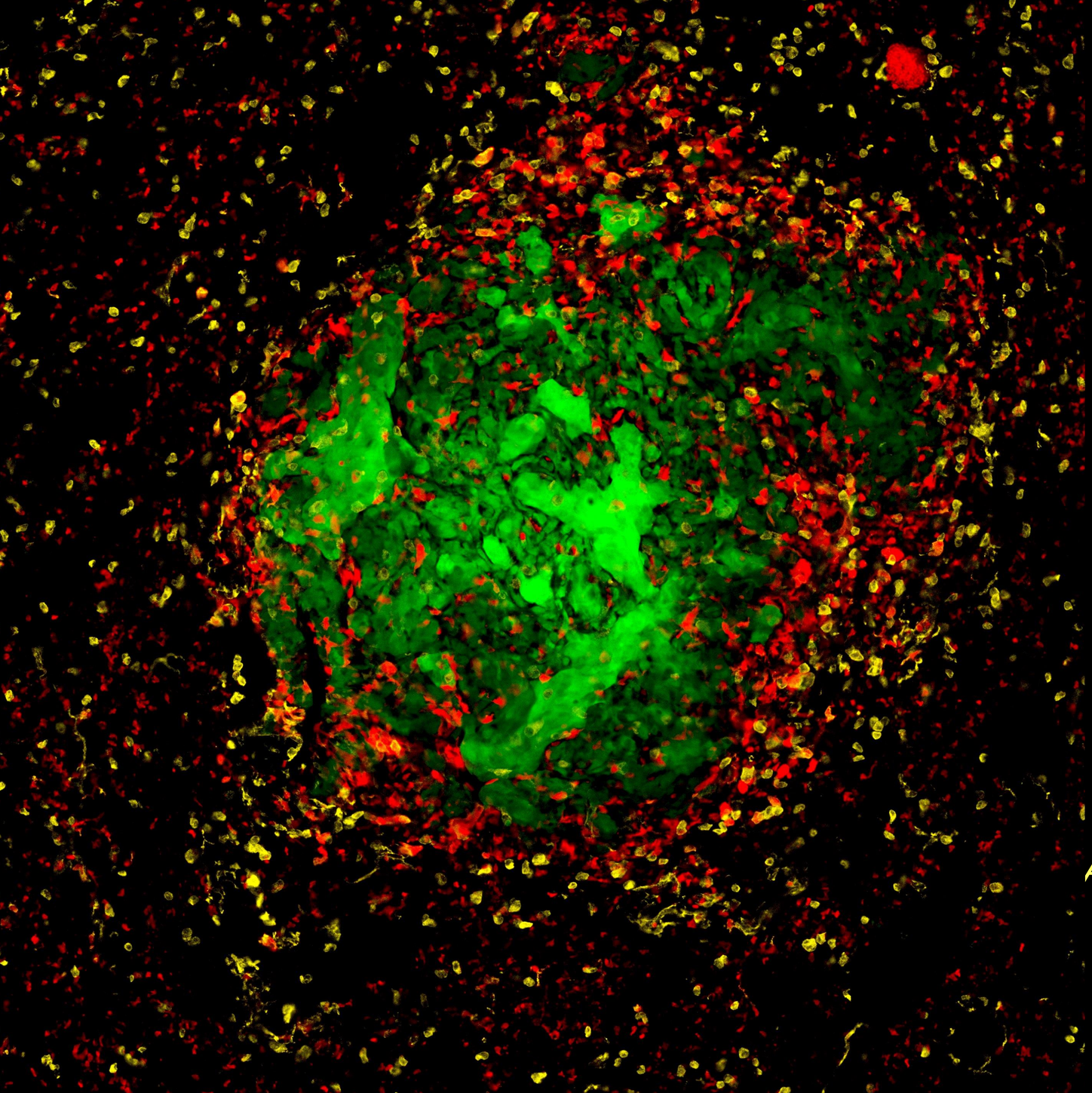


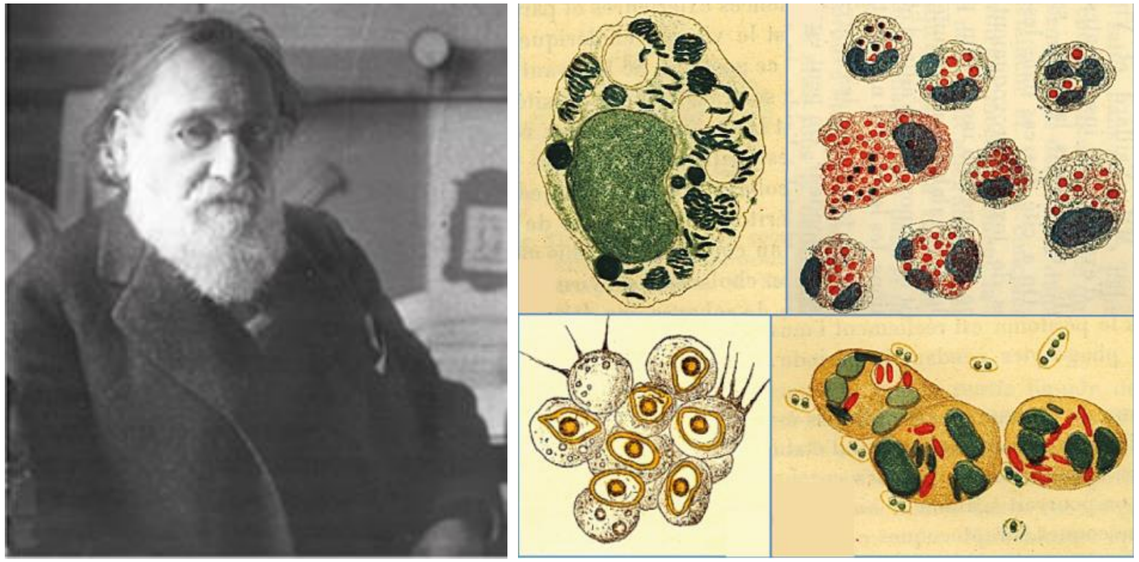
# Tissue-resident macrophages provide a pro-tumorigenic niche to early NSCLC cells

**María Casanova-Acebes, Ph.D.**

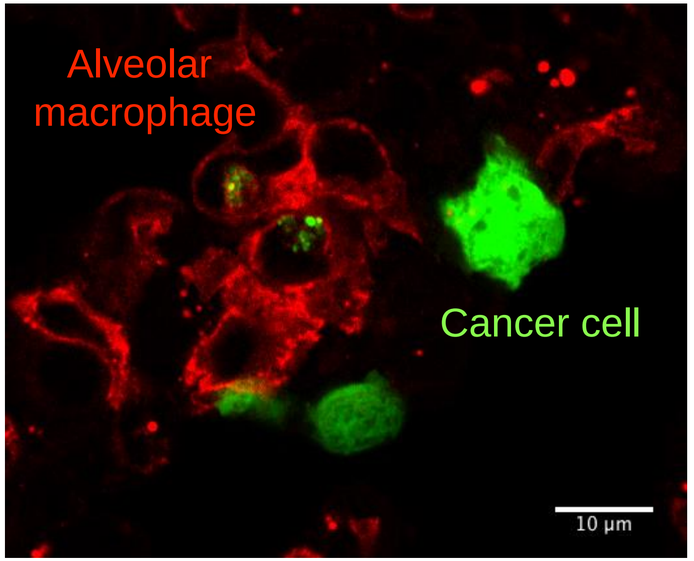
Icahn School of Medicine at Mount Sinai (ISMMS)/
Centro Nacional Investigaciones Oncológicas (CNIO)

SITC Macrophage Biology
for Anti-Tumor Immunity:
*A Deep Dive in Cancer Immunotherapy Targets seminar*
October 7th 2021

# Fundamental aspects of macrophage biology in *2021*



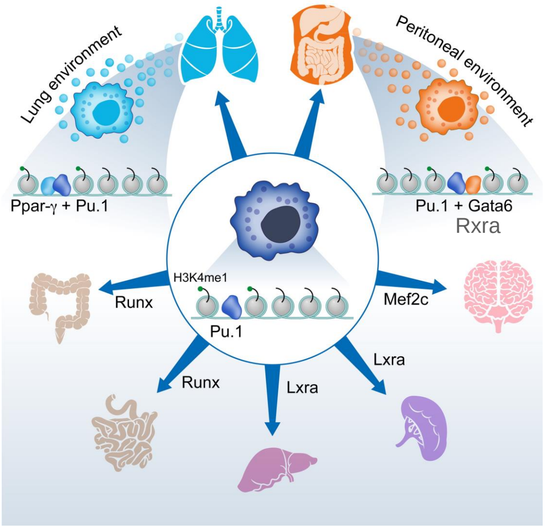
Elie Metchnikoff. 1883, Phagocytosis theory




Highly phagocytic cells in steady-state (immunosuppressive) and disease (immunomodulatory)

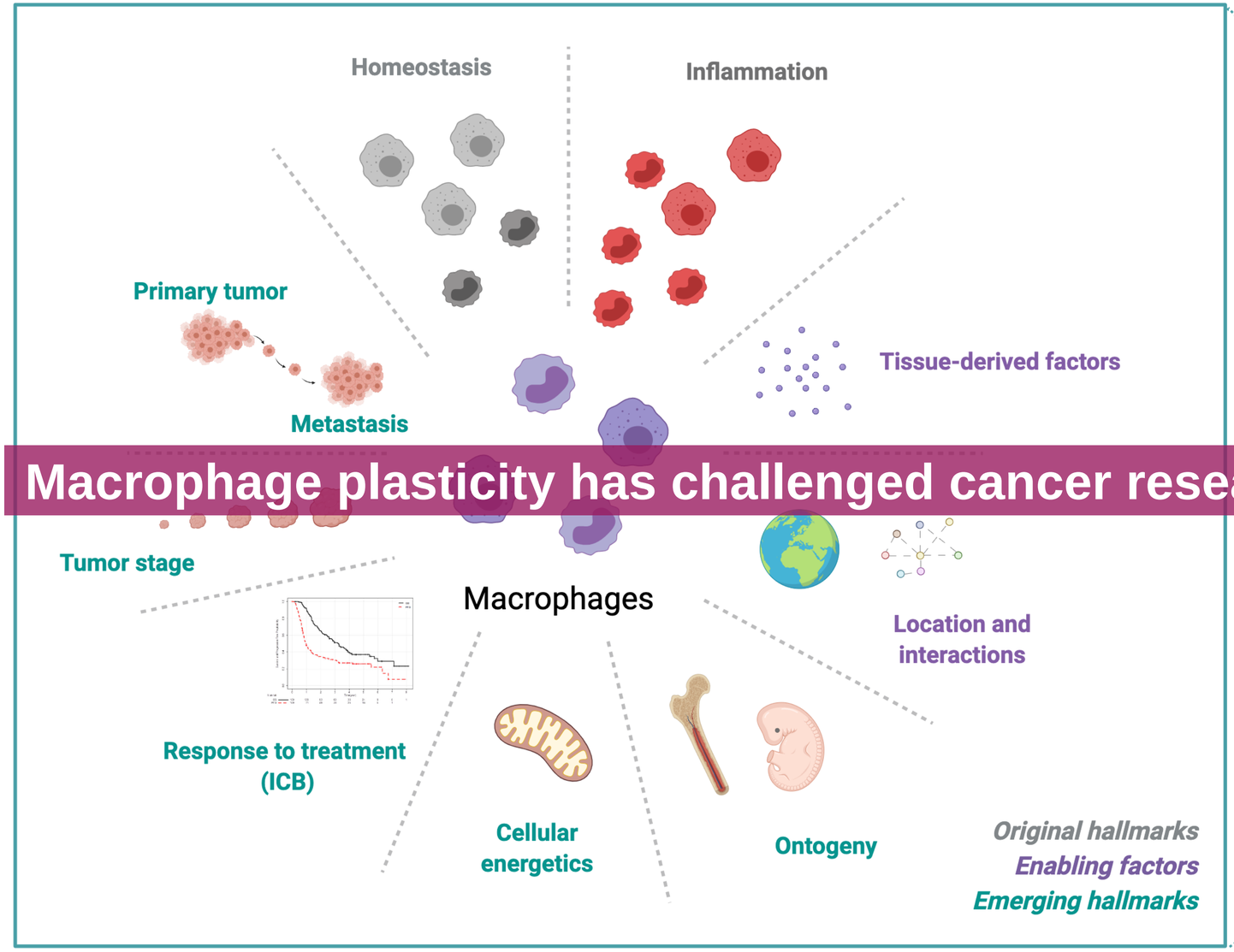
Most heterogeneous lineage of all myeloid cells -> tissular specific cues imprint macrophages

Long-lived (embryonic compartment, self-maintained) vs short-lived (bone-marrow derived compartment, recruitment)




*Lavin, Y et al 2014. Casanova-Acebes et al 2020 & 2021*

# Hallmarks in macrophage heterogeneity and plasticity




**Macrophage plasticity has challenged cancer researchers over the last 40 years**

*Aging*
*Obesity*
*Bacterial infections*
*…*

*Created with BioRender.com*

# Tumor-associated macrophages

★ Largest immune cell compartment in solid tumors

★ Growth, immunosuppression, angiogenesis, invasiveness & metastasis

*Pollard 2004; Boissonnas 2013; Broz 2014; Lewis 2016; Wyckoff 2007; Kitamura 2015; Linde 2018.*

## Tumor and macrophage heterogeneity

★ Organ in which the tumor develops

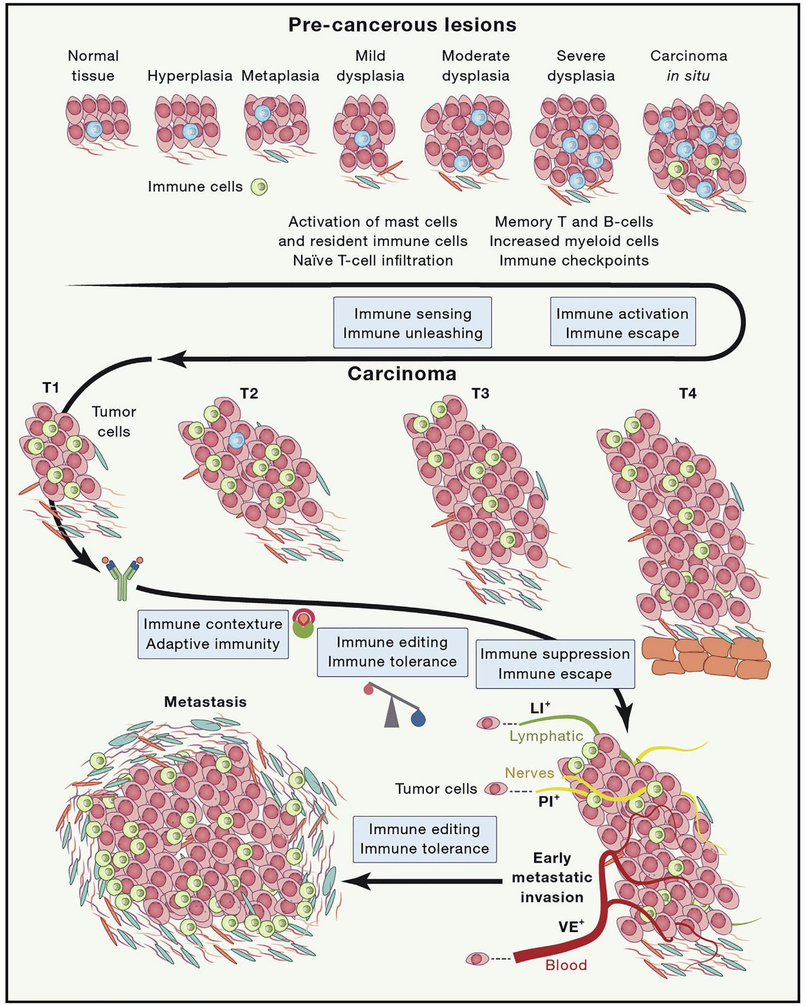
★ **Tumor stage: preneoplastic, early and late lesions**

