# Extracellular vesicles from Staphylococcus aureus follow different pathways to manipulate host cells

Julia Papail, Ligia Prado, Nathalie Daniel, Daniele Vassaux, Yann Le Gouar, Nadia Berkova, Julien Jardin, Svetlana Chabelskaia, Yves Le Loir, Vasco Azevedo, et al.

# Extracellular vesicles from *Staphylococcus aureus* follow different pathways to manipulate host cells

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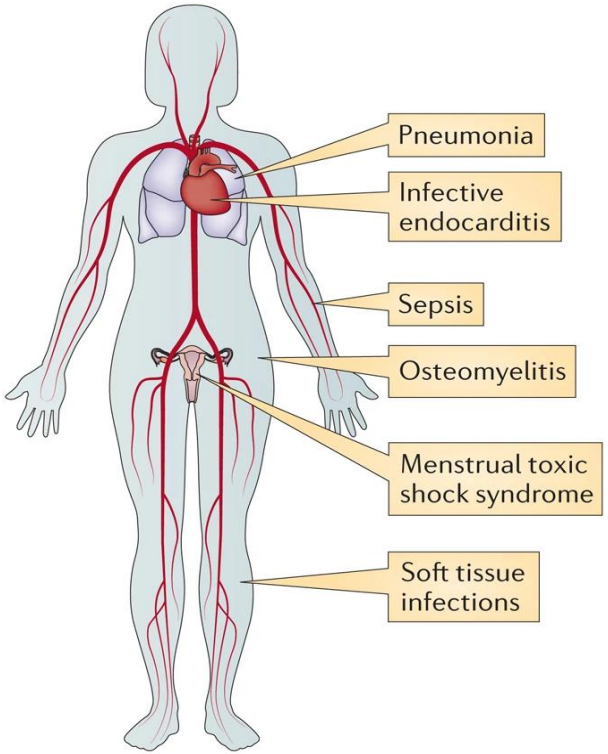
# Conflict of interest declaration

For this presentation,
I declare that I have no conflict of interest.

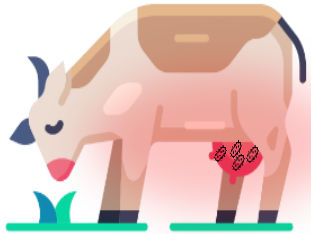
# ✓ *Staphylococcus aureus*

➢ **Human opportunist pathogen**




**Diseases caused by *Staphylococcus aureus***
*Salgado-Pabón W and Schlievert P., 2014*

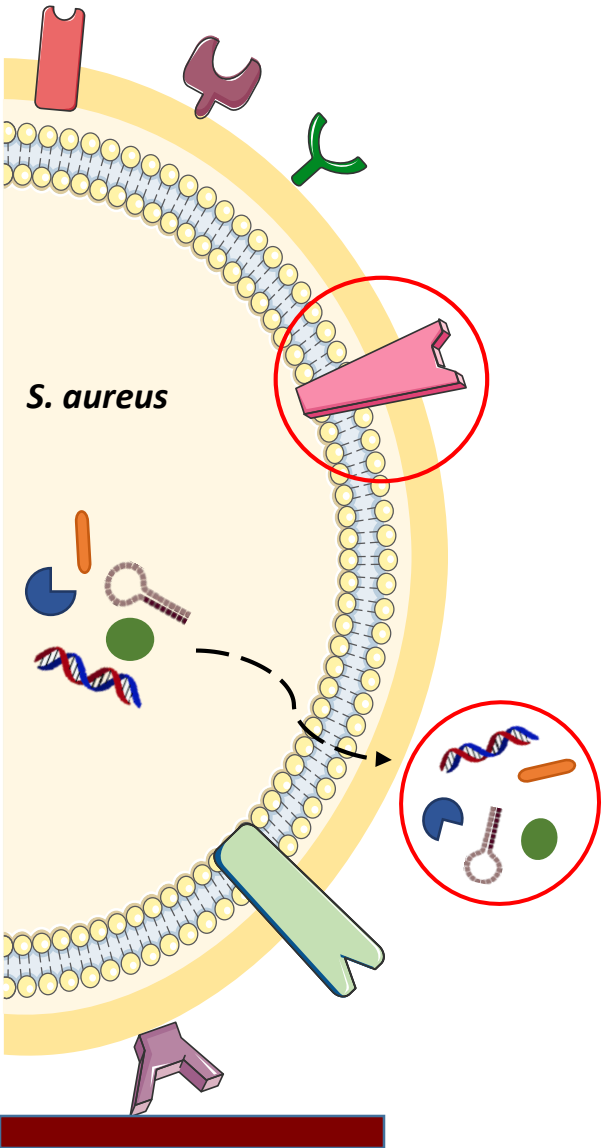
➢ **Significant impact on the veterinary medicine and food fields**
**→ Etiological agent of mastitis**




➢ **The six highly virulent and antibiotic resistant bacterial pathogens**

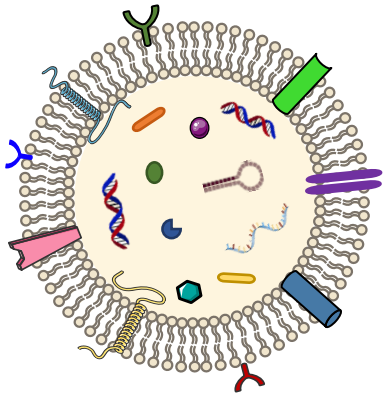
ESKAPE

# ✓ *Staphylococcus aureus* virulence factors




**Surface-exposed proteins**

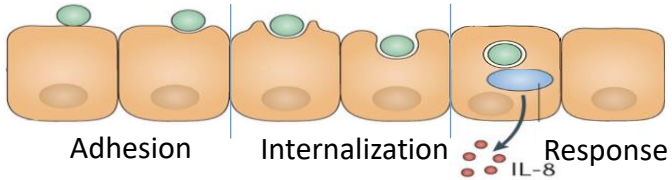
**Extracellular vesicles (EVs)**



**as delivery vehicles**

**Secreted proteins / molecules**

*S. aureus* colonization

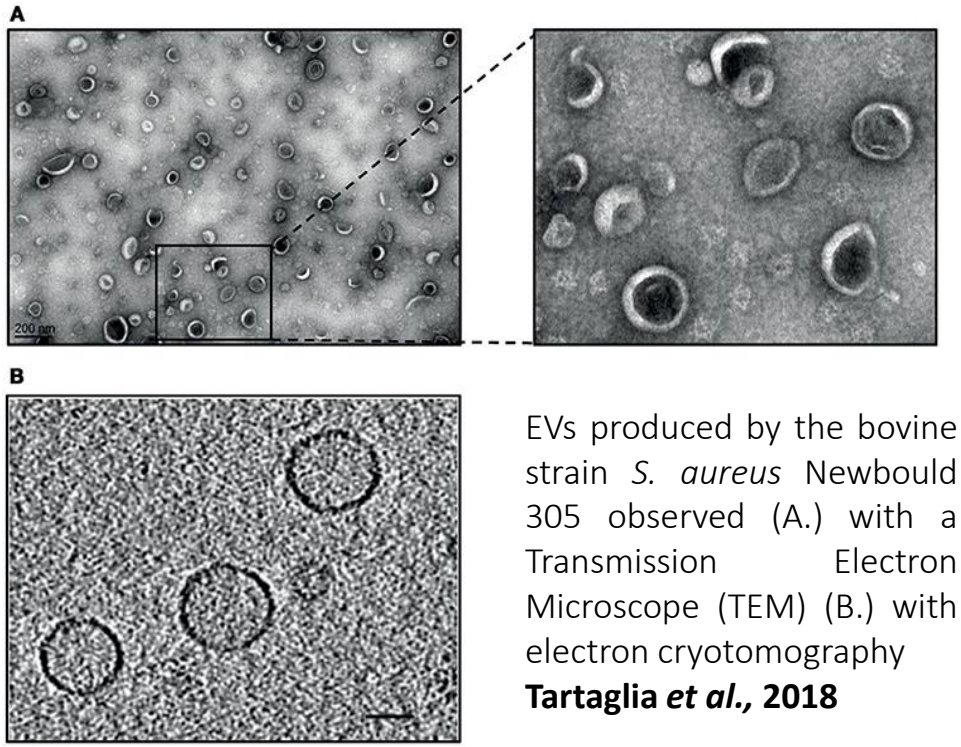



PATHOGENESIS

Disease severity

# ✓ Extracellular vesicles (EVs)

- Lipid bilayer spherical nano-sized particles (30-300 nm) which carry various molecules (eg, lipids, nucleic acids, proteins)
- Vehicles that transport and deliver molecules to local or distant cellular targets
- Imply in cell-to-cell communication and especially in host-pathogen interaction

