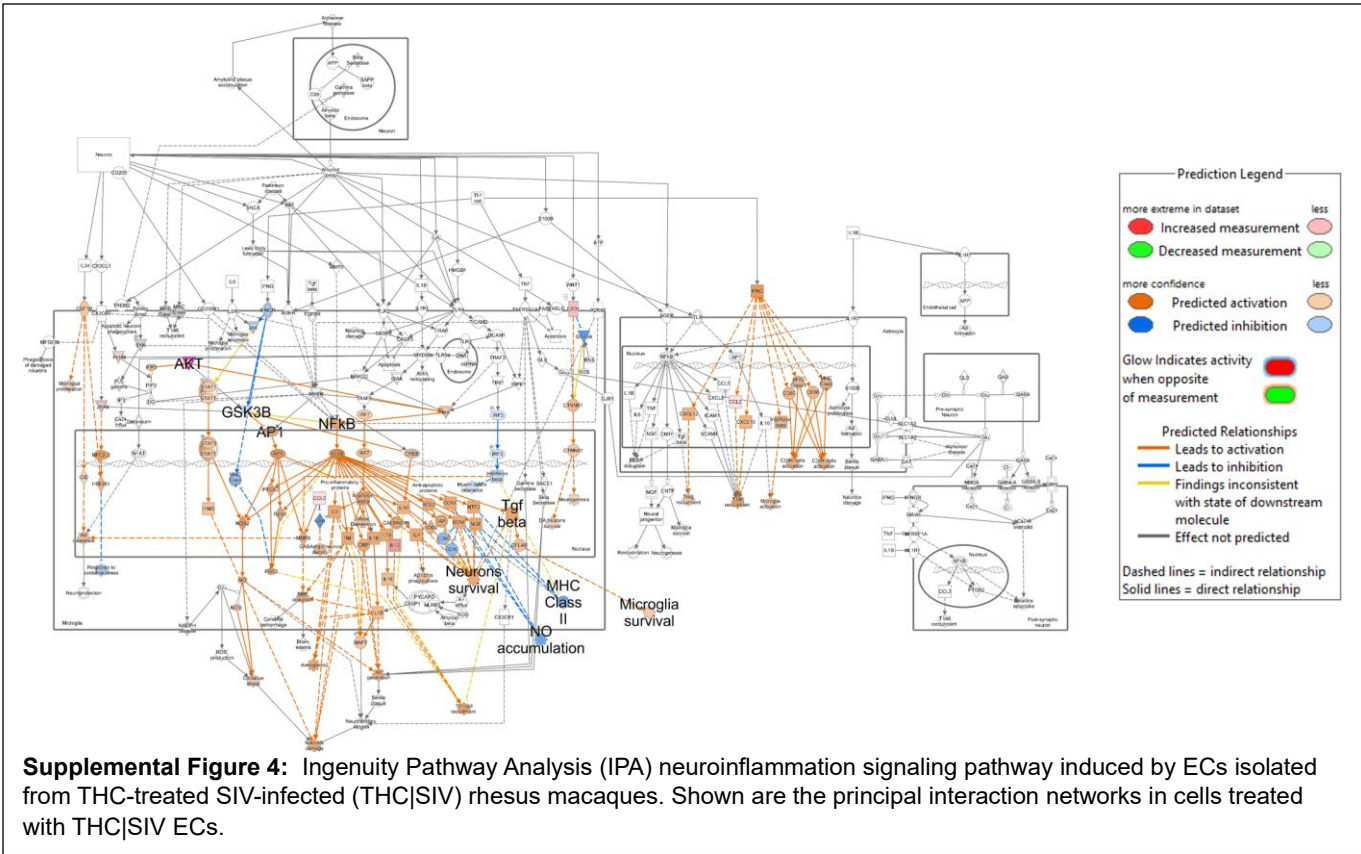
*Corresponding authors

Running title: Extracellular condensates regulate HIV persistence

