HIV-1-Membraneless Organelles: Their Role in Viral Genome Maturation and Immune Evasion

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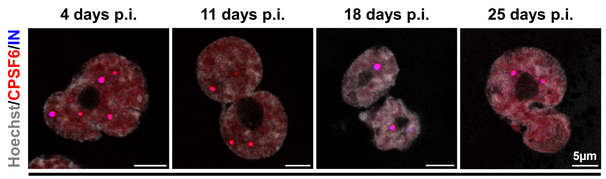
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BACKGROUND AND AIM

HIV-1 establishes unique biomolecular condensates known as HIV-1 membraneless organelles (HIV-1-MLOs) within the nucleus of infected cells. It has been observed that these organelles localize in some nuclear speckles during the early steps of HIV-1 infection. Despite their identification *in vitro*, questions remain regarding their persistence, composition, function, and formation *in vivo*.

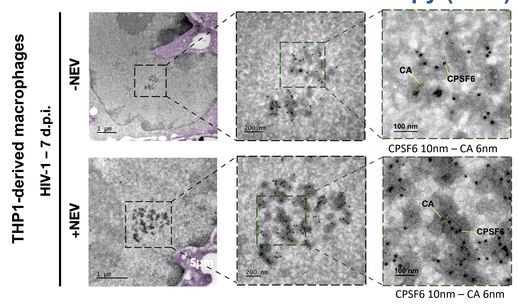
RESULTS

HIV-1 MLOs persist over time during reverse transcription inhibitor treatment

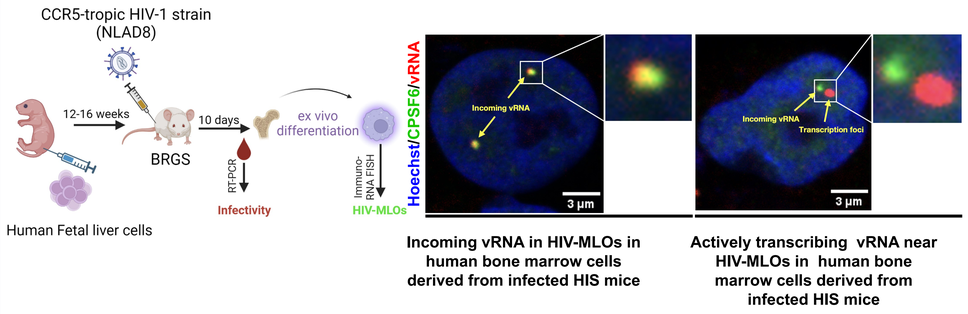


THP1-derived macrophages – HIV-1 infected – Nevirapine [10µM]

First detection of HIV-1 MLOs by Transmission Electron Microscopy (TEM)



First detection of HIV-1 MLOs during acute infection *in vivo*

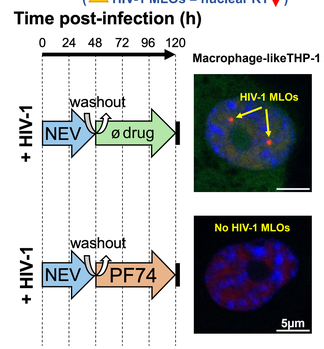


Incoming vRNA in HIV-MLOs in human bone marrow cells derived from infected HIS mice

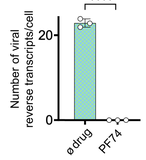
Actively transcribing vRNA near HIV-MLOs in human bone marrow cells derived from infected HIS mice

HIV-1 MLOs are the main site of nuclear RT

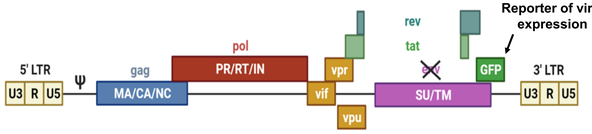
(HIV-1 MLOs = nuclear RT)

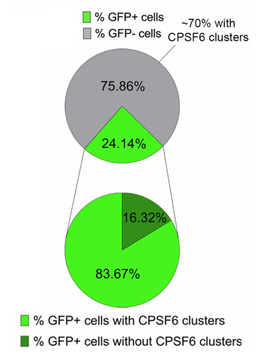


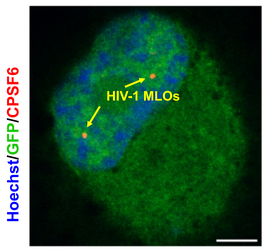
PF74 binds to HIV-1 core at the same site as CPSF6 does, leading to the disassembly of HIV-1 MLOs



