In The Name of GOD

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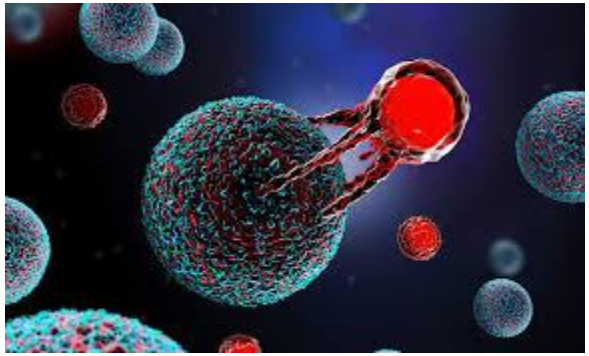


Trogocytosis

Outline of This Presentation

A. Trogocytosis
B. History of Trogocytosis
C. Mechanism of Trogocytosis
D. Trogocytosis-Mediated Signaling
E. Detection of trogocytosis

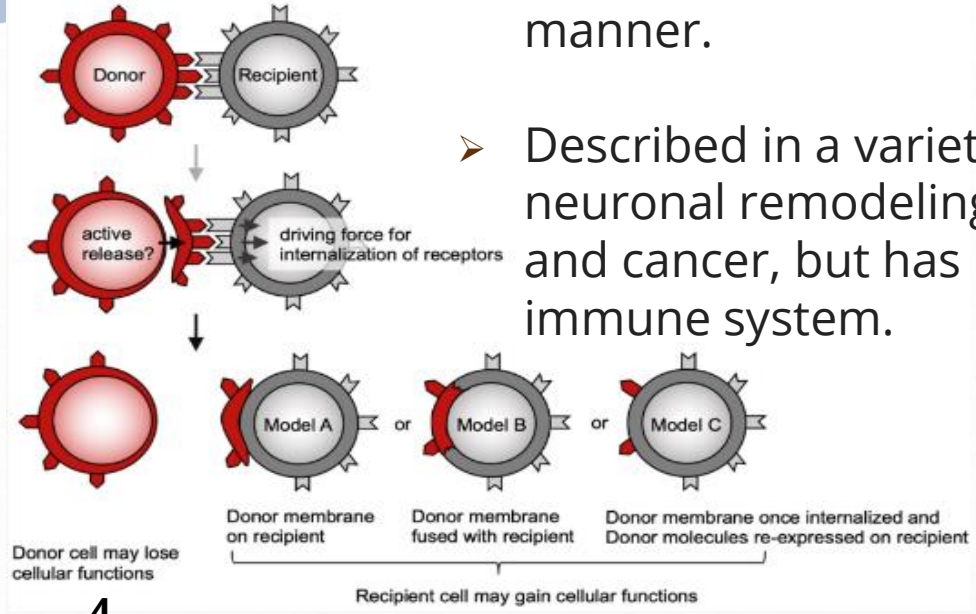
cells

01

What is Trogocytosis?!

Trogocytosis

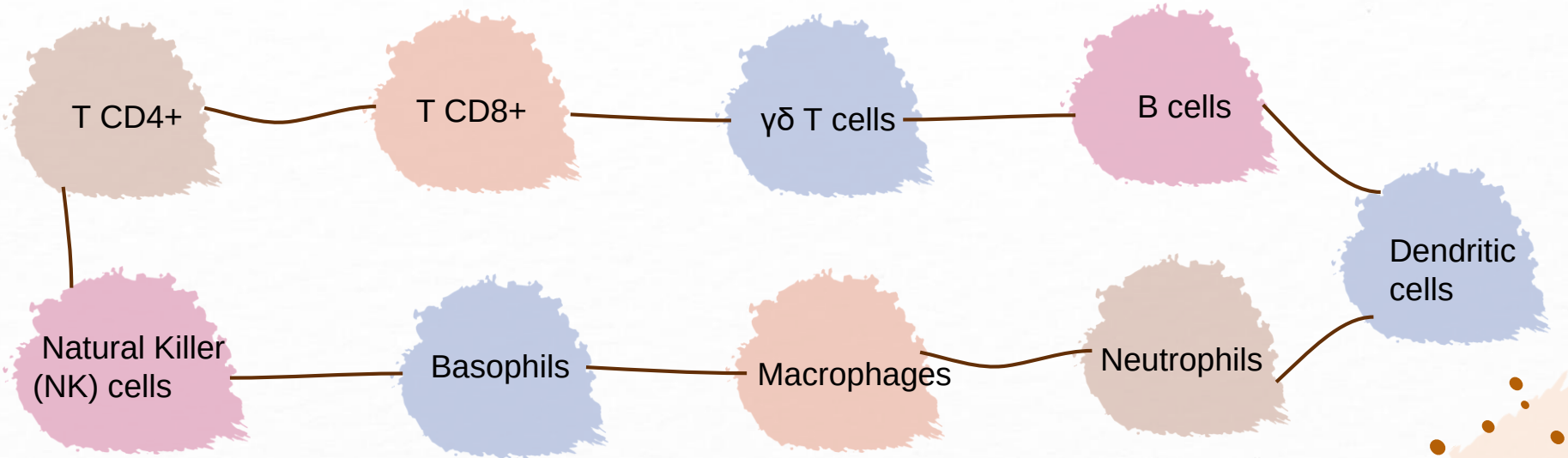
- The **direct, intercellular transfer of membrane and membrane-associated molecules** in a contact-dependent manner.
- Described in a variety of biological settings including neuronal remodeling, fertilization, viral and bacterial spread, and cancer, but has been most widely studied in cells of the immune system.



doi: 10.3390/cells10051155 (2021)

Most studies on trogocytosis have focused on interactions of immune cells.

Trogocytosis has been observed in:



Doi: 10.3390/cells10061478 (2021)

Not limited to immune cells

- **Microglial Presynaptic Remodeling**
- Trogocytosis Of **Oocyte Proteins** By **Sperm** Before Fertilization
- During Embryonic Development
- **Stromal Cell Protein** Trogocytosis By **Cancer Cells**
- **Erythrocyte** Interactions With **Epithelial cells**
- **Parasite** Interactions With **Neutrophils**

- **Cell–cell Spread** Of Intracellular Bacteria Such As **Francisella Tularensis** and **Viral Pathogens**.