## Human tumor macrophages

★ Mostly based on *in-vitro* studies

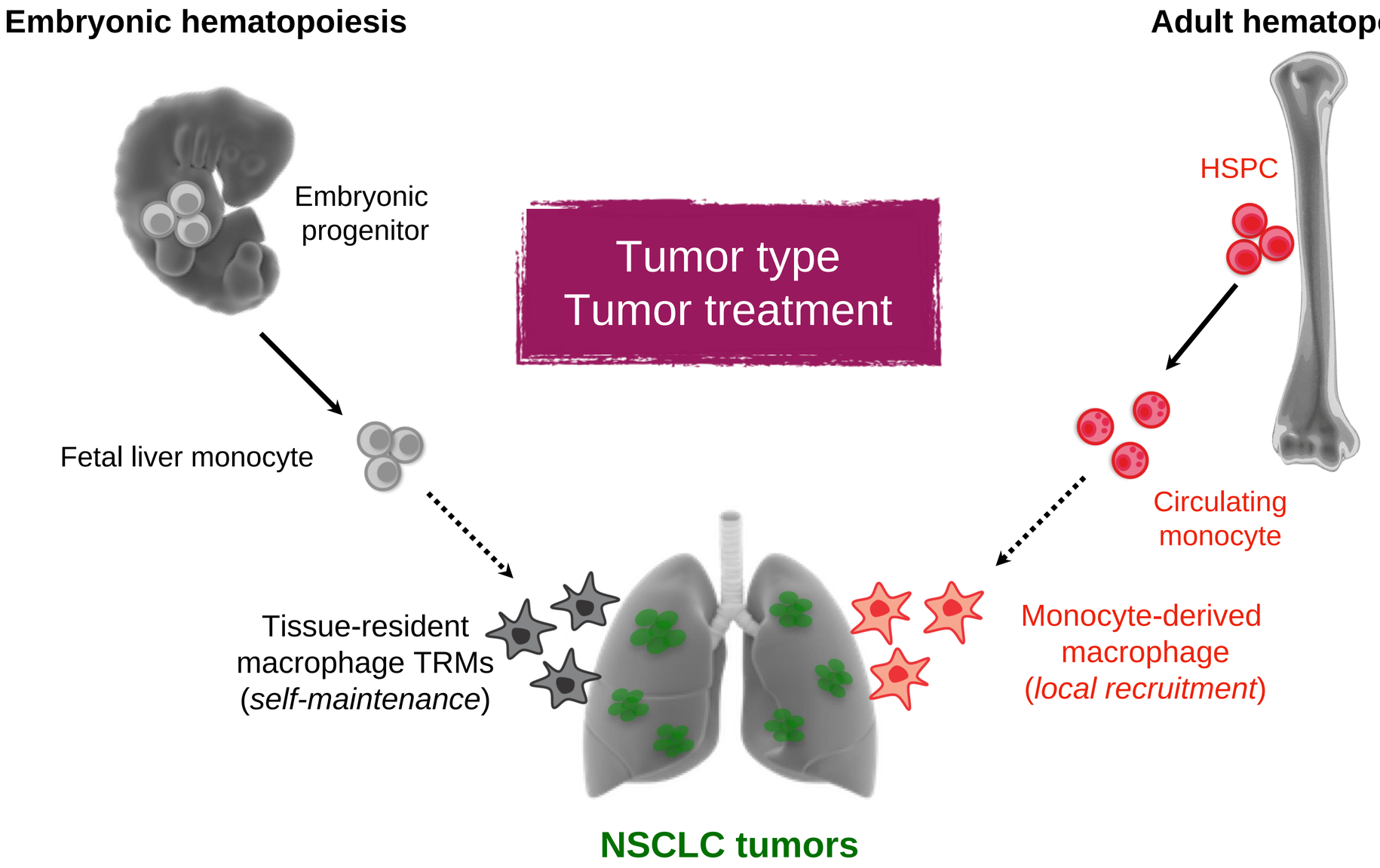
★ M1/M2 paradigm does not recapitulate macrophage function ***in vivo***

★ ***<u>Incomplete definition</u>*: tissue-resident macrophage lineage**

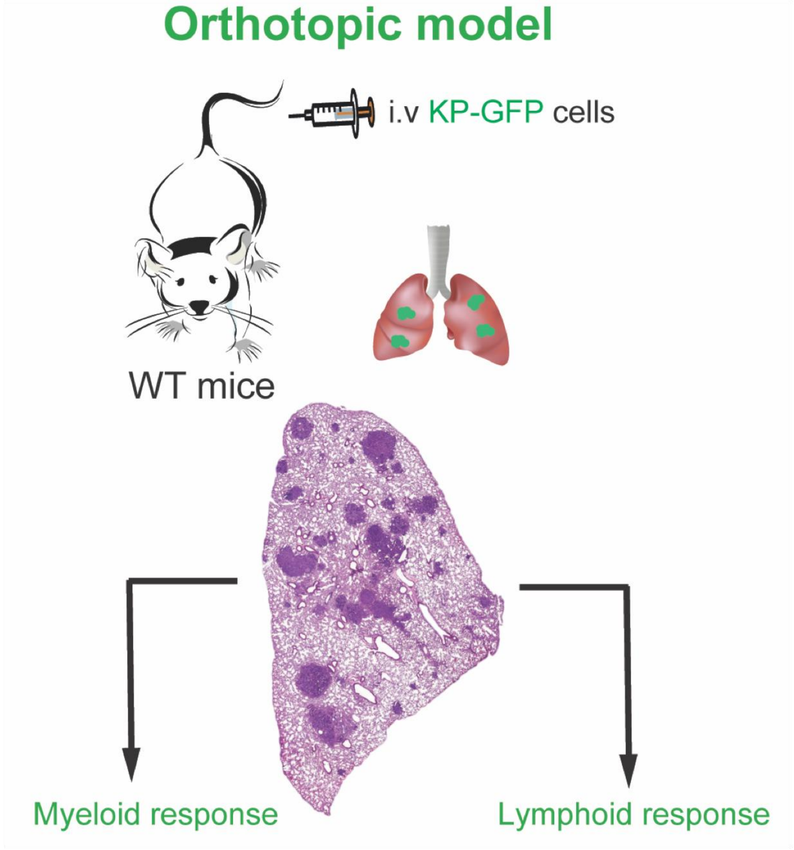


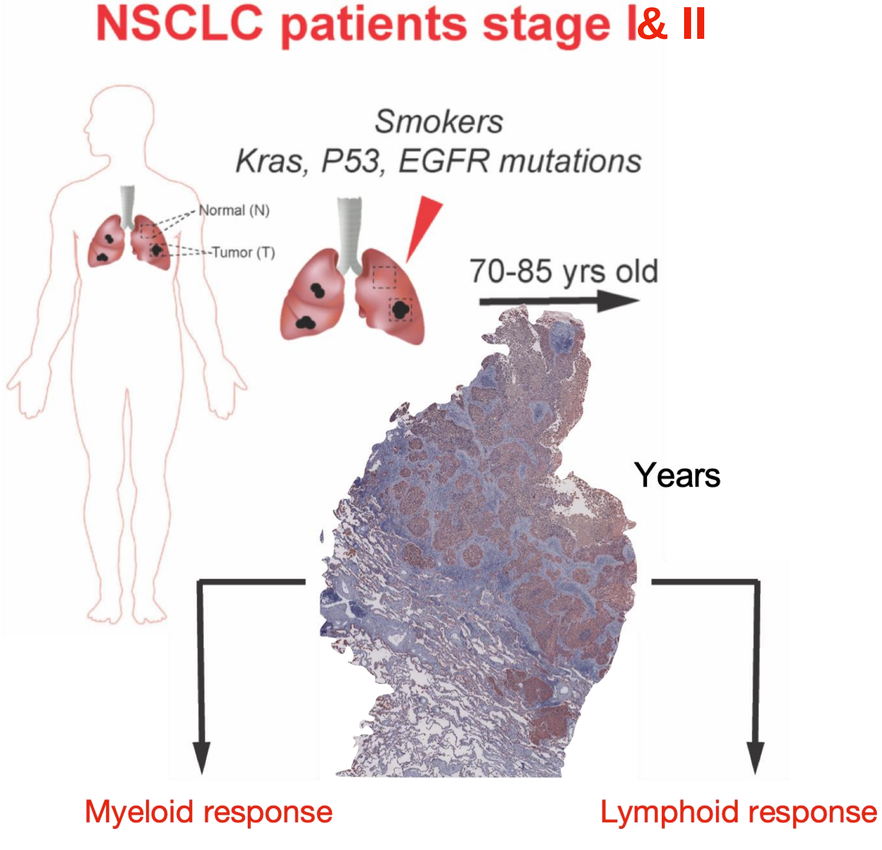

*Milette et al., J.of Pathology 2019;*
*Galon & Bruni., Immunity 2020*

# Macrophages of different origin modulate anti-tumor immunity



# Mouse KP model to study immune response in human NSCLC




**Murine NSCLC tumors**

***K-ras*$^{G12D}$ mutation:** activation of oncogenic allele, sufficient to initiate tumor growth

Deletion of p53: rapid development of adenocarcinomas

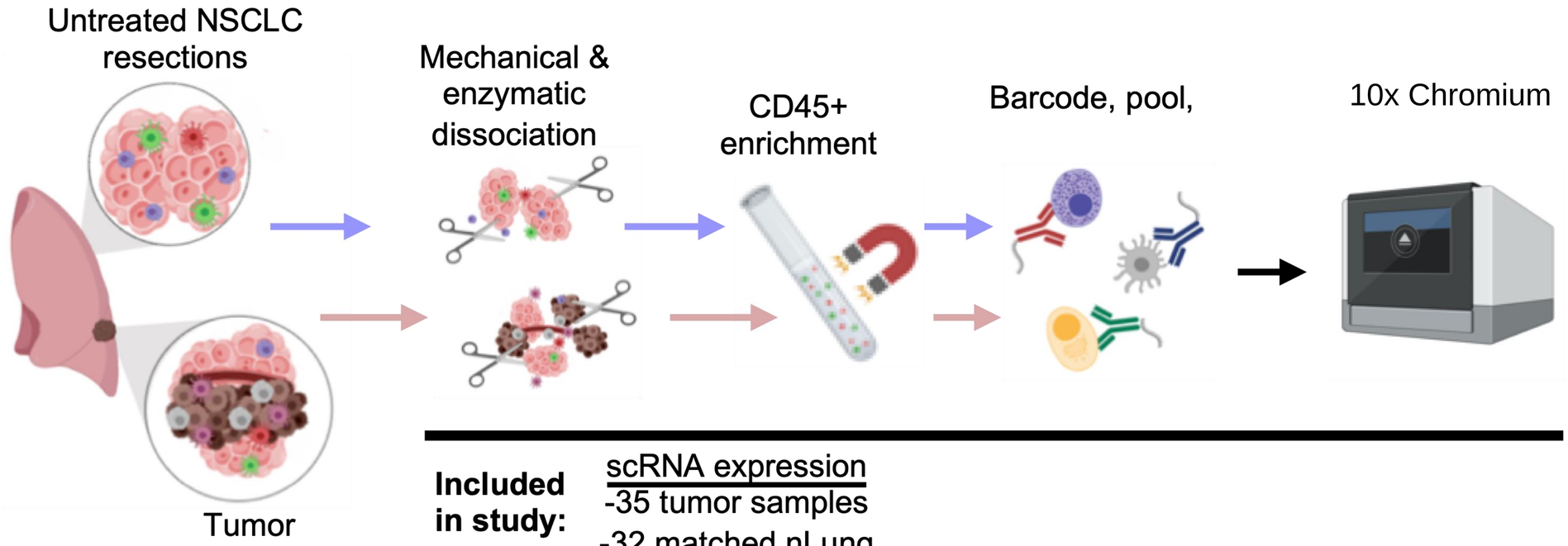
GFP: track tumor growth

**Human NSCLC tumors**

***K-ras*$^{G12D}$ & deletion of p53**

Older patients 70-85 yrs old

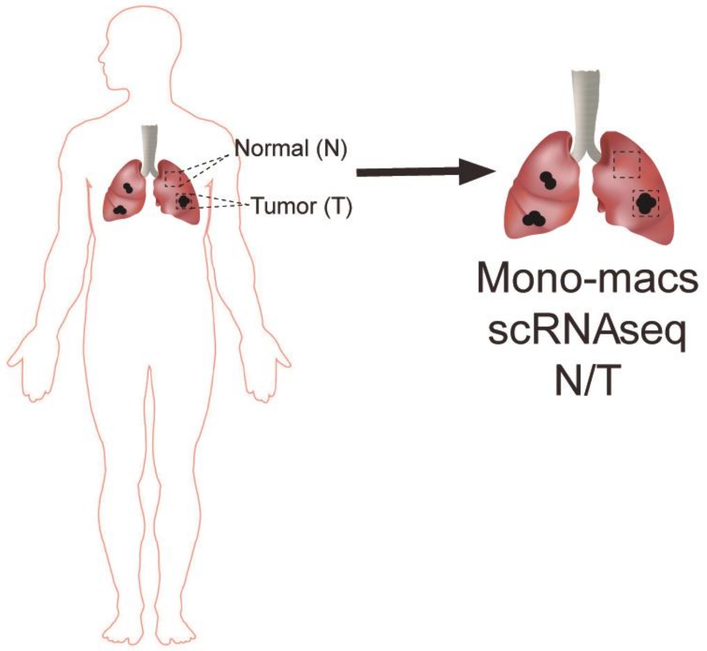
# scRNAseq captures macrophage and monocyte heterogeneity in NSCLC