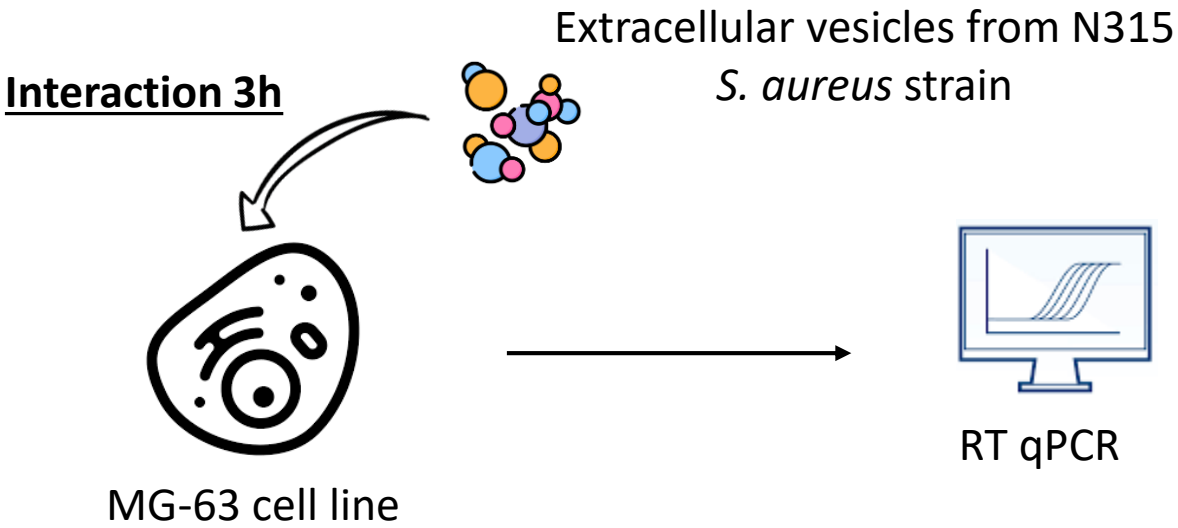



EVs produced by the bovine strain *S. aureus* Newbould 305 observed (A.) with a Transmission Electron Microscope (TEM) (B.) with electron cryotomography
**Tartaglia *et al.*, 2018**

**What is the contribution of extracellular vesicles from *S. aureus* in pathogenesis ?**

# ✓ Role of extracellular vesicles in pathogenesis

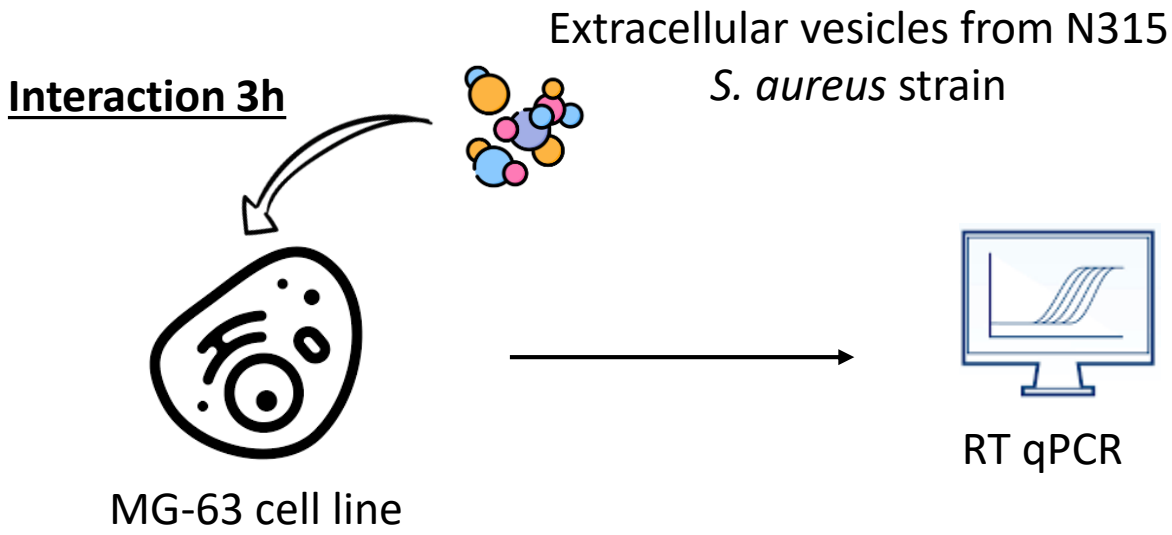
## Impact of EVs on the expression of several inflammatory genes

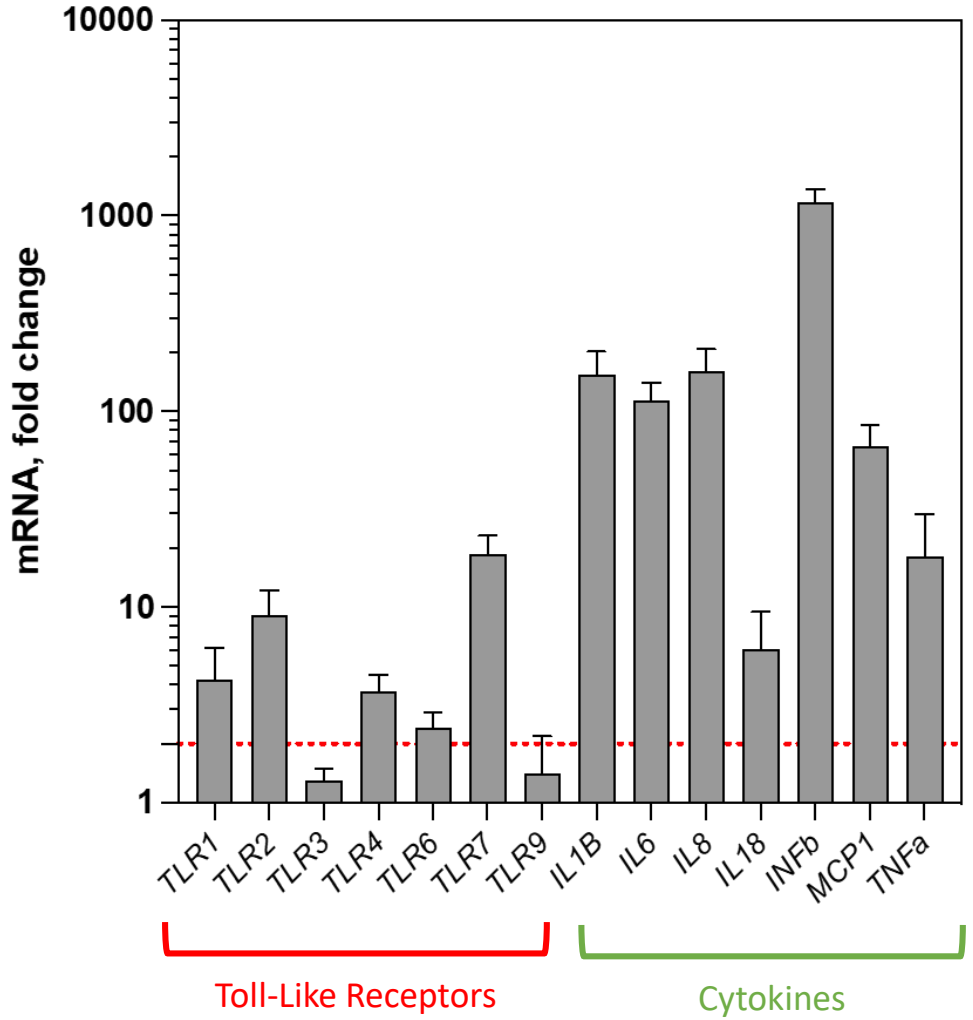



- Human osteoblast-like non-phagocytic cell line
- Taken from patient with osteosarcoma

# ✓ Role of extracellular vesicles in pathogenesis

## Impact of EVs on the expression of several inflammatory genes



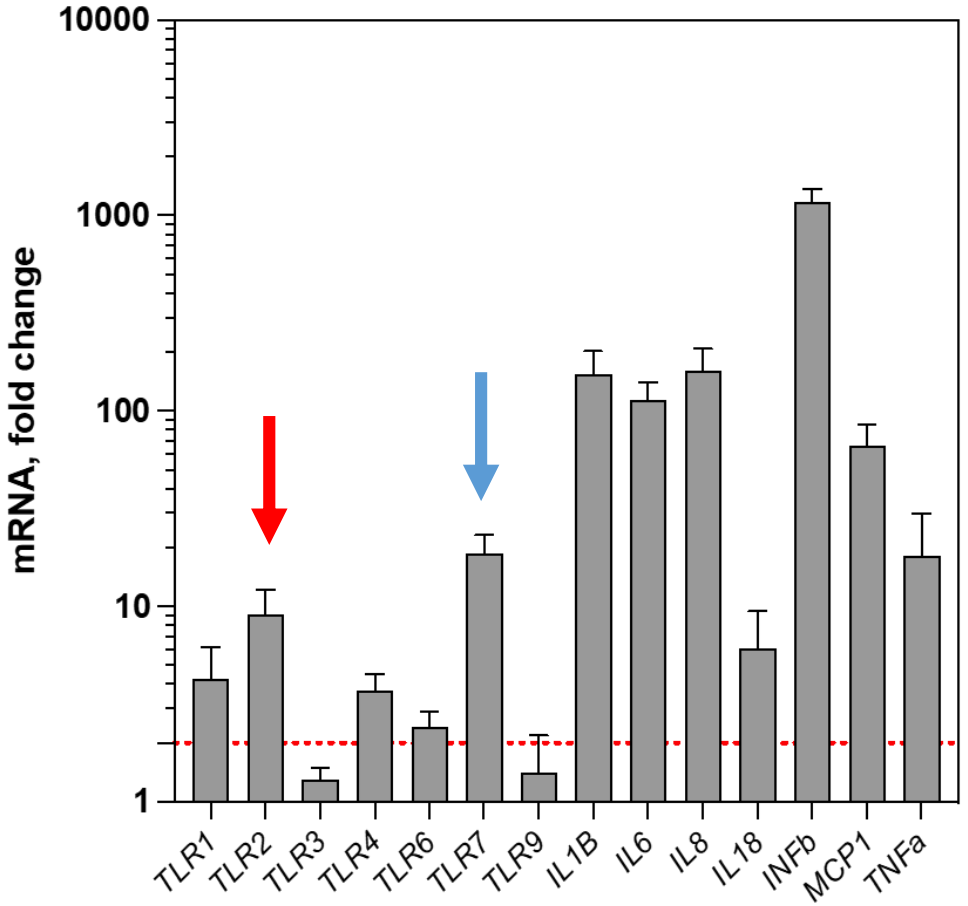

Increased expression of genes coding for :