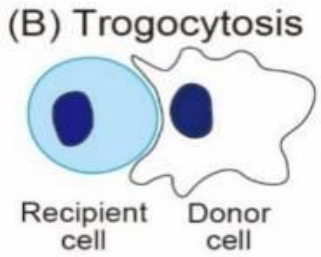
Doi: 10.3390/cells10061478 (2021)

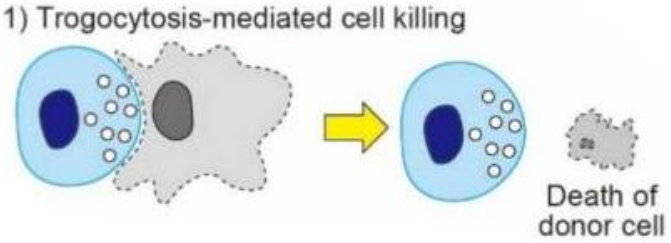
Characteristics of trogocytosis

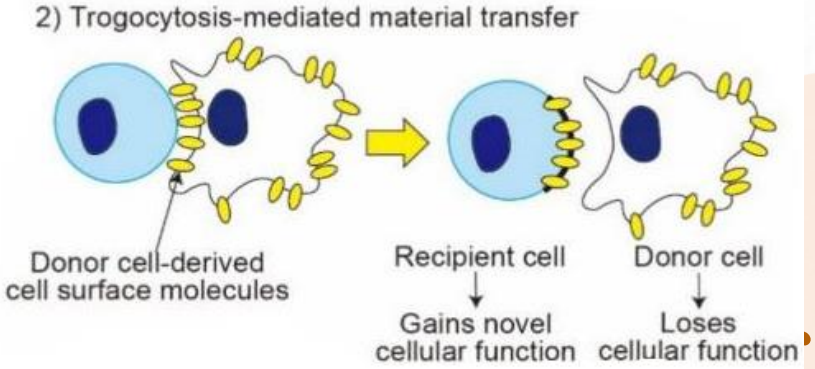
Trogocytosis

1 trogocytosis-mediated cell death

2 trogocytosis-mediated material transfer







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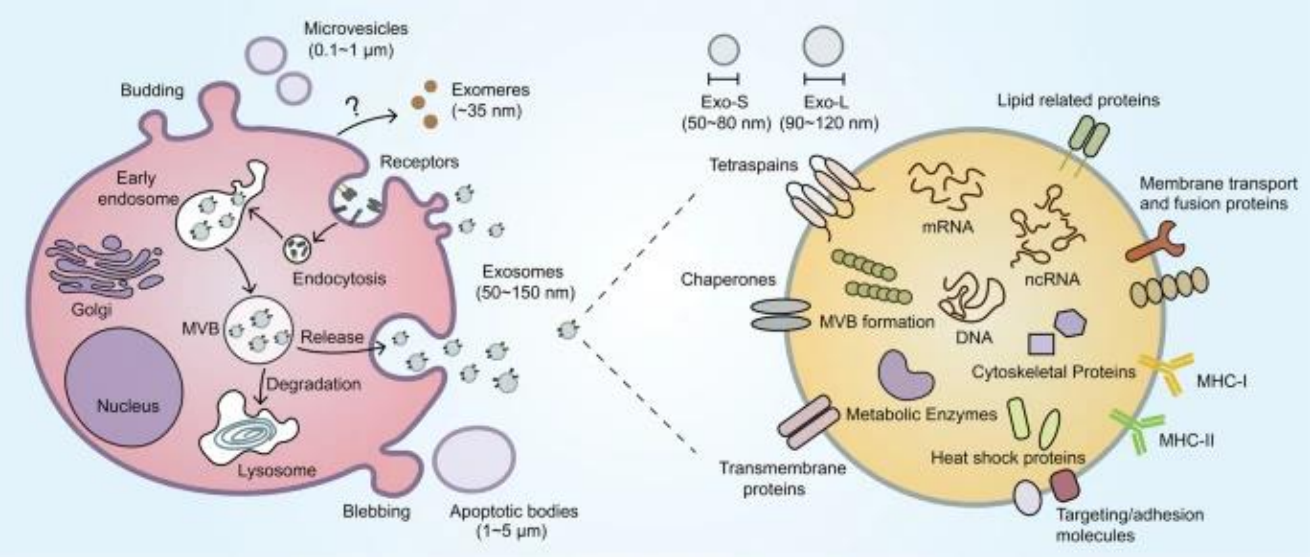
Exosomes vs. Trogocytosis



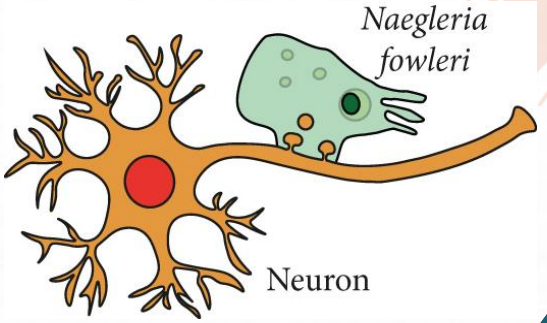
https://doi.org/10.1038/s41423-020-0391-1

02

What is The Brief History of Trogocytosis?!

Brief history of Trogocytosis

- **Brown and colleagues** described the cytopathogenicity of the parasitic amoeba ***naegleria fowleri***



- **Joly and Hudrisier** to describe a **comprehensive model of intercellular transfer** of membrane and membrane molecules between **immune cells.**

Doi: 10.3390/cells10061478 (2021)

Brief history of Trogocytosis

- In 1972, **Cone et al**. detected **allogeneic MHC II** molecules on adoptively transferred **T cells** in mice.
- **Murine T cells do not endogenously express MHC II**.
- In 1973, **Bona et al**. published the first report of **Ag transfer from APCs to lymphocytes.**
- In that paper, they found that **B cells** acquire **LPS from macrophages** in a **B cell receptor-dependent** manner.

Doi: 10.3390/cells10061478 (2021)

Brief history of Trogocytosis

- **Hudson** and **Sprent**, who reported that, **after adoptive transfer** of activated **T cells into allogeneic hosts**, the transferred T cells had **detectable levels of B-cell-derived IgM** on their surface.

- Subsequent reports described the **transfer of MHC:peptide to T cells from APC** both in vitro and in vivo.

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03

What is The Mechanism of Trogocytosis?!

The Current Model of Trogocytosis

- Trogocytosis by CD4+ T cells (and by extension CD8+ T cells and NK cells) is an RRas2- and RhoG-dependent.
- Trogocytosis by RhoG--/-- CD4+ T cells is significantly reduced.