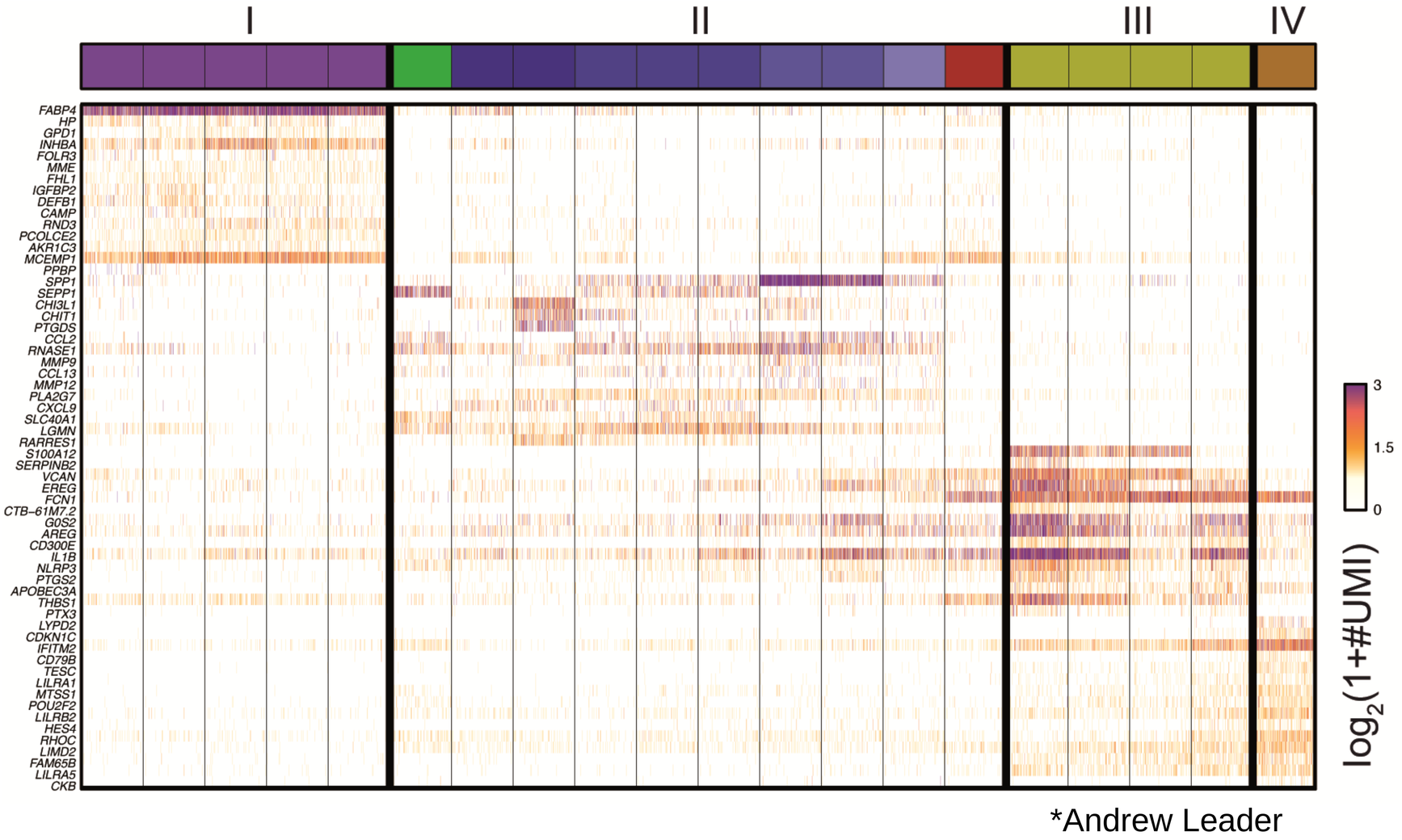

**Included in study:**

scRNA expression
-35 tumor samples
-32 matched nLung

# scRNAseq captures macrophage and monocyte heterogeneity in NSCLC



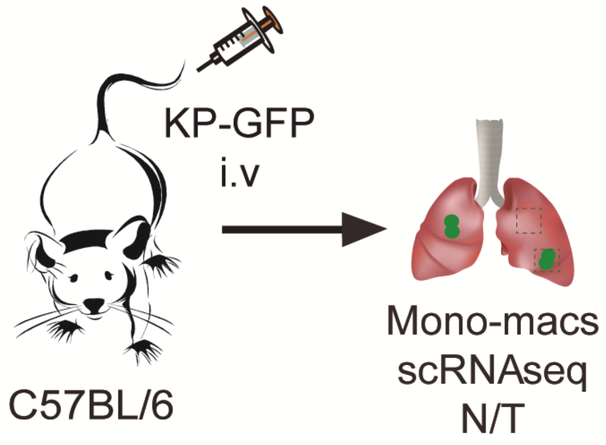

Mono-macs
scRNAseq
N/T

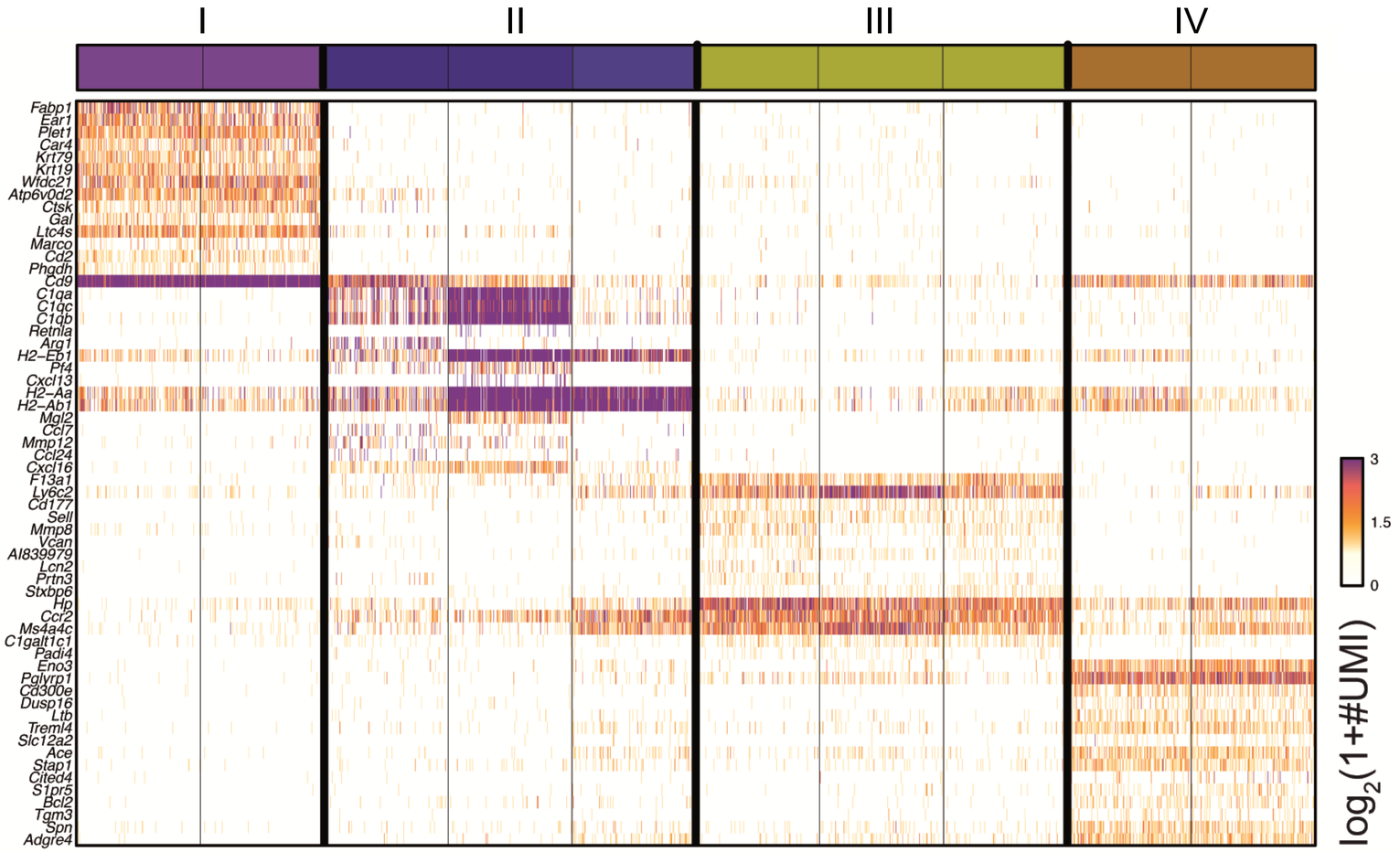



*Andrew Leader

Various intermediate states exist between the so-called M1 and M2 macrophages

# scRNAseq captures macrophage and monocyte heterogeneity in NSCLC



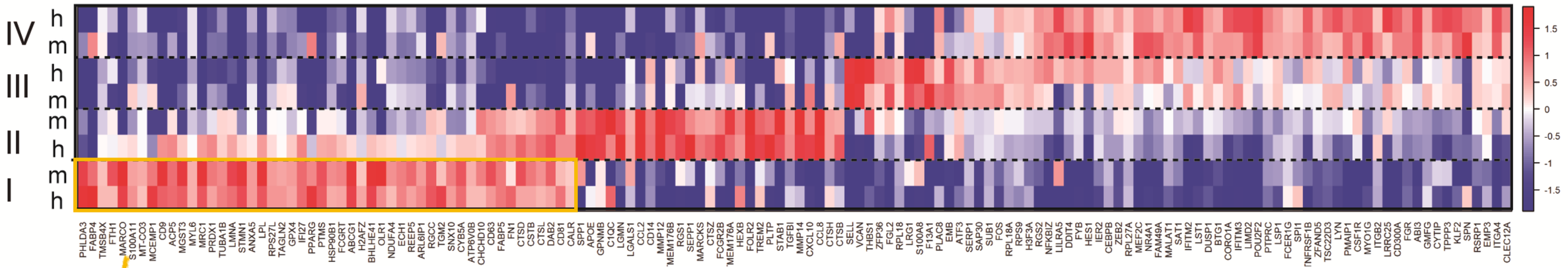




I & II = macrophages

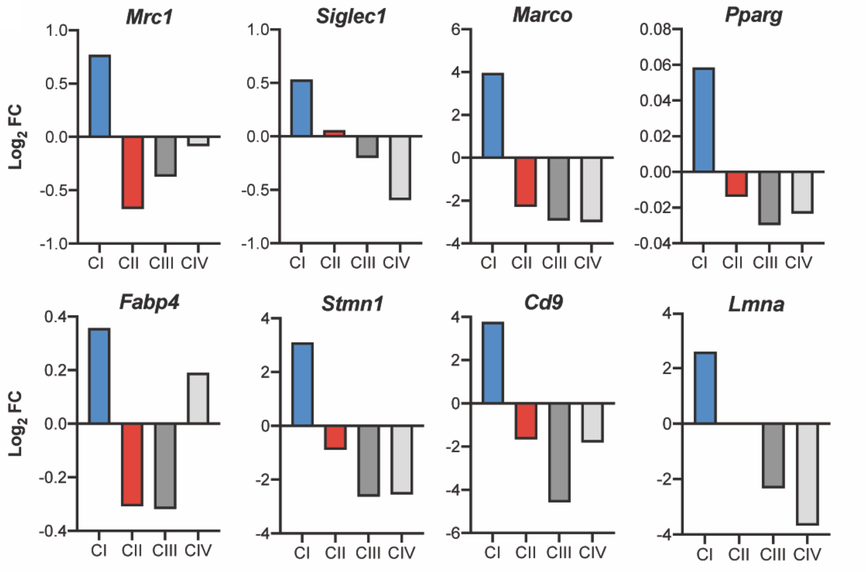
III & IV = monocytes

2 different populations of tumor-associated macrophages are found in NSCLC TME

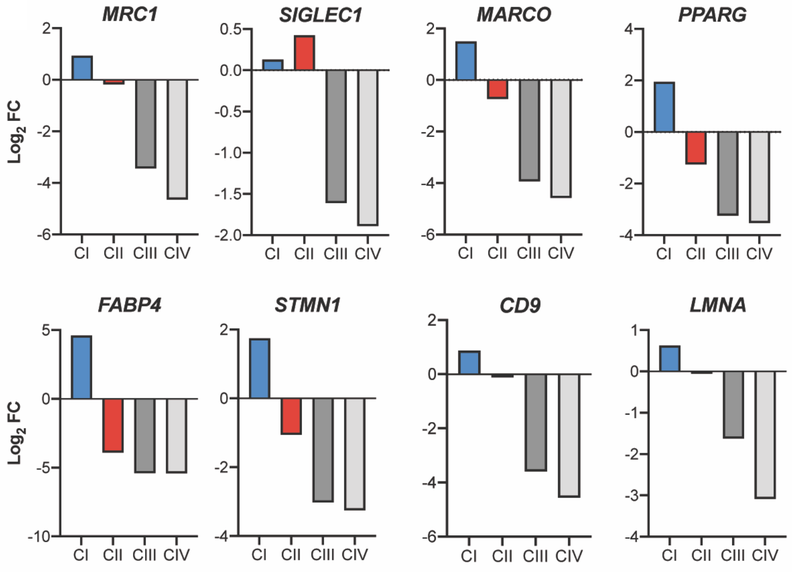
# Modular gene analysis allows the identification of macrophages and monocytes in both species



