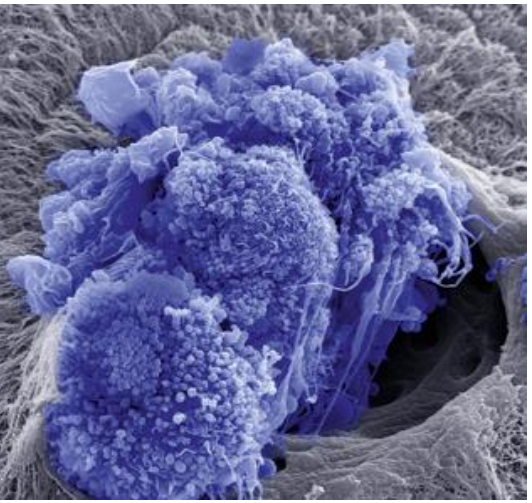
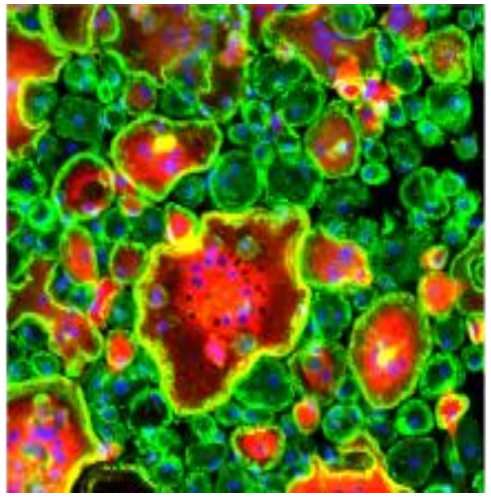
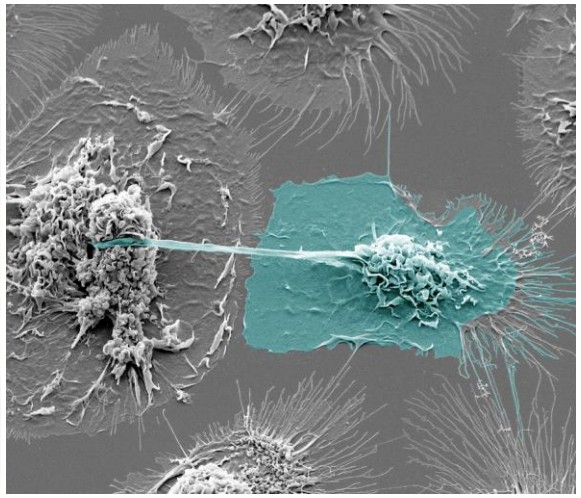
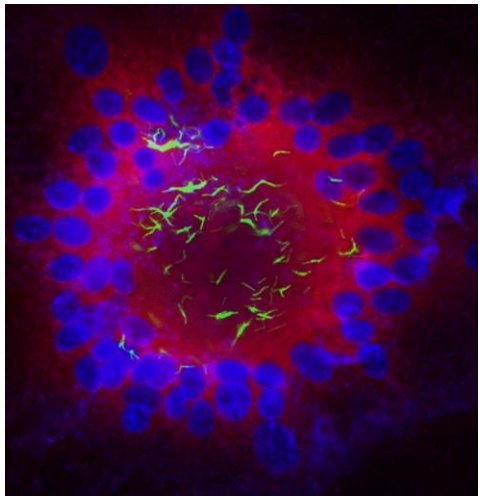
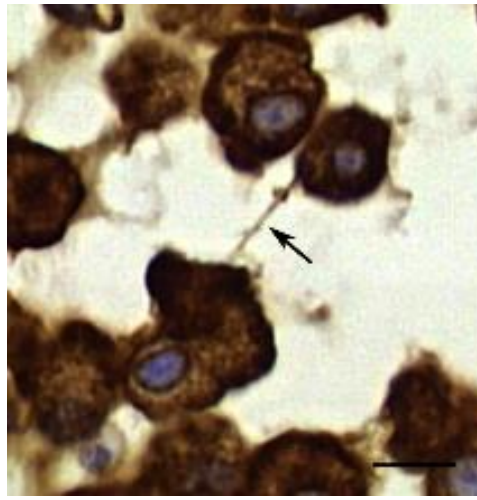
Cell-to-cell transfer of HIV-1 towards macrophages: perspectives in HIV/TB co-infection

Christel Vérollet Inserm

Team leader, with R. Poincloux, *Team « Phagocyte architecture & dynamics »*

IPBS, Toulouse, FRANCE

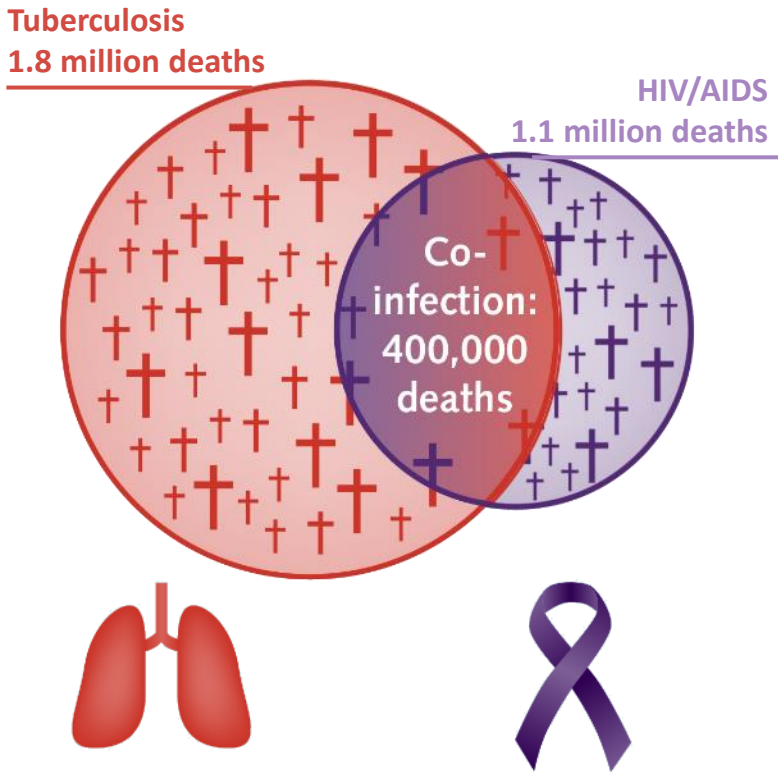
HOT TOPICS IN HIV

Vaccines, immune recovery and eradication

October, 26th, BCN

HIV-1 /*Mtb* co-infection

✱ Clinical synergy between HIV and *Mtb*

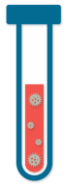


World Health Organization, 2021

✱ TB boosts HIV infection

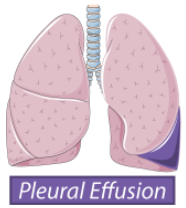
Increase in viral load during TB

✓ Systemic level (Blood)



Goletti D et al. *J Immunol*. 1996

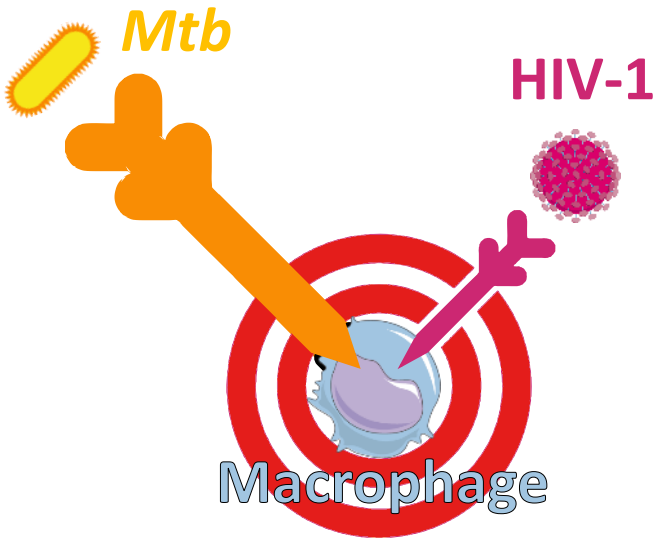
✓ In anatomical sites of co-infection



- ✓ Lungs (BAL and tissue)
- ✓ Pleural Effusion (PE)

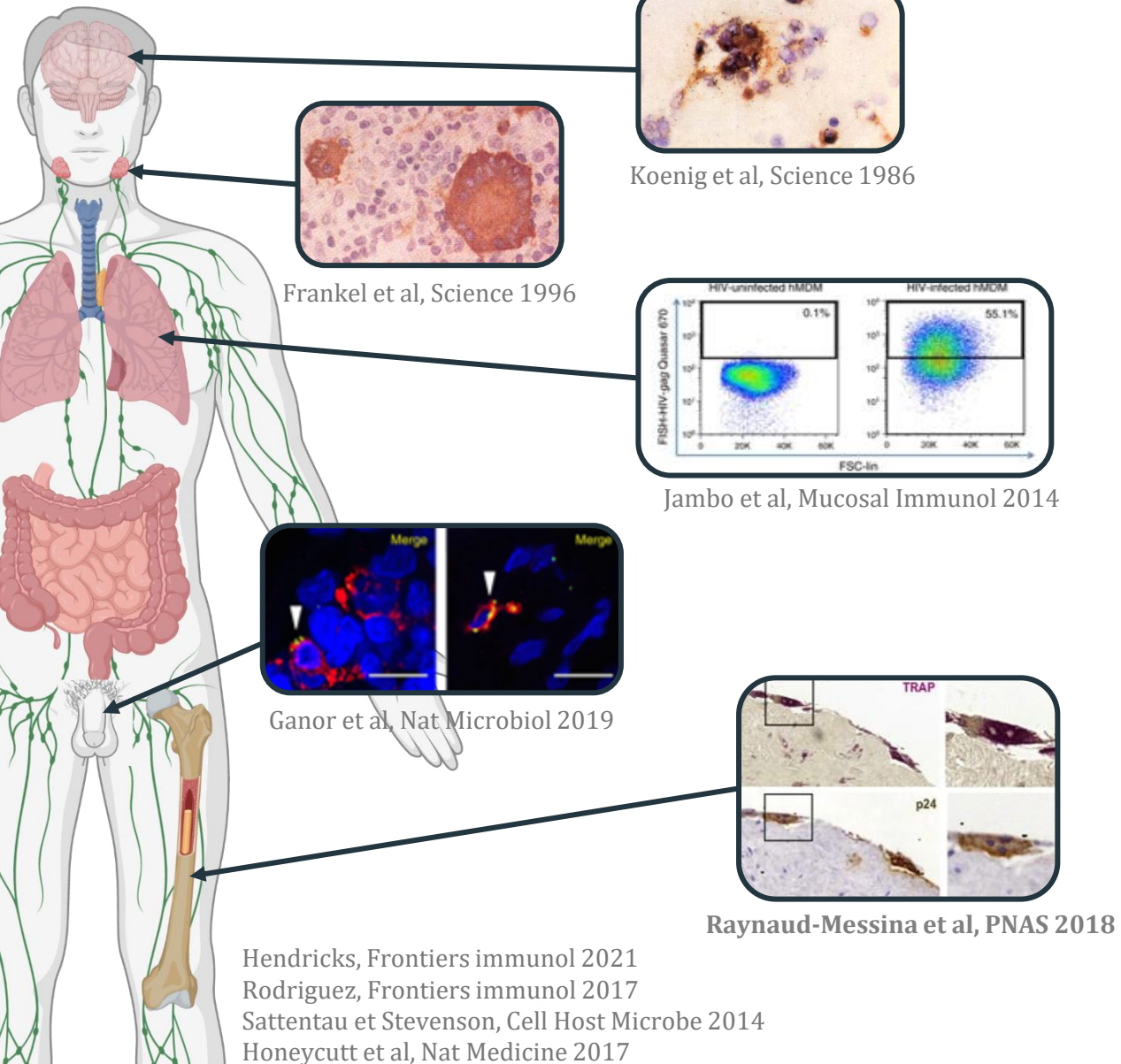
Morris et al. *J Infect Dis.* 1998
Nakata et al, *Am J Respir Crit Care Med.* 1997
Toossi et al . *J Acquir Immune Defic Syndr.* 2001
Collins et al. *J Virol.* 2002
Toossi et al. *Clin Exp Immunol.* 2011

✱ A role for macrophages ?



Lugo-Villarino*, Verollet C.*, et al. *Front Immunol.* 2011

Macrophages in HIV-1 pathogenesis



Koenig et al, Science 1986

Frankel et al, Science 1996

Jambo et al, Mucosal Immunol 2014

Ganor et al, Nat Microbiol 2019

Raynaud-Messina et al, PNAS 2018

Hendricks, Frontiers immunol 2021
Rodriguez, Frontiers immunol 2017
Sattentau et Stevenson, Cell Host Microbe 2014
Honeycutt et al, Nat Medicine 2017

✱ Infected macrophages

- ✓ Long life upon infection, no cytopathic effect
- ✓ Replicate and produce high level of viruses
- ✓ **Found in all tissues of HIV+ patients,**
- ✓ **often as multinucleated giant cells (MGC)**

✱ Virus dissemination

- ✓ HIV-1 increases macrophage tissue infiltration

Vérollet et al, Blood 2015; Vérollet, J Immunol, 2010

✱ Persistent viral tissue reservoirs

Mechanism(s) of macrophage infection ?

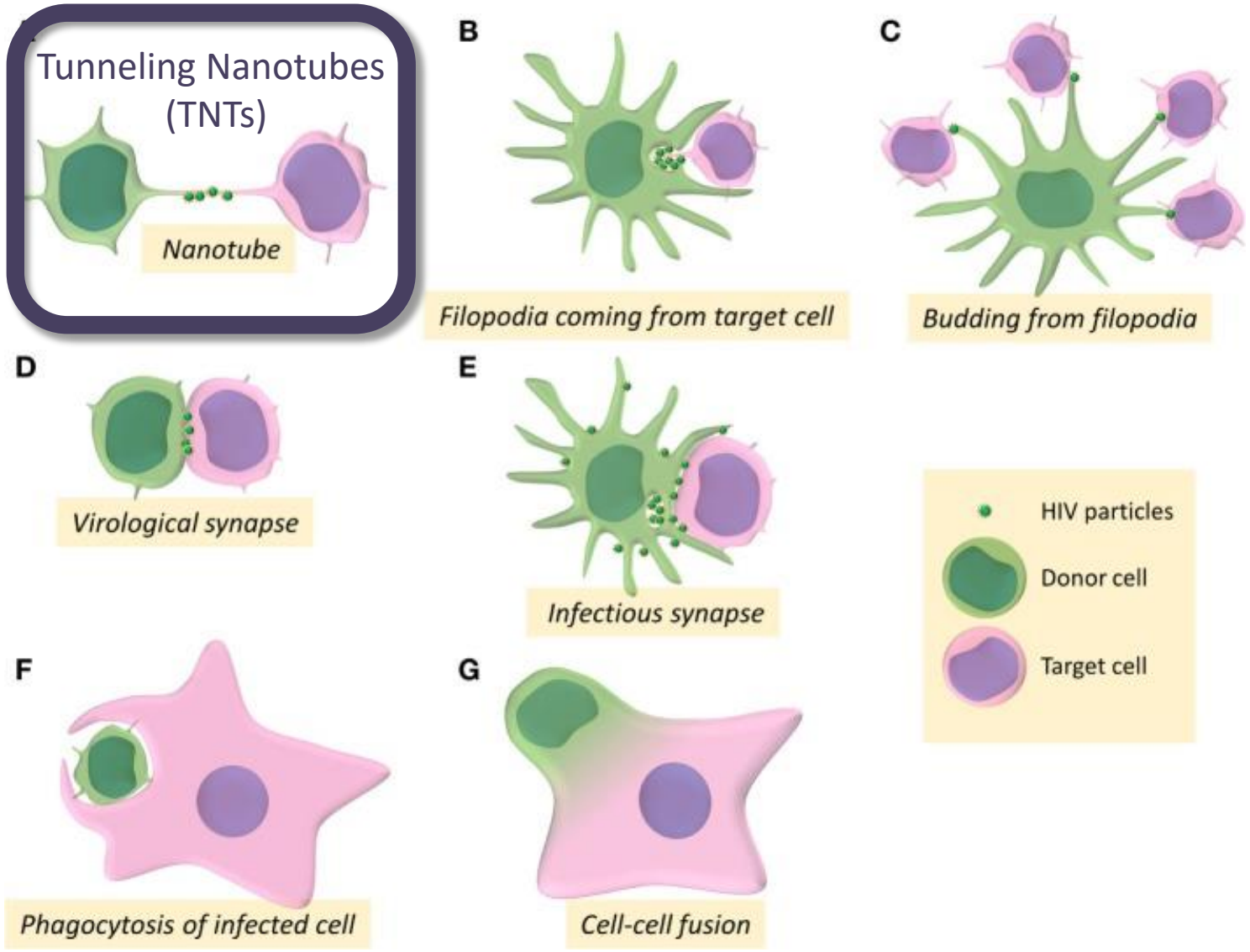
Part I- Macrophage infection by intercellular transfer *via* TNTs

- ✓ Tissue macrophages are poorly susceptible to HIV-1, especially with cell-free viral particles

✱ Cell-to-cell transfer of HIV-1

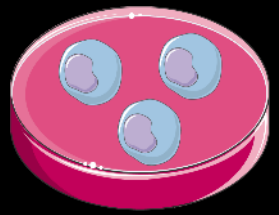
- ✓ Rapid and efficient
- ✓ Escapes innate immune responses
- ✓ **Main mode of infection *in vivo***

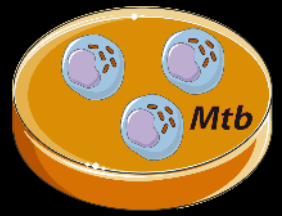
Dufloo *et* al, 2018
Bracq *et* al, 2018
Dupont & Sattentau, 2020
Bracq et al, Front Immunol 2018
Calantone et al., Immunity, 2014
Izquierdo-Useros N et al., 2014
.....