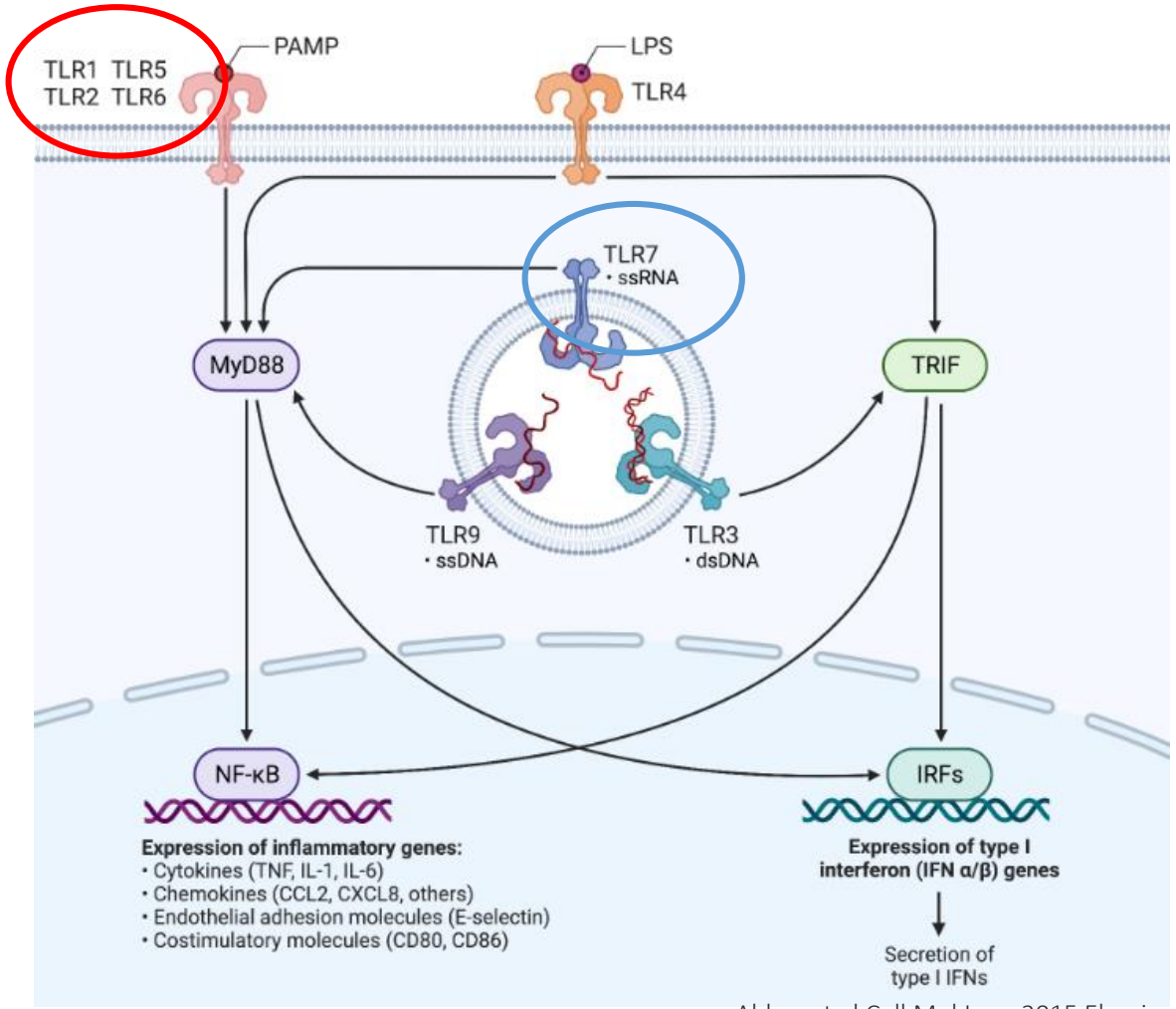
- ✓ Cytokines
- ✓ Toll-Like Receptors (TLR)

→ **N315-derived EVs induce the expression of various *TLR* and immune genes in MG-63 cells**

