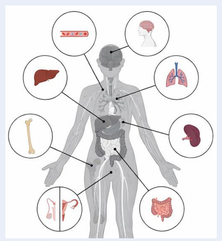
Placenta as a Model to Study Immune Dysfunction and HIV-1 Reservoirs in Macrophages Under ART

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BACKGROUND



Tissue-resident macrophages (TRMs) are widespread and may serve as HIV reservoirs during ART, yet remain poorly accessible.

Term placenta: readily available and unique source of TRMs called **placental macrophages** (**PMs**); embryonic-derived **Hofbauer cells (HBCS**) and monocyte-derived **decidual macrophages (dMs**). PMs circumvent the limitation to TRMs access for HIV-1 mechanistic studies. PMs are key **regulators of placental homeostasis.** They are targeted by various pathogens, contributing to **increased adverse pregnancy outcomes.** The involvement of PMs in HIV-associated adverse outcomes is not fully defined.

PMs with a self-renewal phenotype support integrative HIV-1 infection *in vitro*, indicating that term placenta offers a unique model to mechanistically study mechanisms of HIV reservoir latency/reactivation in long-lived TRMs in view of their pharmacological targeting.

RESULTS

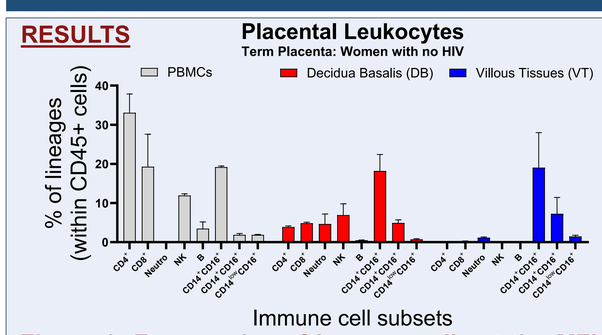


Figure 1: Frequencies of immune cells at the MFI of term placenta from pregnant PWOH. Maternal and fetal placental leukocytes were analyzed by flow cytometry to identify major immune cell subsets

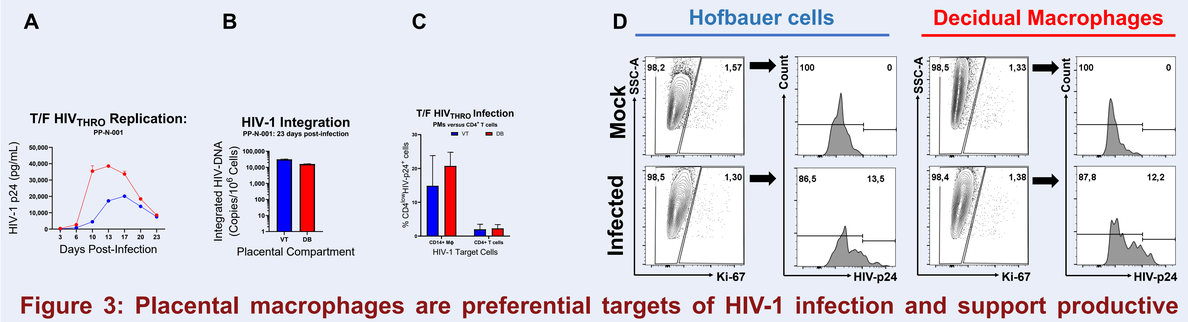
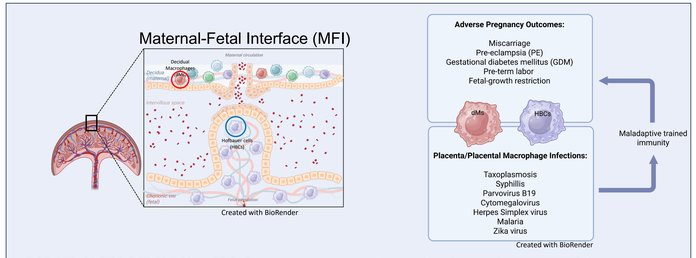


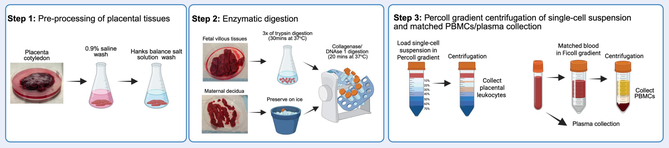
Figure 3: Placental macrophages are preferential targets of HIV-1 infection and support productive viral replication *in vitro*. **(A)** HIV-1 replication kinetics of *in* vitro T/F HIV-1 THRO infected fetal (blue) and maternal (red) placental leukocytes from pregnant PWOH. **(B)** Quantification of integrated HIV-DNA, **(C)** flow cytometric assessment of productively infected PMs and CD4⁺ T cells and **(D)** assessment of HIV-infection of proliferating Ki-67⁺ CD14⁺ PMs in Mock and HIV conditions two weeks post-infection.