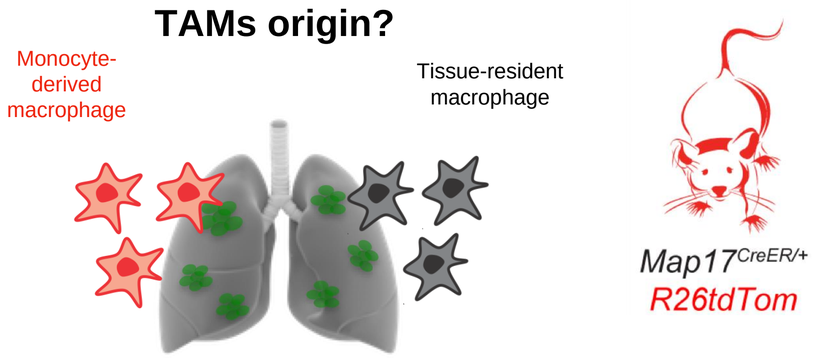

MOUSE




HUMAN

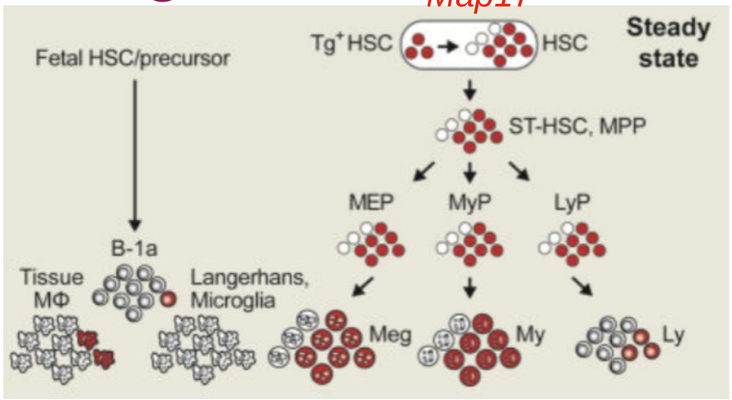


# Fate-mapping of blood-derived immune cells delineates macrophage origin in the lung



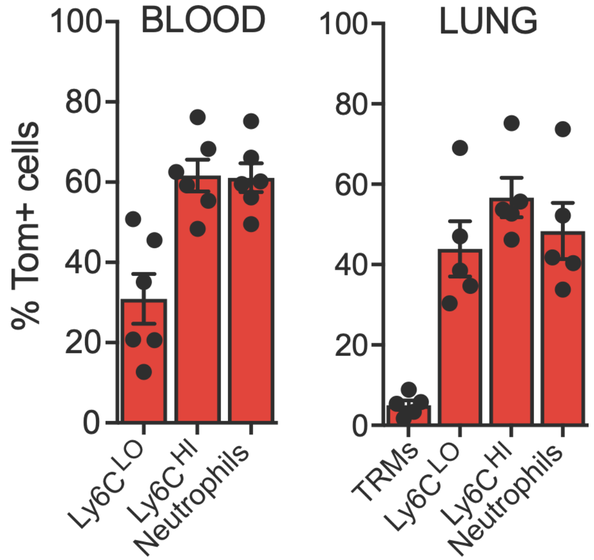


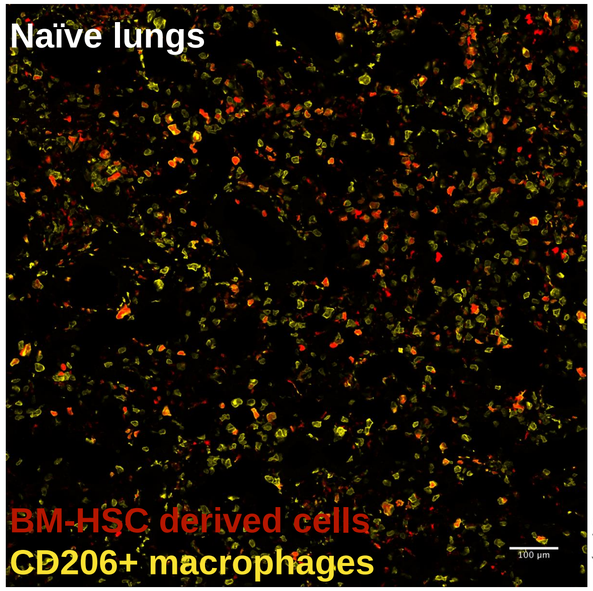





**Only BM-derived progeny** will generate **Td-Tomato+ myeloid cells**

Collaboration with **Boris Reizis, NYU**

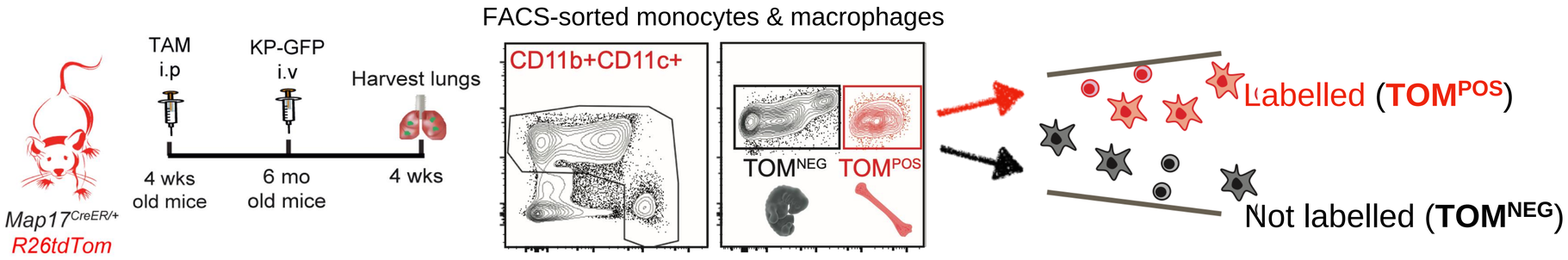


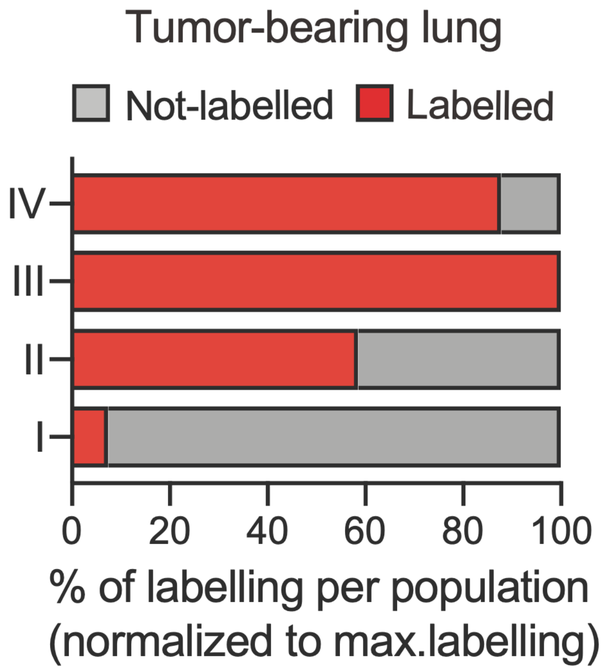



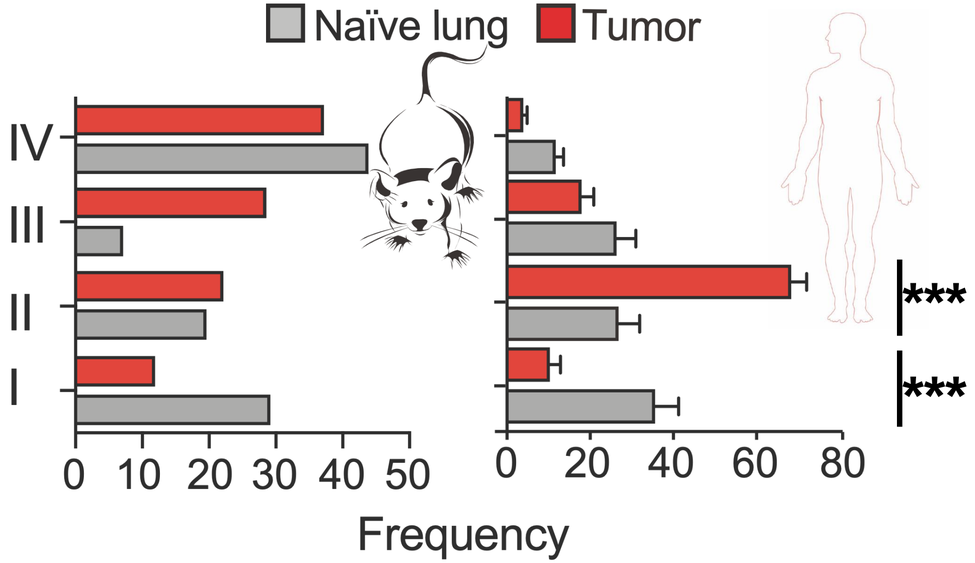

Sawai et al.,, 2016
Yona et al., 2013

Bone marrow derived cells barely contribute to tissue-resident macrophages in naïve lungs

# scRNAseq of lineage-traced adult macrophages revealed 2 ontogenically distinct macrophage populations in NSCLC lesions



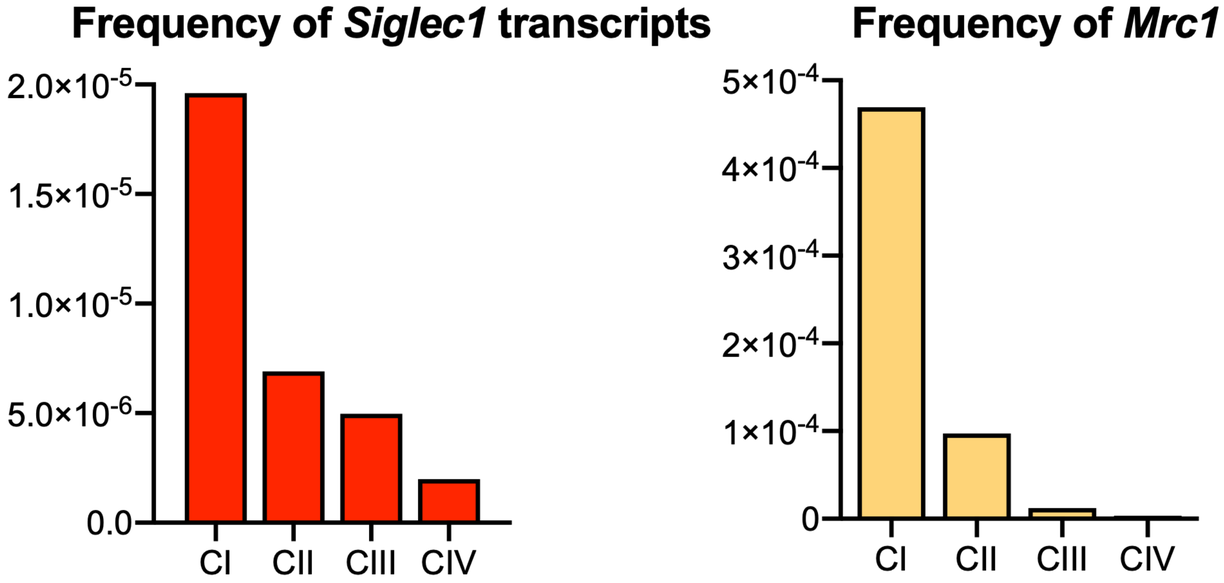




I: TRMs
II: MoMacs
III: Inflammatory monocytes
IV: Patrolling monocytes

# scRNAseq identification of specific markers for macrophage subsets to probe its function in the TME

*Siglec1*=CD169 protein
*Mrc1*=CD206 protein

**CD169$^{Cre\text{-}LSL\text{-}R26tdTomato}$**

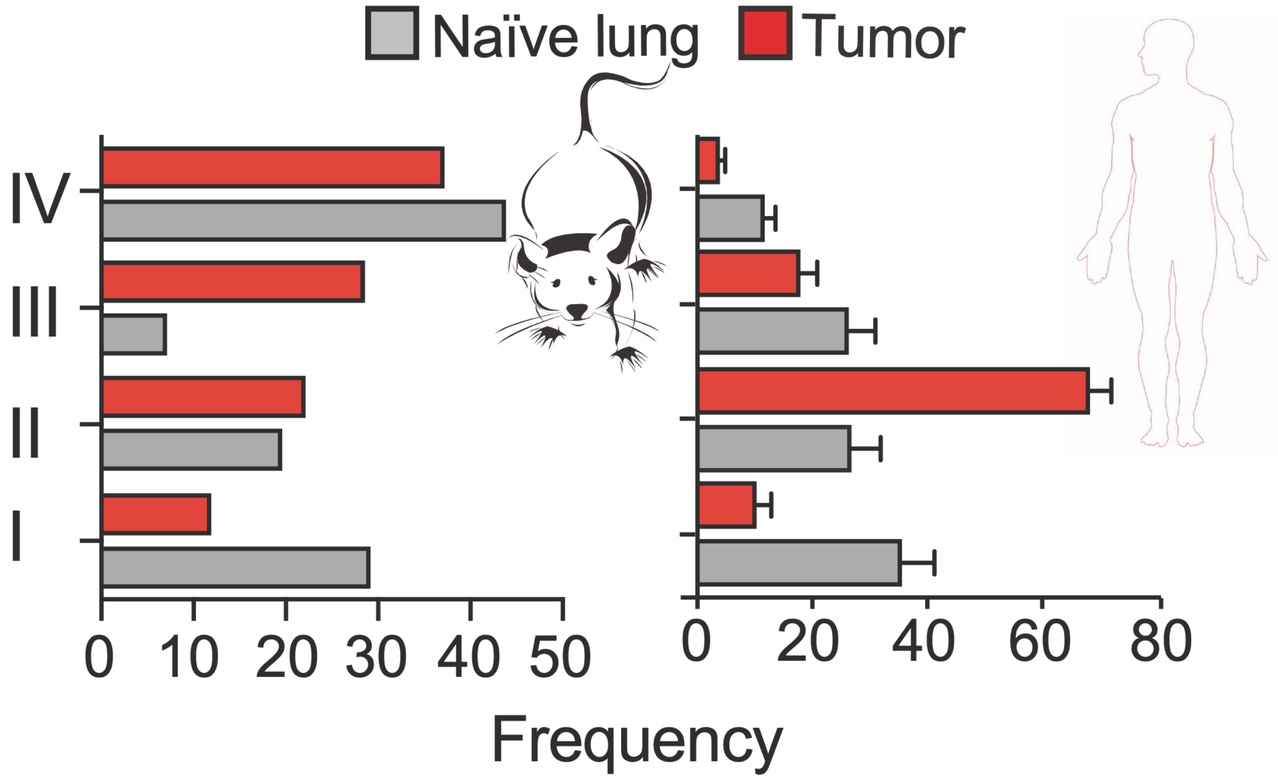




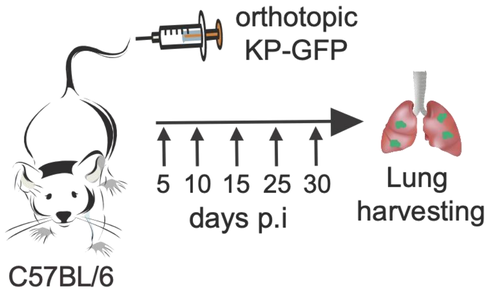


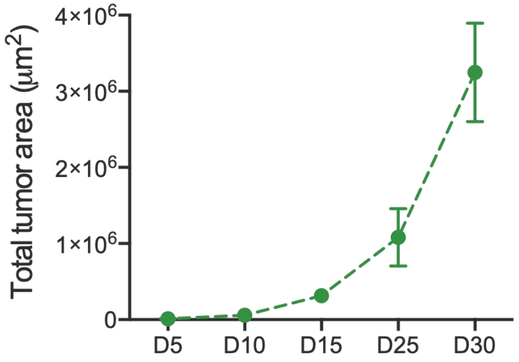

CD169 and CD206 identify TRMs in murine KP lesions

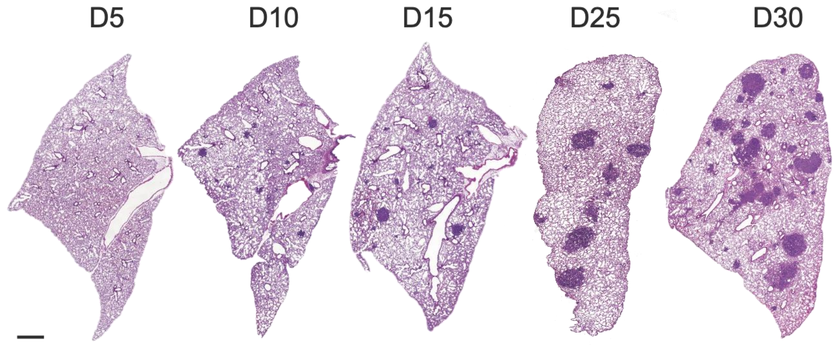
# TRM compartment is reduced in NSCLC lesions



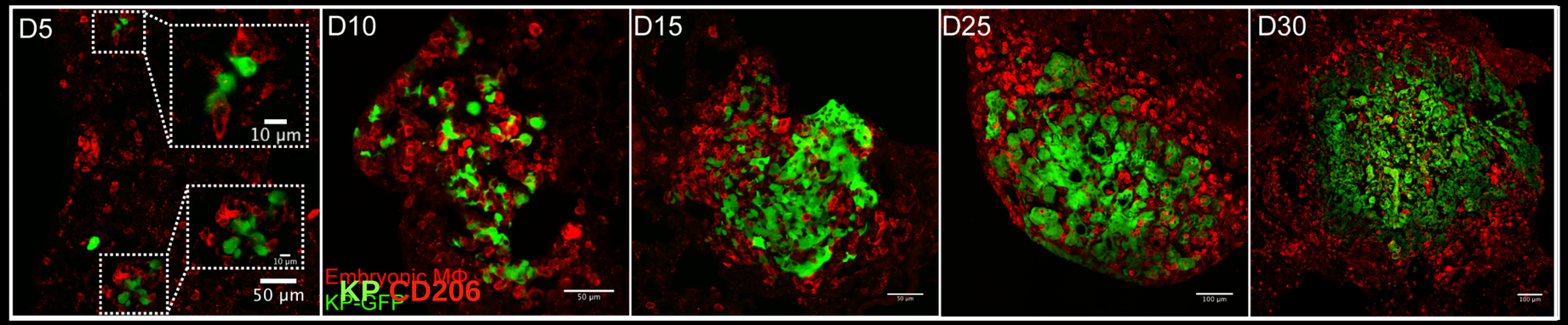
# Early interactions of tumors cells occur with TRMs, which become redistributed at the periphery of the tumors