From Han et al., J Leuk biol, 2022

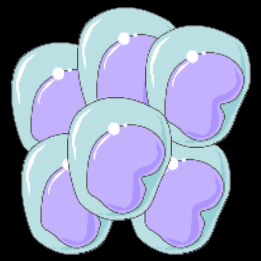
In vitro model to mimic **TB-associated microenvironment**





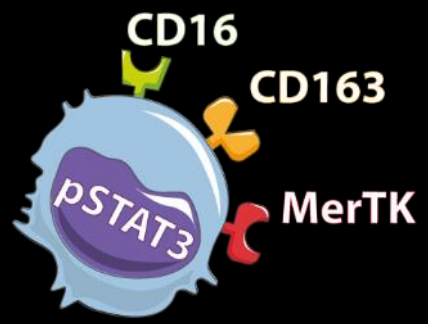
CmCTR
Conditioned medium of ***uninfected*** *macrophages*

CmMTB
Conditioned medium of ***Mtb-infected*** *macrophages*



Primary human monocytes

CmMTB



Glycolytic activity > OXPHOS

TB-induced macrophages
Their abundance ***in vivo*** *correlates with TB disease severity*

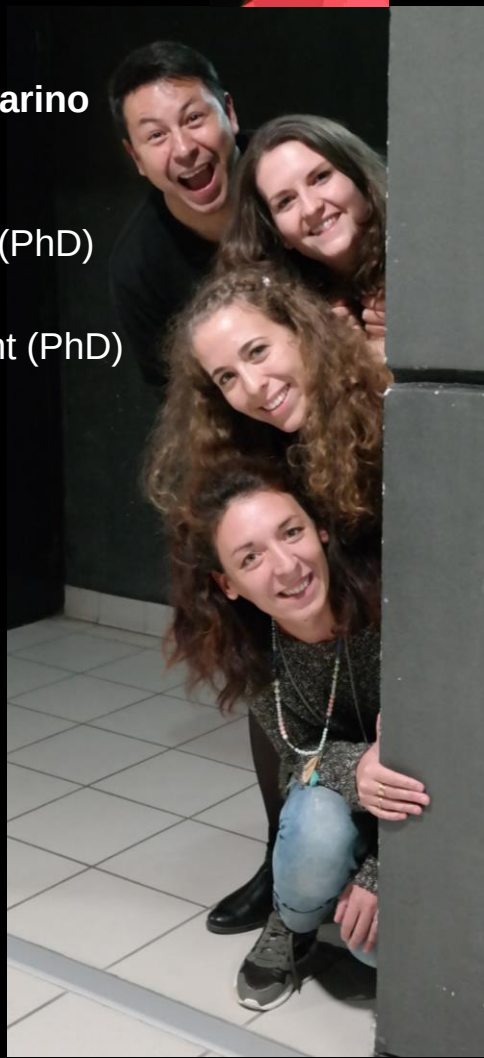
Lastrucci et al., *Cell Research*, 2015

Zoï Vahlas (PhD)

Geanncarlo Lugo-Villarino

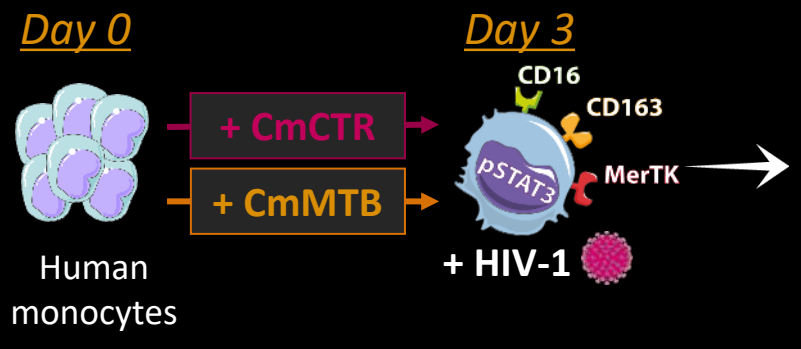
Maeva Dupont (PhD)

Shanti Souriant (PhD)

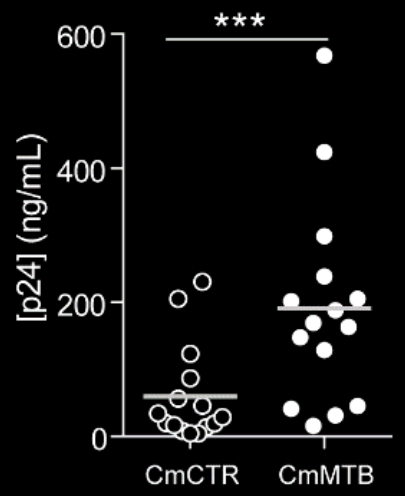


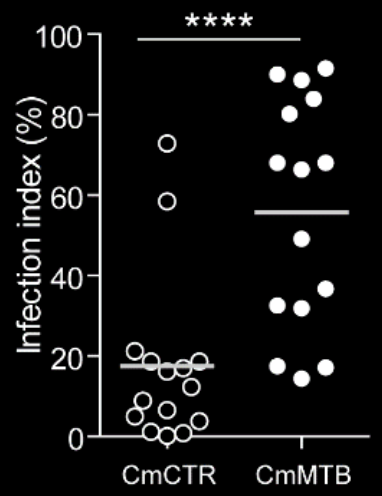
Collaboration : O. Neyrolles (IPBS)

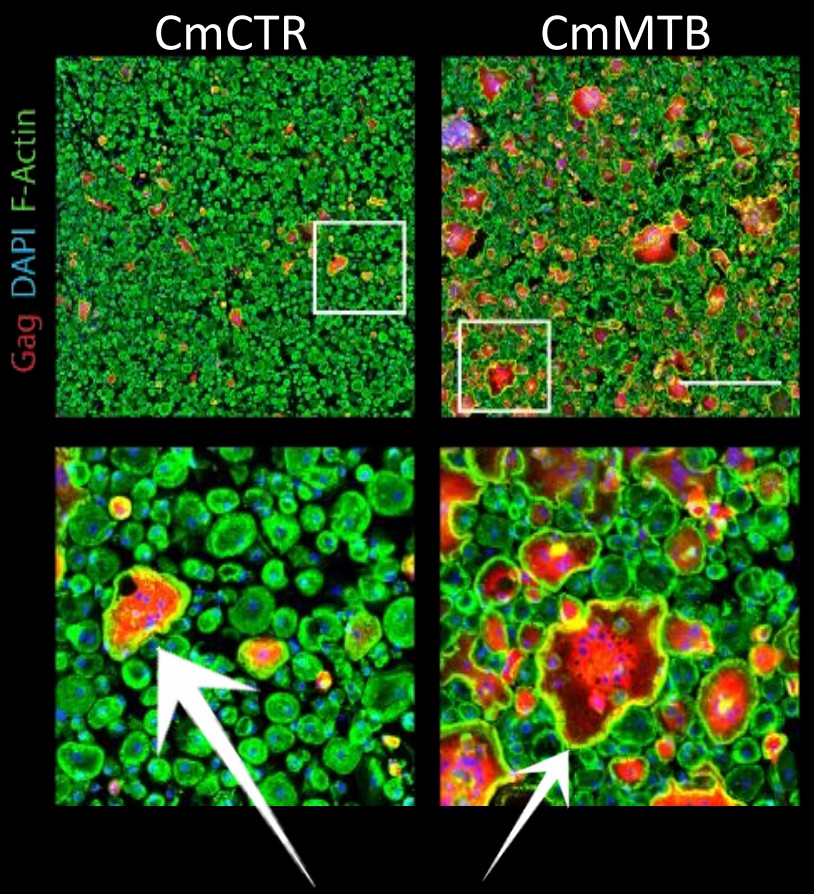
TB-associated microenvironment **increases HIV-1 infection** of macrophages



- ✓ HIV-1 production (p24 ELISA)
- ✓ HIV-1 infection (IF)







Multinucleated giant cells

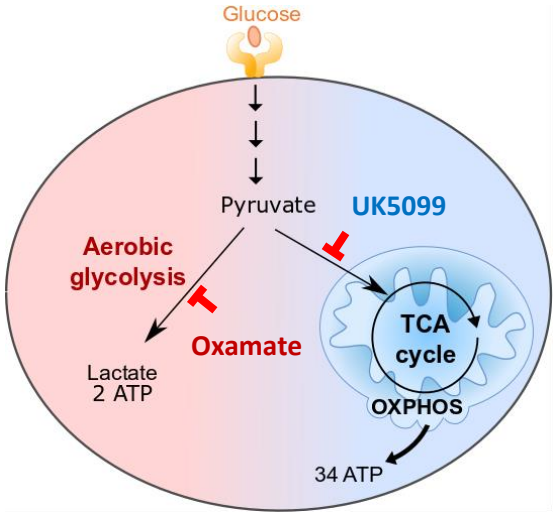
Hallmark of HIV infection of macrophages

Vérollet *et al, J Immunol.* 2010
Vérollet *et al, Blood.* 2015
Oreinstein *et al.,* Immunol., 2001

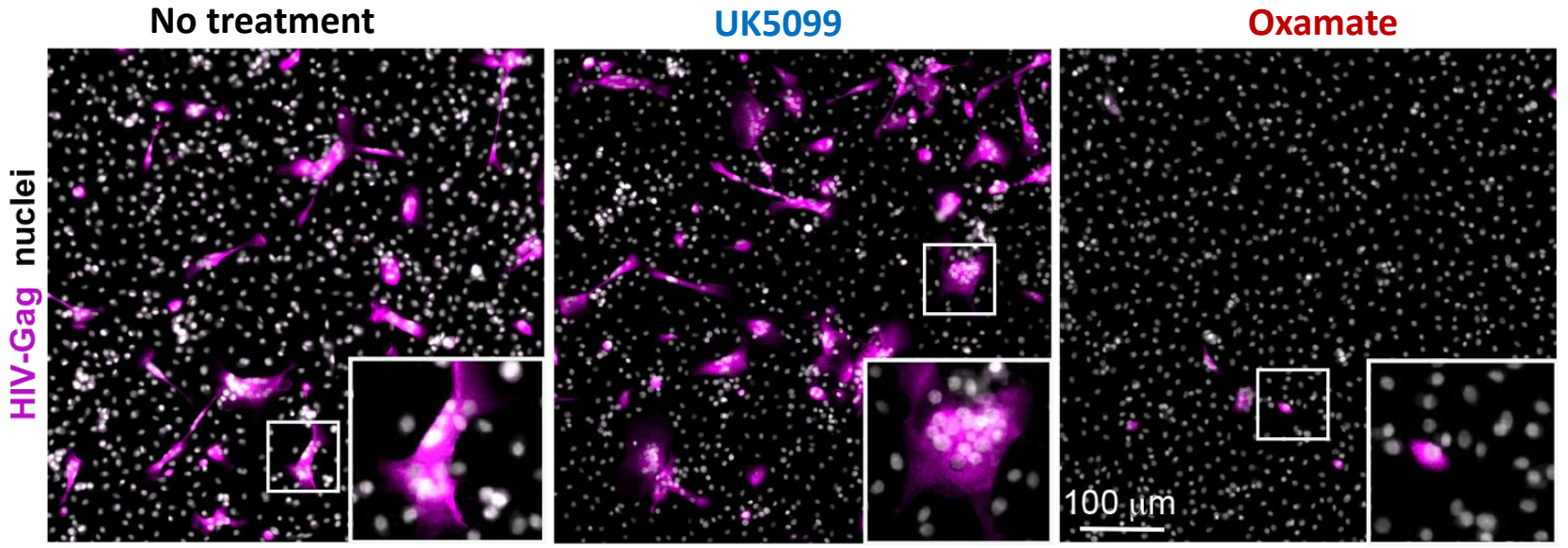
Souriant S *et al. Cell Reports, 2019*
Dupont M *et al., eLife, 2020*