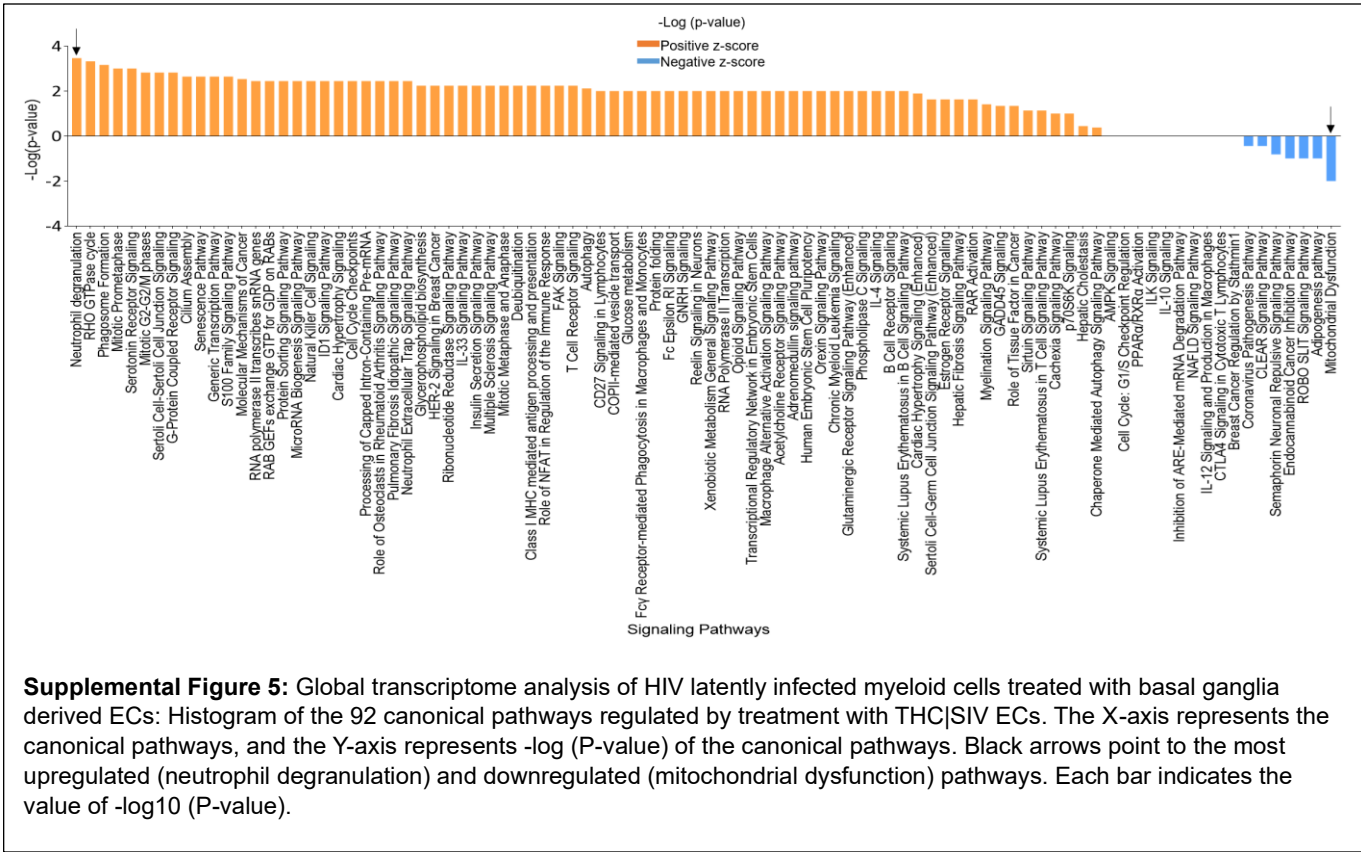
Supplementary Figures
Figures S1 – S9



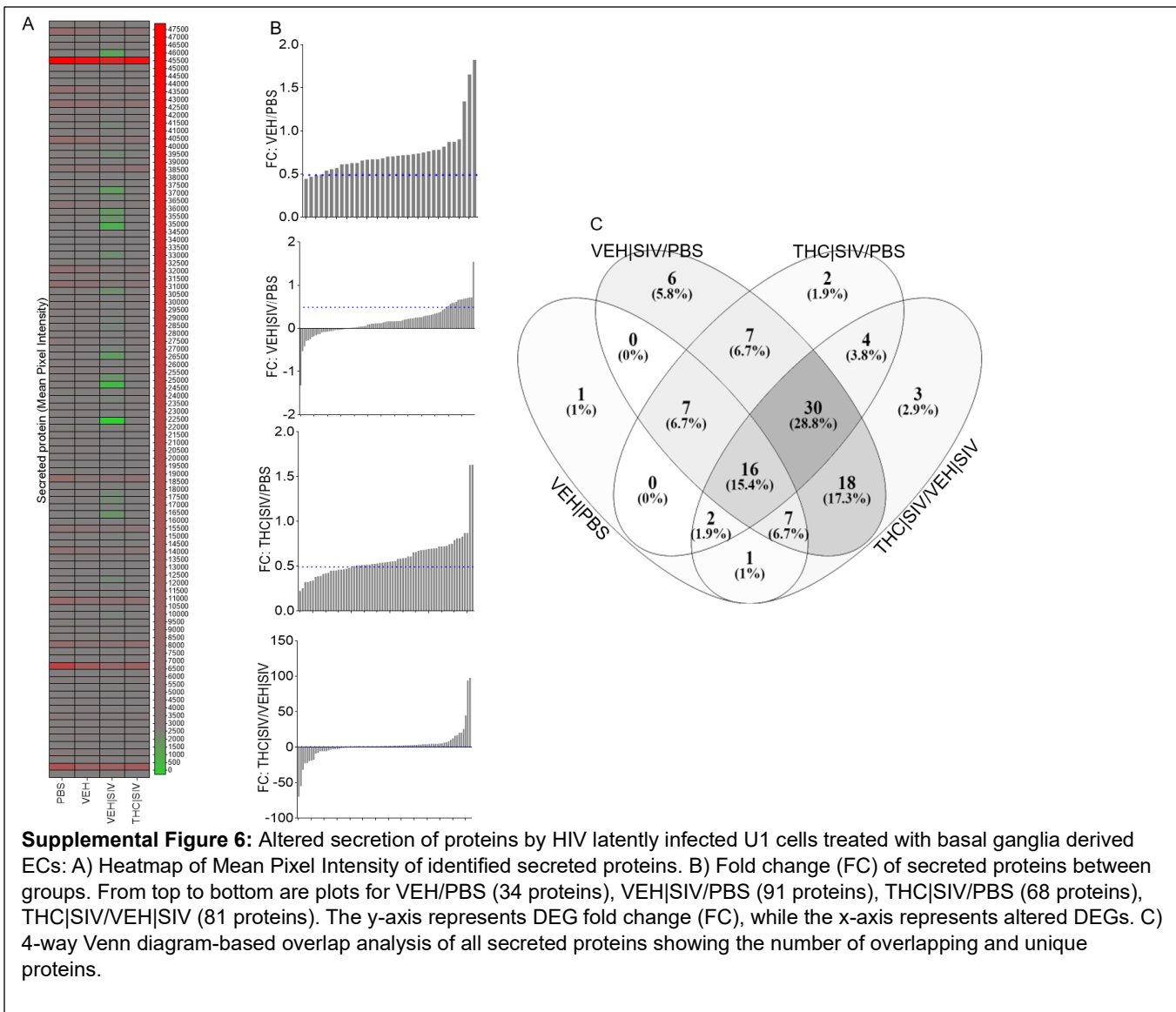