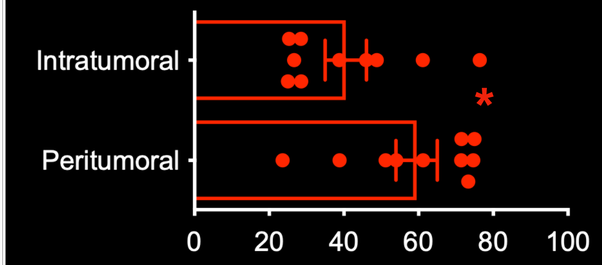


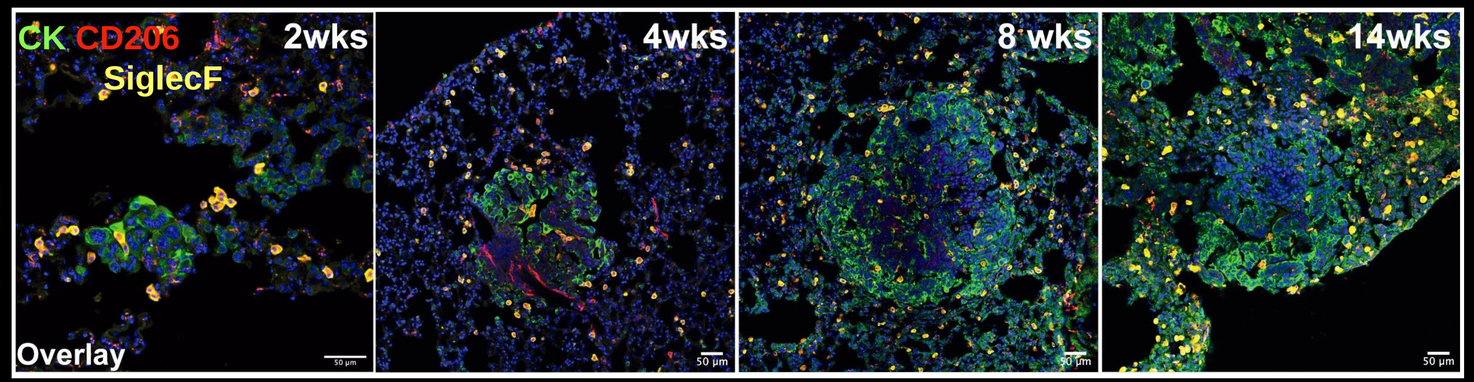

KP orthotopic

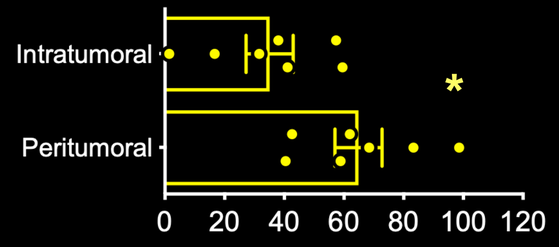



% TRMs in D30 tumors

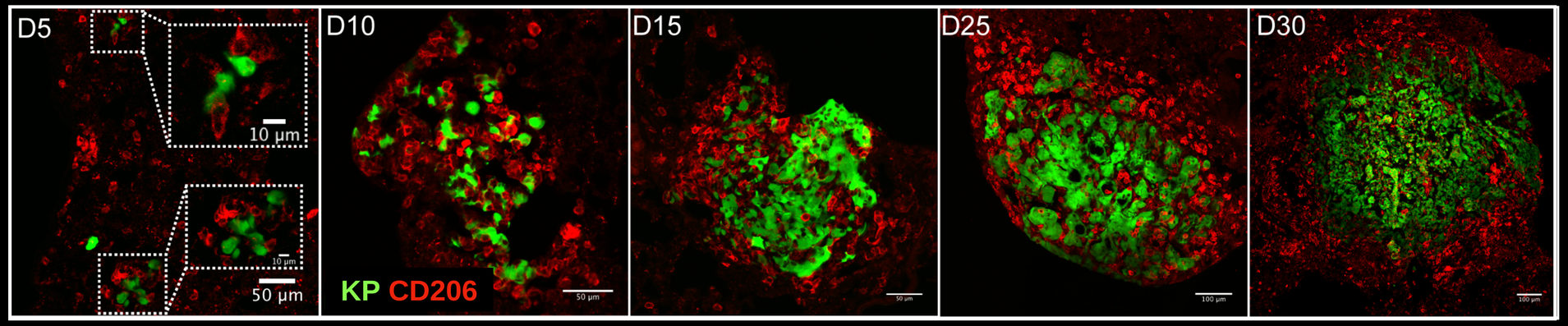
KP GEMM

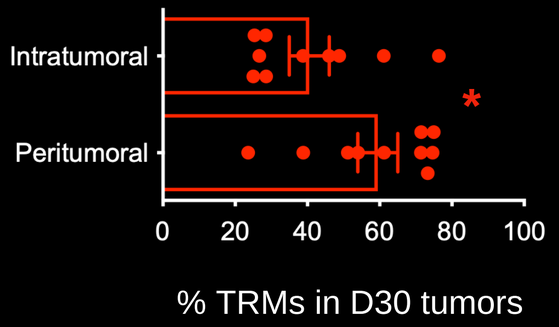


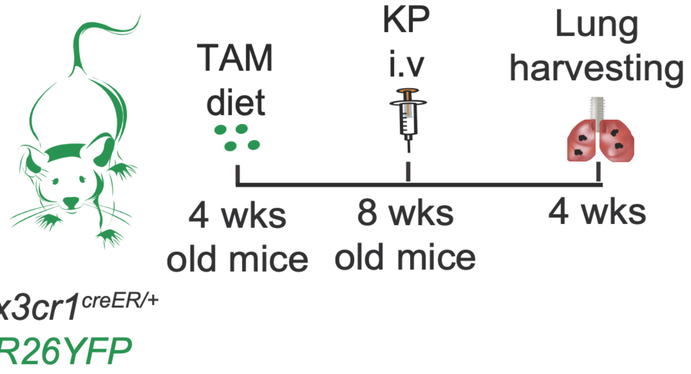



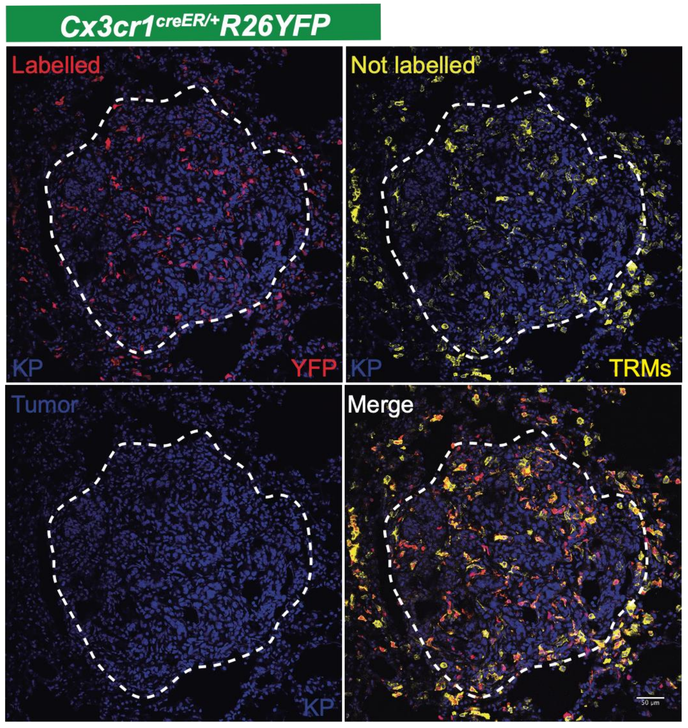

% TRMs in D100 tumors

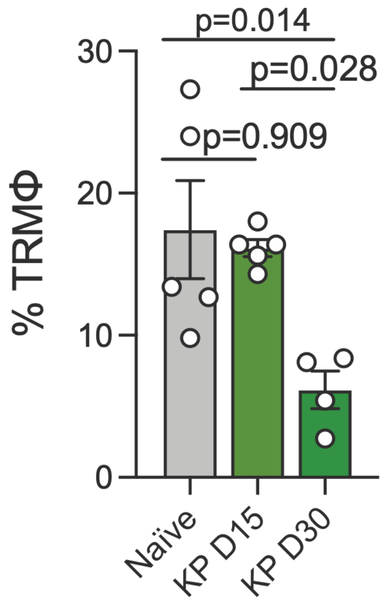
# Macrophage choreography in NSCLC: on time, in place

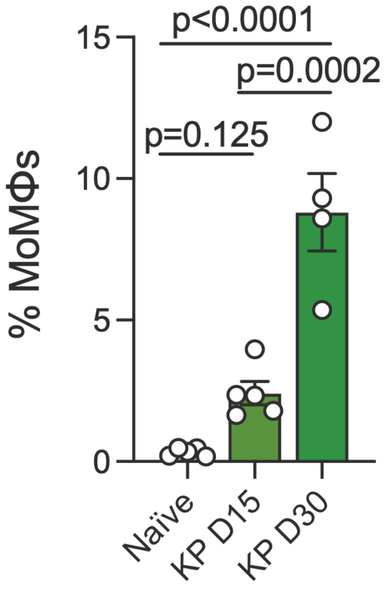


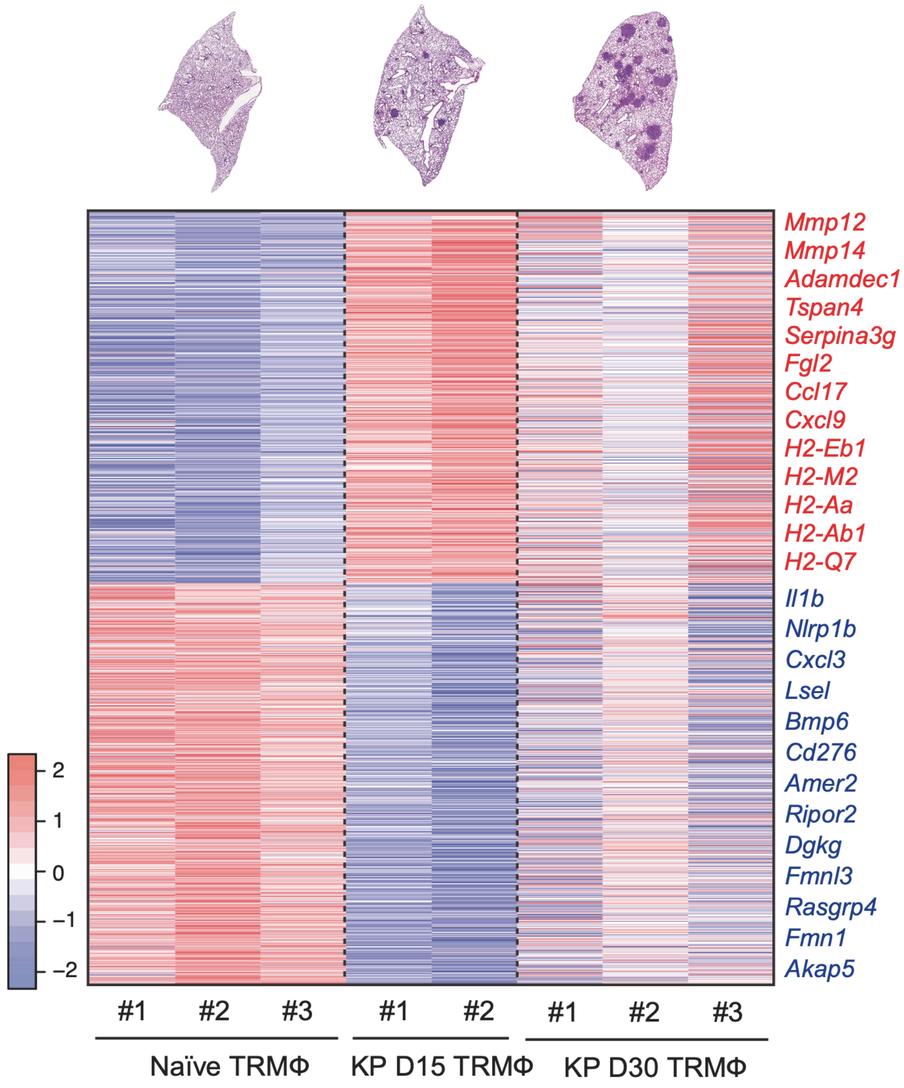



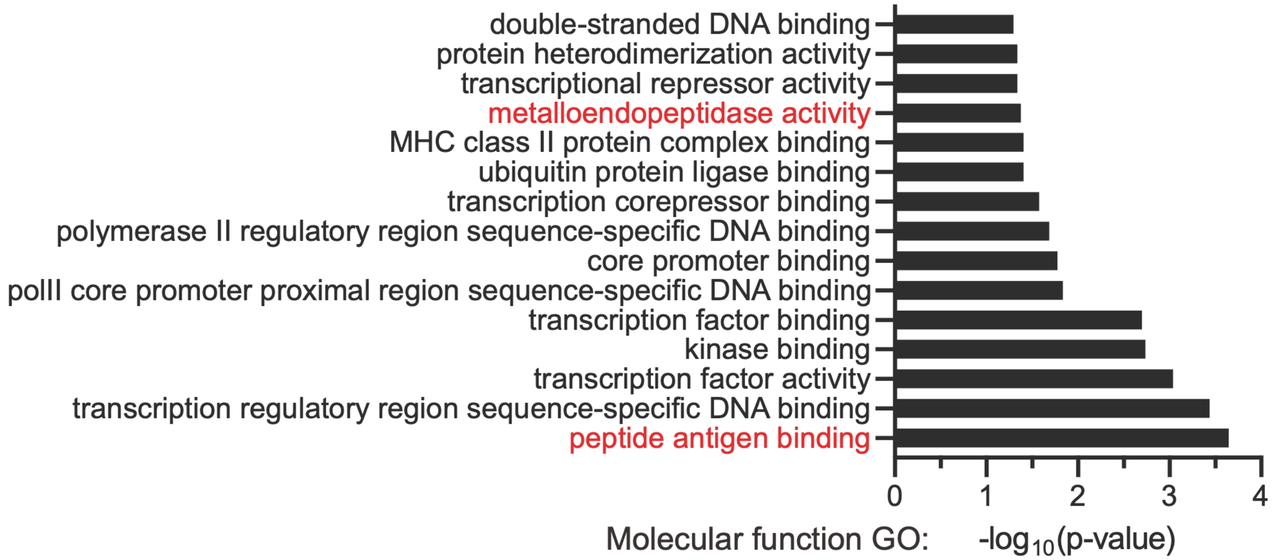



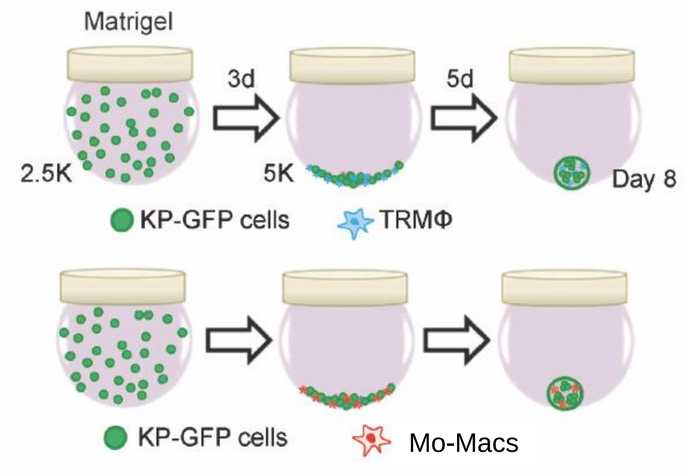
# TRMs acquire a remodeling and antigen presentation program in response to early tumor growth