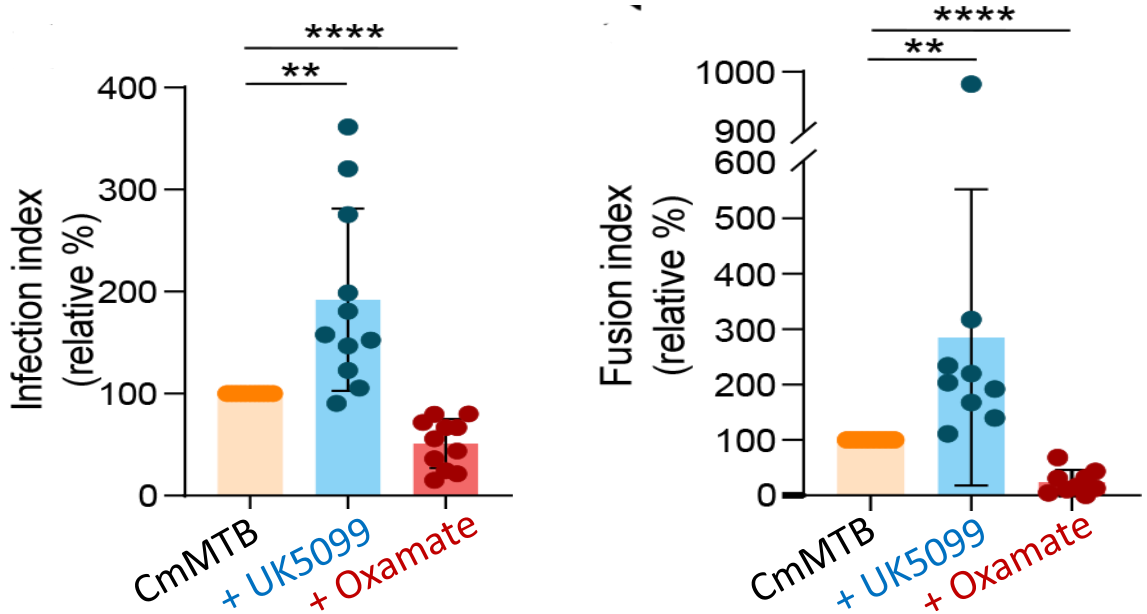
Glycolysis favors HIV-1 infection of TB-induced macrophages

cmMTB-treated cells



or modulation of Glucose in CmMTB

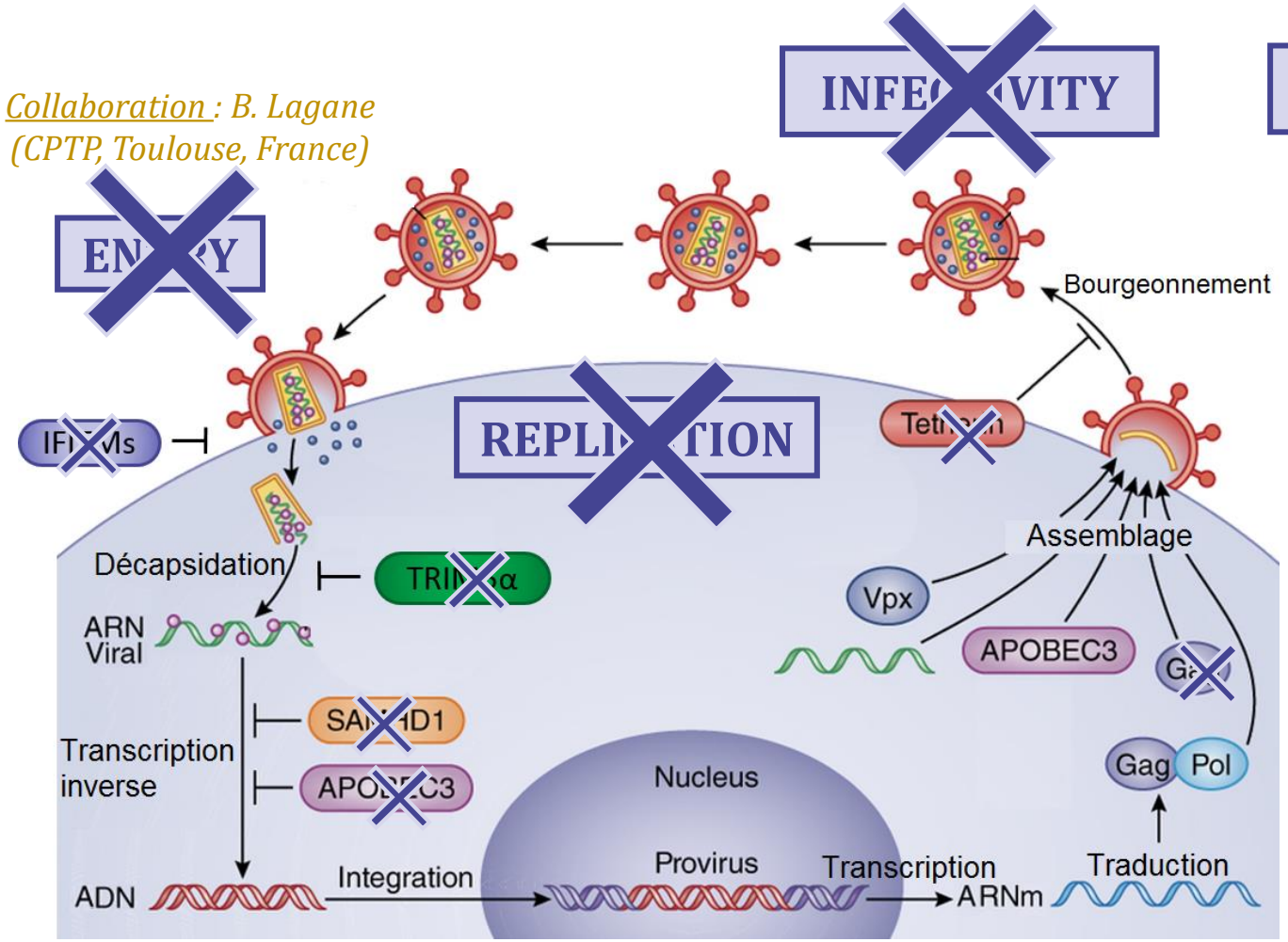




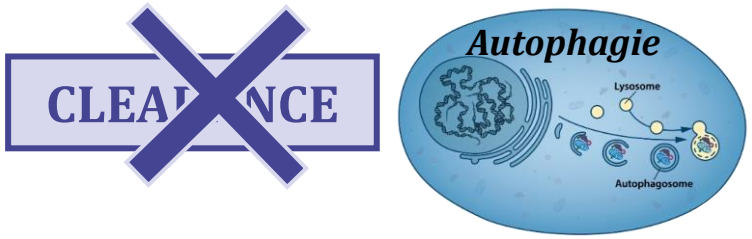
Vahlas *et al.*, *in preparation*

What is modified in TB-induced macrophages?

Collaboration : B. Lagane (CPTP, Toulouse, France)



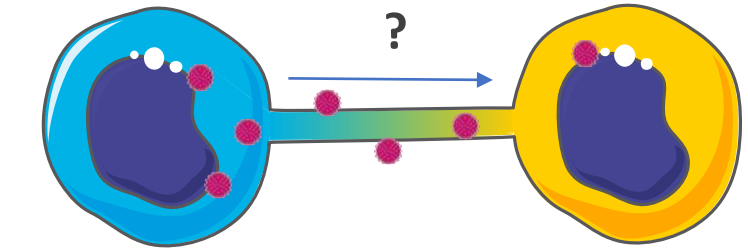
Adapté de Simon *et al*. *Nature Immunology*. 2015



Collaboration : I. Vergne (IPBS, Toulouse, France)

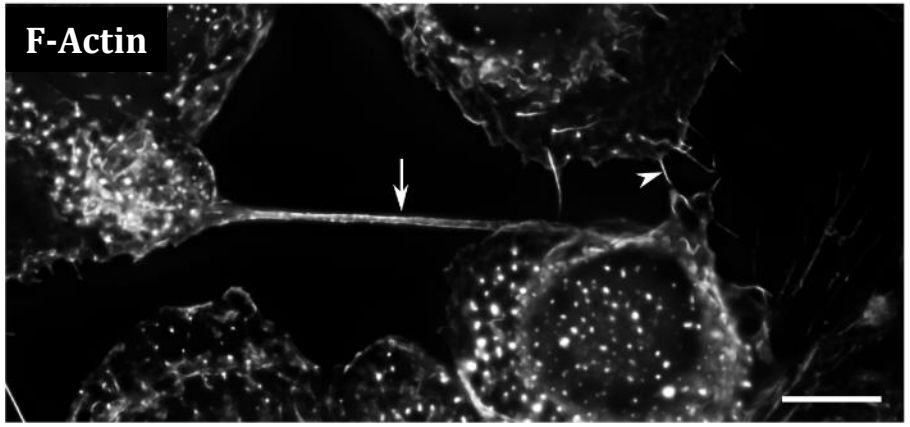
CELL-TO-CELL TRANSFERT

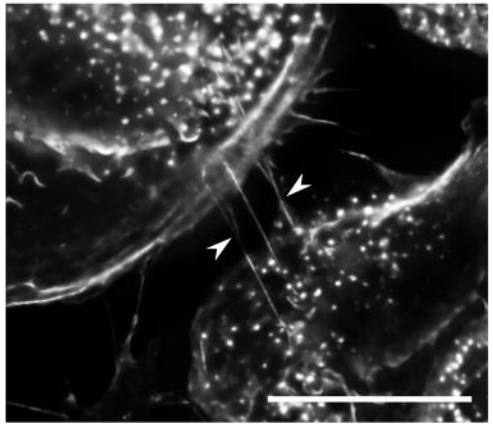
✱ Tunneling nanotubes (TNT)

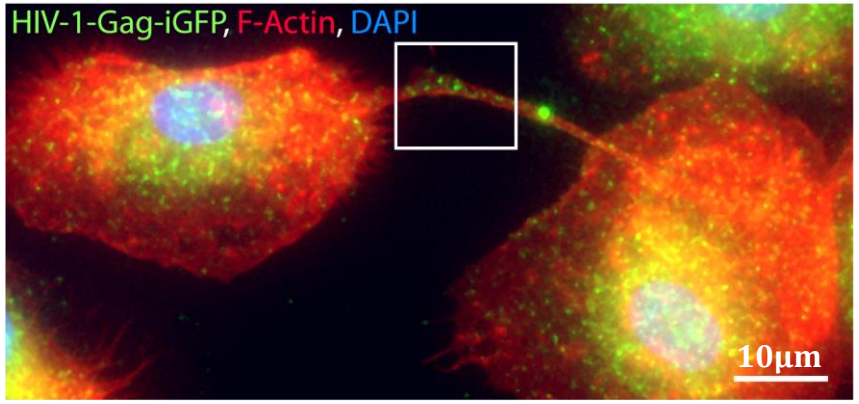


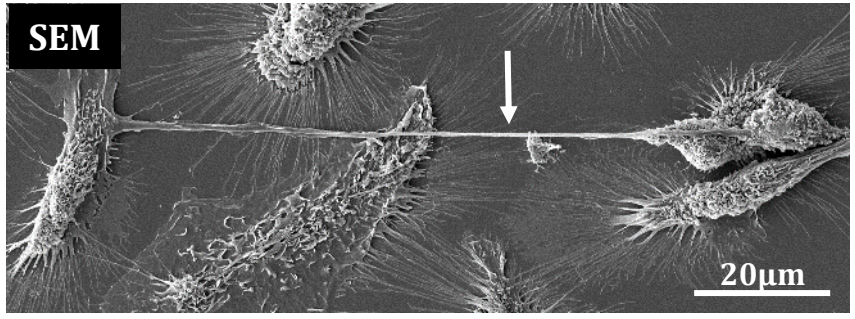
Okafo *et al.*, J Immunol, 2020
Eugenin *et al., Cell. Immunol.*, 2009
Dupont *et al, Frontiers Immunol.* 2018

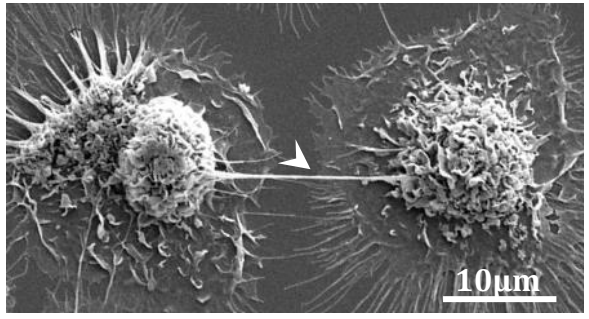
Tunneling nanotubes are enhanced in TB-induced macrophages

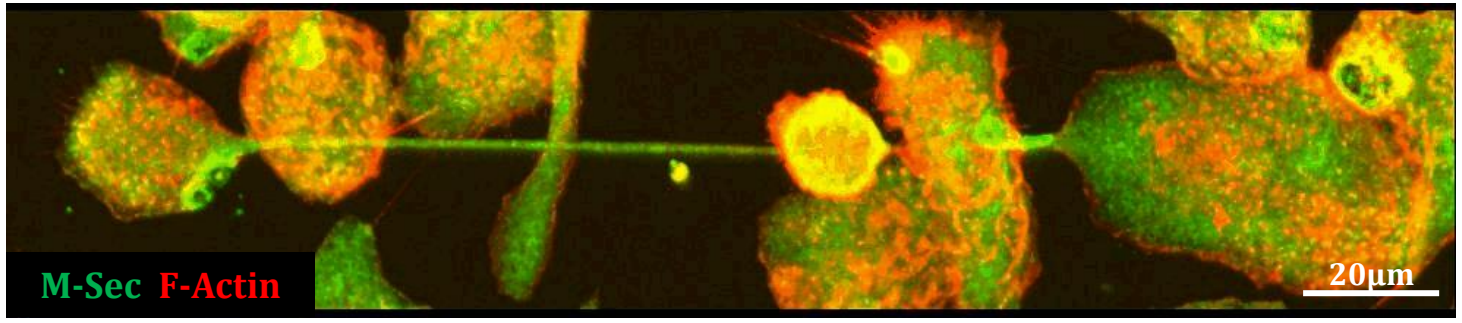


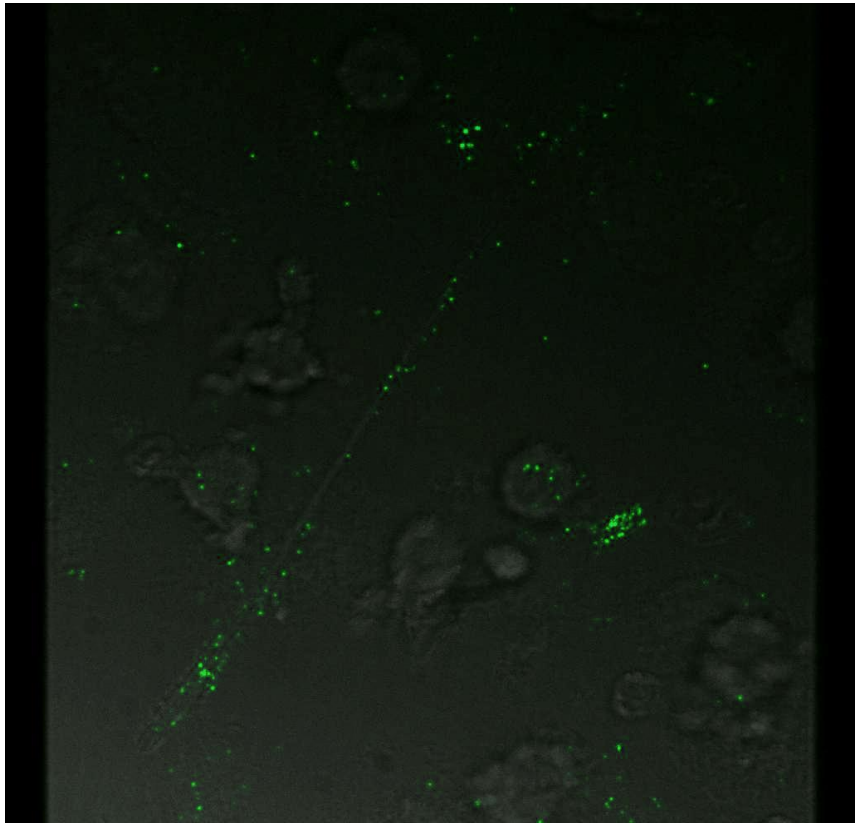










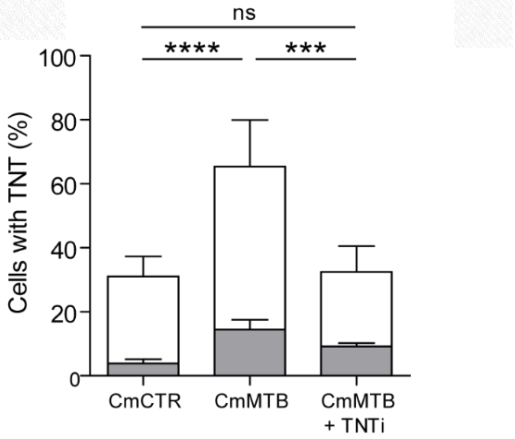


Dupont *et al, Frontiers Immunol.* 2018

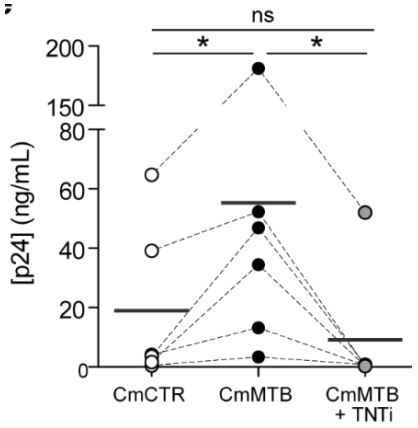
TB boosts HIV-1 infection of macrophages through Glycolysis-dependent TNT formation

✱ TNTs induced by a TB-derived microenvironment boost HIV-1 production …..