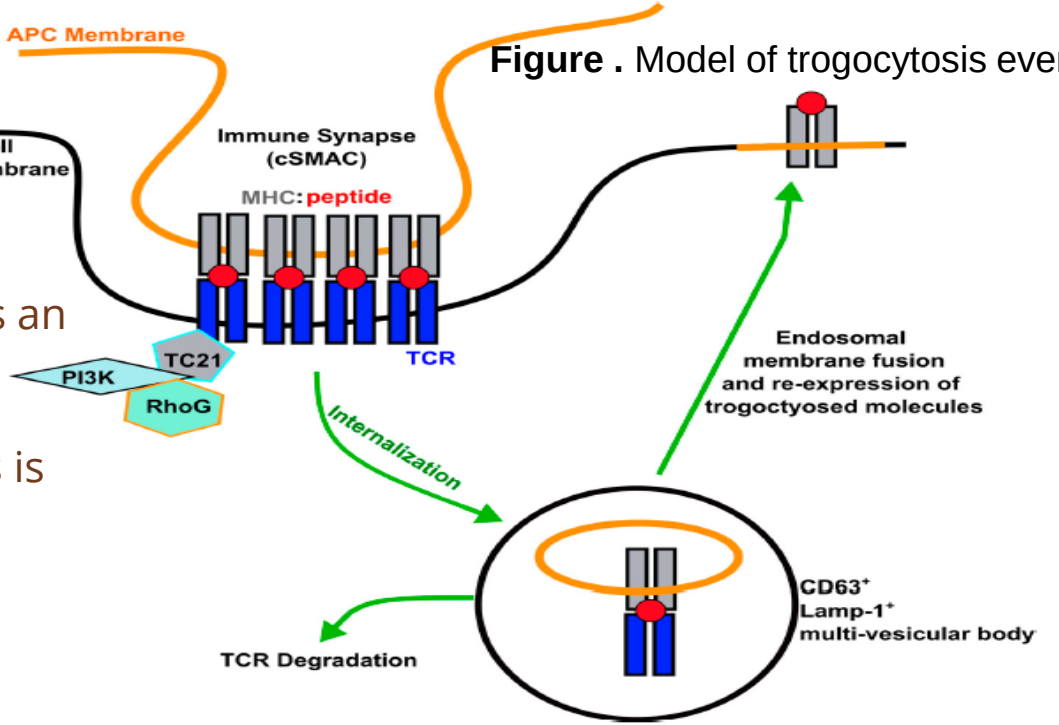

Figure . Model of trogocytosis event

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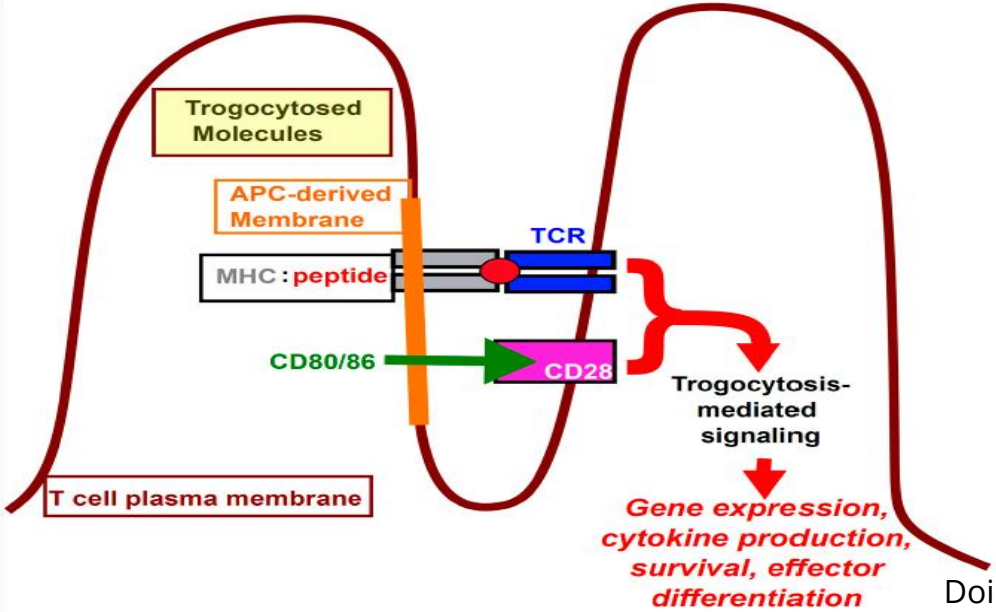
What is The Trogocytosis-Mediated Signaling?!

Simplified schematic of CD4+ T cell trogocytosis-mediated signaling

- Trogocytosis by CD4+ T cells is dependent on the affinity of the TCR for MHC:peptide



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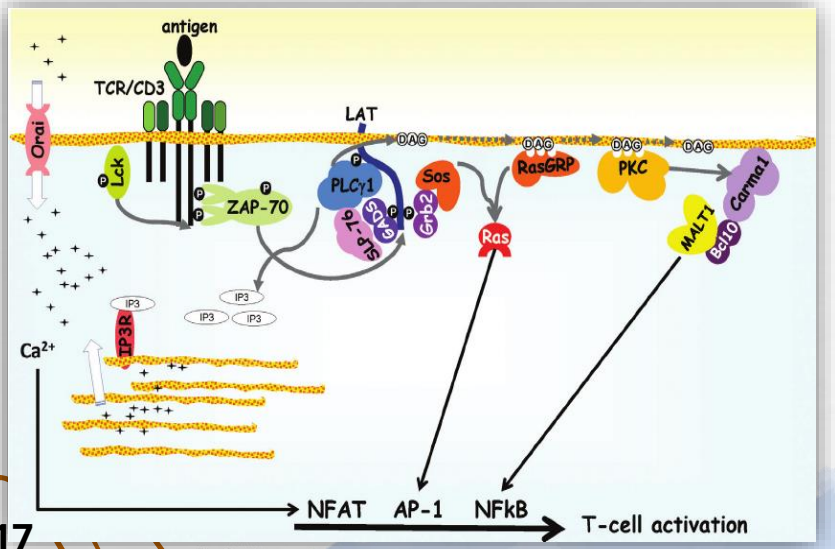
TCR signaling

- **Blocking ZAP-70** significantly reduces the rate of trogocytosis
- Similarly,**MEK/ERK inhibitor** or the **Src family kinase inhibitor** significantly inhibits trogocytosis



➢Trogocytosis requires **actin cytoskeletal rearrangement**.
Actin polymerization inhibitors **latrunculin or cytochalasin D** severely limits trogocytosis.

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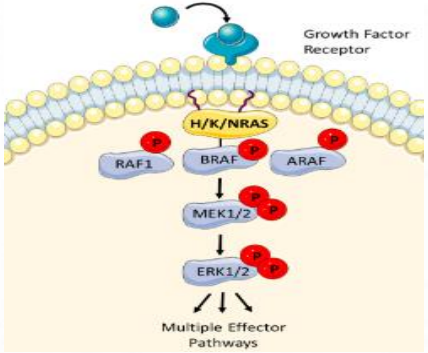
Transmission in Thorogocytosis

- Early studies found that, it **did not** include **transfer** of **cytoplasmic contents** between cells.



- **H-Ras**

- In both **T cells and NK cells**

Intracellular signaling → Elevated **phosphorylated ERK 1/2**
Increased IFN, TNF, and **Proliferation**, and **enhanced NK killing**

Doi: 10.3390/cells10061478 (2021)
Doi: 10.3390/genes12040553 (2021)

NK cell trogocytosis

NK cell trogocytosis, from both activating and inhibitory immune synapses , is largely mediated by receptor–ligand interactions

Ly49A mediates the trogocytosis of **Db**

NKG2D mediates the trogocytosis of **MICA , MICB , viral ligands** , and **Rae-1** from target cells