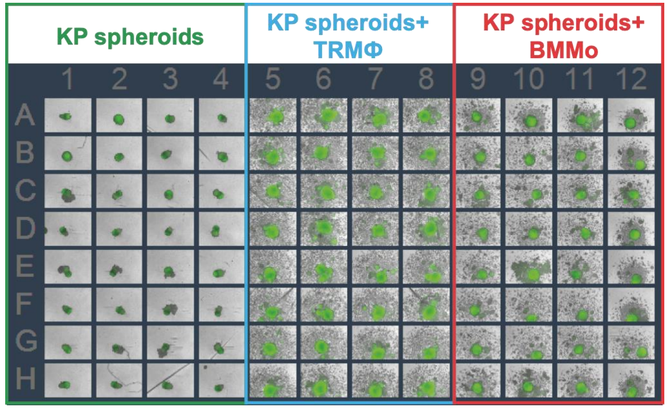


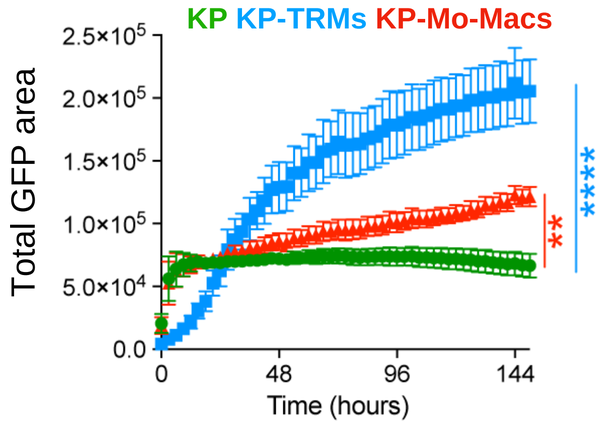

1322 DEGs at early stage



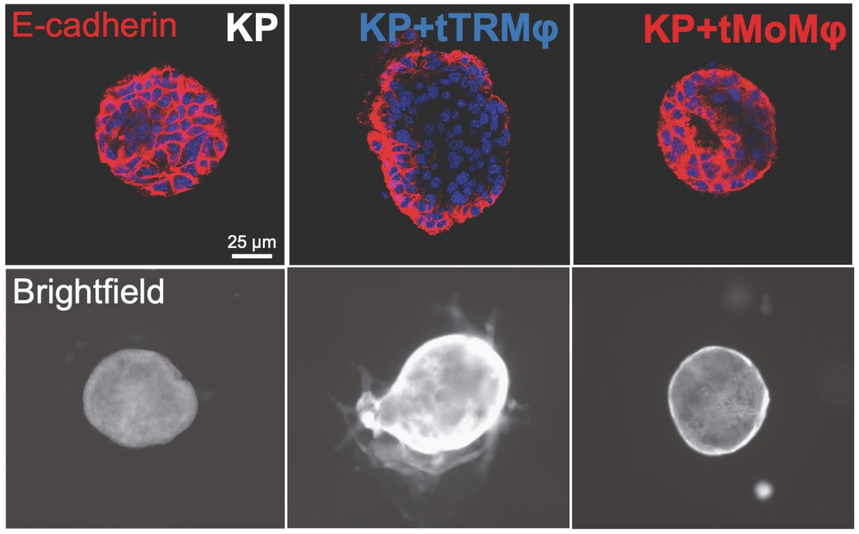
# TRMs promote an EMT phenotype in 3D-spheroids



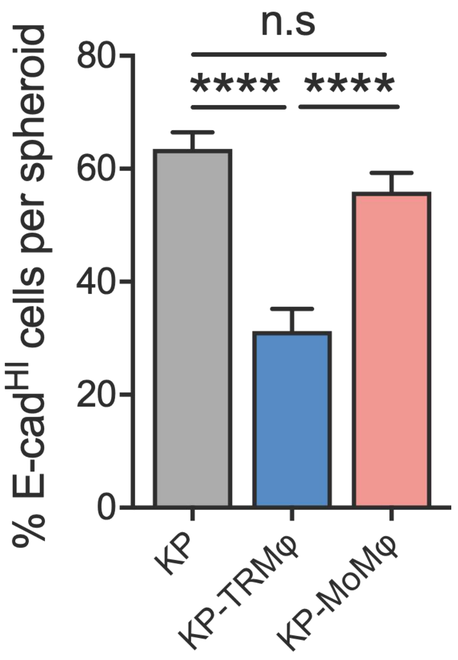



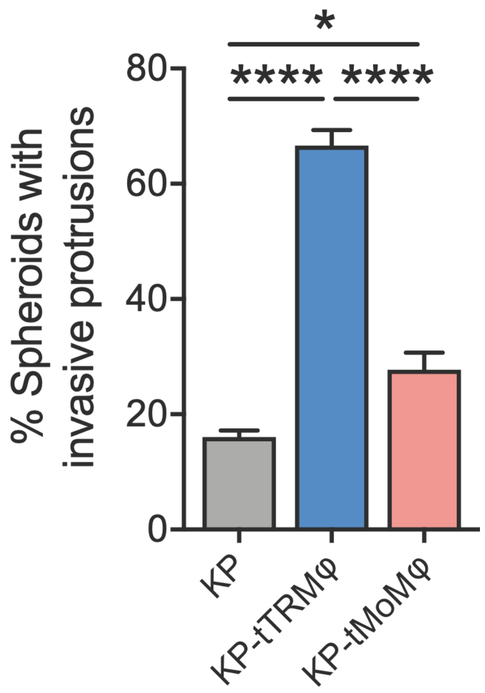




Jovan Nikolic & Philippe Benaroch, Institute Curie



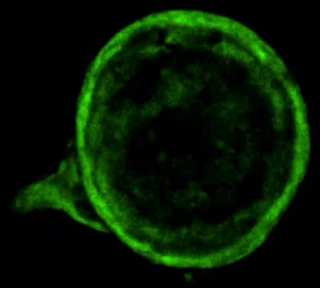

Erica Dalla & Julio Aguirre-Ghiso, MSSM

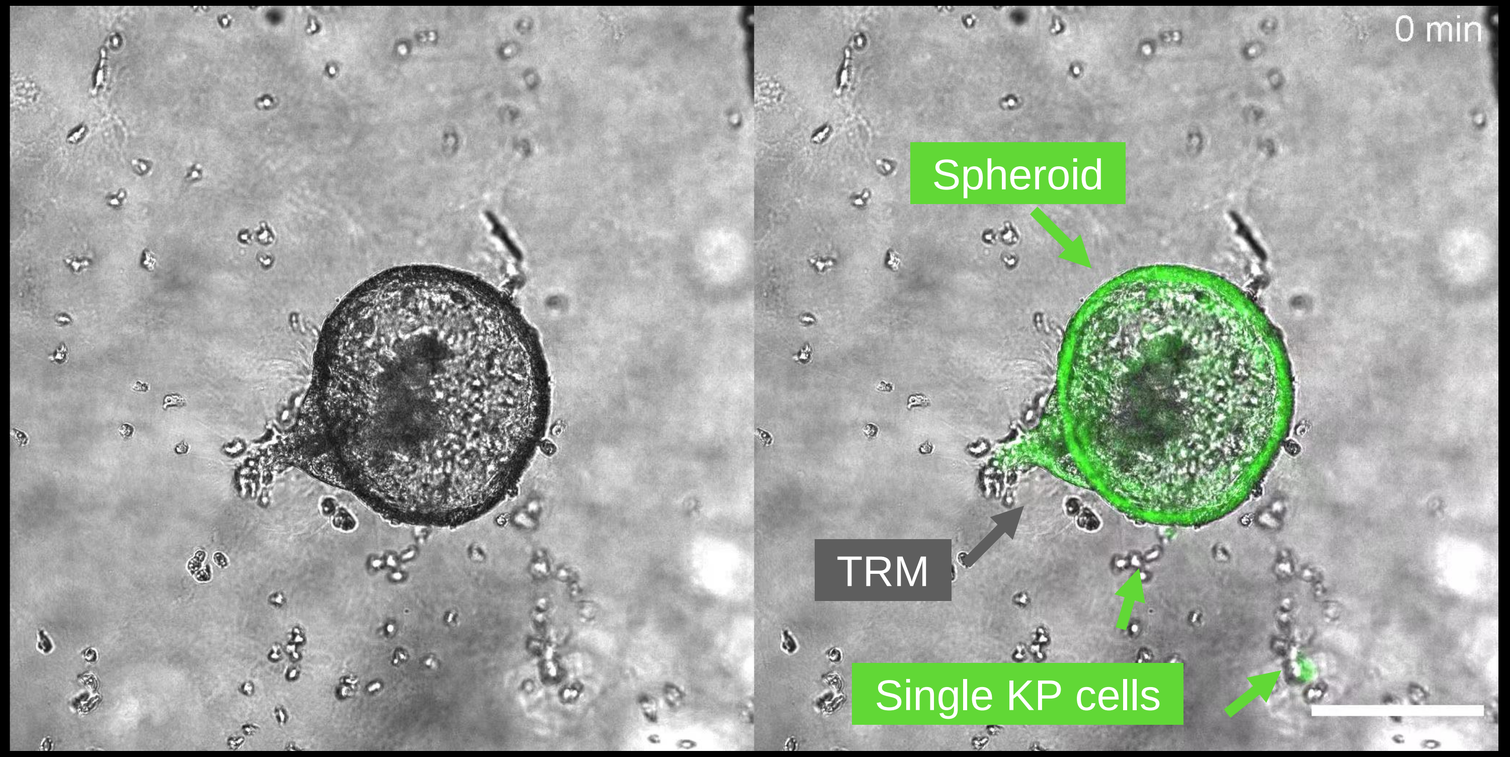


# TRMs promote an EMT phenotype in 3D-spheroids

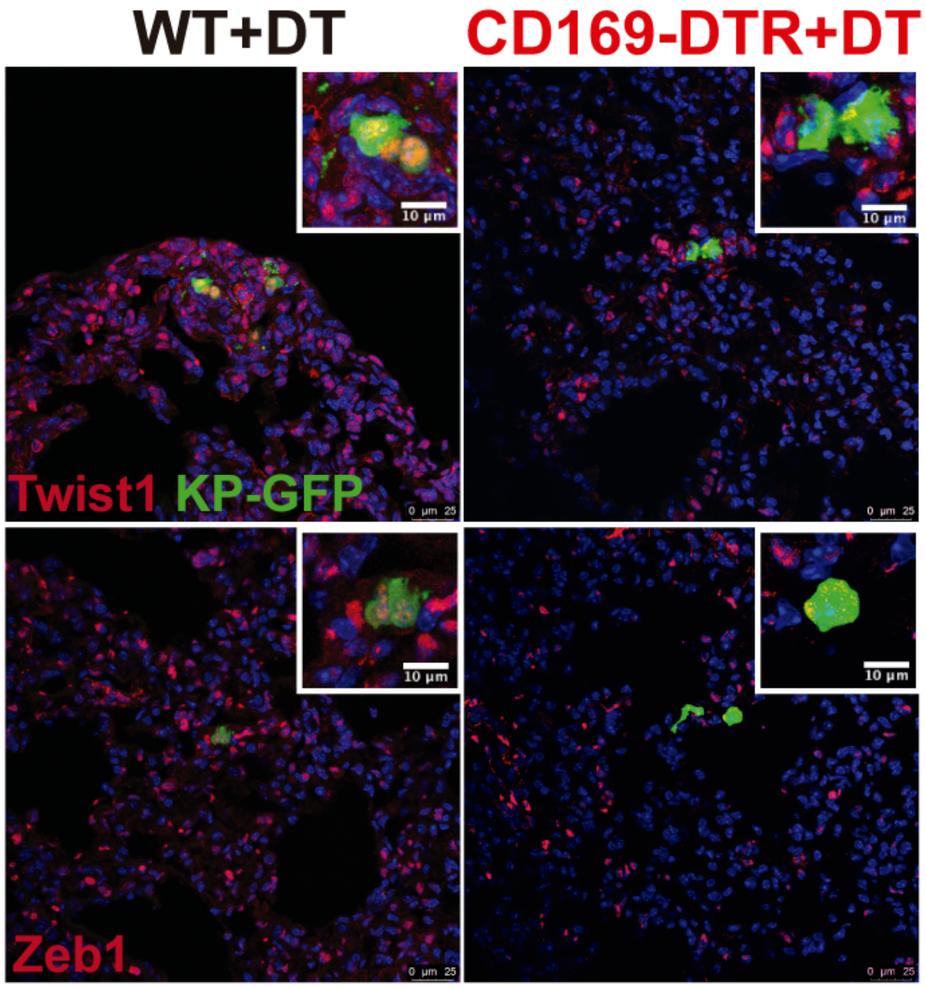
**TRMs-KP spheroid**



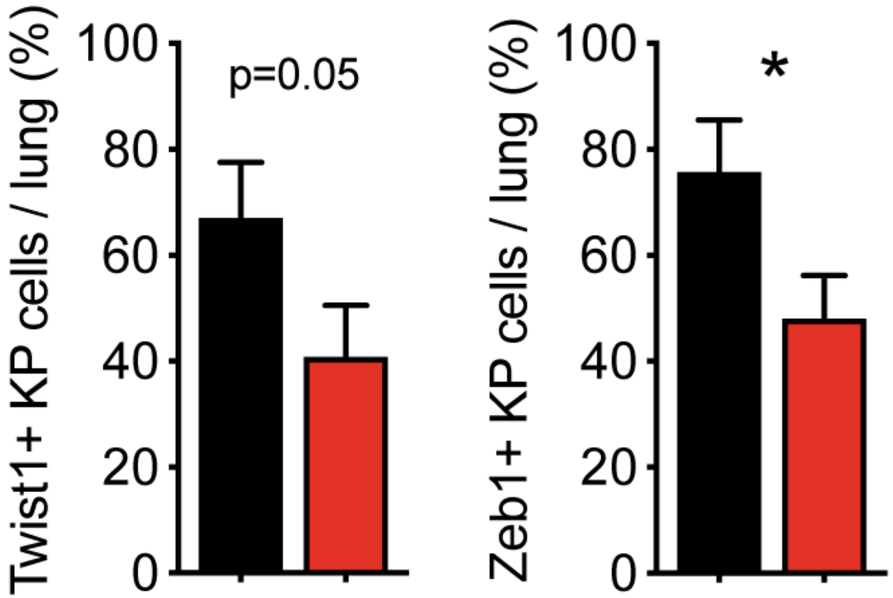


# TRMs promote an EMT phenotype in vivo

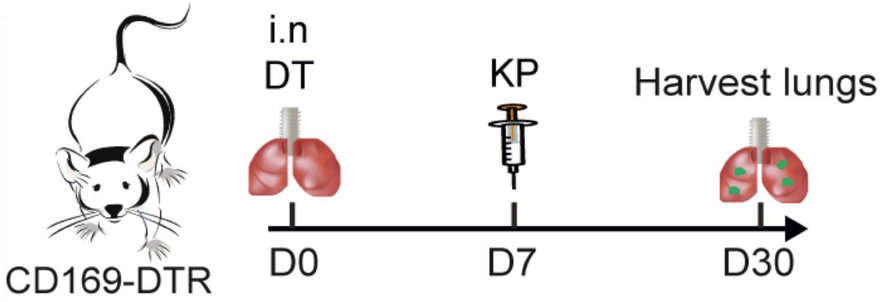
CD169DTR + DT = depletion of tissue-resident macs