# ✓ Role of extracellular vesicles in pathogenesis

## Impact of EVs on the expression of several inflammatory genes



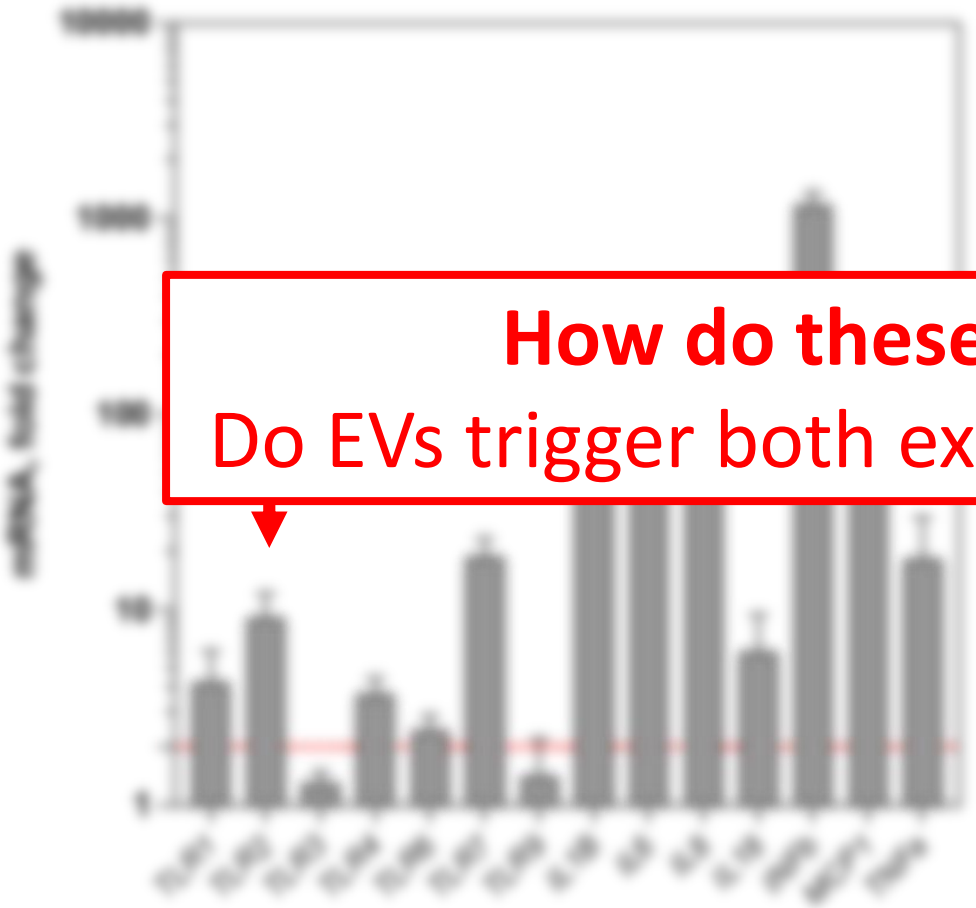



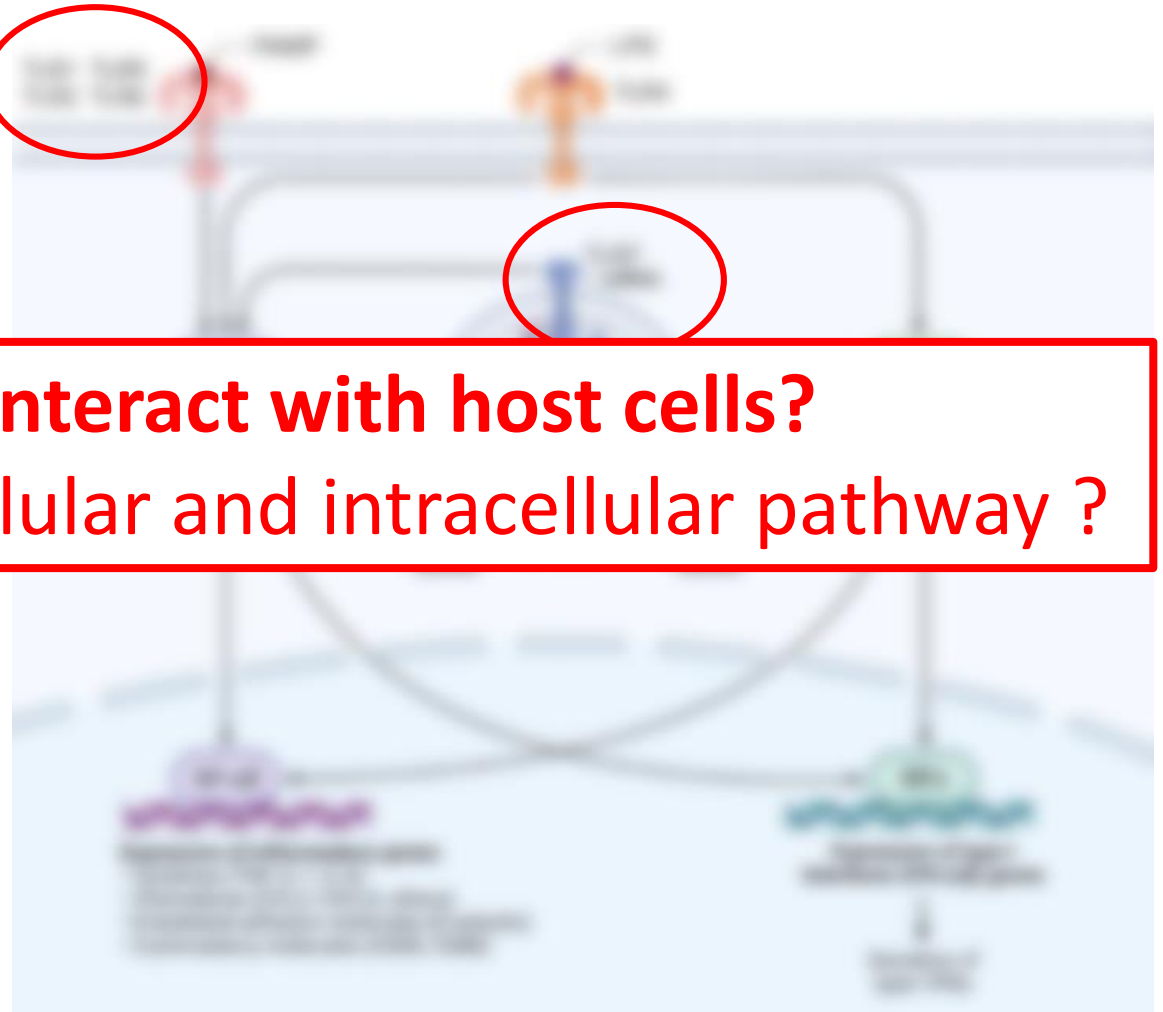

Abbas et al Cell Mol Imm 2015 Elsevier

→ **N315-derived EVs induce the expression of various *TLR* and immune genes in MG-63 cells**

# ✓ Role of extracellular vesicles in pathogenesis

Impact of EVs on the expression of several inflammatory genes





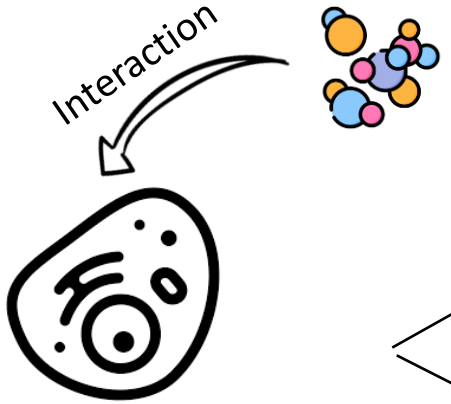
**How do these EVs interact with host cells?**

Do EVs trigger both extracellular and intracellular pathway ?

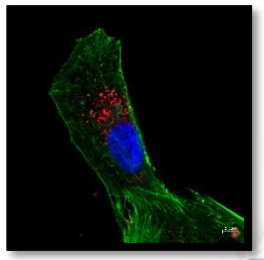
→ **N315-derived EVs induce the expression of various *TLR* and immune genes in MG-63 cells**

# ✓ Methods

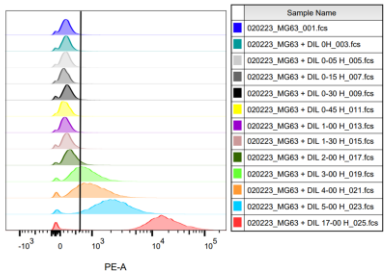
DiI-labelled Extracellular vesicles from N315 strain