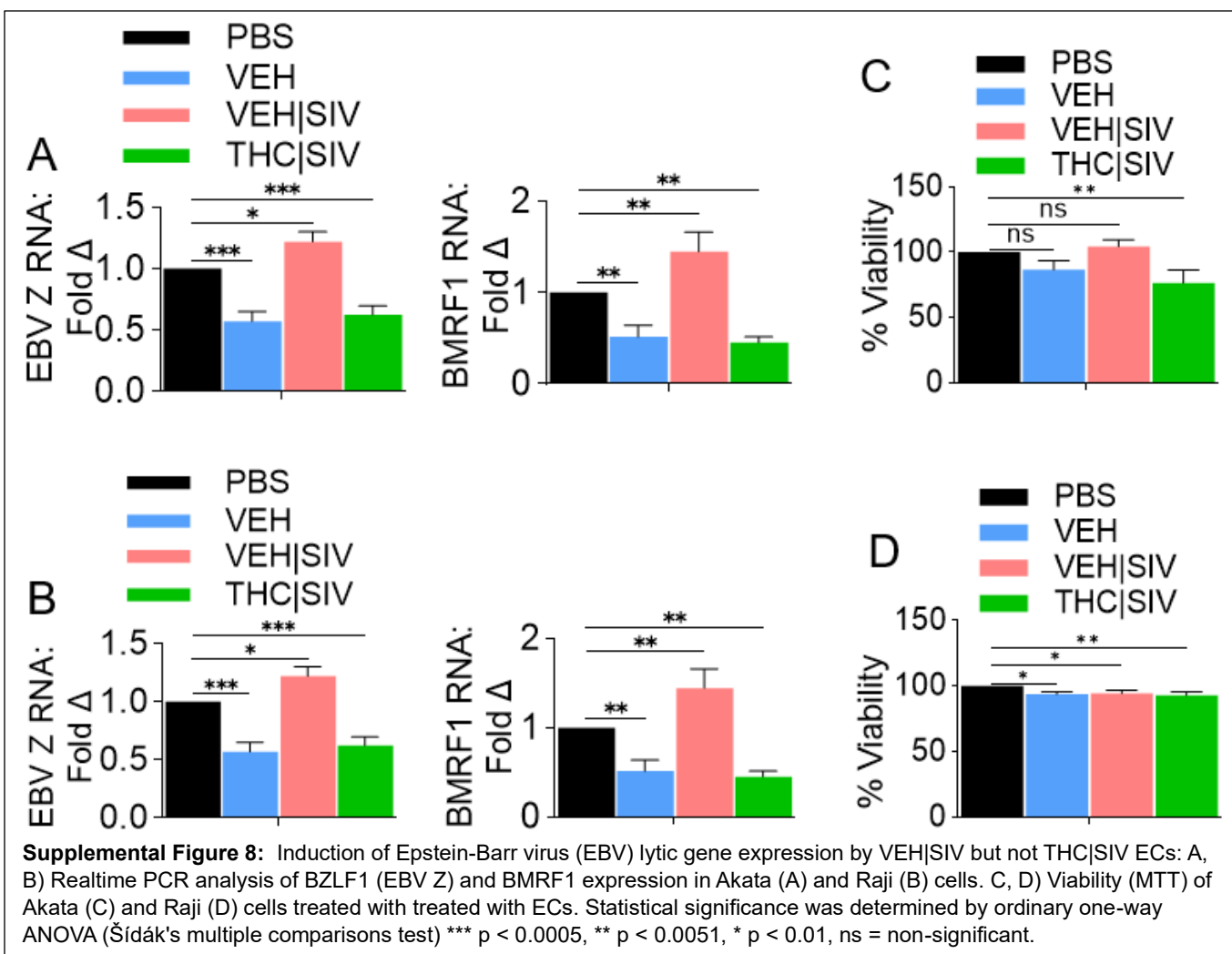
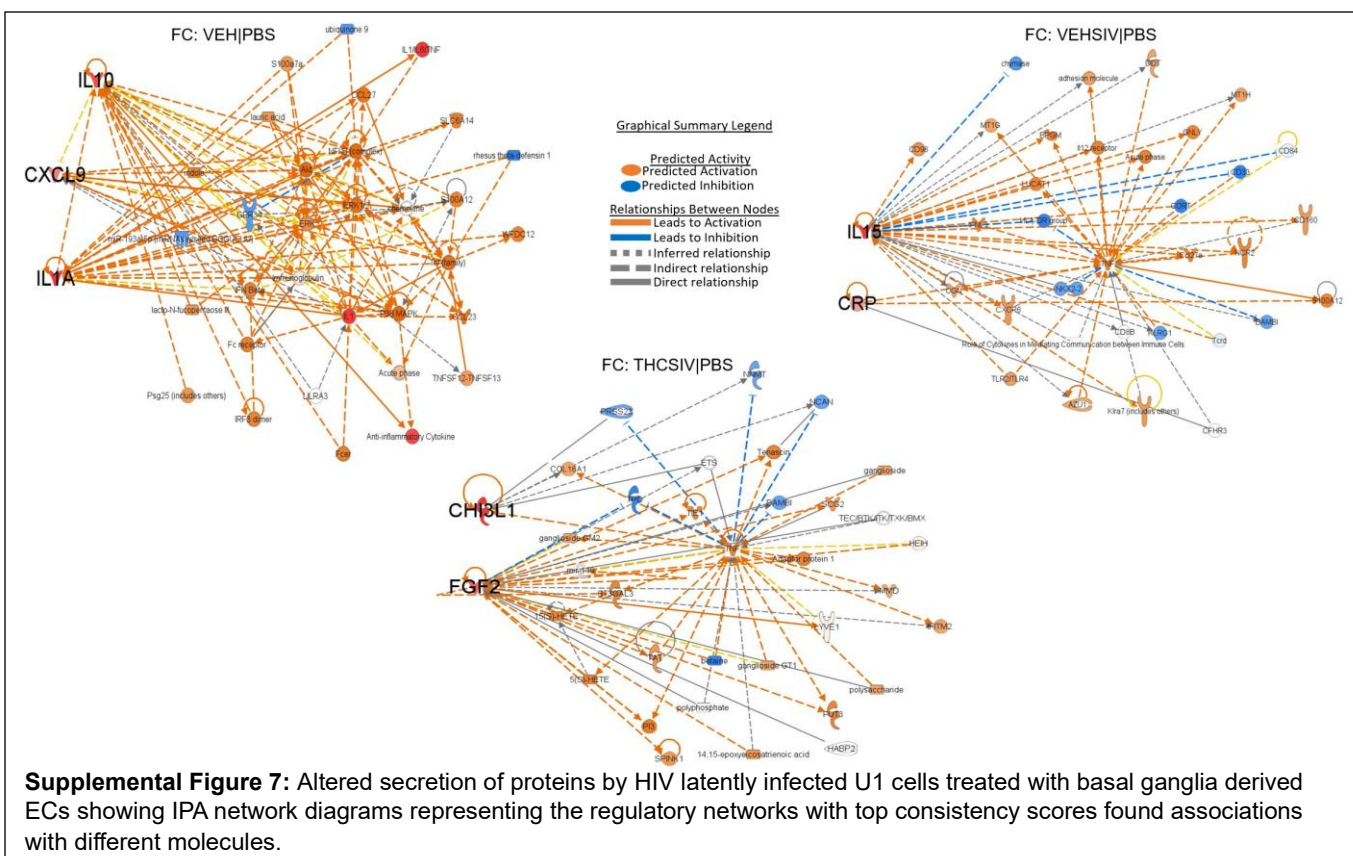


Supplemental Figure 4: Ingenuity Pathway Analysis (IPA) neuroinflammation signaling pathway induced by ECs isolated from THC-treated SIV-infected (THC|SIV) rhesus macaques. Shown are the principal interaction networks in cells treated with THC|SIV ECs.