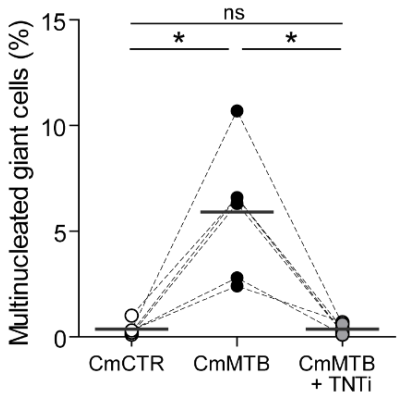
↘ TNT formation



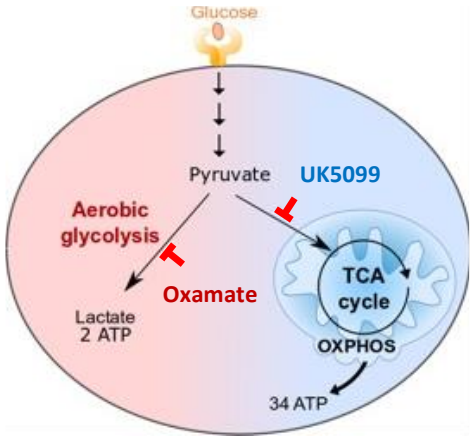
↘ HIV-1 production



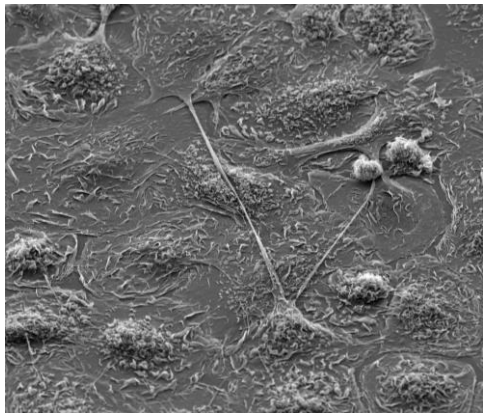
↘ MGC formation



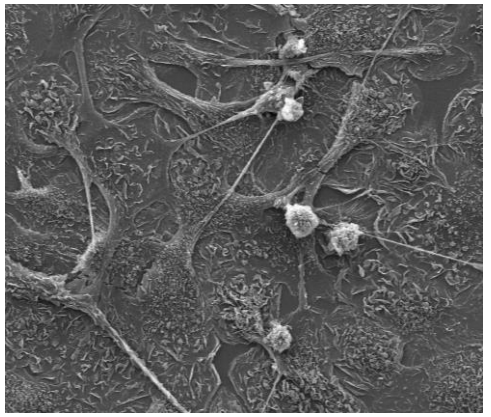
✱ … in a glycolytic dependent manner



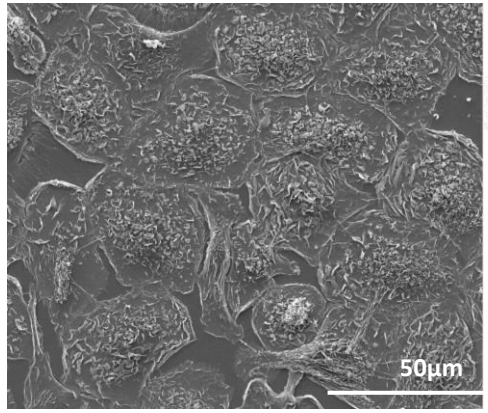
CmMTB treatment



+ UK5099

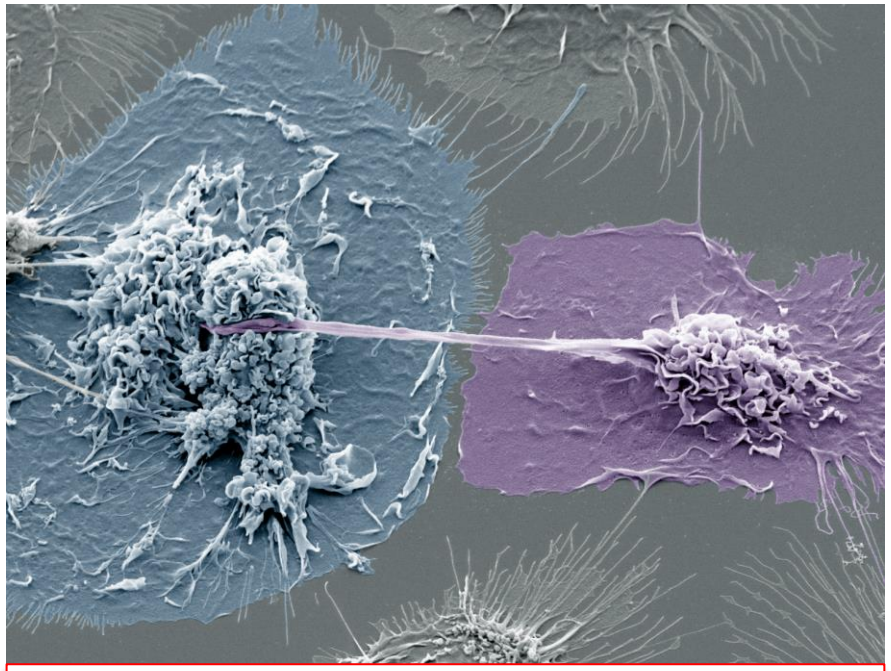


+ Oxamate

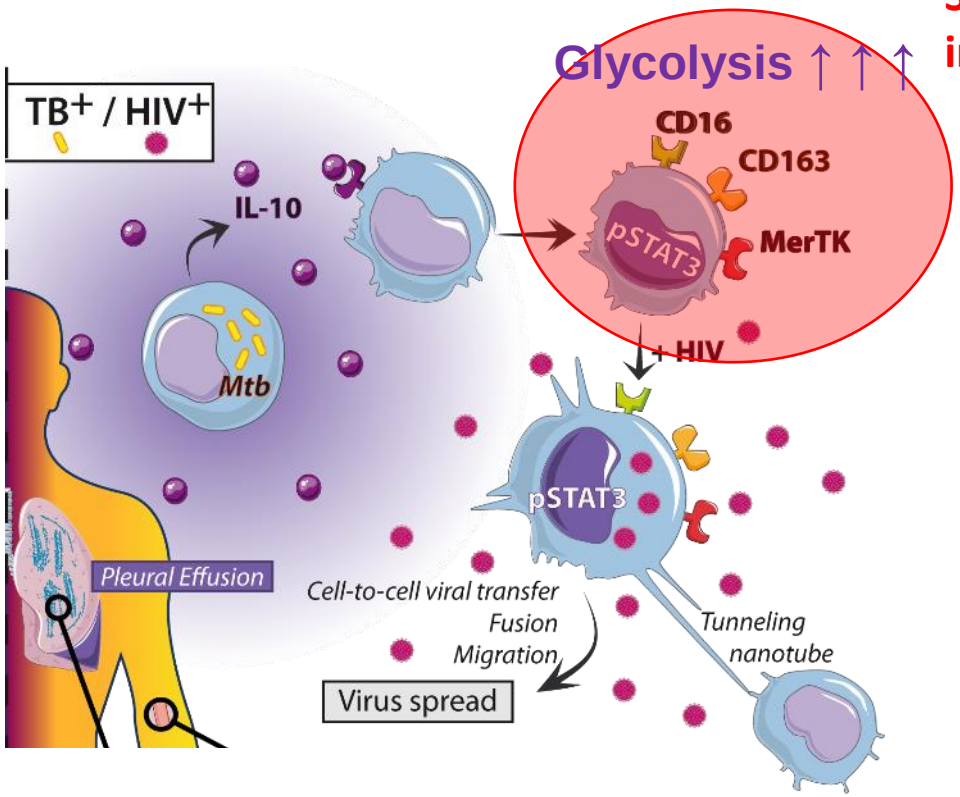


Vahlas *et al.*, *in preparation*

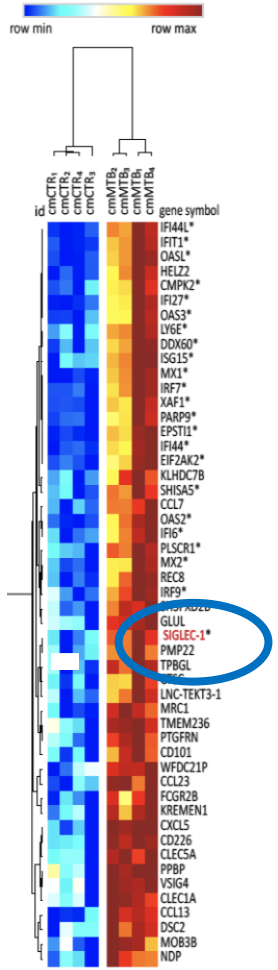
TB exacerbates HIV-1 infection through glycolytic-dependent TNT formation in macrophages



Tunneling nanotubes



Specific gene expression in these cells ?

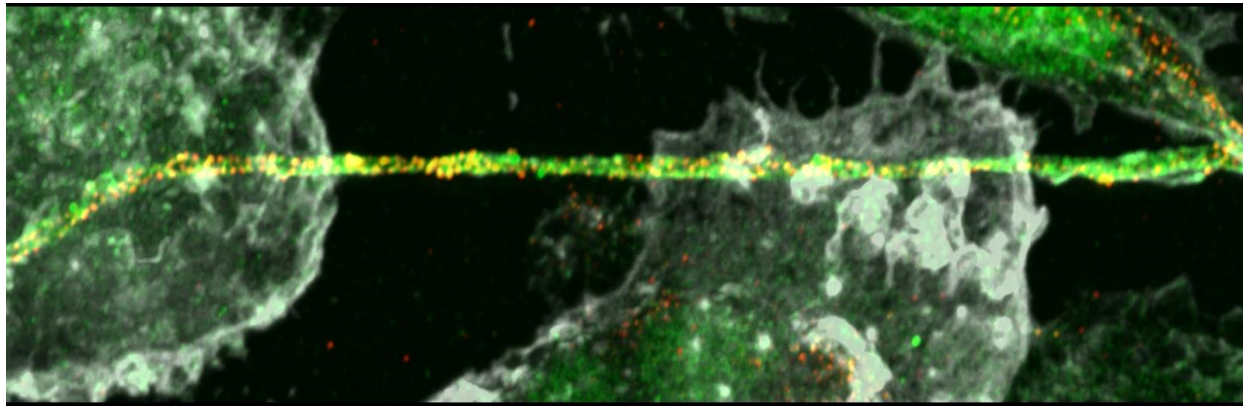


Dupont M *et al., eLife, 2020*
Dupont M *et al., L Leuk Biol, 2022*
Souriant S *et al. Cell Reports, 2019*

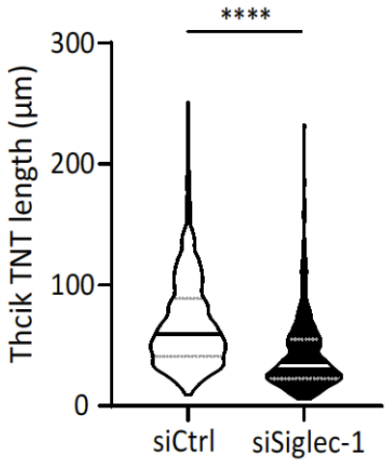
Izquierdo-Useros N *et al., 2014*
Perez-Zsolt D *et al., 2019;*
Raïch-Regué D et al., 2023; …

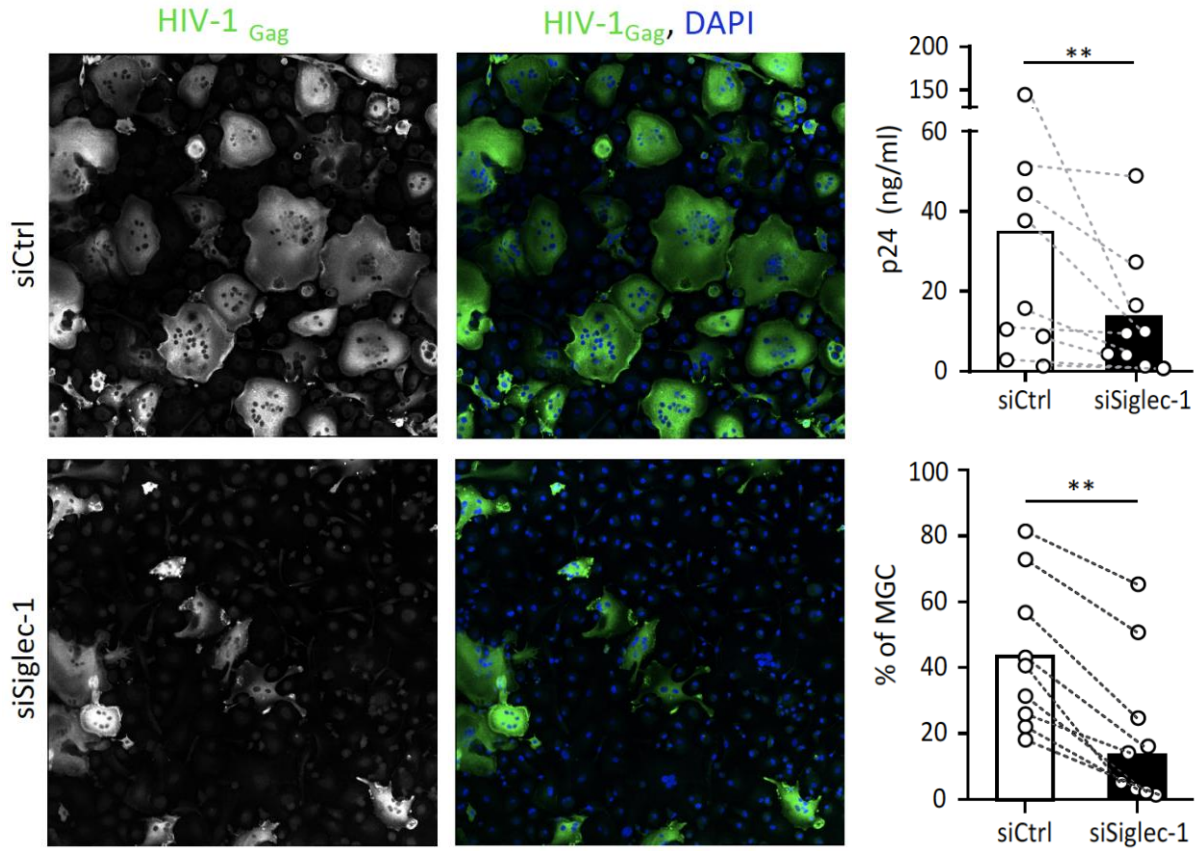
Siglec-1 is crucial for TNT stability and TB-driven HIV-1 exacerbation

★ **Siglec-1 localizes on long and stable TNTs, and participates in HIV-induced fusion of macrophages**