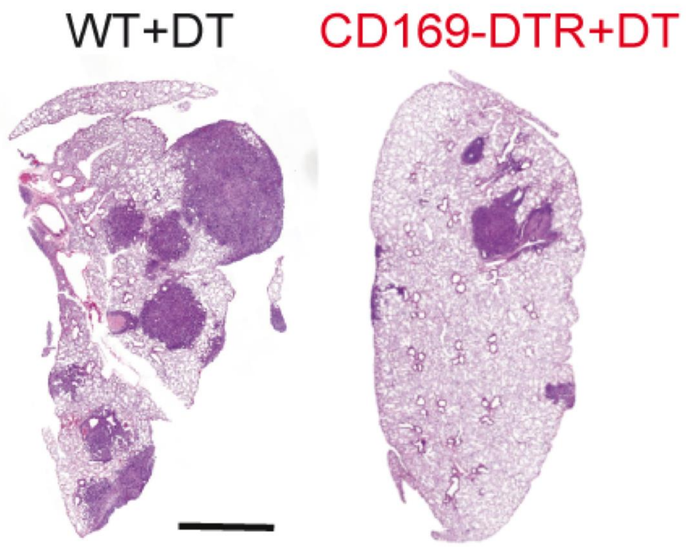


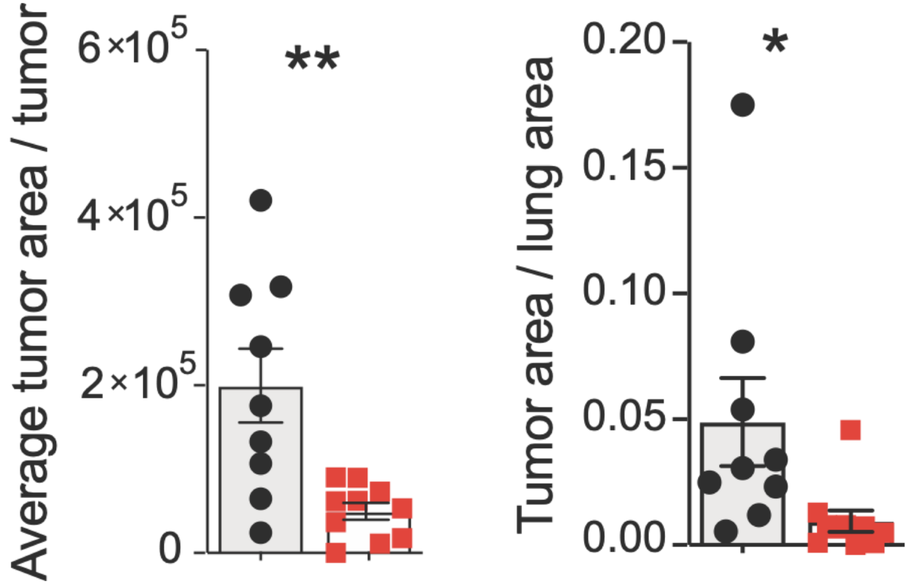

D5 lesions



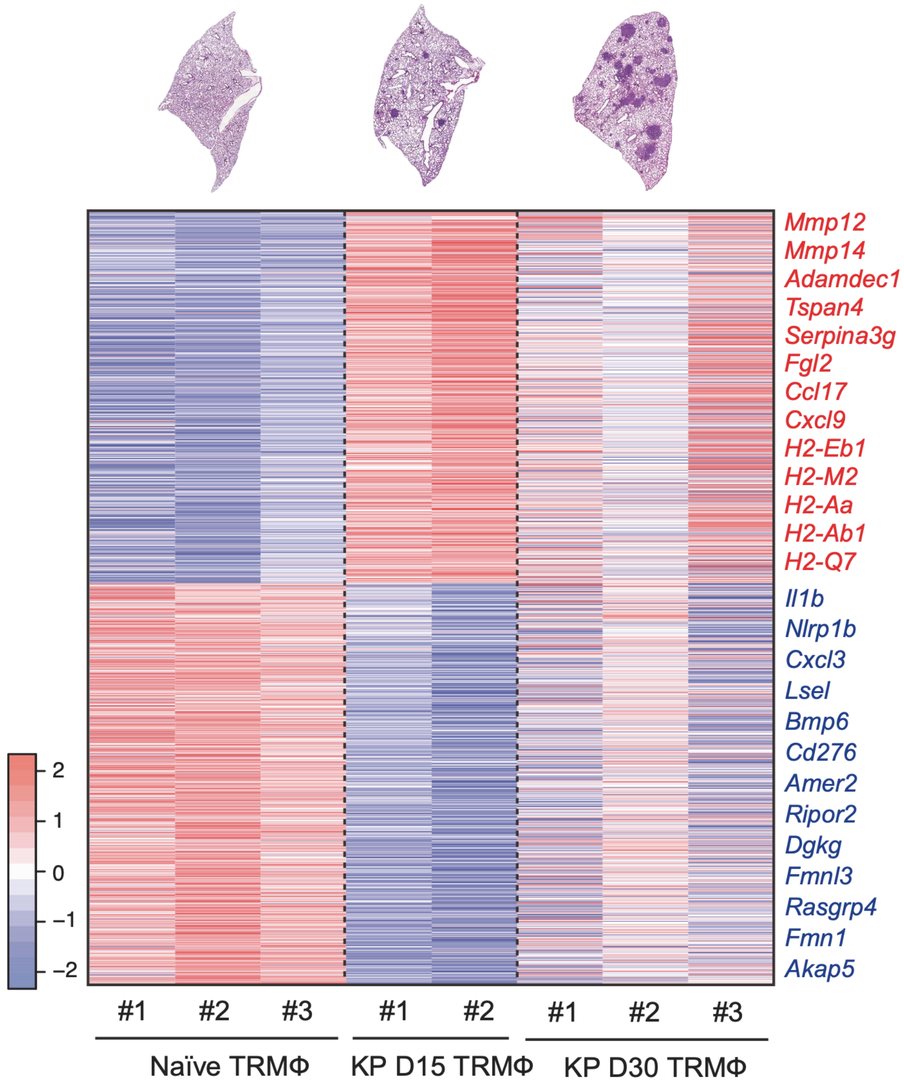
# Depletion of TRMs pre-tumor implantation reduces lung metastasis



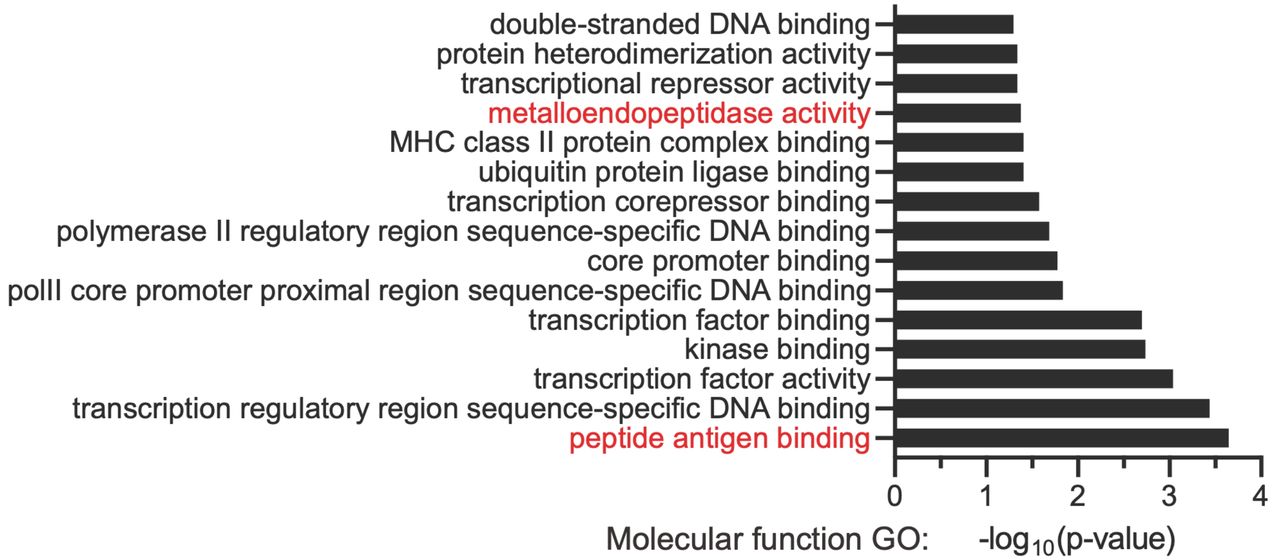



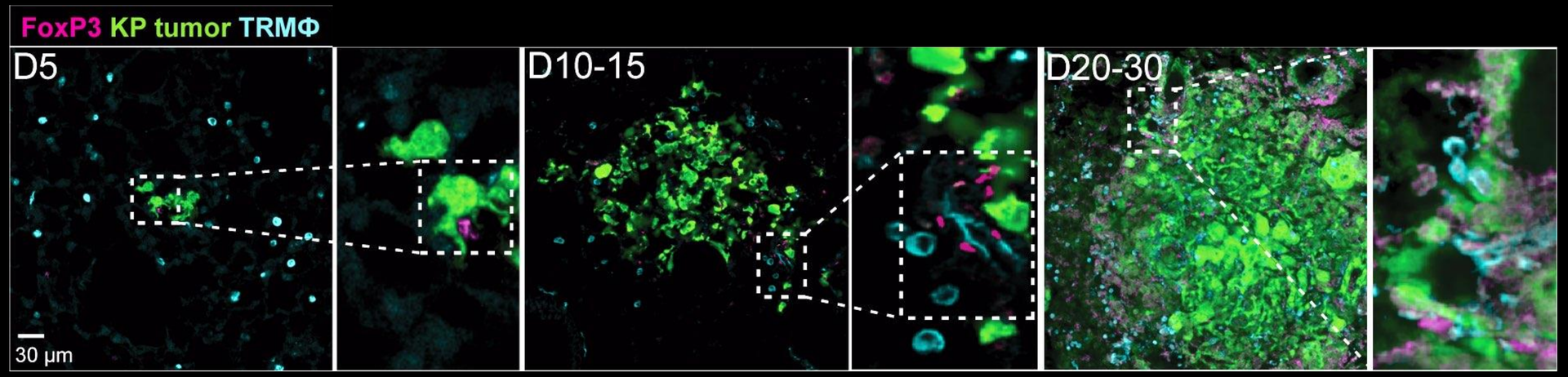
# TRMs acquire a remodeling and antigen presentation program in response to early tumor growth



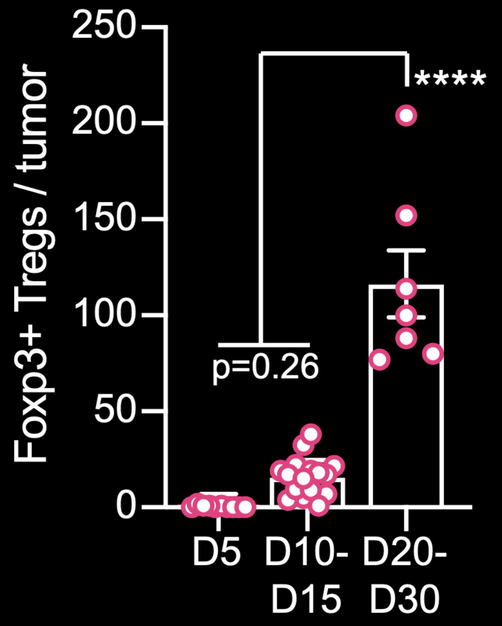

1322 DEGs at early stage

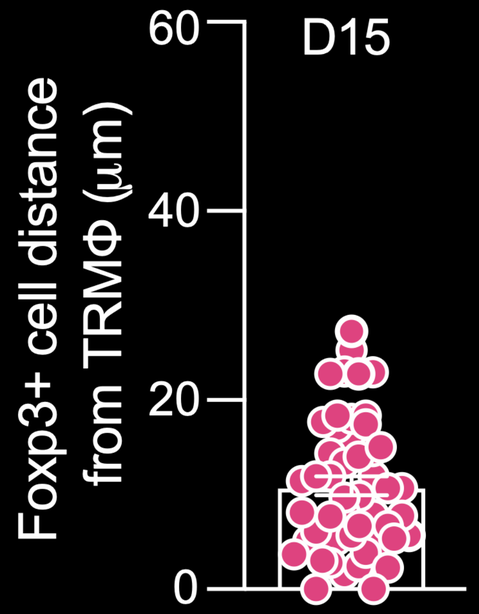


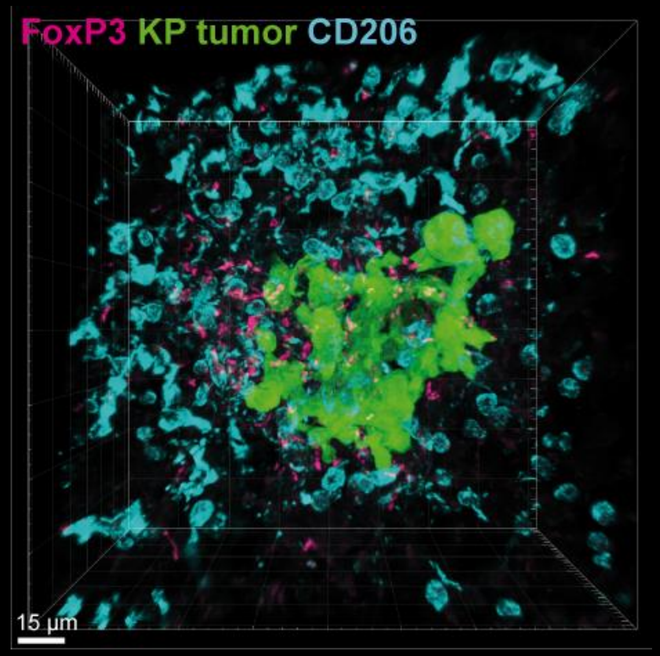
# TRMs create an early immunosuppressive TME that favor tumor progression



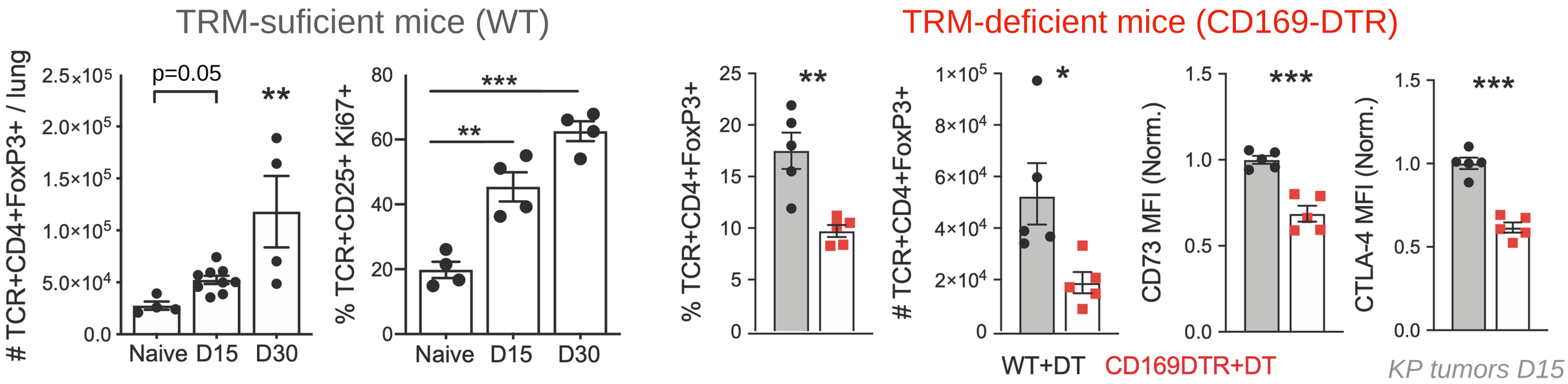

Cleared KP lungs



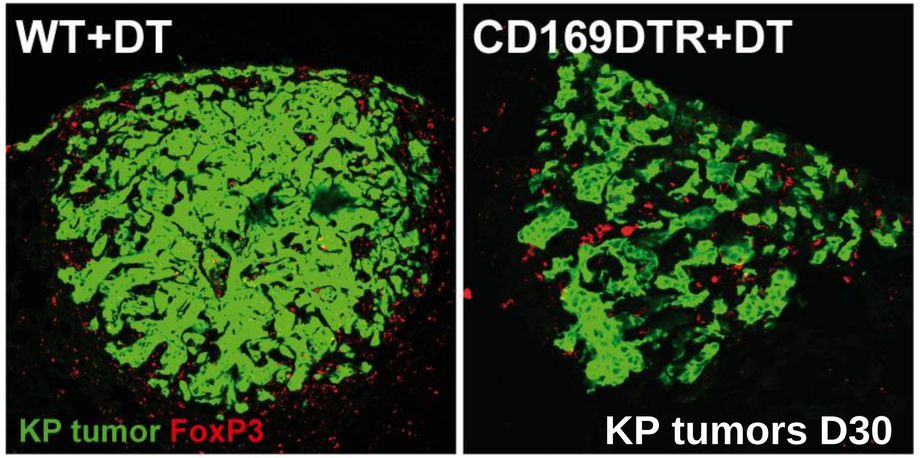



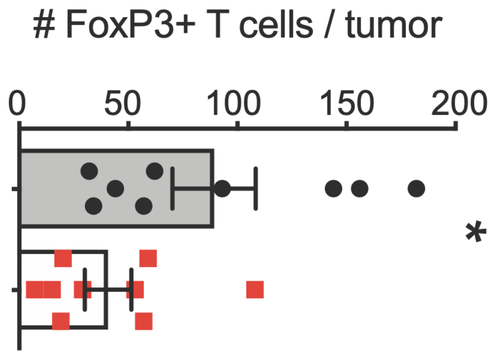
# TRMs create an early immunosuppressive TME that favor tumor progression