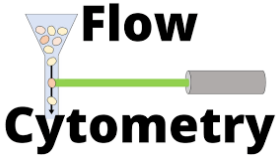



MG-63 cell line



Confocal Microscopy

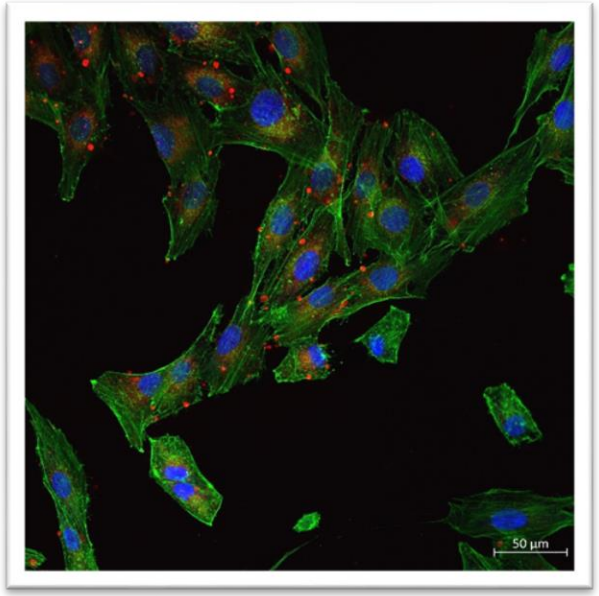


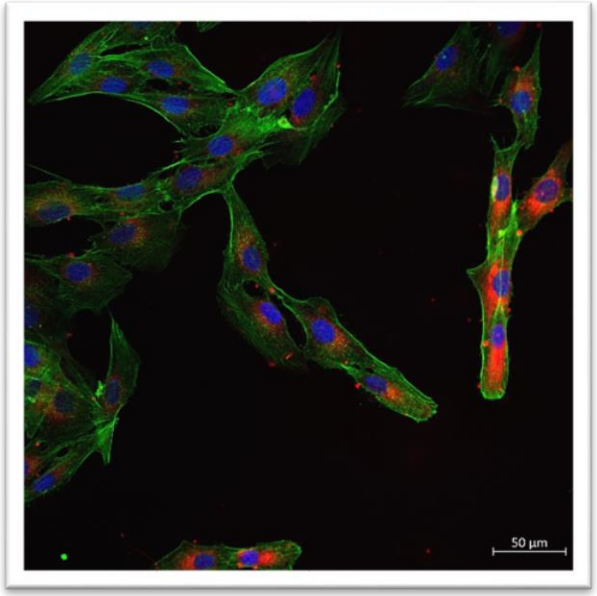
Flow Cytometry

# ✓ Internalization of EVs by MG-63 cells

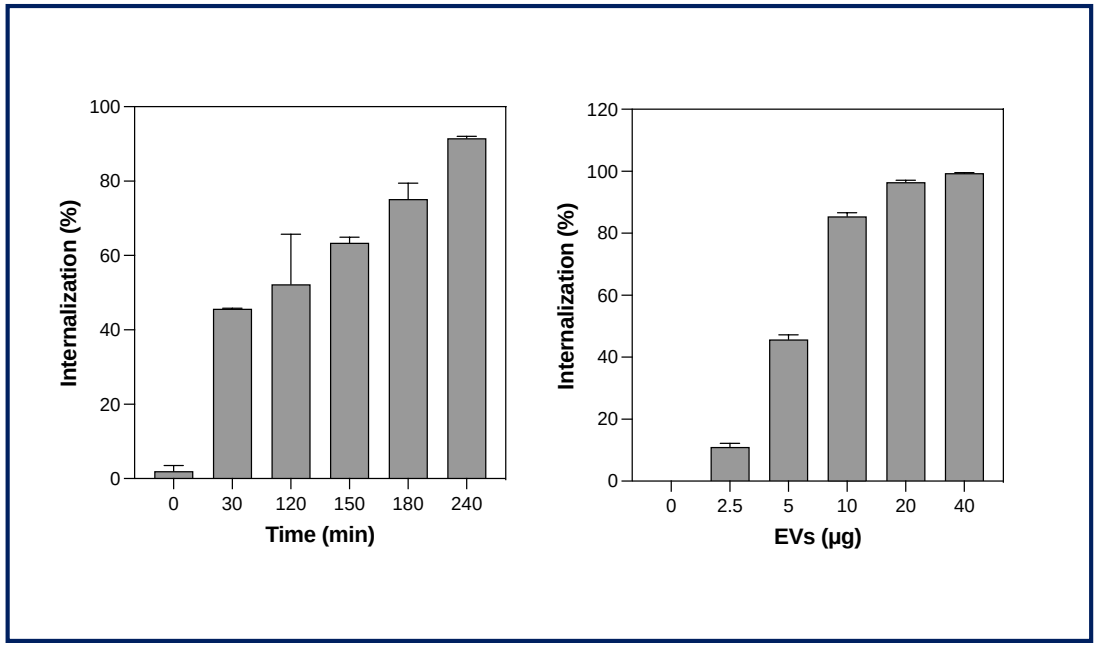
+ EVs at 3 h of incubation




+ EVs at 17 h of incubation



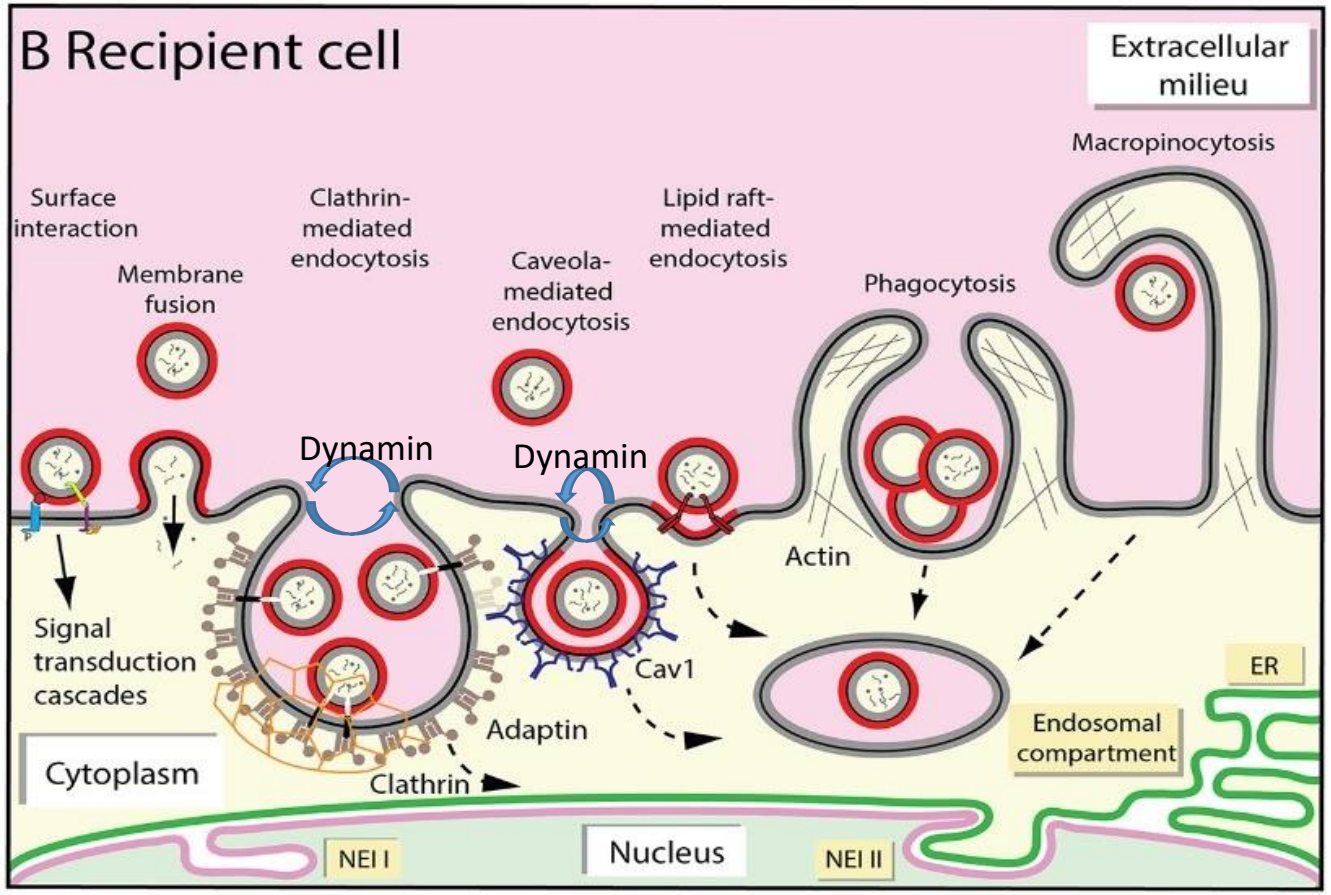

***Unpublished results***




→ ***S. aureus*** **N315-derived EVs are internalized by MG-63 cells**

- **in a dose-dependent manner**
- **in a time-dependent manner**

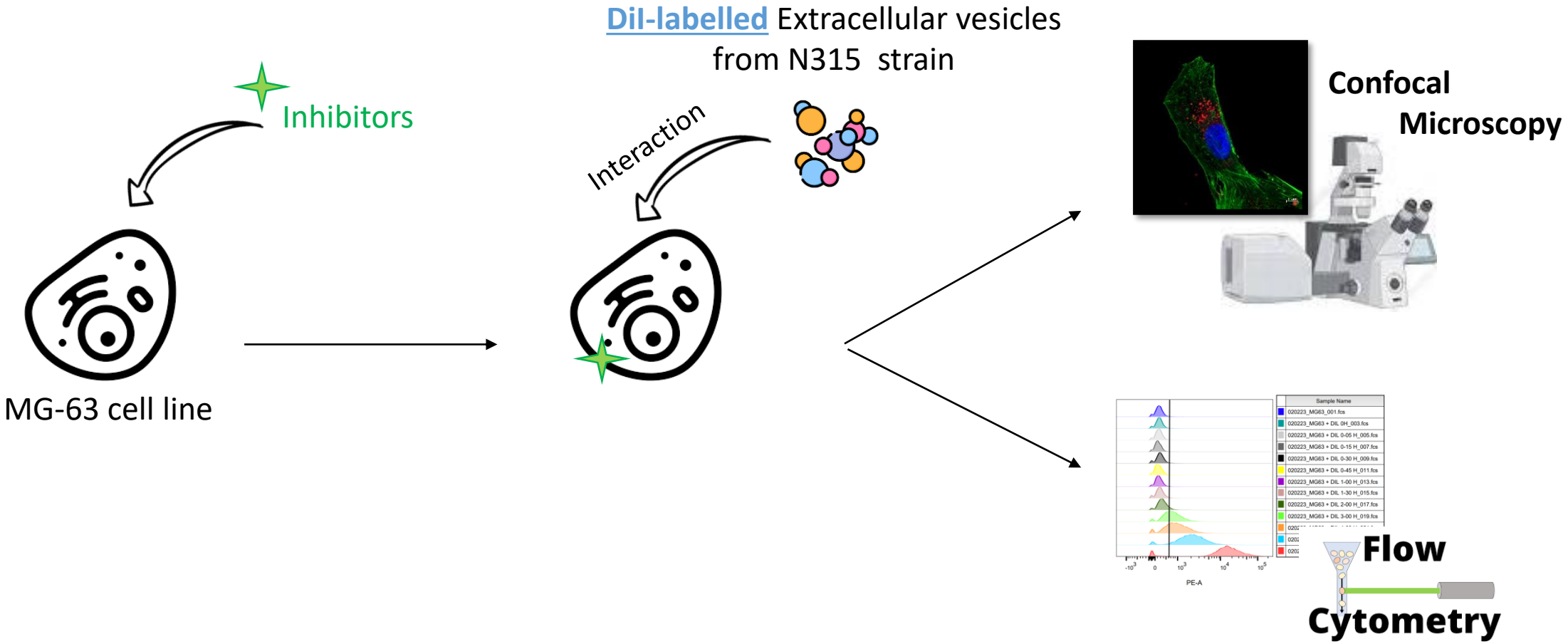
# ✓ Internalization of EVs by MG-63 cells