Siglec-1 HIV-1_{Gag} WGA

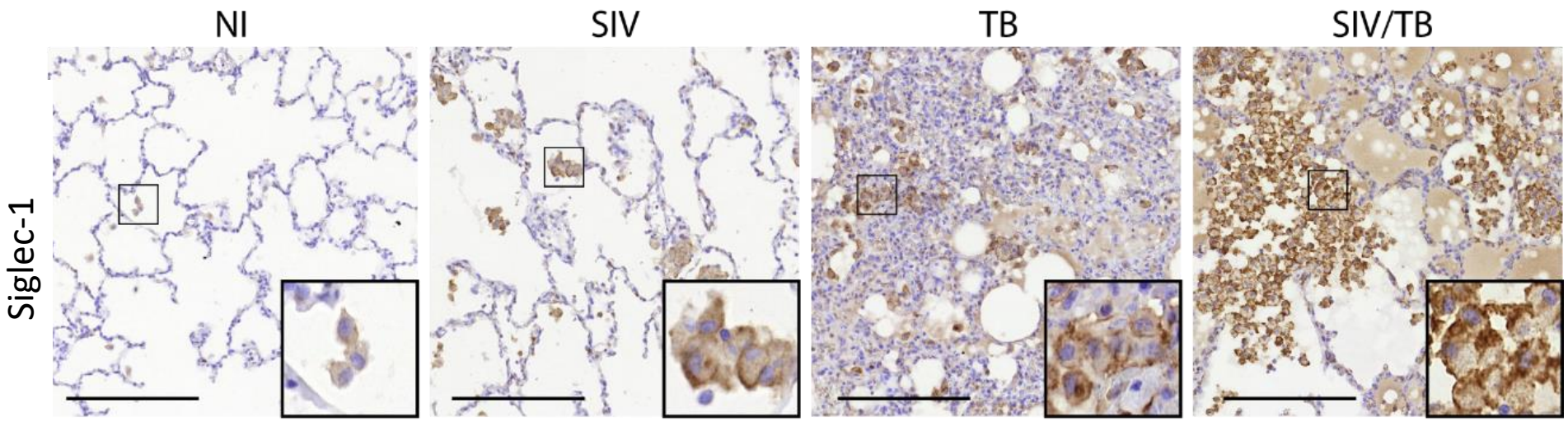


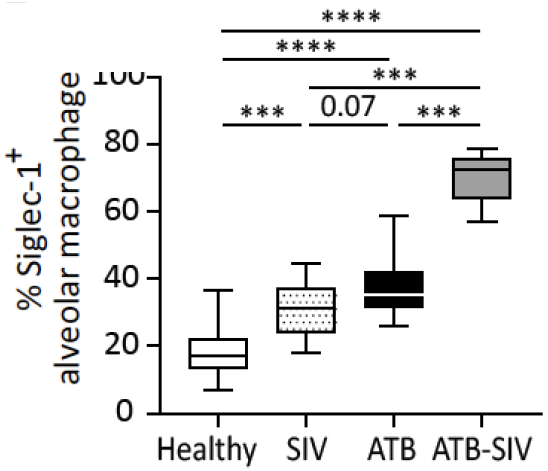


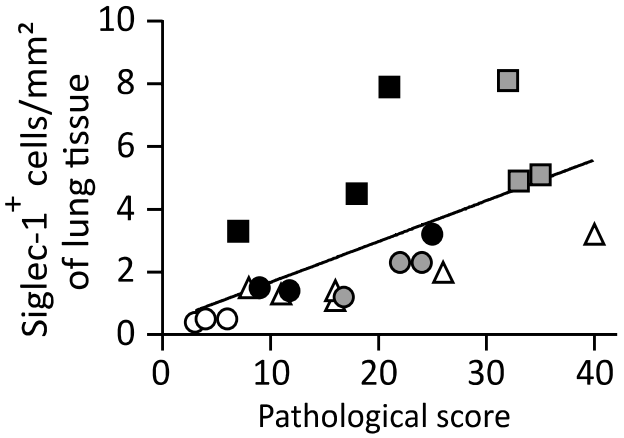
Dupont M *et al., eLife, 2020*

In vivo relevance of Siglec-1$^+$ M(TB) macrophages in co-infection

Rhesus macaques
SIV / TB active

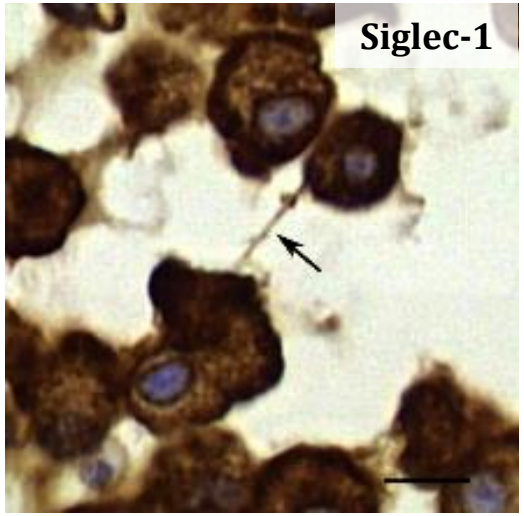




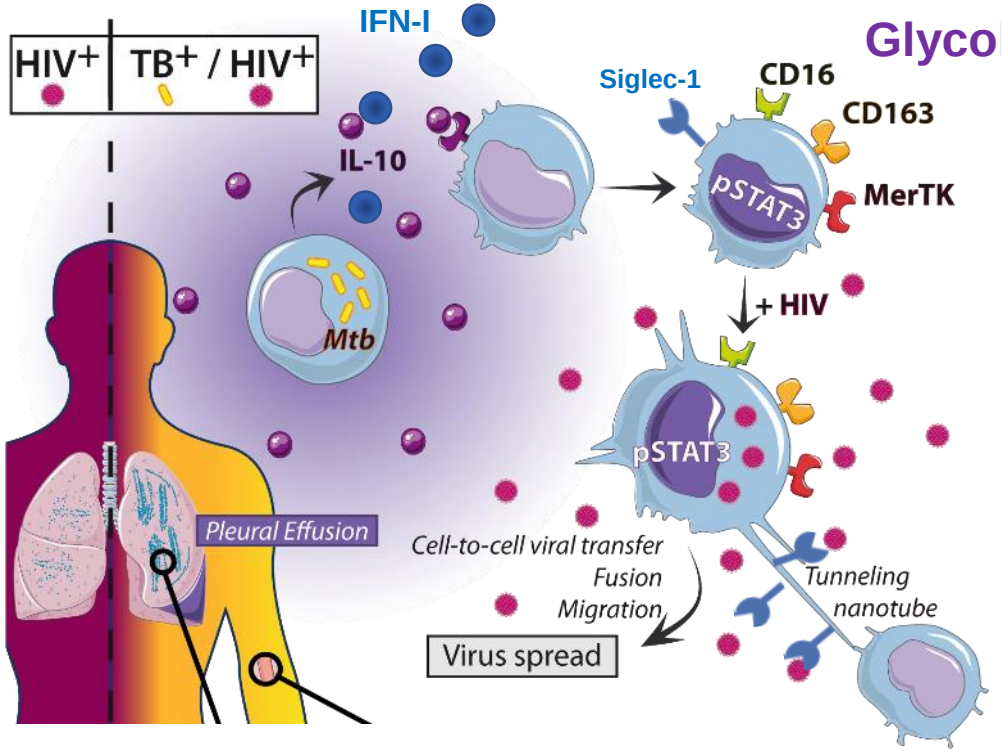


Spearman coefficient
r = 0.7261
p-value <0.001

○ Healthy △ SIV
● LTB ◎ LTB-SIV
■ ATB ▣ ATB-SIV



Conclusion part I- Role of macrophages in HIV/Mtb co-infection



Glycolysis ↑ ↑ ↑

- **Identification of glycolytic TB-induced macrophages**
- **Cellular (TNT) & molecular (Siglec-1) mechanisms involved in HIV-1 exacerbation by Mtb**

Glycolysis is key for TNT formation in these cells

- *In vivo* relevance of these macrophages

Collaboration : L. Balboa (Buenos Aires, Argentina – IRP CNRS)

Souriant S *et al. Cell Reports, 2019*
Dupont M *et al., eLife, 2020*
Dupont M *et al., J Leuk Biol, 2022*
Vahlas *et al., in preparation*

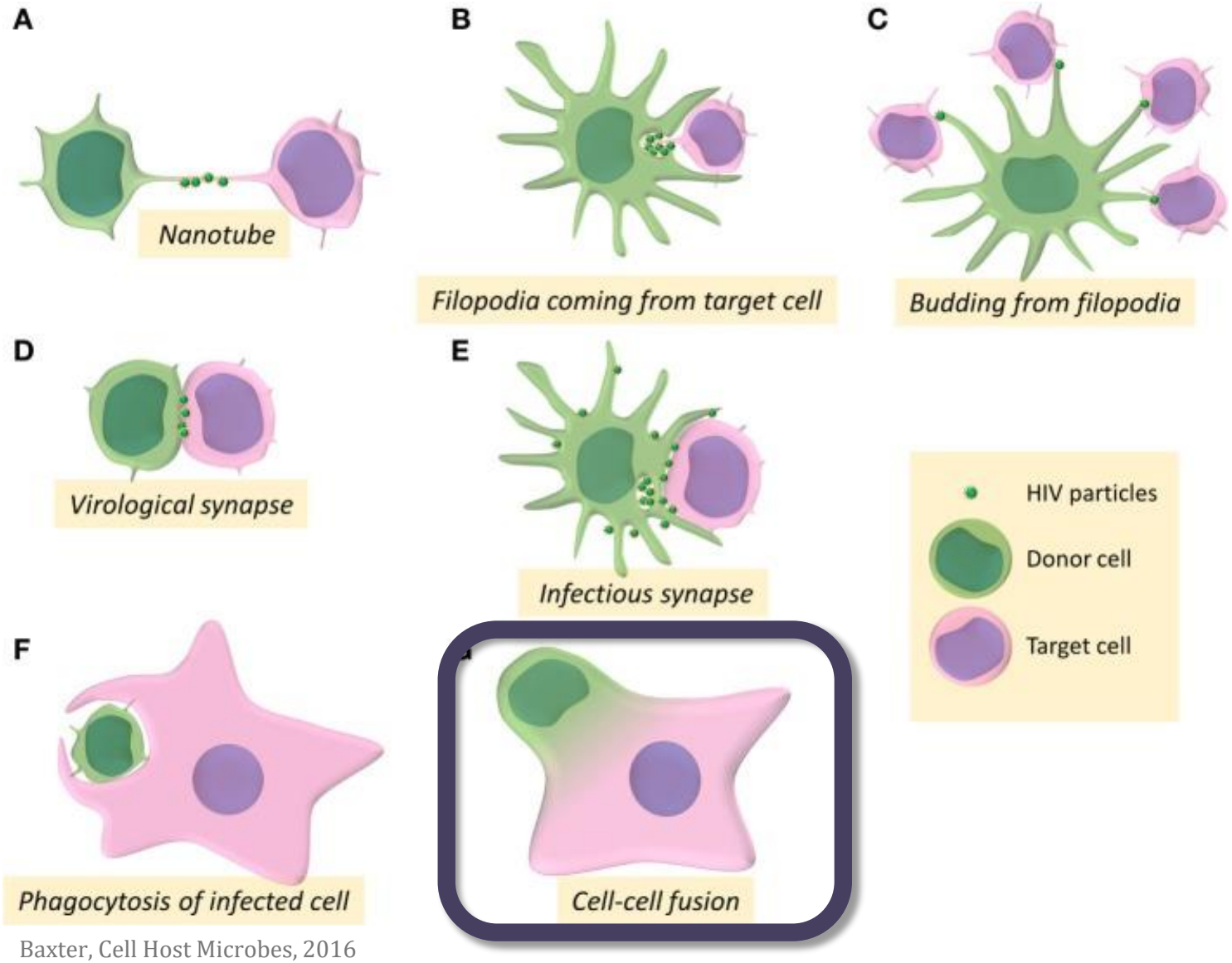
Part II- Macrophage infection by transfer from CD4+ T cells

✱ Cell-to-cell transfer of HIV-1

- ✓ Rapid and efficient
- ✓ Escapes innate immune responses
- ✓ **Main mode of infection *in vivo***

T cell material in HIV+ macrophages, rare M-tropic viruses

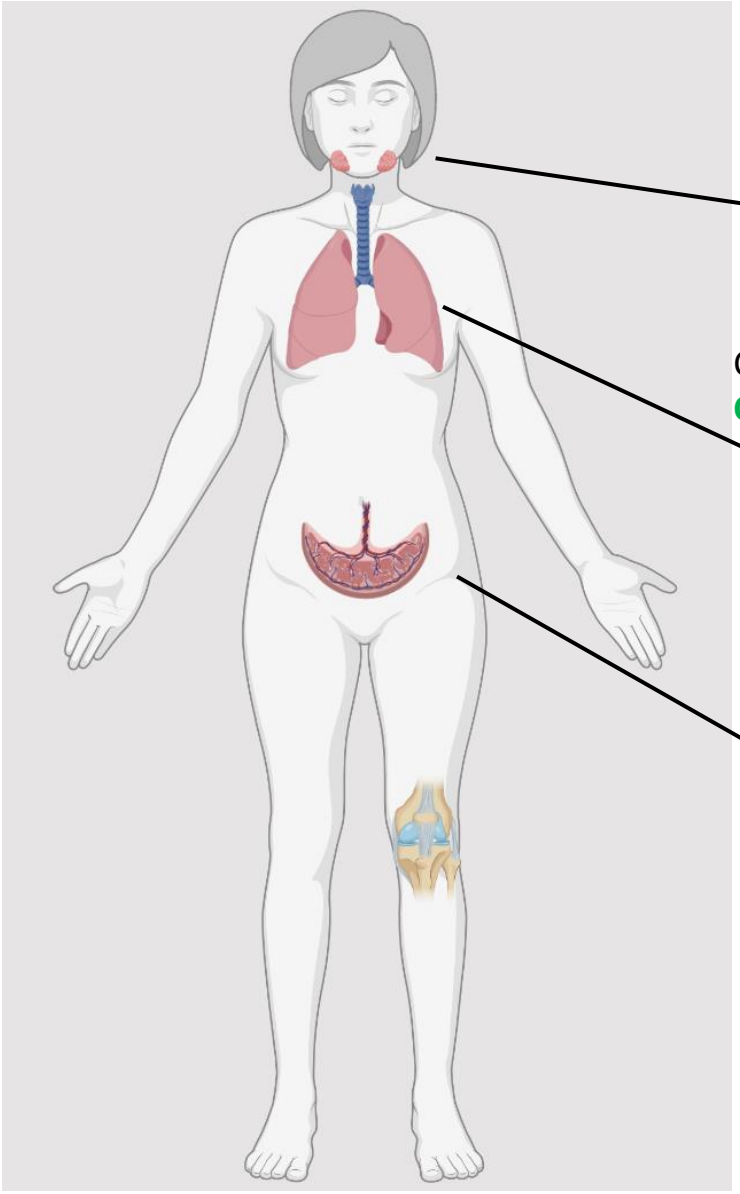
Rémi Mascarau (PhD 2022)