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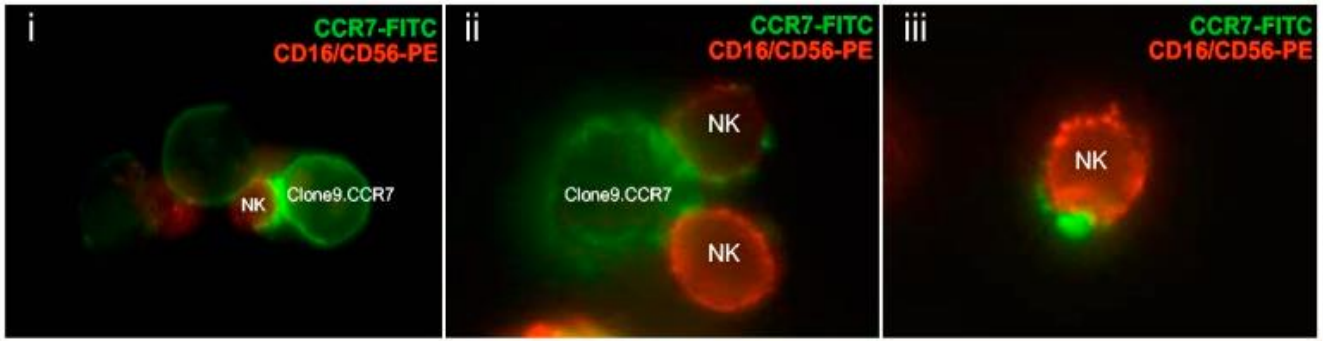
Example of NK cell trogocytosis

- Chemokine receptor **CCR7**



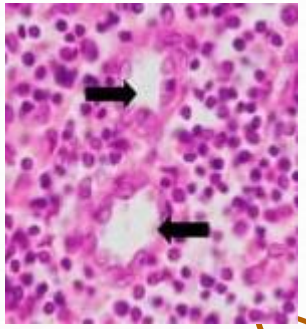
After adoptive transfer into nude mice, these CCR7+ NK cells migrated to lymph nodes at a significantly higher rate than the CCR7– NK cells.

doi: 10.1182/blood-2011-11-389924

Example of trogocytosis in other cell

Trogocytosis is the transfer of CCR5 from peripheral blood mononuclear cells to endothelial cells during transendothelial migration.

It is hypothesized that this may allow the endothelial cells to be infected by a macrophage-tropic strain of HIV-1.



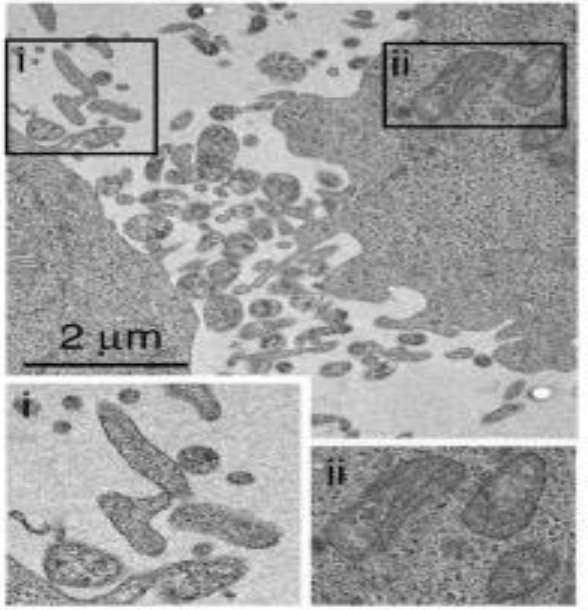
Doi: 10.3390/cells10061478 (2021)

05

How is trogocytosis diagnosed?!

Detection of trogocytosis

- The detection of trogocytosis by **Flow cytometry**
- By transmission electron microscopy (**TEM**)



Doi: 10.1038/s41467-018-06090-8 (2018)
Doi: 10.3390/cells10061478 (2021)

OPEN ACCESS Freely available online

PLOS ONE

T Cells at the Site of Autoimmune Inflammation Show Increased Potential for Trogocytosis

Bettina Haastert¤, Richard J. Mellanby, Stephen M. Anderton, Richard A. O'Connor*

Medical Research Council/University of Edinburgh Centre for Inflammation Research, Queen's Medical Research Institute, Edinburgh, United Kingdom

The activation state of the cell impacts trogocytosis, as activated T cells are much more efficient at performing trogocytosis than naive T cells.

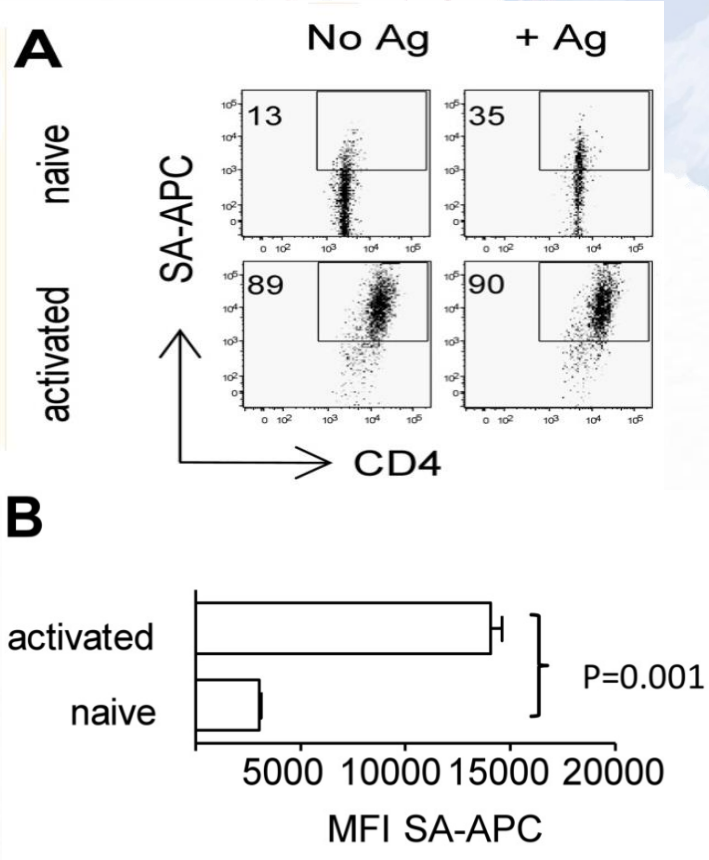
- **Increased size** of activated cells
- **Increased avidity** of the activated cell (increased expression of **adhesion** and **costimulatory** , such as **CD28, LFA-1, and CD44**)

Doi:10.1371/journal.pone.0081404

Con.

- B10.PL, B10.PLxC57BL/6, and C57BL/6 mice were used as a source of APCs and T cell.
- Trogocytosis assay
- Antibodies and FACS analysis

- Surface staining used the antibodies



Membrane acquisition by highly activated T cells is antigen independent.