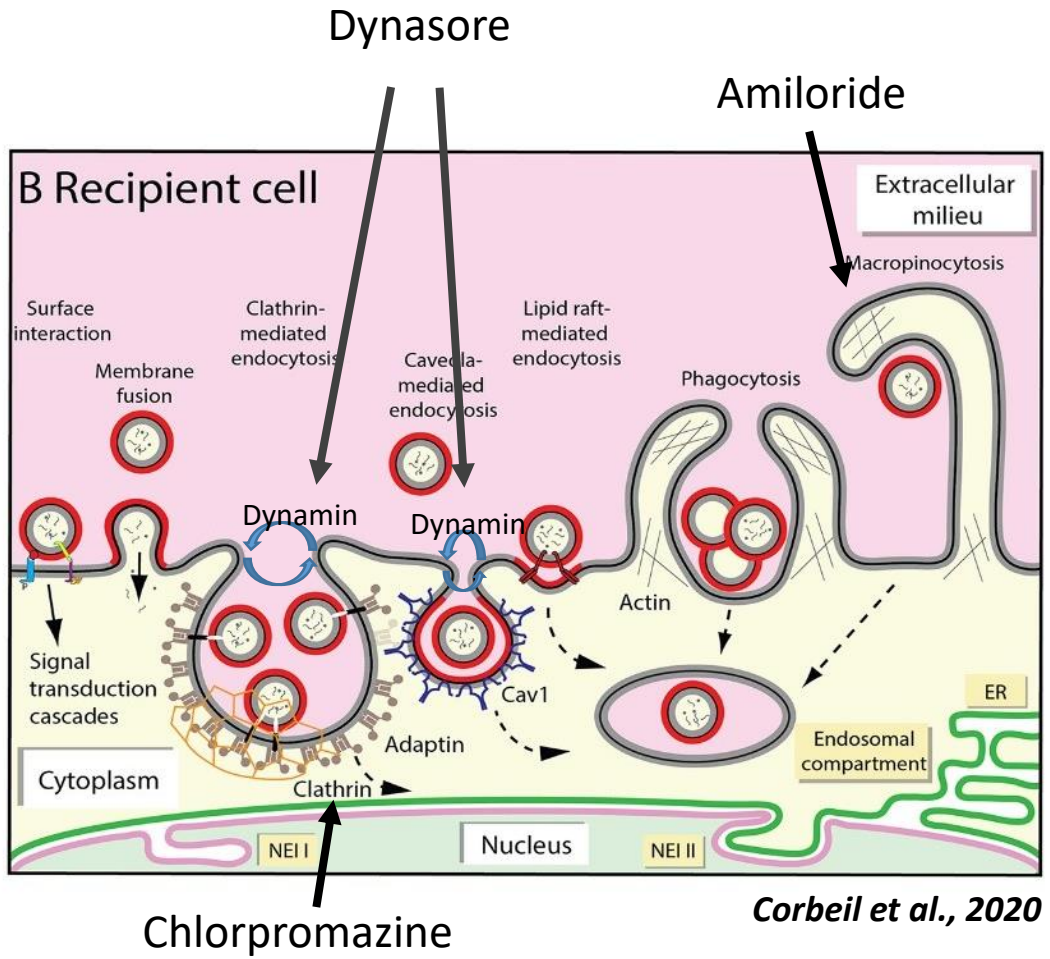

*Corbeil et al., 2020*

→ **Several different pathways of internalization are possible**

# ✓ Methods

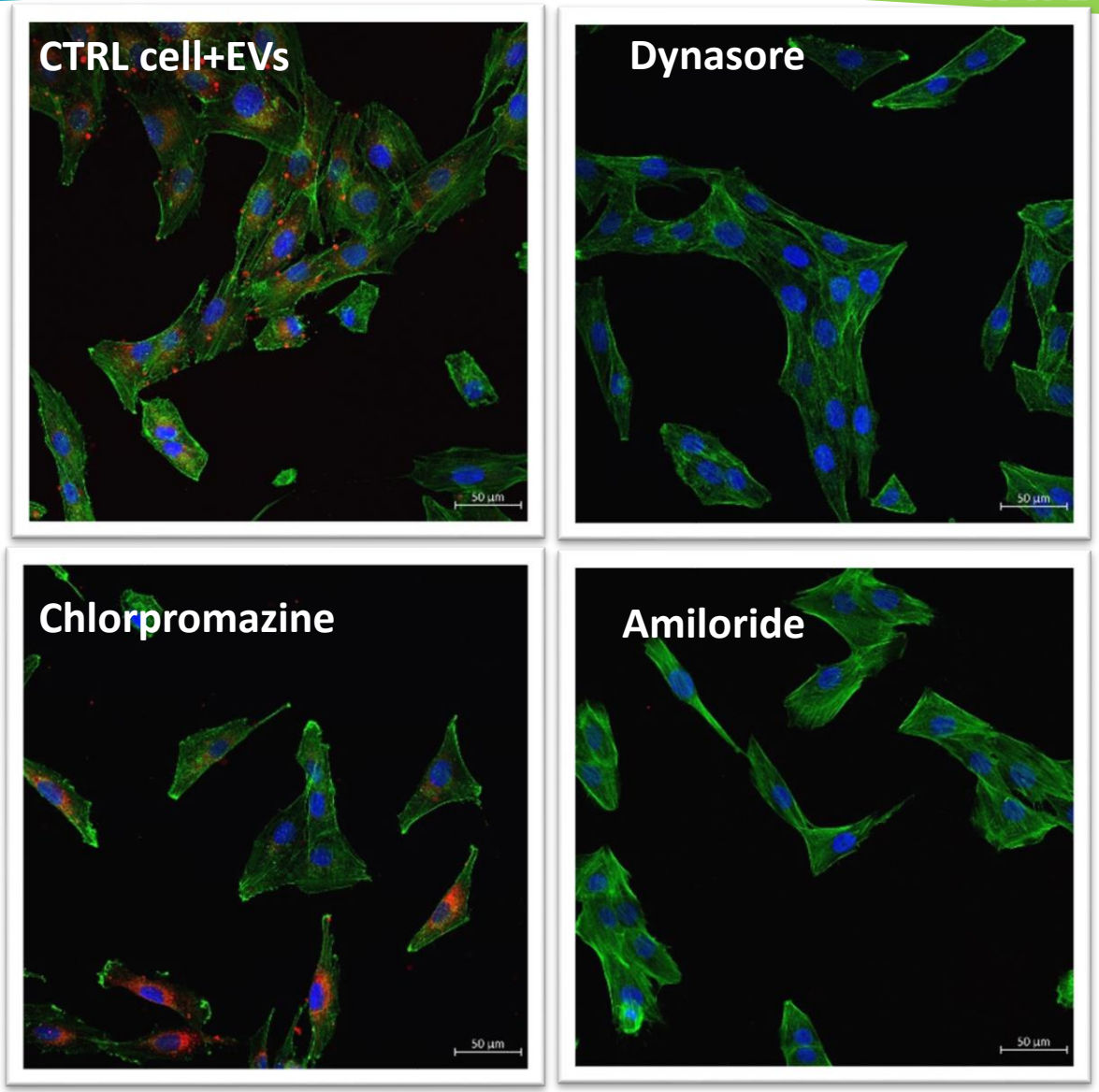

DiI-labelled Extracellular vesicles from N315 strain
Inhibitors
Interaction
MG-63 cell line
Confocal Microscopy
Flow Cytometry

# ✓ Internalization of EVs by MG-63 cells




*Corbeil et al., 2020*

→ **Internalization of N315 EVs can dependent upon dynamin-mediated endocytosis and macropinocytosis**




*Unpublished results*

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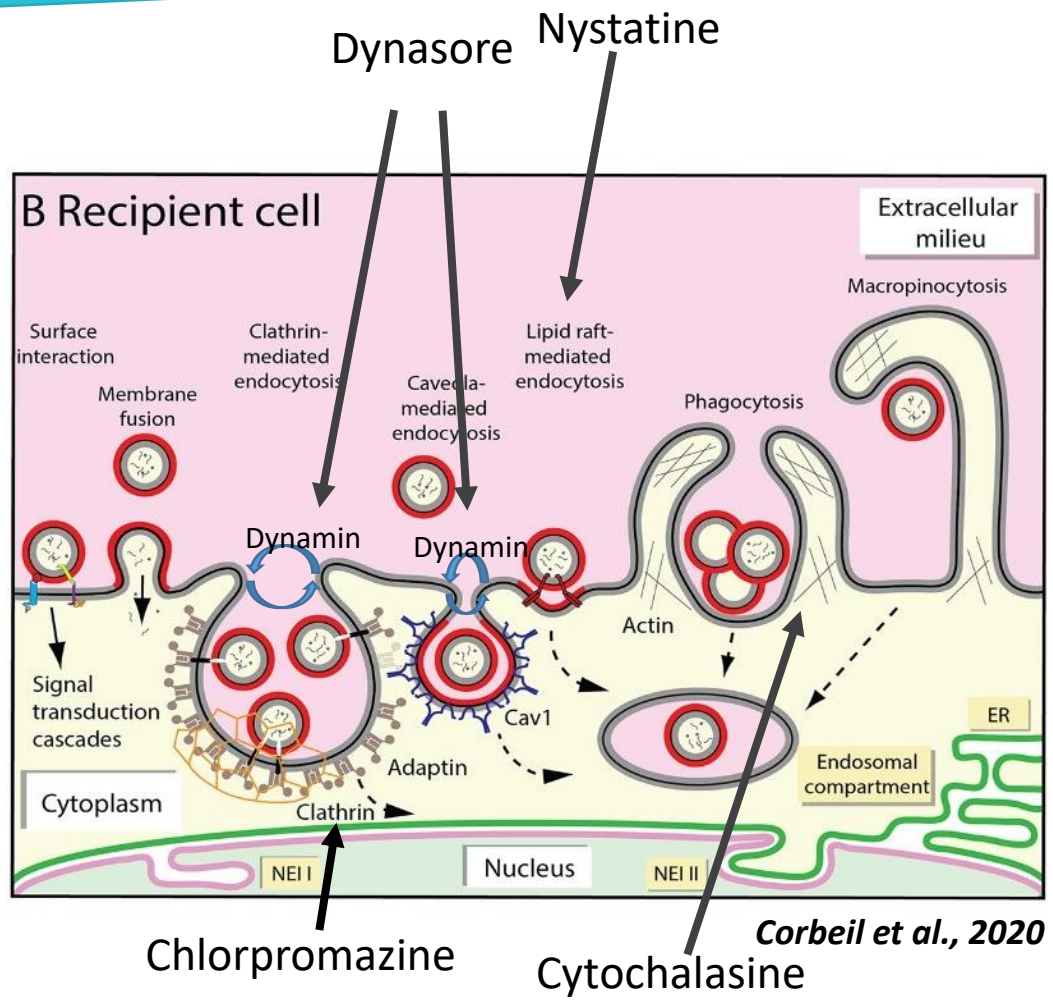
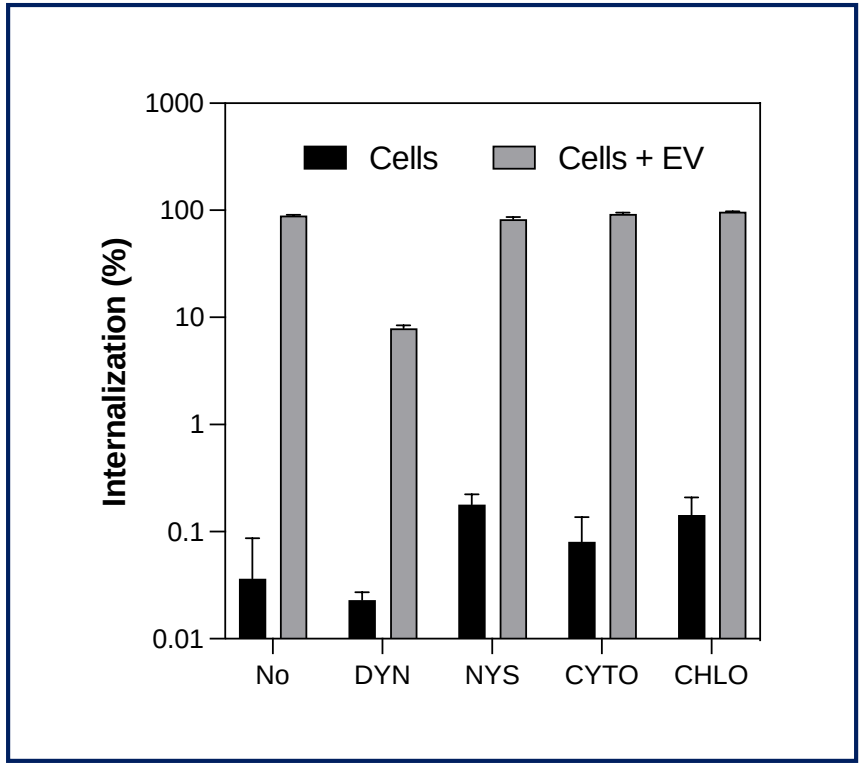