Baxter, Cell Host Microbes, 2016

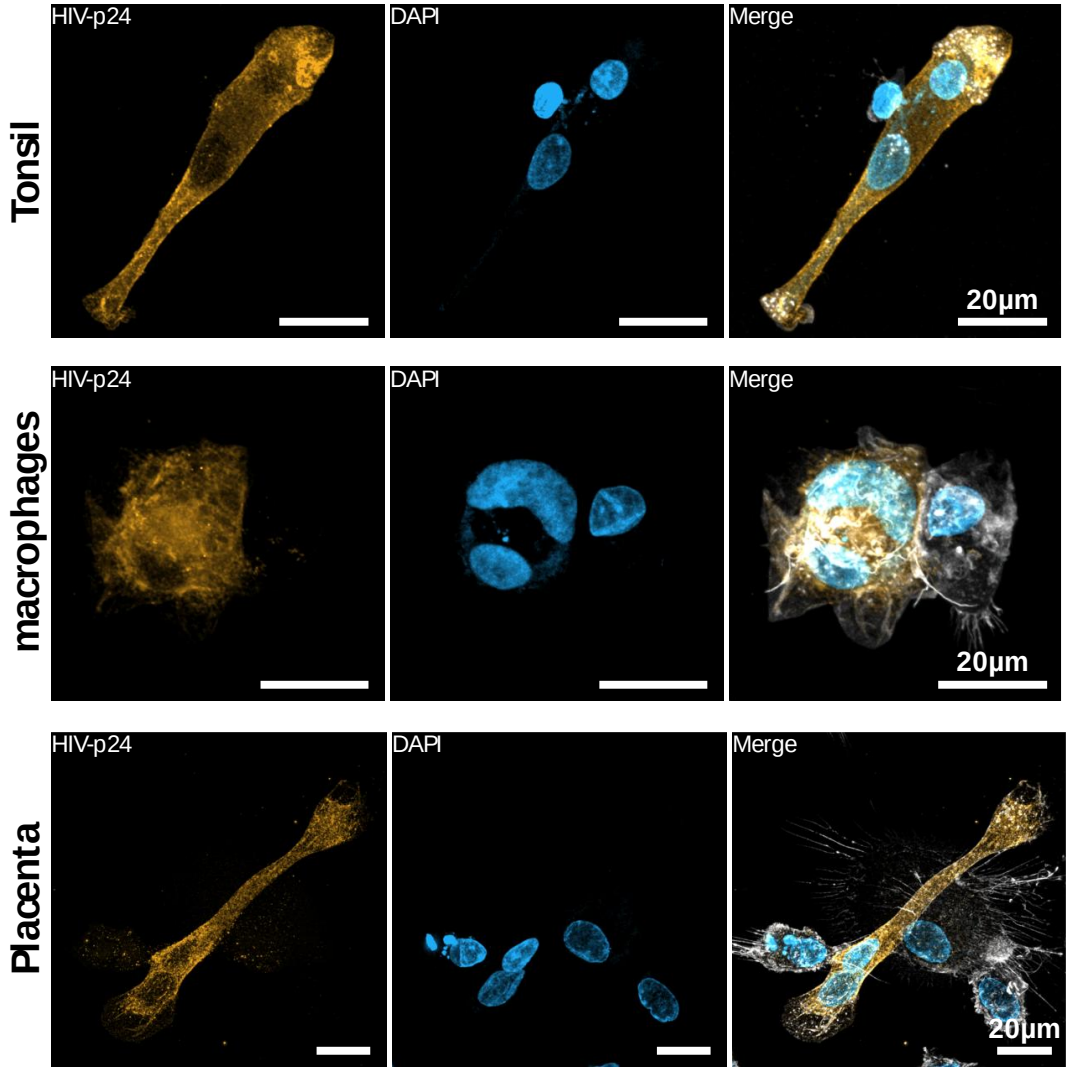
From Han et al., J Leuk biol, 2022

Cell fusion occurs with several **human tissue macrophages**



Coculture 24h-
CD4 T- HIV-2d

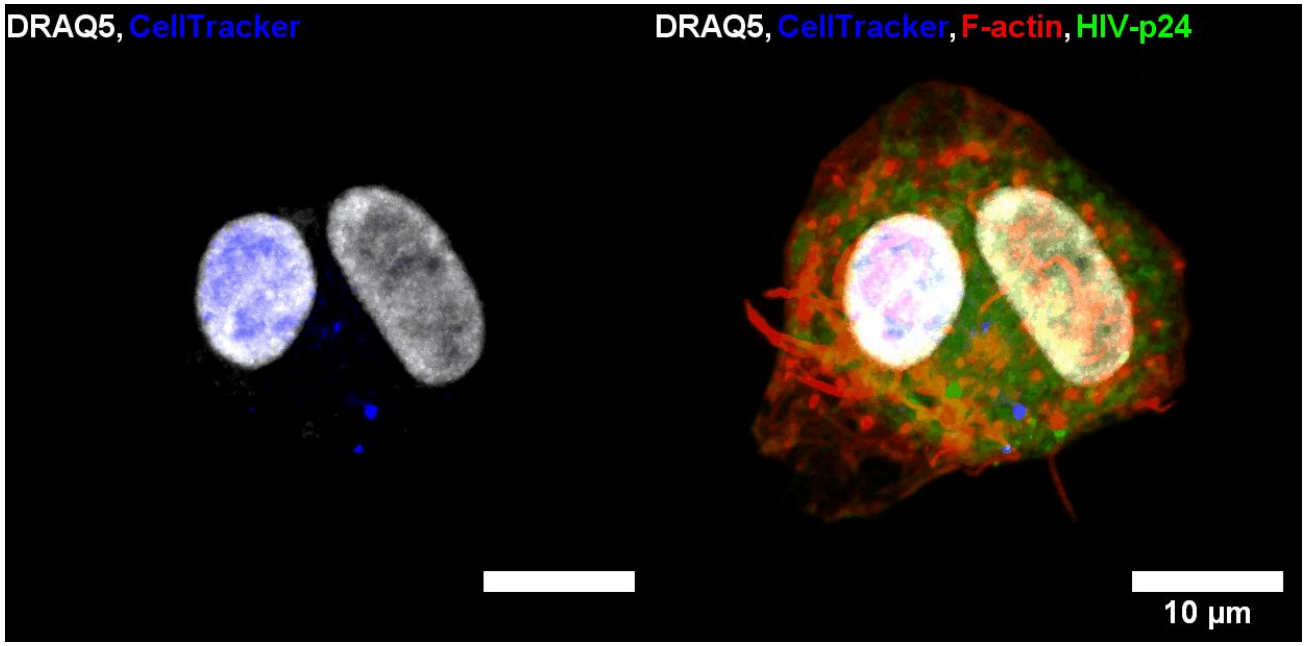
✱ ***Ex vivo* infected purified macrophages**



Collaborations: Dr. Guibert (CHU, Toulouse); B. Lagane & N. Jabrane (Infinity, Toulouse)

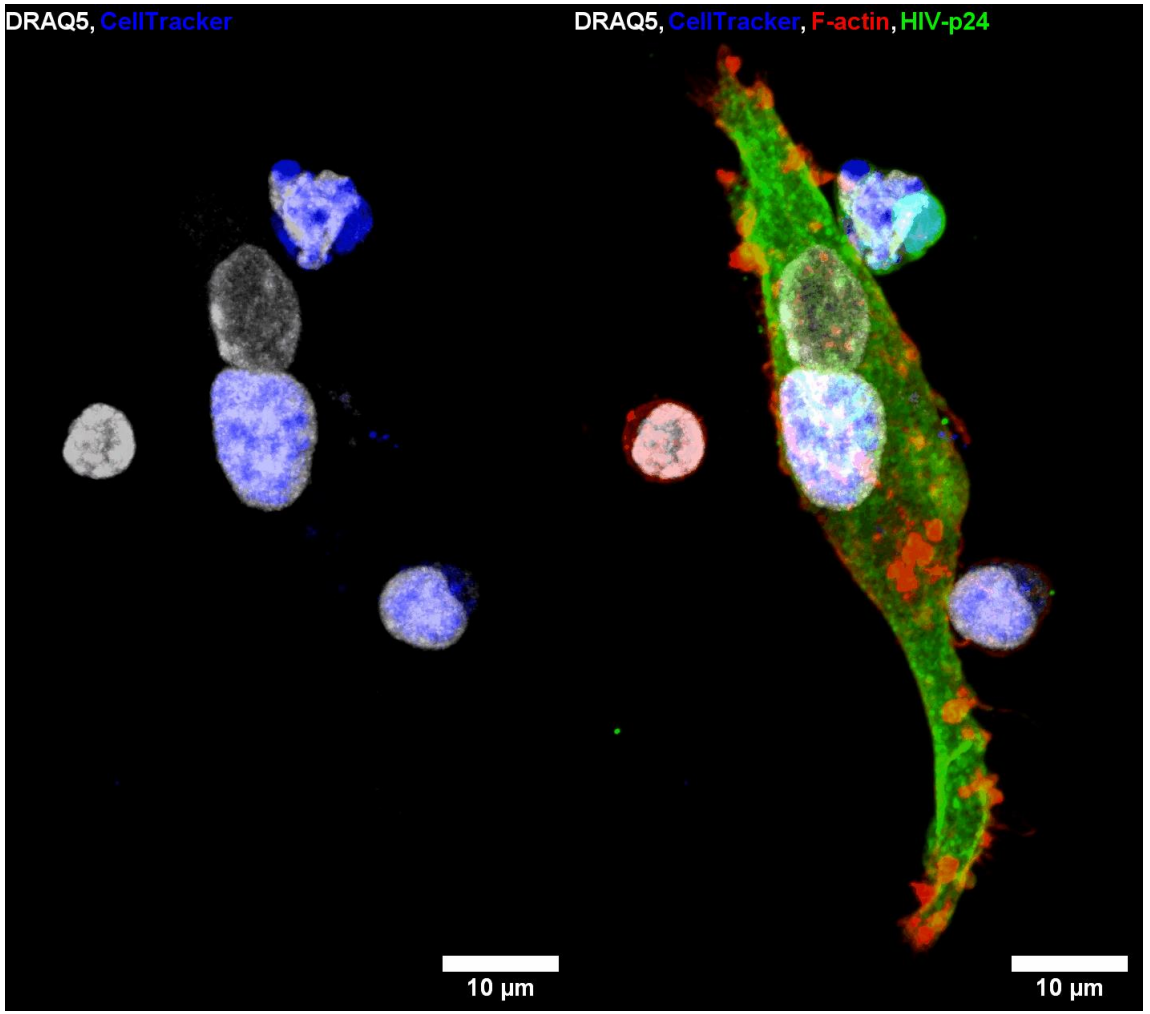
Heterotypic cell fusion occurs with several human tissue macrophages

Alveolar macrophage (Lung)



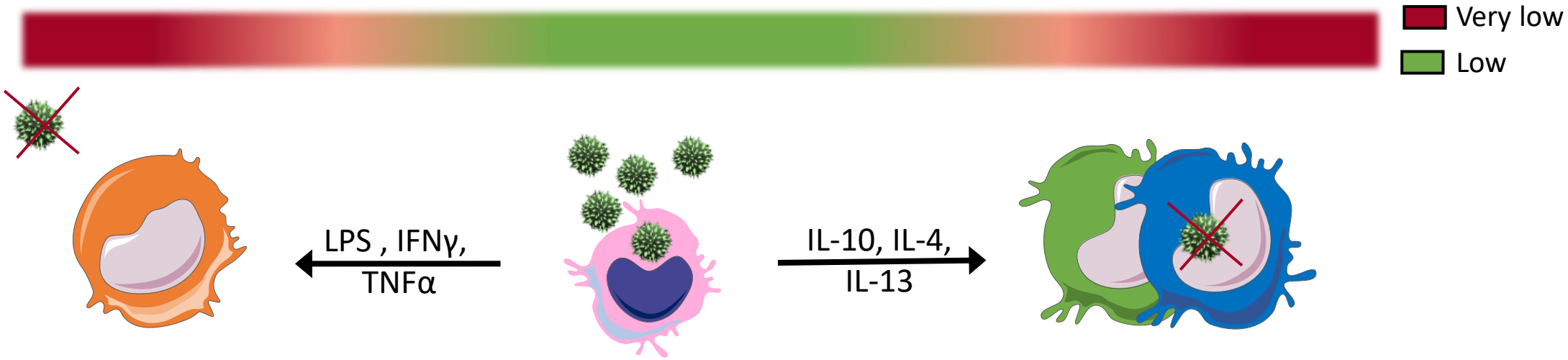
Nuclei from the CD4 T cell

Macrophage from lymphoid organ (Tonsil)



Macrophage polarization in cell-free infection

Macrophages susceptibility to cell-free infection



M1 Pro-inflammatory macrophages

Inhibition of viral DNA synthesis and viral integration

M0

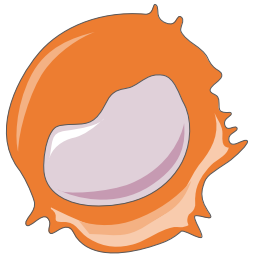
M2 Anti-inflammatory macrophages

Inhibition of reverse transcription and viral translation

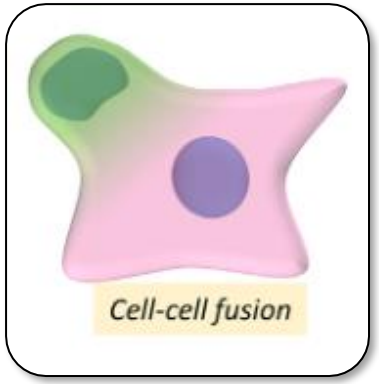
Cassol et al, J immunol 2009
Cassol et al, AIDS 2013
Cassetta, AIDS 2013

Macrophage polarization and cell-to-cell transfer of HIV-1?

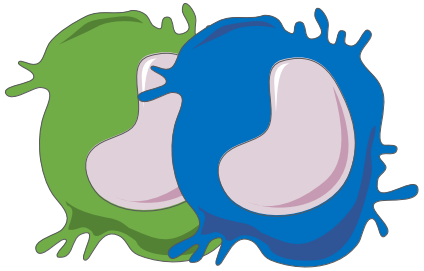
?



LPS , IFNγ, TNFα



IL-10, IL-4, IL-13



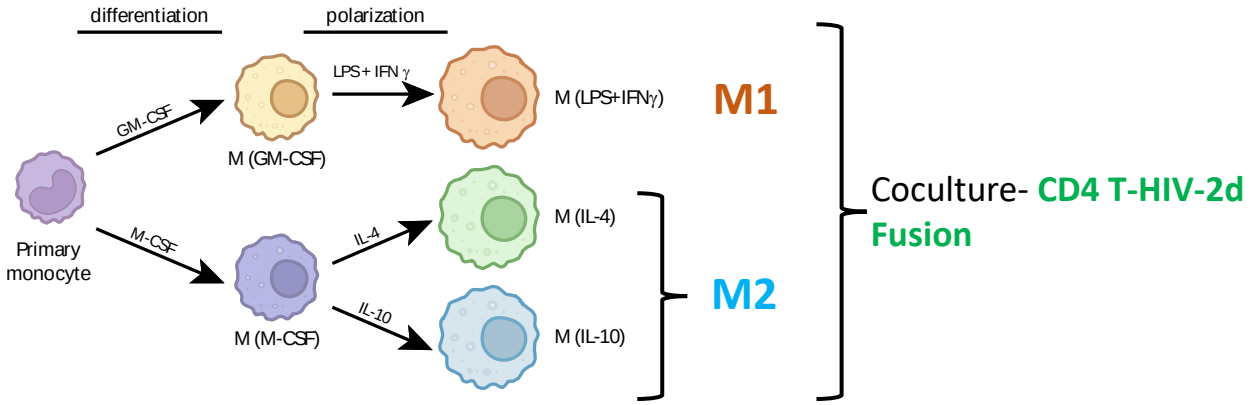
M1 Pro-inflammatory macrophages

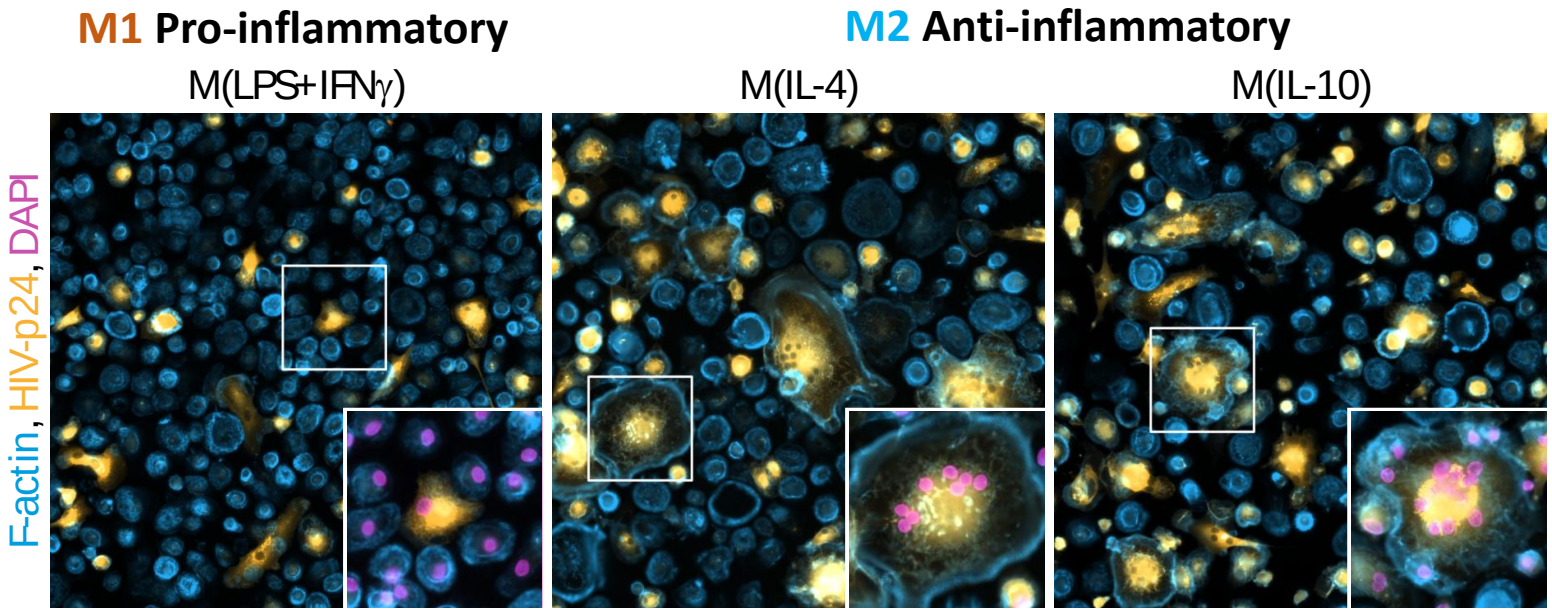
Inhibition of viral DNA synthesis and viral integration