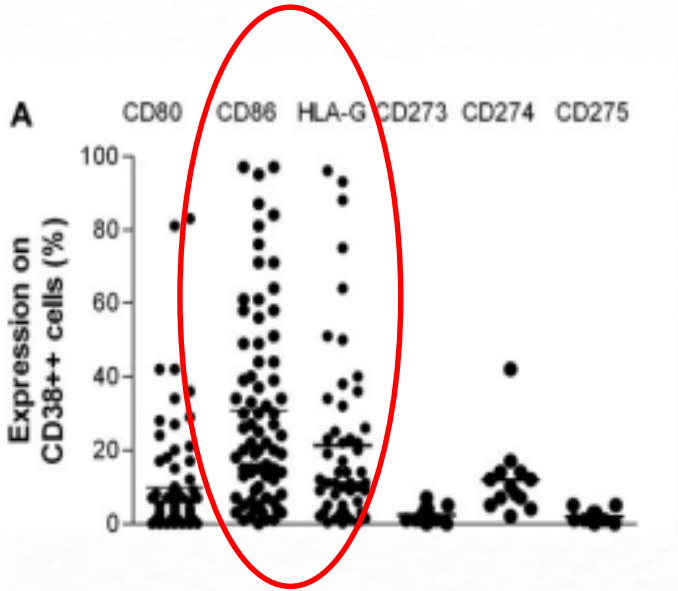
Doi:10.1371/journal.pone.0081404

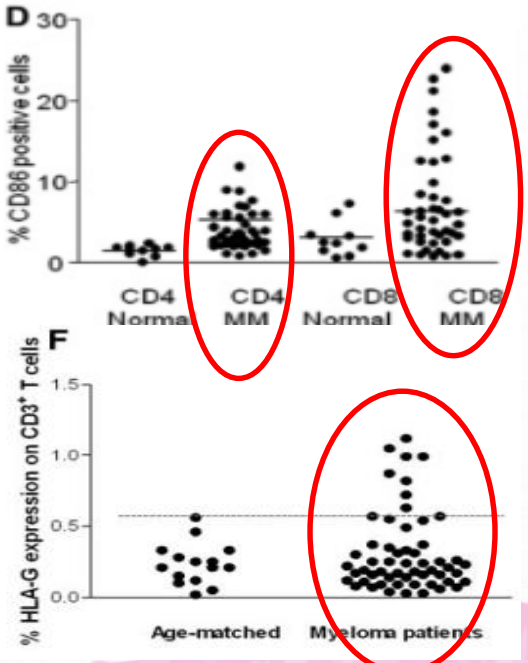
DOI: 10.1182/blood-2020-03-416792

CD86$^+$ or HLA-G$^+$ can be transferred via trogocytosis from myeloma cells to T cells and are associated with poor prognosis

Ross Brown,[1] Karieshma Kabani,[1] James Favaloro,[1] Shihong Yang,[1] P. Joy Ho,[1] John Gibson,[1] Phillip Fromm,[2] Hayley Suen,[1] Narelle Woodland,[3] Najah Nassif,[3] Derek Hart,[2] and Douglas Joshua[1]

[1]Institute of Haematology, Royal Prince Alfred Hospital, Sydney, Australia; [2]Dendritic Cell Biology and Therapeutics, Australian and New Zealand Army Corps (ANZAC) Research Institute, Sydney, Australia; and [3]School of Medical and Molecular Biosciences, University of Technology, Sydney, Australia





nature medicine

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KIR-based inhibitory CARs overcome CAR-NK cell trogocytosis-mediated fratricide and tumor escape

Ye Li[1,2], Rafet Basar[1], Guohui Wang[1], Enli Liu[1], Judy S. Moyes[1], Li Li[1], Lucila N. Kerbauy[1,3,4], Nadima Uprety[1], Mohsen Fathi[5], Ali Rezvan[5], Pinaki P. Banerjee[1], Luis Muniz-Feliciano[1], Tamara J. Laskowski[1], Emily Ensley[1], May Daher[1], Mayra Shanley[1], Mayela Mendt[1], Sunil Acharya[1], Bin Liu[1], Alexander Biederstädt[1,6], Hind Rafei[1], Xingliang Guo[1], Luciana Melo Garcia[1], Paul Lin[1], Sonny Ang[1], David Marin[1], Ken Chen[7], Laura Bover[8,9], Richard E. Champlin[1], Navin Varadarajan[5], Elizabeth J. Shpall[1], Katayoun Rezvani[1,*]

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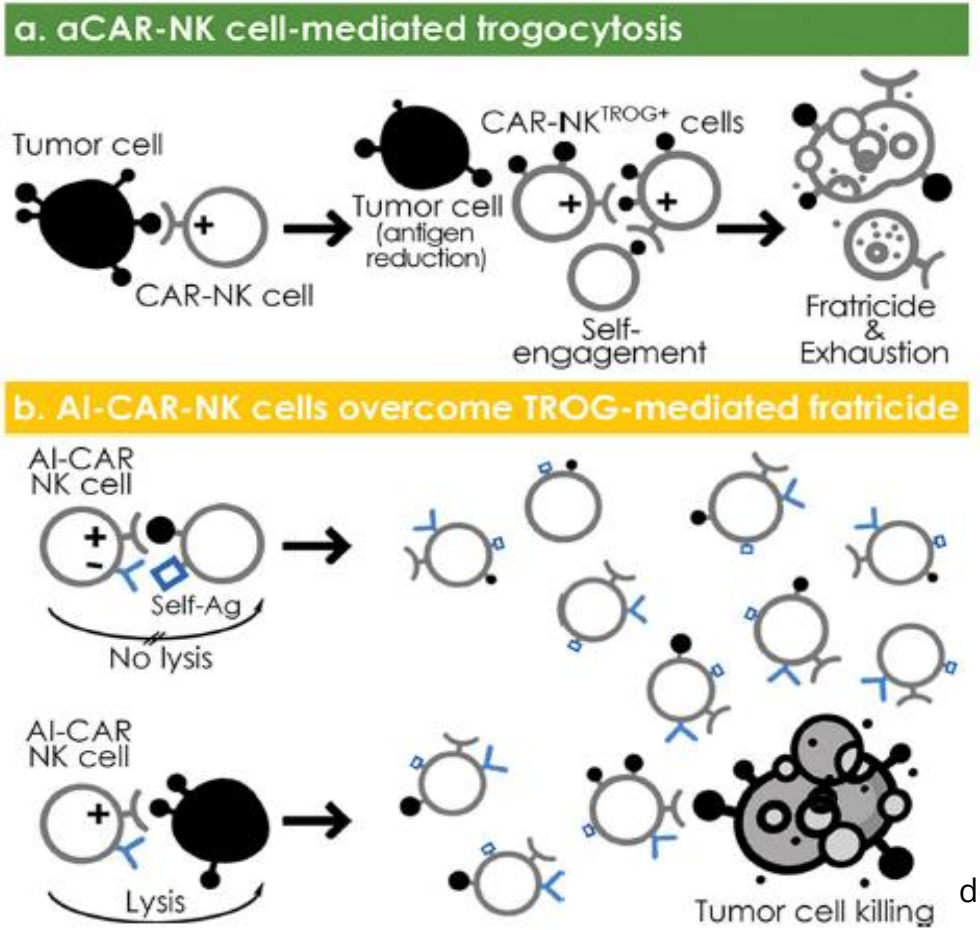
[5]Department of Chemical and Biomolecular Engineering, University of Houston, Houston, TX, USA

[6]Department of Medicine III: Hematology and Oncology, Technical University of Munich, Munich, Germany

CAR activation in NK cells promoted transfer of the CAR-cognate-antigen from tumor to NK cells.