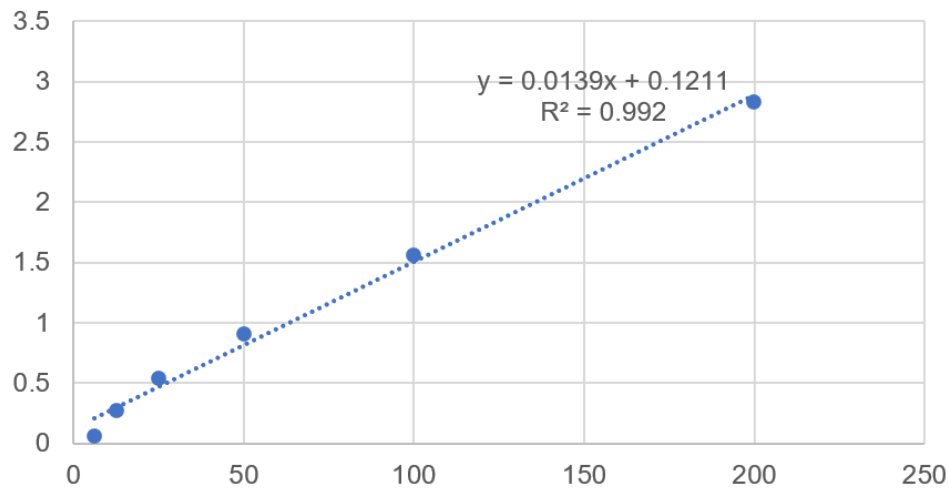


Supplemental Figure 5: Global transcriptome analysis of HIV latently infected myeloid cells treated with basal ganglia derived ECs: Histogram of the 92 canonical pathways regulated by treatment with THC|SIV ECs. The X-axis represents the canonical pathways, and the Y-axis represents $-\log(P\text{-value})$ of the canonical pathways. Black arrows point to the most upregulated (neutrophil degranulation) and downregulated (mitochondrial dysfunction) pathways. Each bar indicates the value of $-\log_{10}(P\text{-value})$.





HIV gag p24 ELISA Standard



Raw values for HIV gag p24

	Donor 1 (D1)			Donor 2 (D2)			Donor 3 (D3)			Donor 4 (D4)		
PBS	35.4	54.9	57.2	45.2	40.9	45.2	29.1	37.0	19.1	23.3	20.2	21.2
PMA	91.2	86.4	87.5	48.1	48.9	48.8	37.5	37.5	30.4	35.5	35.3	26.8
VEH	48.9	47.6	45.4	33.5	28.9	40.7	20.3	42.6	25.2	12.6	18.8	10.1
VEH SIV	87.6	85.8	84.6	56.3	55.7	56.1	34.7	36.9	34.2	41.0	40.5	31.6
VEH SIV	21.2	22.1	22.4	20.4	20.7	19.0	10.1	9.9	7.3	10.9	12.1	6.3

Supplemental Figure 9: Production of extracellular HIV gag p24 by ELISA: HIV replication was measured by quantifying the accumulation of HIV gag p24 in PBMC culture supernatants by ELISA (HIV-1 p24 ELISA, Xpress Bio Cat#XB-1000)). Top) Standard and Bottom) Raw p24 values extrapolated from the standard curve.

Supplemental Table 1: List of genes with Raw TPMs P values ≥ 1 .

Supplemental Table 2: List of Differentially expressed genes (DEGs) identified using p-value (unpaired t-test) < 0.05 , p-adj (FDR < 0.05).

Supplemental Table 3: List of genes identified with Venn overlap analysis.