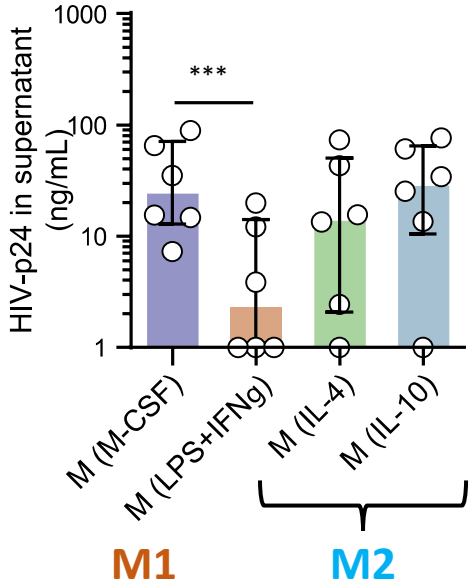
M2 Anti-inflammatory macrophages

Inhibition of reverse transcription and viral translation

Pro-inflammatory macrophage activation reduces HIV-1 transmission

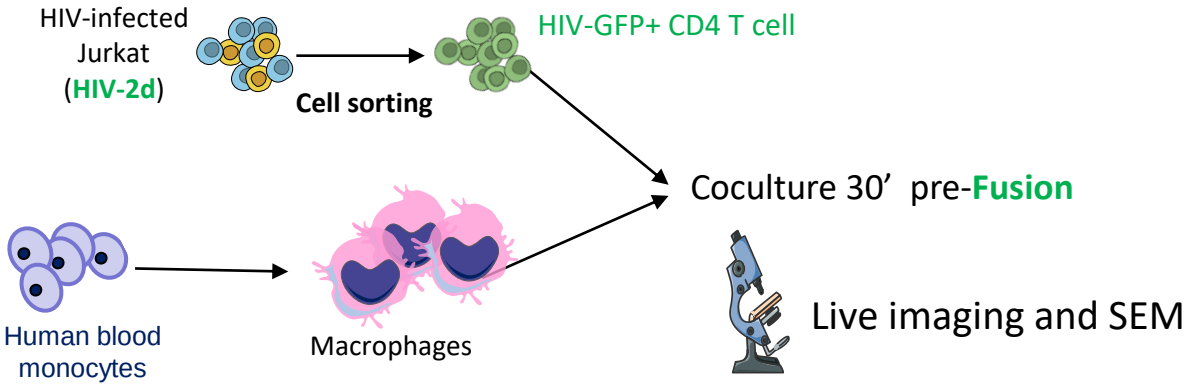


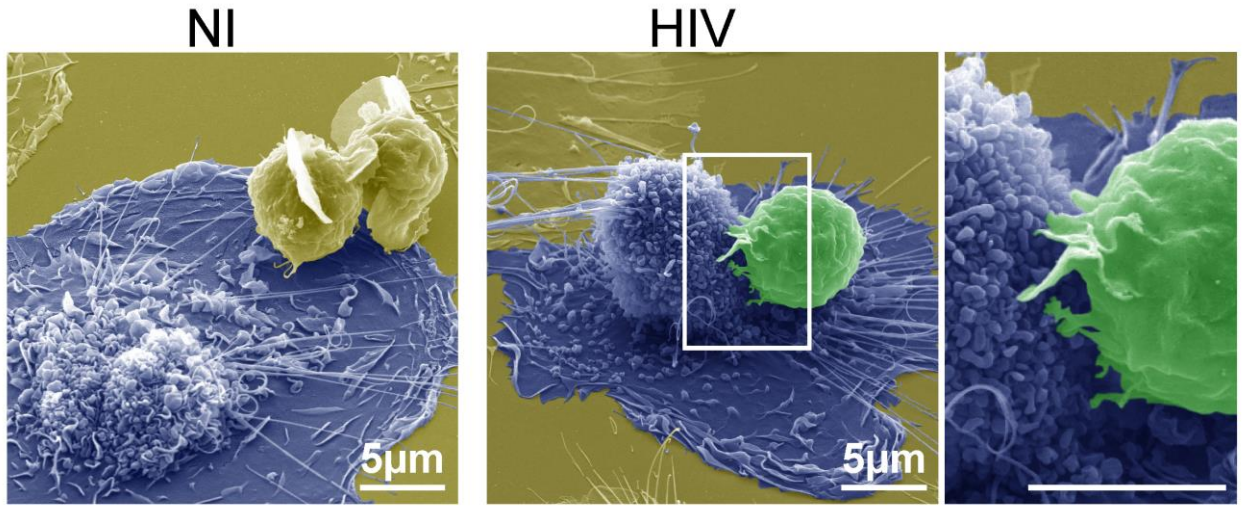


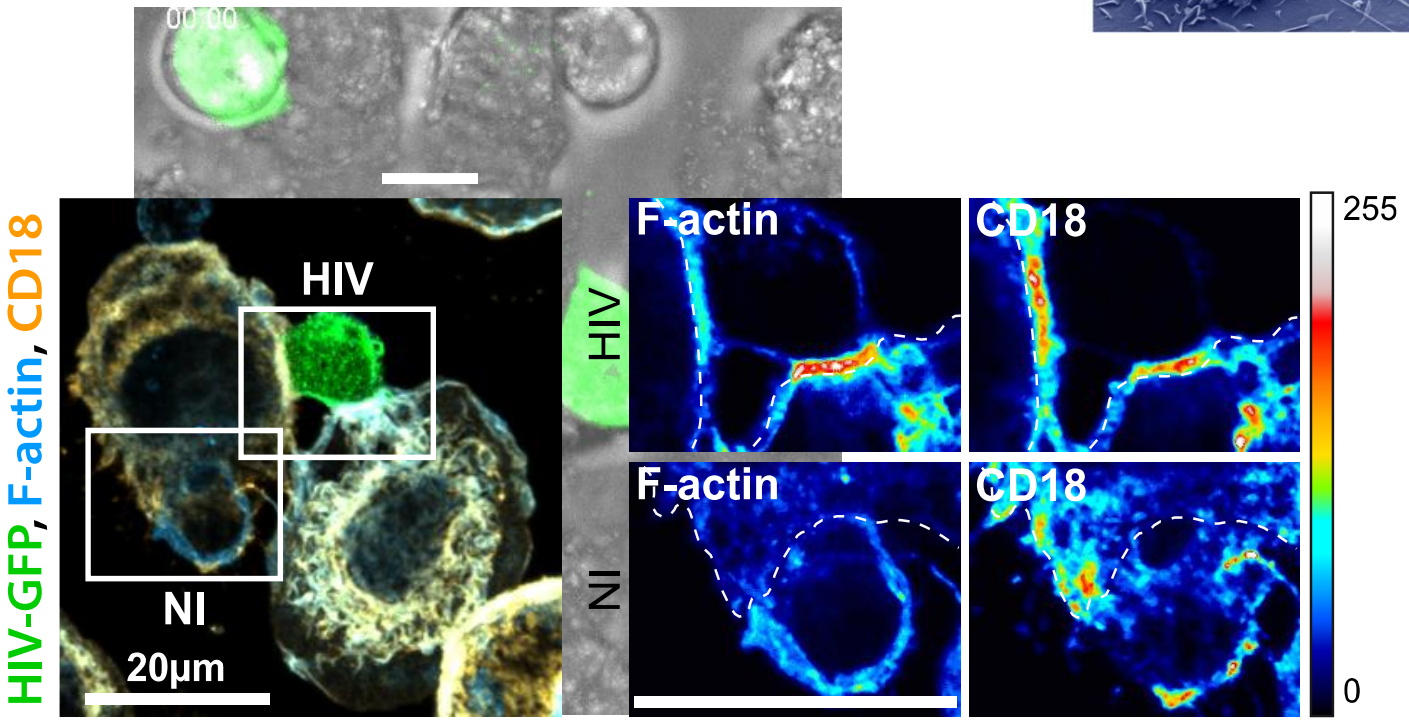


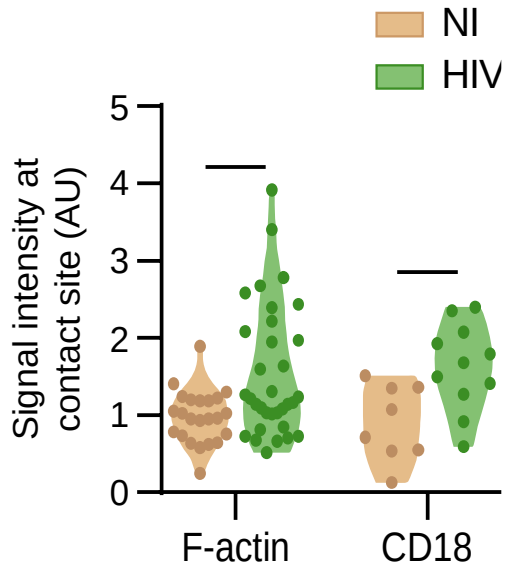
Mascarau et al, J Cell Biol 2023

Characterization of the « **fusogenic contact** » between infected CD4+ T cell and macrophages