*Corbeil et al., 2020*

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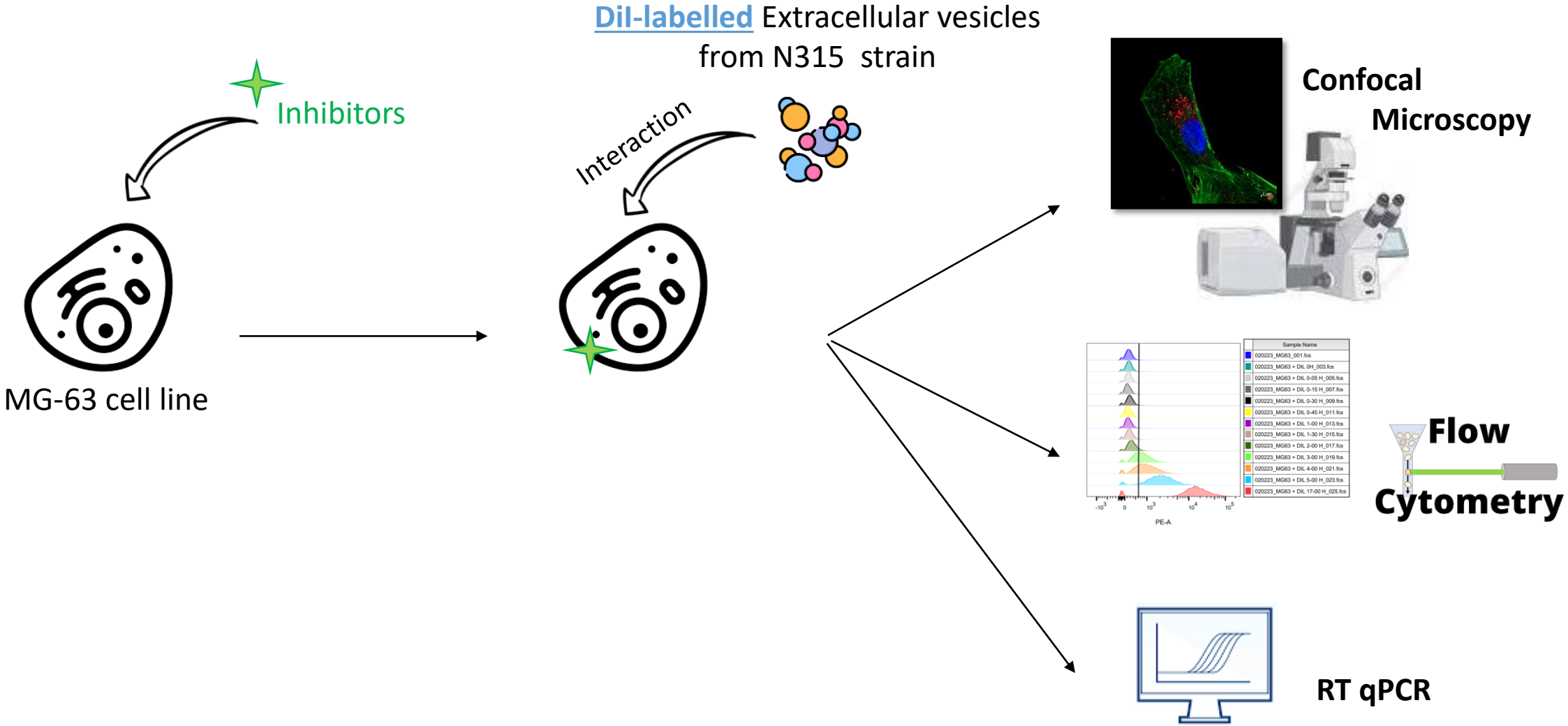
# ✓ Methods


DiI-labelled Extracellular vesicles from N315 strain
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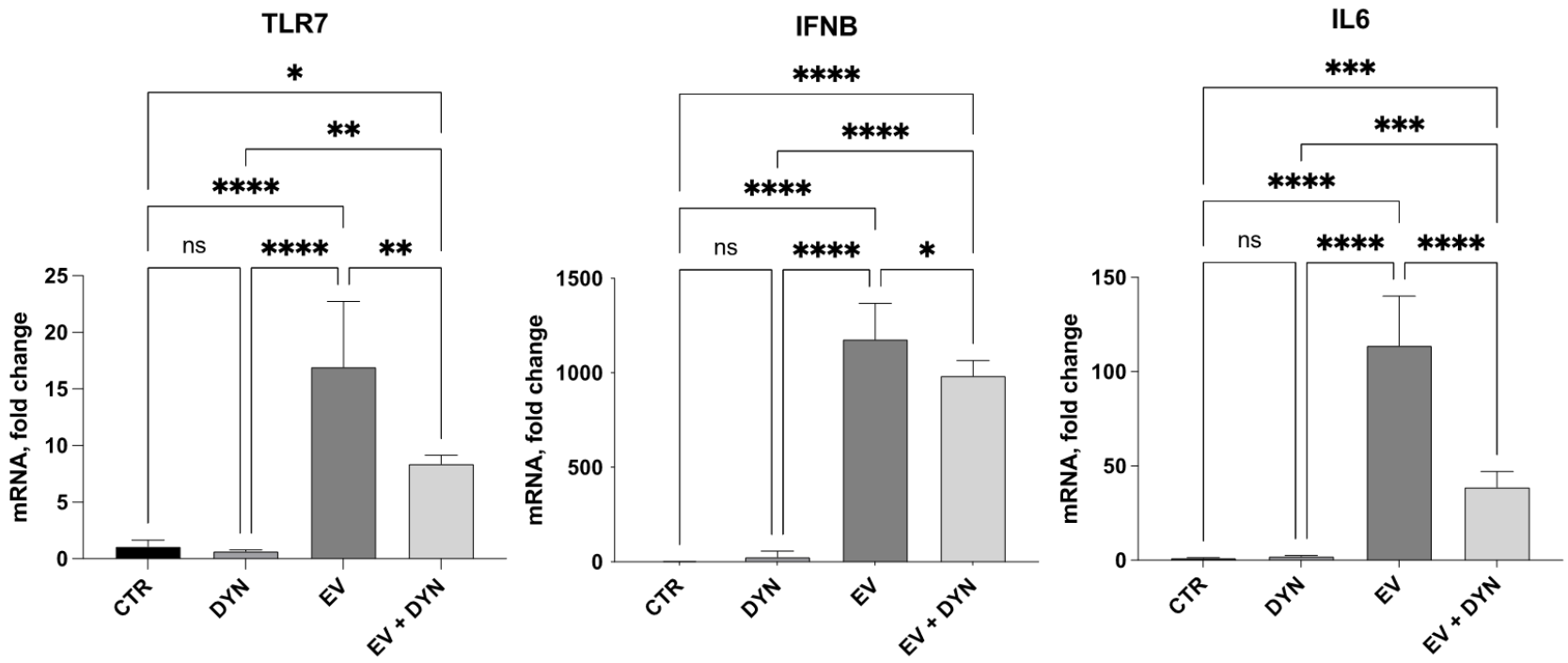
Flow Cytometry
RT qPCR

# ✓ Modulation of genes expression




→ The induced expression mediated by EVs of *TLR7, IFNβ* and *IL-6* genes is dependent on the internalization of EVs