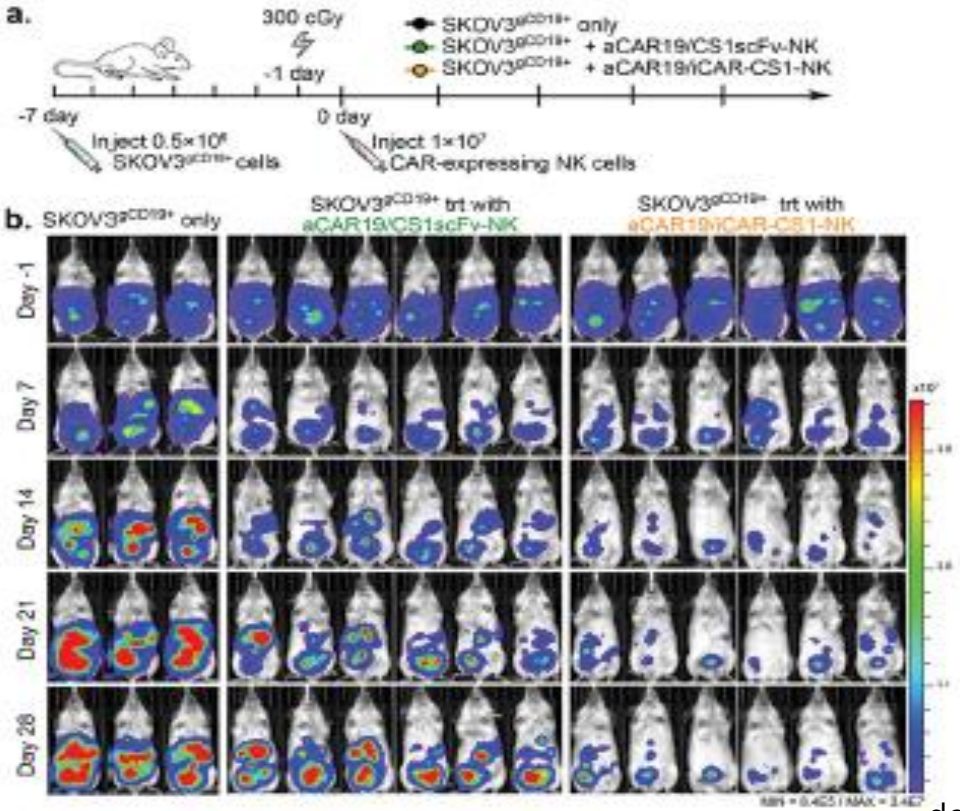
I. lower tumor antigen density
II. induced self-recognition and continuous CAR-mediated engagement

Con.



doi:10.1038/s41591-022-02003-x.

AI-CAR expressing NK cells exert superior *in vivo* anti-tumor activity in a SKOV3gCD19+ ovarian cancer model.



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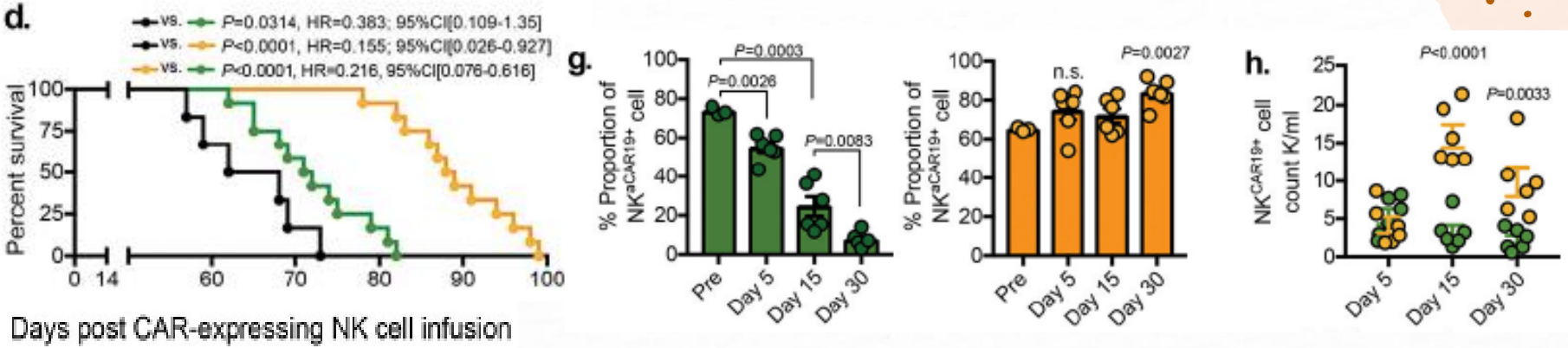
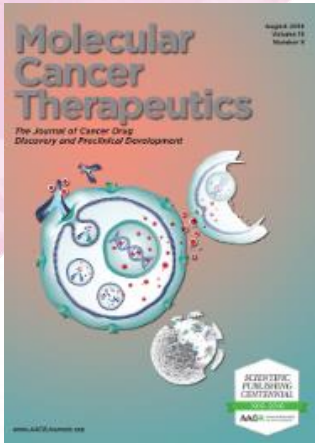


Percentage of live CAR19+ NK cells in the peripheral blood after infusion of aCAR19 (green, left) or aCAR19/iCAR-CS1 (orange, right) NK cells

doi:10.1038/s41591-022-02003-x.



Macrophage-mediated trogocytosis leads to death of antibody-opsonized tumor cells

Ramraj Velmurugan[a,b,c], **Dilip K. Challa**[a,b,c], **Sripad Ram**[d], **Raimund J. Ober**[a,e,*], and **E. Sally Ward**[a,b,*]