HYPOTHESIS: PMs are targets of abortive and/or integrative HIV-infection in PWH receiving ART and drive maladaptive trained immunity during pregnancy, thus disrupting placental homeostasis.

METHODS

- Leukocytes isolation from term placenta of people with HIV (PWH) receiving ART and people without HIV (PWOH)



- Flow cytometric characterization of placenta leukocytes
- *In vitro* HIV infection of placental leukocytes from PWOH
- HIV-p24 quantification by ELISA and flow cytometry
- Nested real-time PCR quantification of integrated HIV-DNA

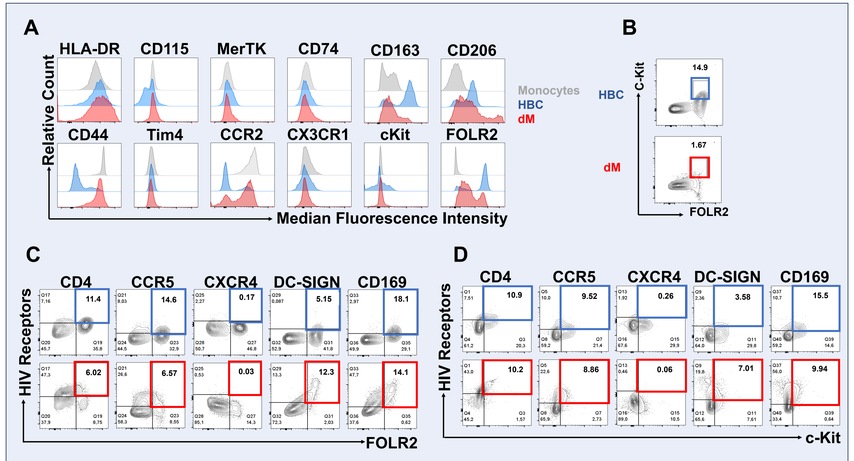


Figure 2: Phenotypic characterization of placental macrophages from the DB and VT compartments. **(A)** Flow cytometric analyses of TRM markers on CD14⁺ HBCs (blue) and dMs (red) relative to monocytes. **(B)** A subset of FOLR2⁺c-Kit⁺ PMs is present in both maternal and fetal compartments and exhibits proliferative capacity. **(C-D)** Surface expression of HIV entry and trans-infection–associated receptors on FOLR2⁺ and c-Kit⁺ PMs.

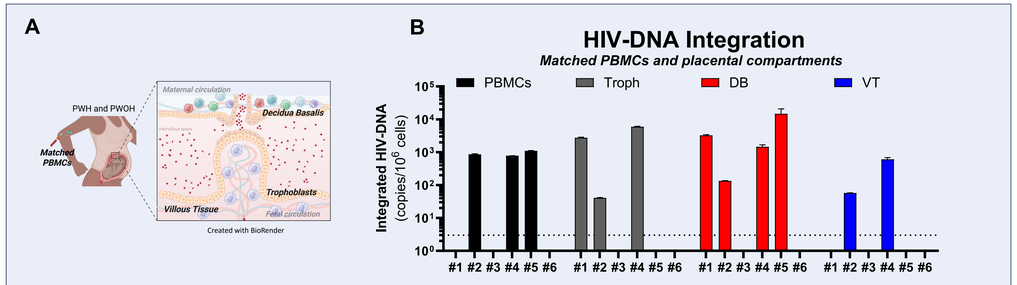


Figure 4. Placental leukocytes harbor integrated HIV-DNA in PWH receiving ART during the last months of pregnancy. **(A)** Matched DB, VT, trophoblasts and PBMCs from ART-treated PWH (n=6), were used for the detection of **(B)** integrated HIV-DNA by nested real-time PCR. The results are depicted as the number of proviral HIV copies normalized to the number of cells.

CONCLUSIONS

- PMs with self-renewal capacity are highly susceptible to productive HIV-1 infection *in vitro* and may serve as a model to study HIV reservoirs in long-lived TRMs.
- Placental tissues from the fetal side harbor integrated HIV-DNA in 2/6 PWH receiving ART. HIV replication competency in these compartments remain to be elucidated.

PLAIN LANGUAGE SUMMARY

The placenta provides a unique opportunity to study tissue-resident macrophages from people living with HIV. Studying placental macrophages can improve our understanding of their role in HIV-1 persistence and disease.

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