→ Several pathways can be used by EVs to interact with host cells

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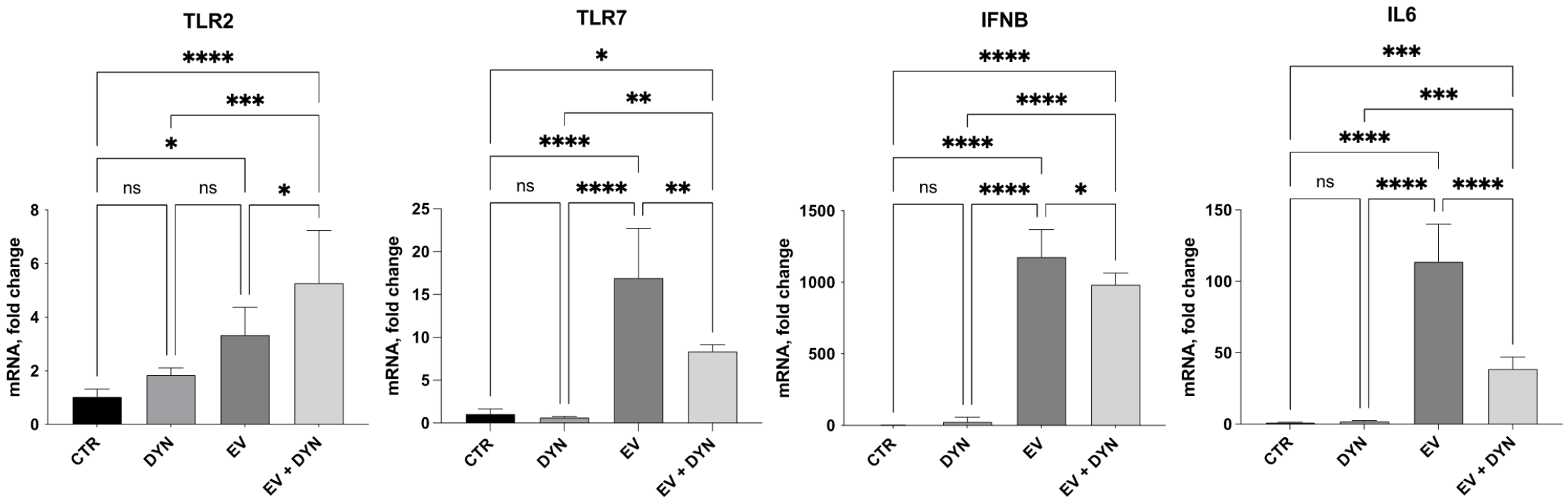


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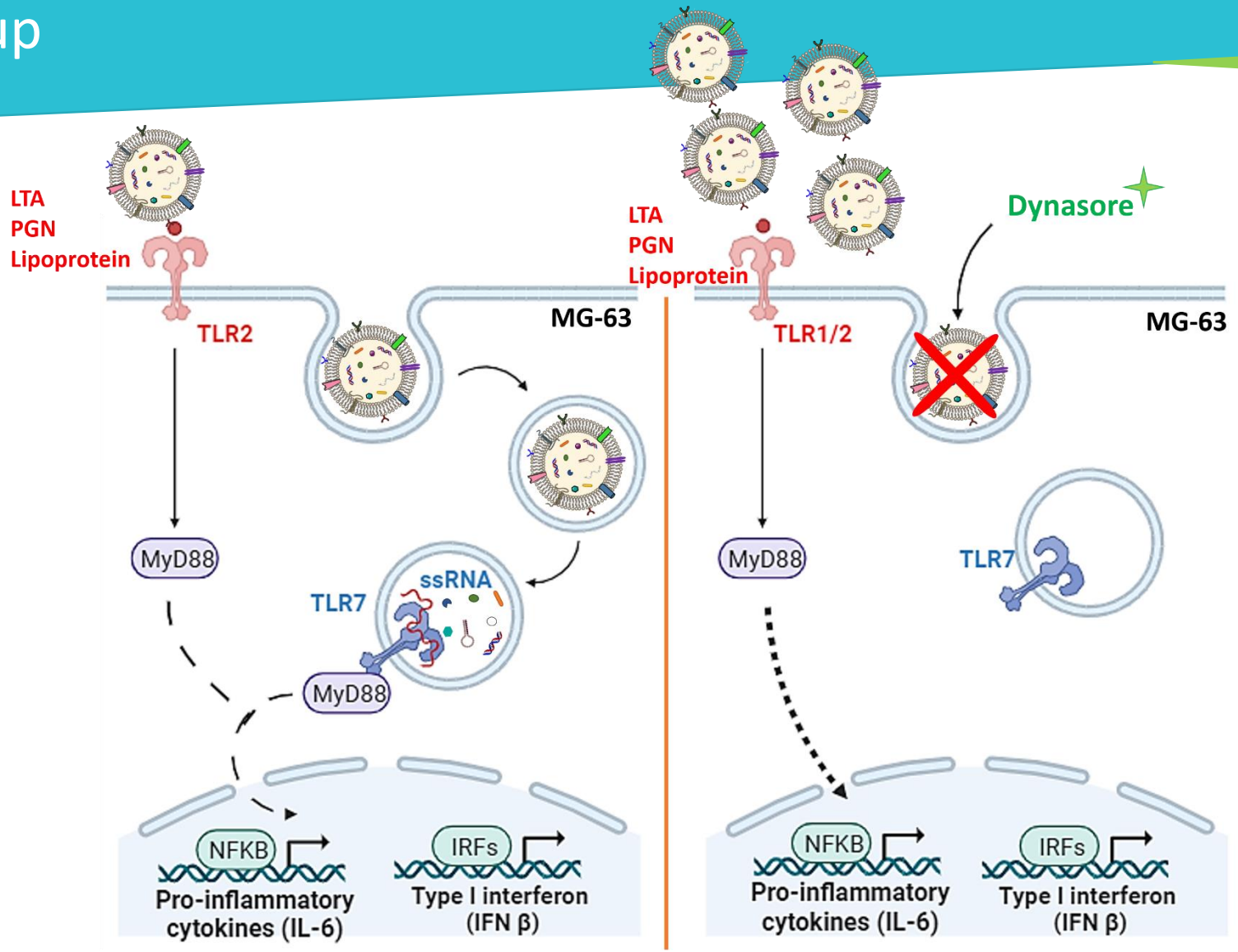
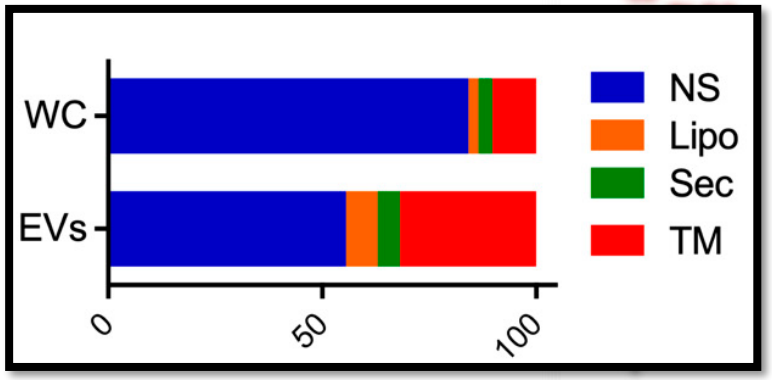
# ✓ To sum up



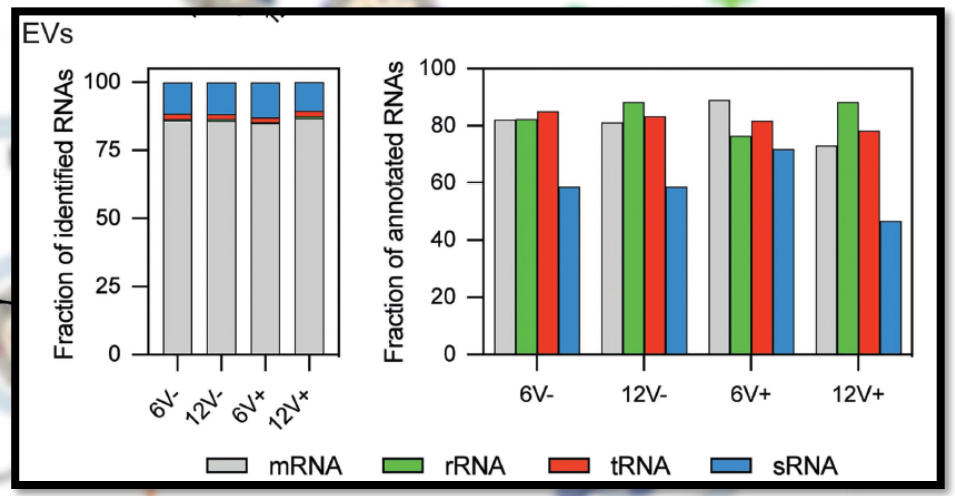

→ N315-derived EVs can trigger both extracellular and intracellular signaling pathway in host cells

→ Several EV components (proteins, RNA) can be involved in the interaction with host cells

# ✓ To sum up




*Luz and al., 2022*

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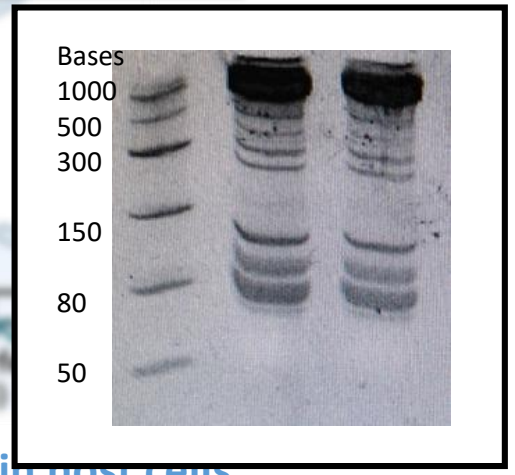


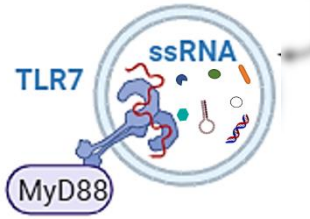


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# Thanks


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