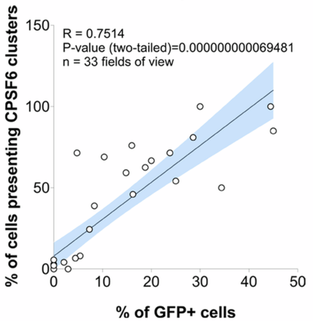
HIV-1 MLOs abundance correlates with viral infectivity





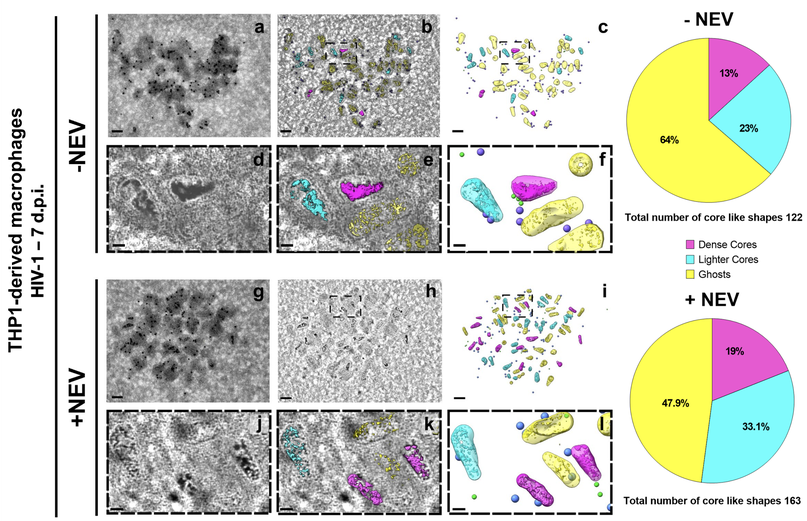


THP1-derived macrophage HIV-1 infected (4 days p.i.)



Despite the persistence of CPSF6 clusters within these cells, only 24% of them, mainly carrying CPSF6 clusters, progresses to a productive infection. A positive correlation between the percentage of cells presenting CPSF6 clusters and GFP-positive cells (more than 80% of GFP+ cells show CPSF6 clusters) has been shown.

HIV-1 MLOs contain different viral core morphologies – Tomograms

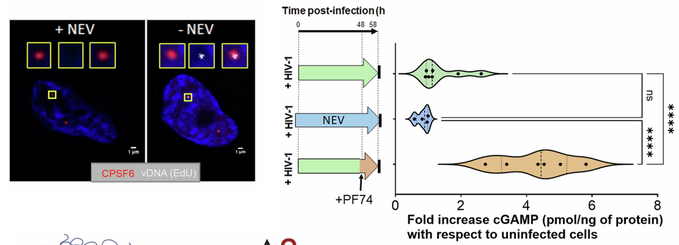


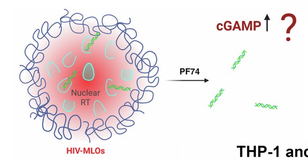
Hypothesis: Dense = Before RT; Lighter = During RT; Ghost = After RT & PIC exit

Three categories of viral cores have been identified inside HIV-1 MLOs based on their intensity signals relative to the surrounding background. Ghosts can also be detected, although in smaller numbers, under the nevirapine condition. This suggests that some may form during the viral journey or release their contents into the MLOs.

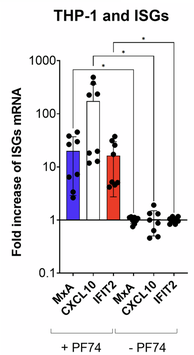
HIV-1 MLOs shield viral genomes from innate immunity

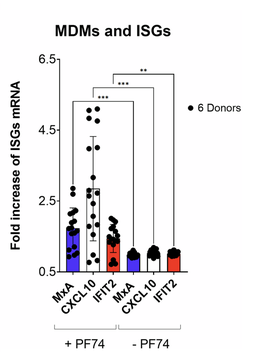
(HIV-1 MLOs = cGAS activation = ISG)



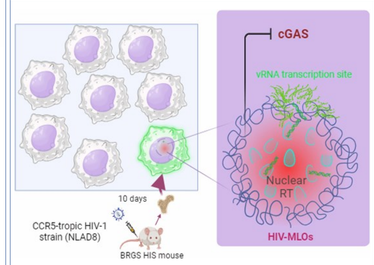


The disassembly of HIV-1 MLOs using a high dose of PF74 at 48 hours post-infection (p.i.), after the virus has already entered the nucleus, leads to the exposure of newly synthesized dsDNA. This activates cGAS and triggers a cascade of cytokines.





CONCLUSIONS



Acknowledgments

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