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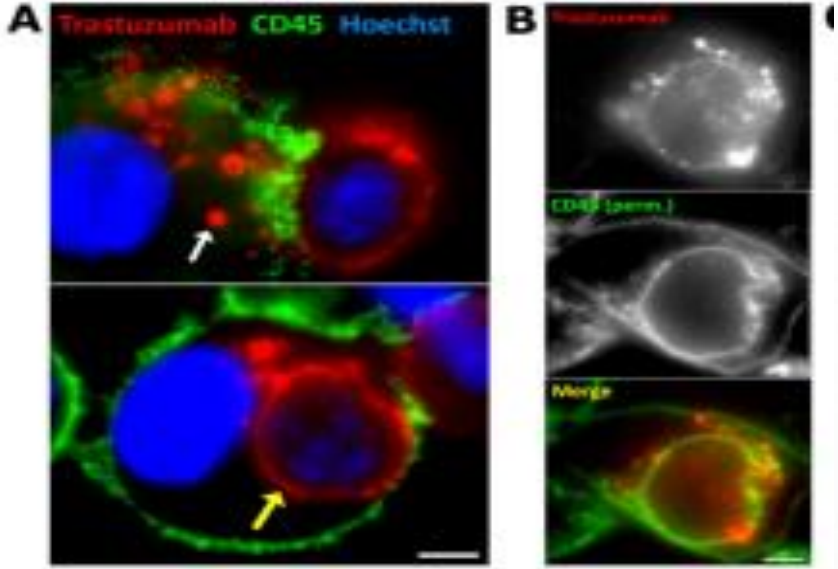
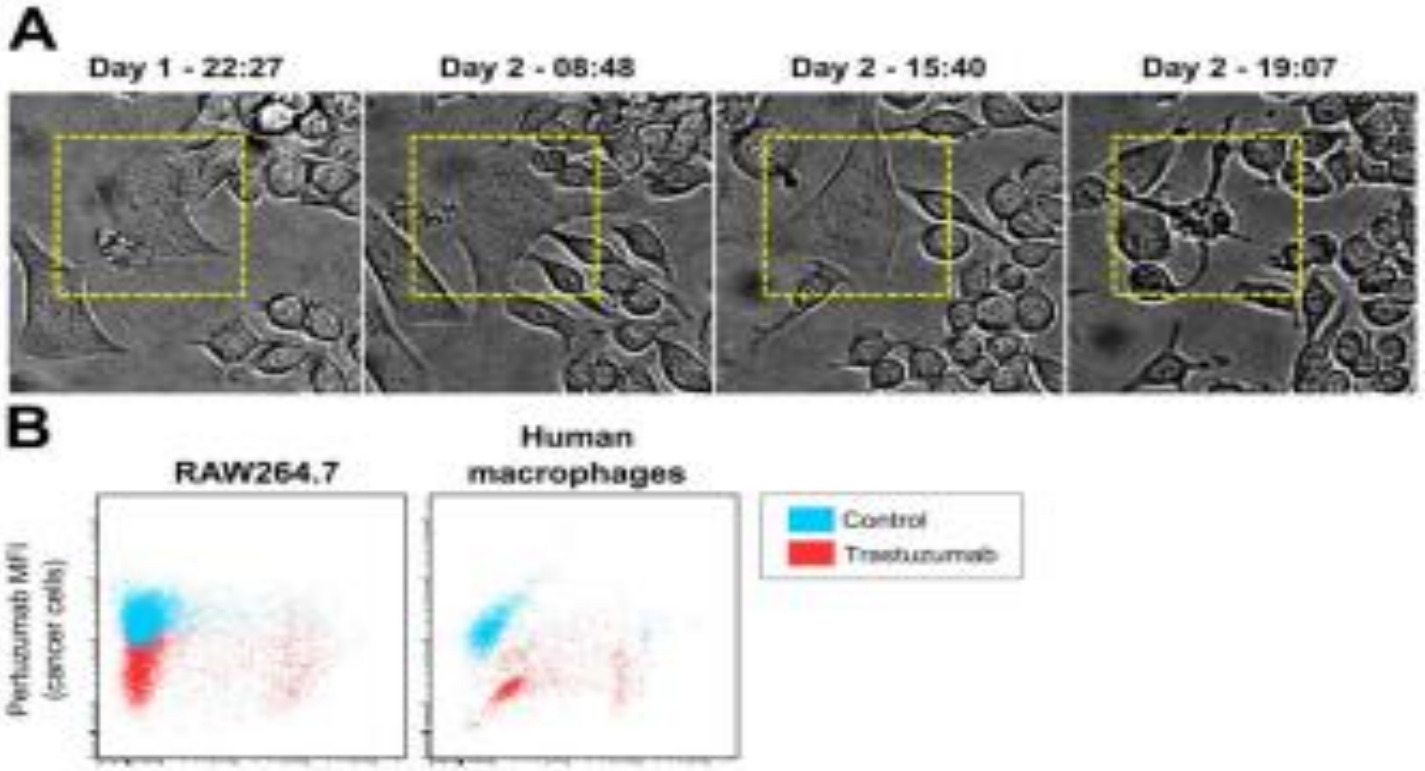


- The murine macrophage cell lines RAW264.7
- the human breast cancer cell lines MDA-MB-453.

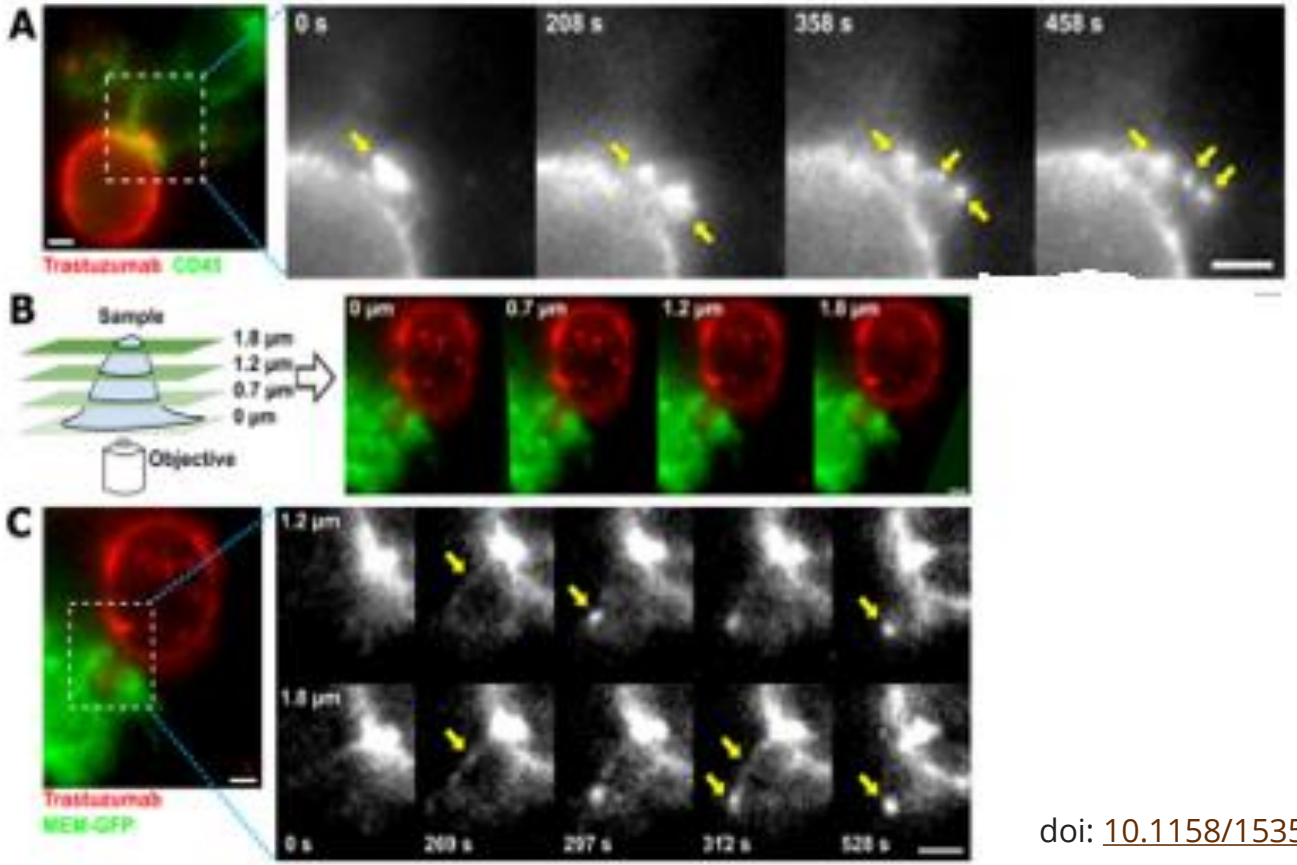
J774A.1 and RAW264.7 macrophages exhibit different phagocytic activities.

doi: 10.1158/1535-7163.MCT-15-0335



RAW264.7 and human monocyte-derived macrophages induce similar levels of apoptosis in opsonized cancer cells.

doi: 10.1158/1535-7163.MCT-15-0335



doi: 10.1158/1535-7163.MCT-15-0335

33 Fluorescence microscopy analyses using MUM reveals that trogocytosis involves tubular extensions of the cancer cells.

Challenges

- Cells are **tolerant** to limited amounts of trogocytosis

- T cells also **capture nonspecific mhc**, e.g., CD4+ T cells capture MHC class I molecules or irrelevant MHC molecules.