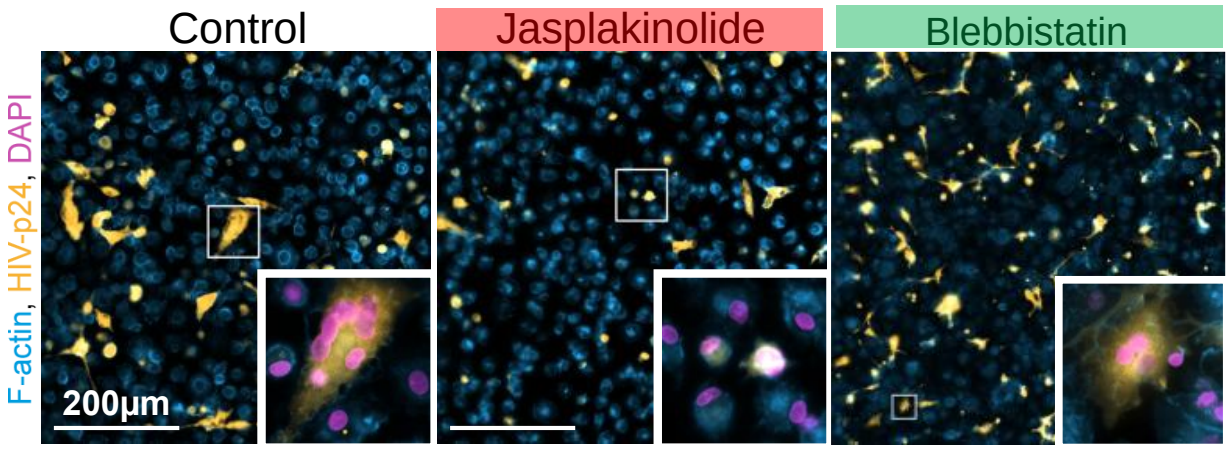


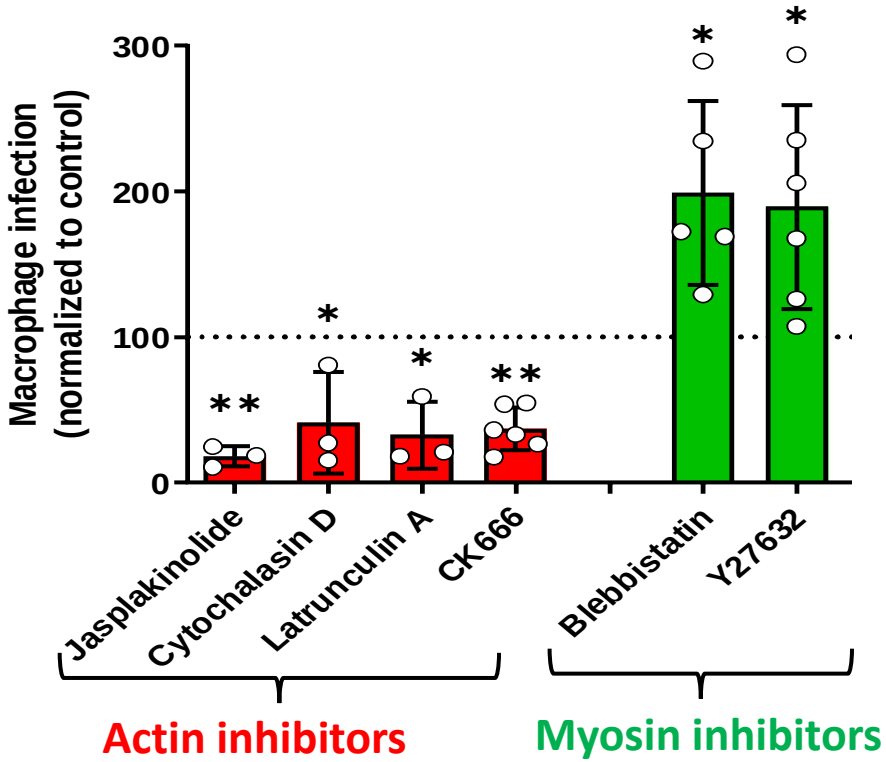


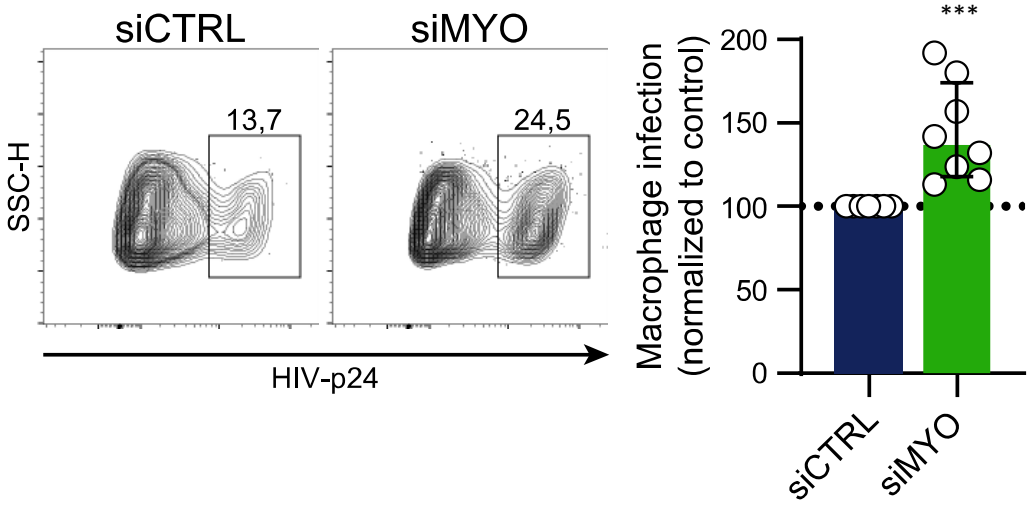




Actomyosin contraction inhibits macrophage infection by fusion







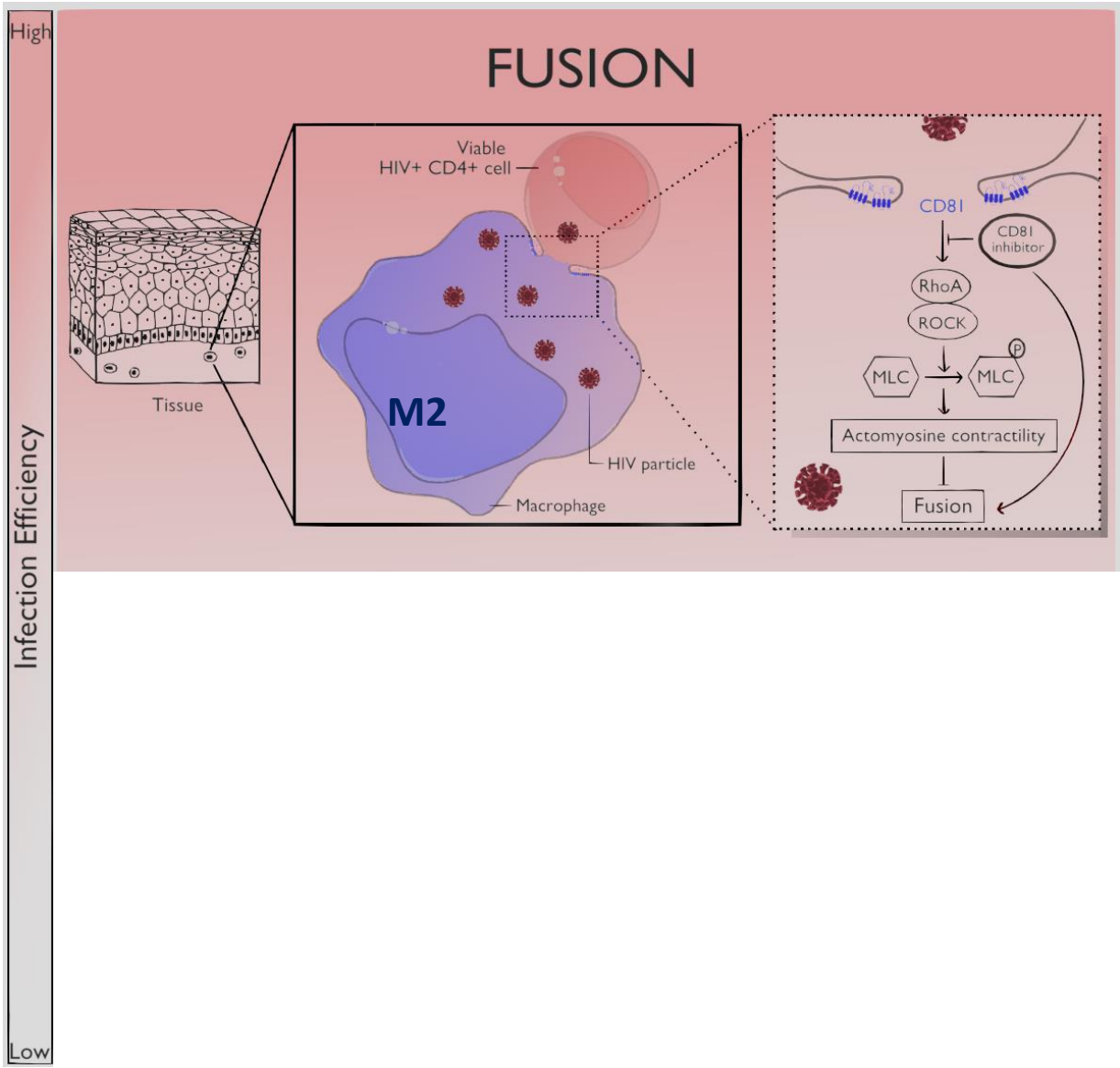
Mascarau et al, J Cell Biol 2023

Conclusion part II- Macrophage infection by fusion with CD4 T cells

CELL FUSION with CD4+ T lymphocytes:

- **is inhibited by the CD81/ROCK/RhoA/myosin axis**
- **is the most efficient for macrophage infection**
- **is influenced by macrophage activation profiles and CD4T cell fate**
- **occurs with several tissue macrophages**

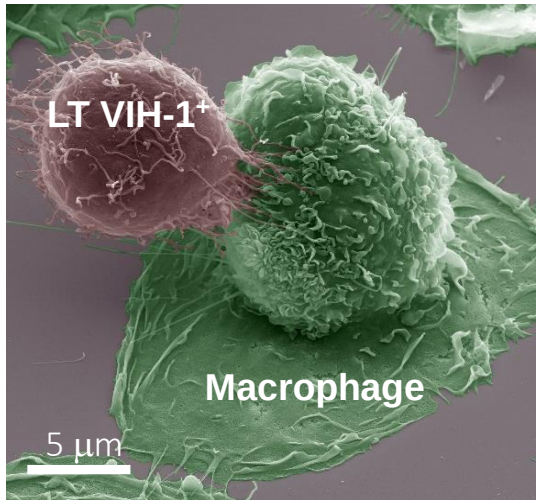
➢ ***Favoring persistent HIV reservoirs***

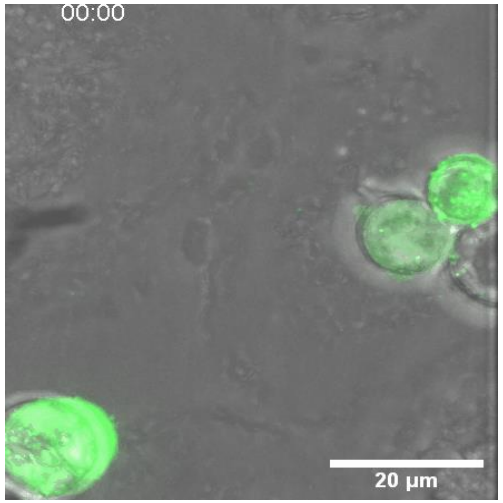


Mascarau et al, J Cell Biol 2023

Work in progress …

✱ A new mode of infection of macrophages by virus transfer from infected CD4 T lymphocytes





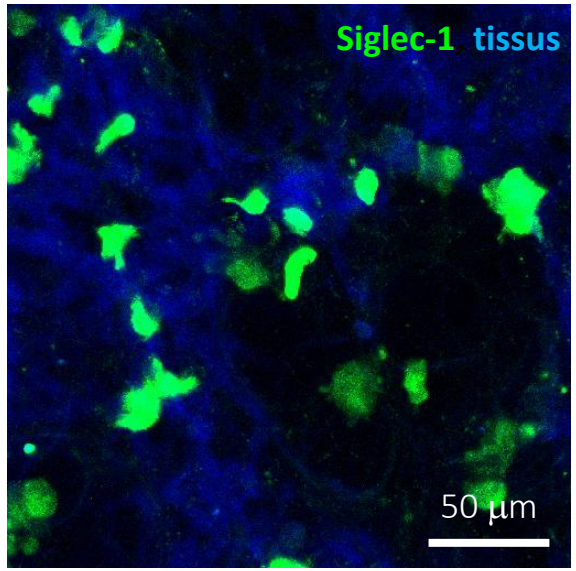
Mascarau et al, J Cell Biol 2023

Modulation by TB and metabolism ?

Natacha Faivre
(phD student)

✱ Role of Siglec-1⁺ macrophages in TB ?

- Mtb dissemination by Siglec-1⁺ macrophages
  Benet et al, J Extracell Vesicles 2021
- Cell-to-cell communication (TNT *in vivo*)



Intravital microcopy:
lung of Mtb-infected mice

BSL-3

Siglec-1-Cre-mTmG
Siglec-1 KO

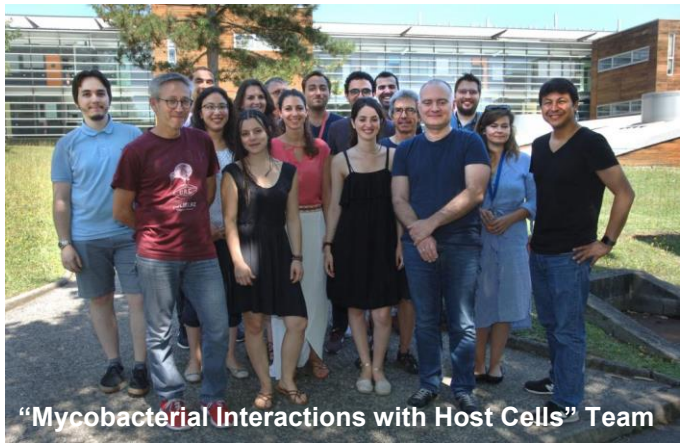
Sarah Monard
(phD student)

Collaboration : O. Neyrolles (IPBS, Toulouse, France)

THANKS !

* **Christel Vérollet & Renaud Poincloux's Team** (IPBS, Toulouse)
  - **Shanti Souriant (PhD, IRP)**
  - **Remi Mascarau (PhD)**
  - Zoï Vahlas (post-doc ANRS)
  - Sarah Monard (PhD student, IRP)
  - Karine Pingris
  - Brigitte Raynaud-Messina

* **Olivier Neyrolles' team** (IPBS, Toulouse)
  - **Geanncarlo Lugo-Villarino**
  - **Maeva Dupont (PhD, IRP)**
  - Sarah Monard (PhD student, IRP)
  - Stella Rousset

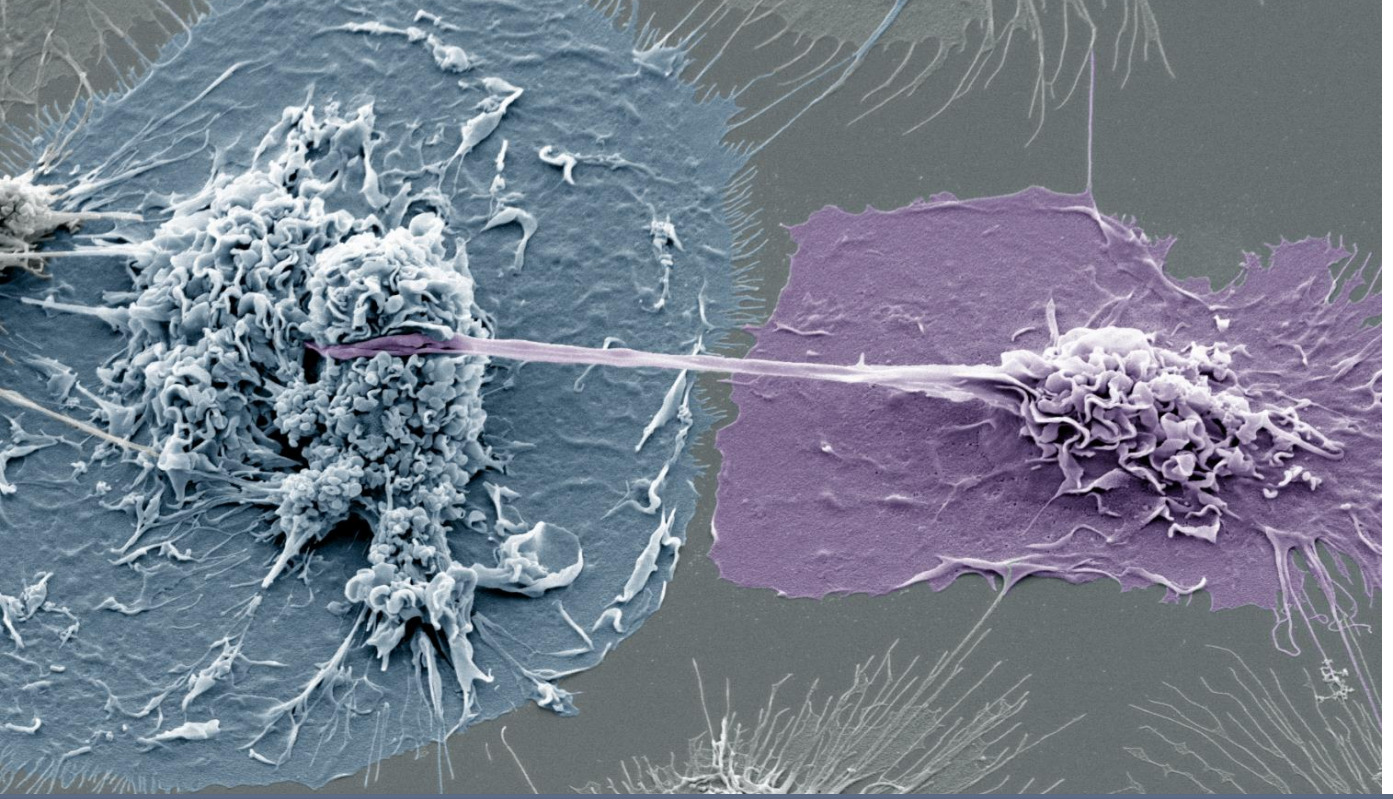


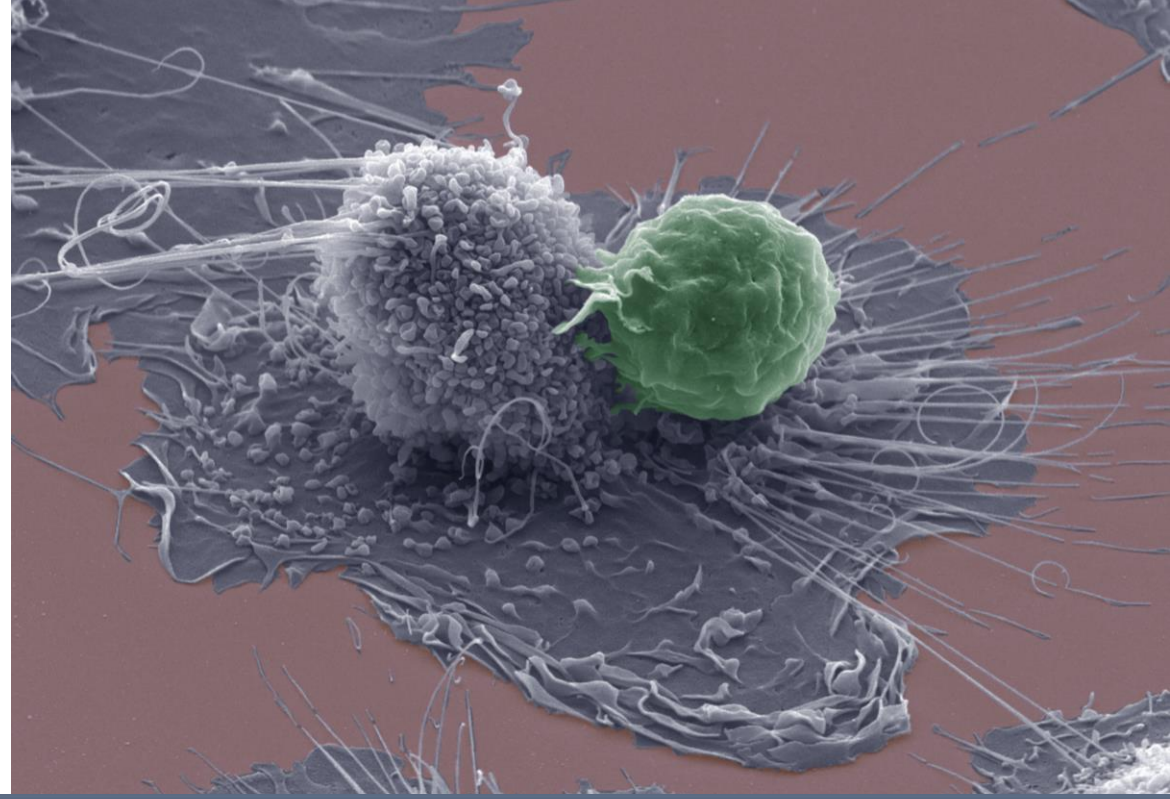
* **Lucina Balboa's team** (CONICET, Buenos Aires, Argentina)
  - Mariano Maio
  - José Luis Marin

International Research Project
IM-TB/HIV

* **Others collaborators:** M. Kuroda, T. Mempel, N. Izquierdo-Useros, S. Benichou, B. Lagane, …

* **Fundings**





THANKS !