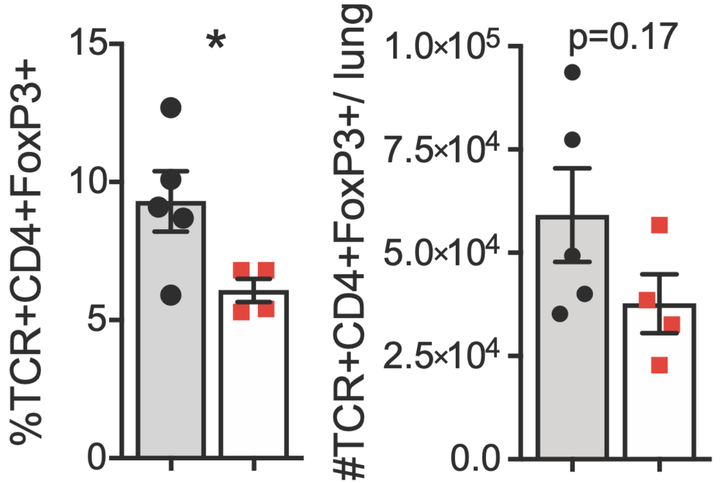


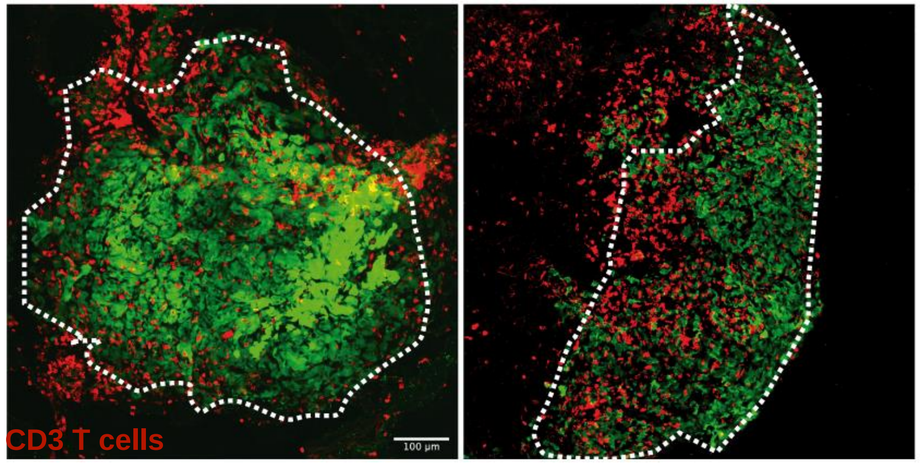
# TRMs create an early immunosuppressive TME that favor tumor progression

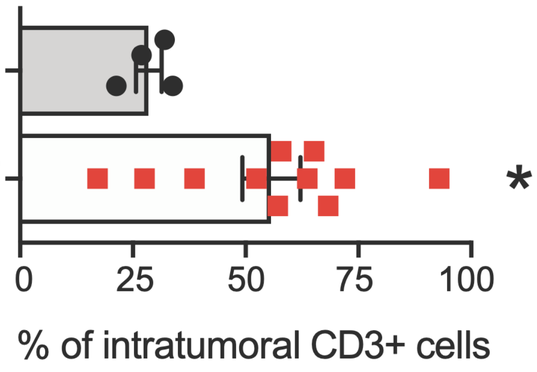
**Depletion PRE-tumor implantation**

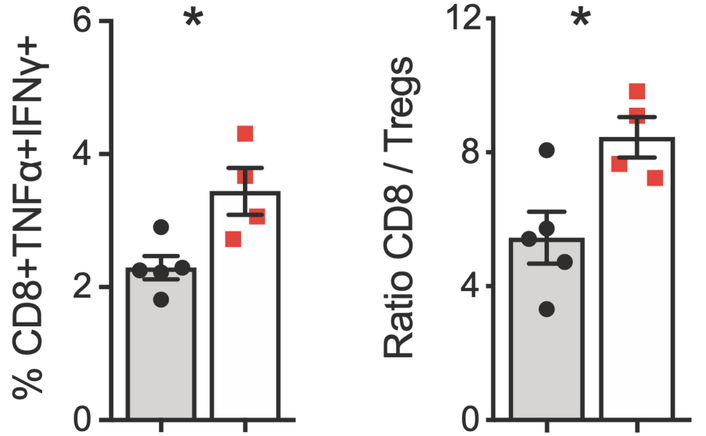


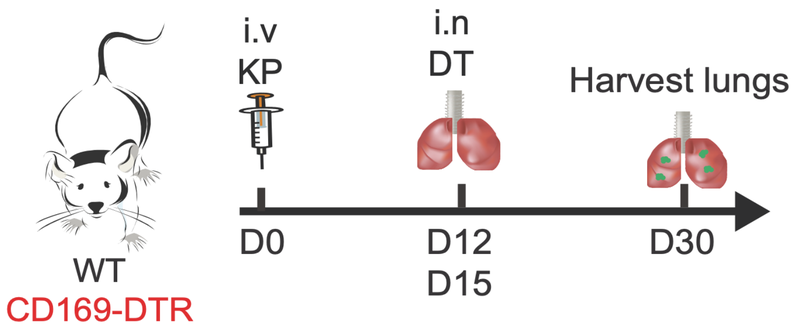



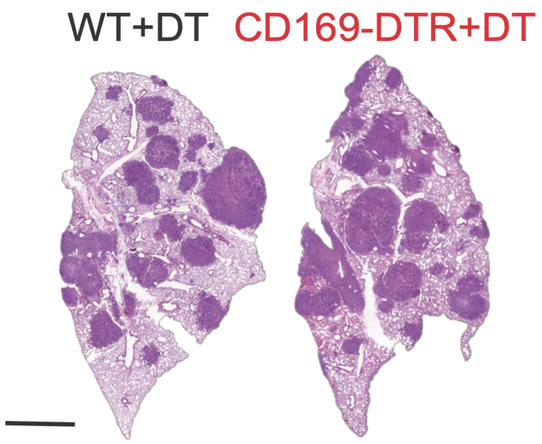



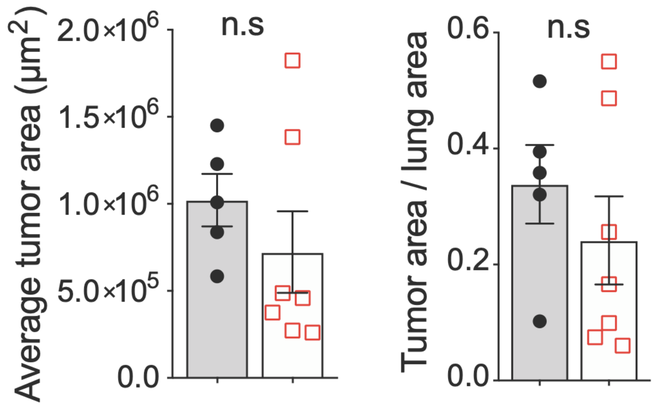



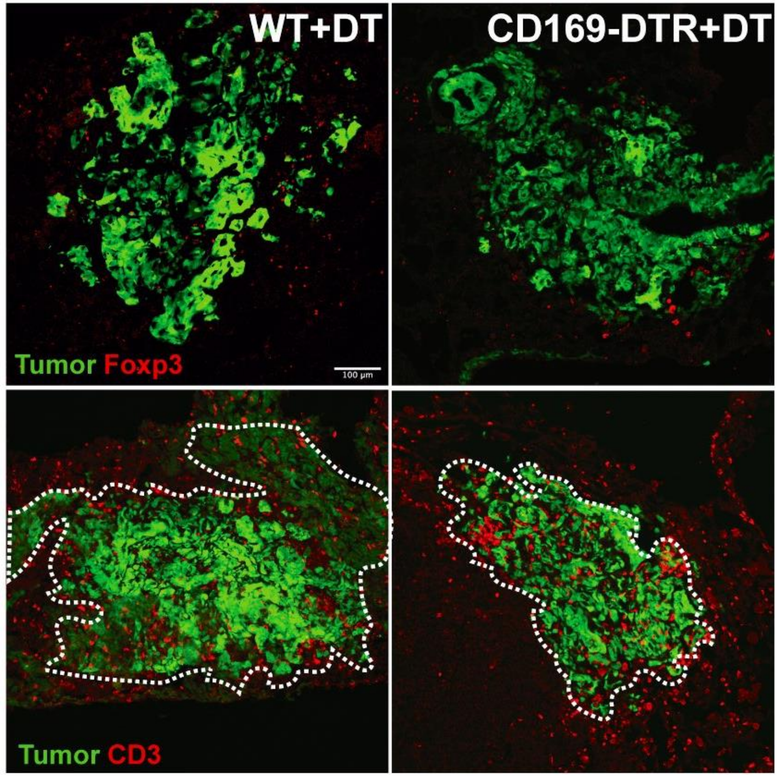
# TRMs create an early immunosuppressive TME that favor tumor progression

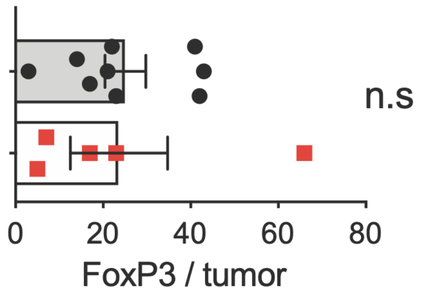
**Depletion POST-tumor implantation**

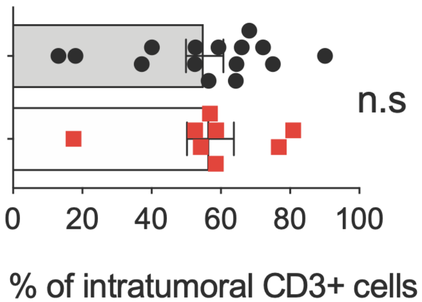


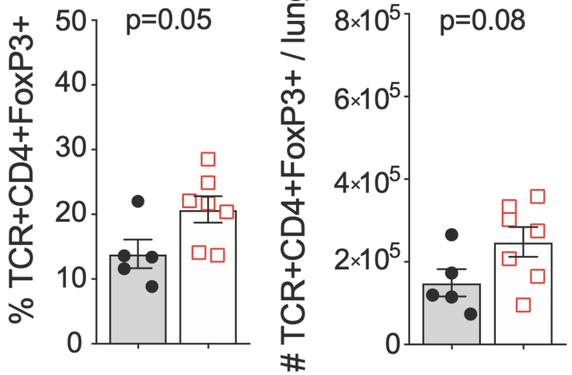



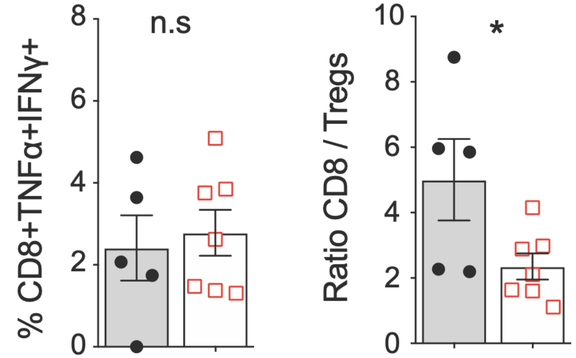



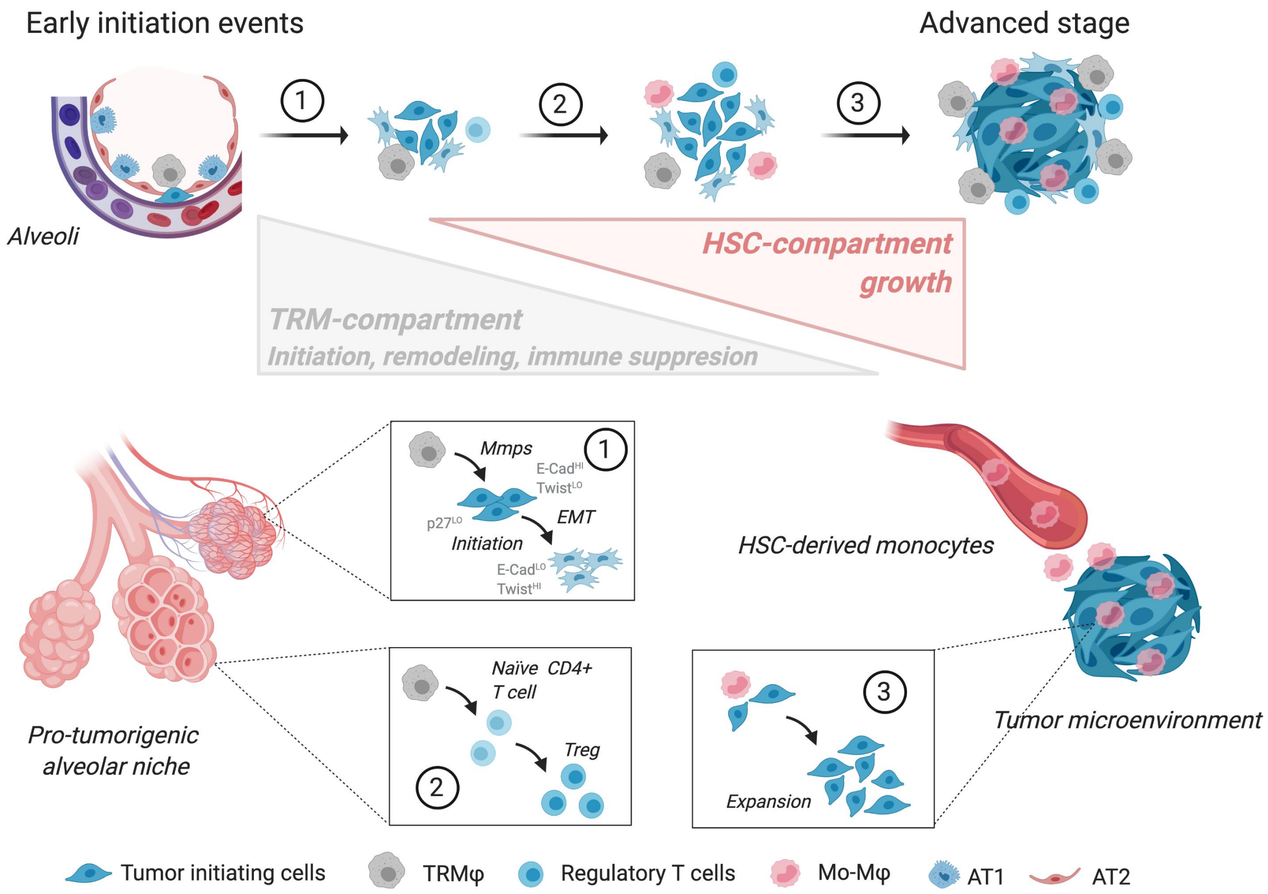



# Take home message




*Nature, June 2021*

TRMs are the first ones to interact with tumoral cells promoting cancer cell invasiveness and early T reg expansion

Different waves of ontogeny distinct macrophages accumulate in tumor lesions

Adult monocytes cannot give rise to tissue-resident macrophages even when recruited to tissues, importance of understanding the biology of TRMs (on a tissue-specific manner)

Our results establish that TRM are mostly relevant during tumor inception

Clinical relevance: early intervention activating the TRM compartment in order to reduce their tolerogenic poten

# Acknowledgements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Catherine Sawai & Boris Reizis, NYU
Jovan Nikolic & Philippe Benaroch,
Institut Curie (France)
Christine Moussion (Genentech)

Flow Cytometry & Microscopy Cores
Human Immune Monitoring Core



AACR American Association for Cancer Research

HFSP HUMAN FRONTIER SCIENCE PROGRAM
FUNDING FRONTIER RESEARCH INTO COMPLEX BIOLOGICAL SYSTEMS

Mount Sinai MSSM MOUNT SINAI SCHOOL OF MEDICINE

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