- Resistance to therapeutic antibodies:

can lead to antigenic modulation, which can **reduce the efficacy of mab-based therapies**.

- Adverse role in the immune system: trogocytosis can **accelerate the spread of certain pathogens among macrophages** and participate in **autoimmunity**.

Conclusion

- Trogocytosis has been observed in **various immune and non-immune cells**, and confers antigen-presenting activity in non-APCs, leading to the modulation of T cell immune responses.
- Trogocytosis-positive T cells and NK cells can present antigens to bystander cells and modulate immune responses.
- Regulation of Trogocytosis could be a good target for treating a wide variety of diseases.

Reseferences

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2. Mercer, F.; Ng, S.H.; Brown, T.M.; Boatman, G.; Johnson, P.J. Neutrophils kill the parasite Trichomonas vaginalis using trogocytosis. PLoS Biol. 2018, 16, e2003885.
3. Macrophage-Mediated Trogocytosis Leads to Death of Antibody-Opsonized Tumor Cells. DOI: 10.1158/1535-7163.MCT-15-0335
4. T Cells at the Site of Autoimmune Inflammation Show Increased Potential for Trogocytosis doi: 10.1371/journal.pone.0081404
5. CD86+ or HLA-G+ can be transferred via trogocytosis from myeloma cells to T cells and are associated with poor prognosis DOI: 10.1182/blood-2012-03-416792

Reseferences

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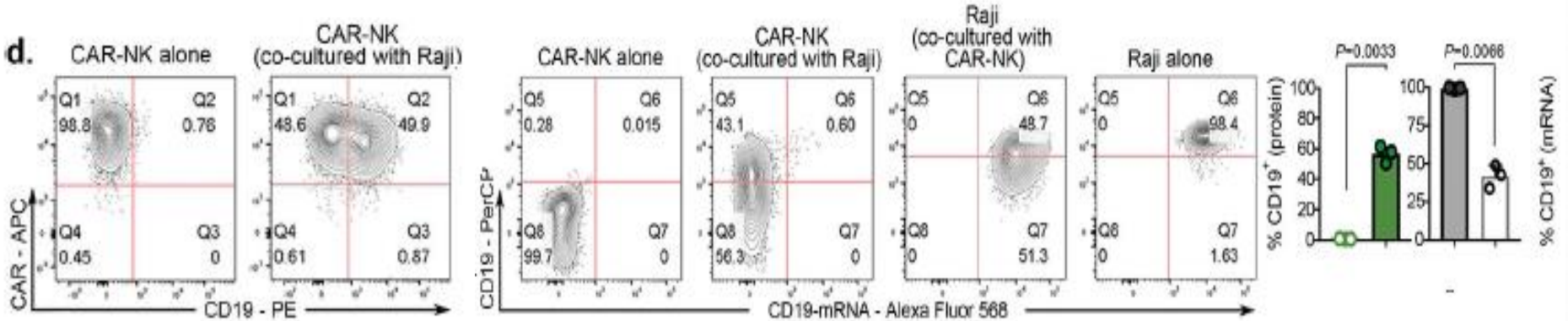
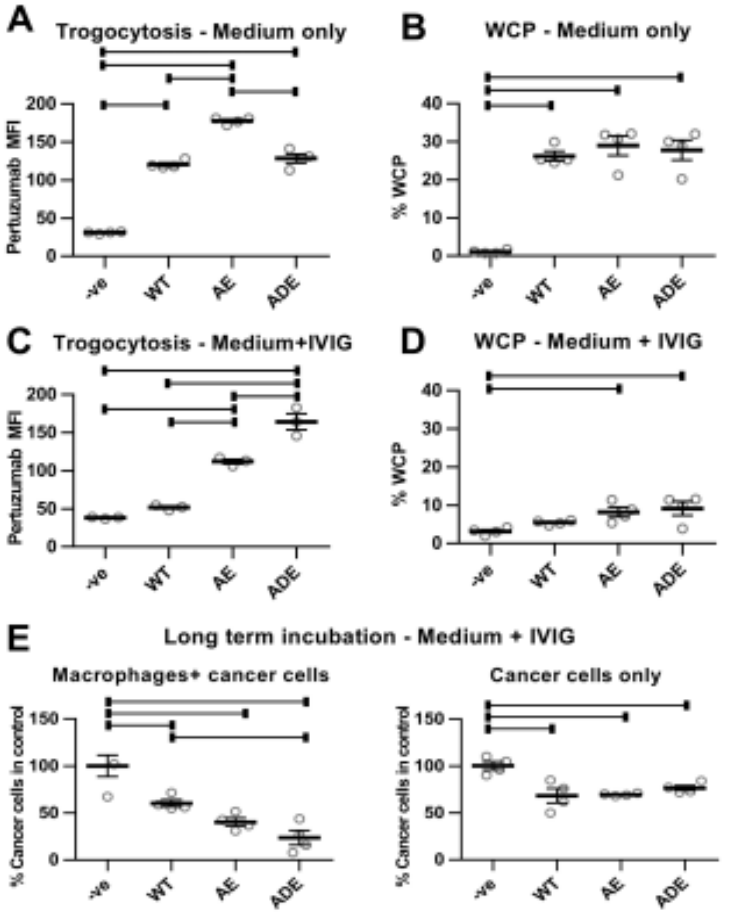
10. Gnawing Between Cells and Cells in the Immune System: Friend or Foe? A Review of Trogocytosis https://doi.org/10.3389/fimmu.2022.791006

Thanks For Your Attention



d. Flow cytometric analyses show expression of CD19at the protein level (left) and the mRNA level (middle) in CAR-NK cells cultured alone, Raji cells cultured alone, and CAR-NK cells co-cultured with Raji cells for 5 mins (representative of 3 donors). Inset numbers indicate percentages of cells within the indicated gated regions. Bar graphs show the summary data for each marker (right)

Antibodies with enhanced affinity for activating FcγRs have increased trogocytic and cellkilling activity.



Hudrisier et al. also reported that FcR is trogocytosed by CD4+ T cells and it efficiently binds to immune complexes, although it does not induce detectable intracellular signaling.