New Mechanisms of Collateral Formation: The Role of Perivascular Cells and Extracellular RNA in Arteriogenesis

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Busan-South Korea, 2015

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Reactions and Players in Arteriogenesis

Arteriogenesis: Formation of collateral vessels upon ischemia
(Elisabeth Deindl, Markus Sperandio - Munich)

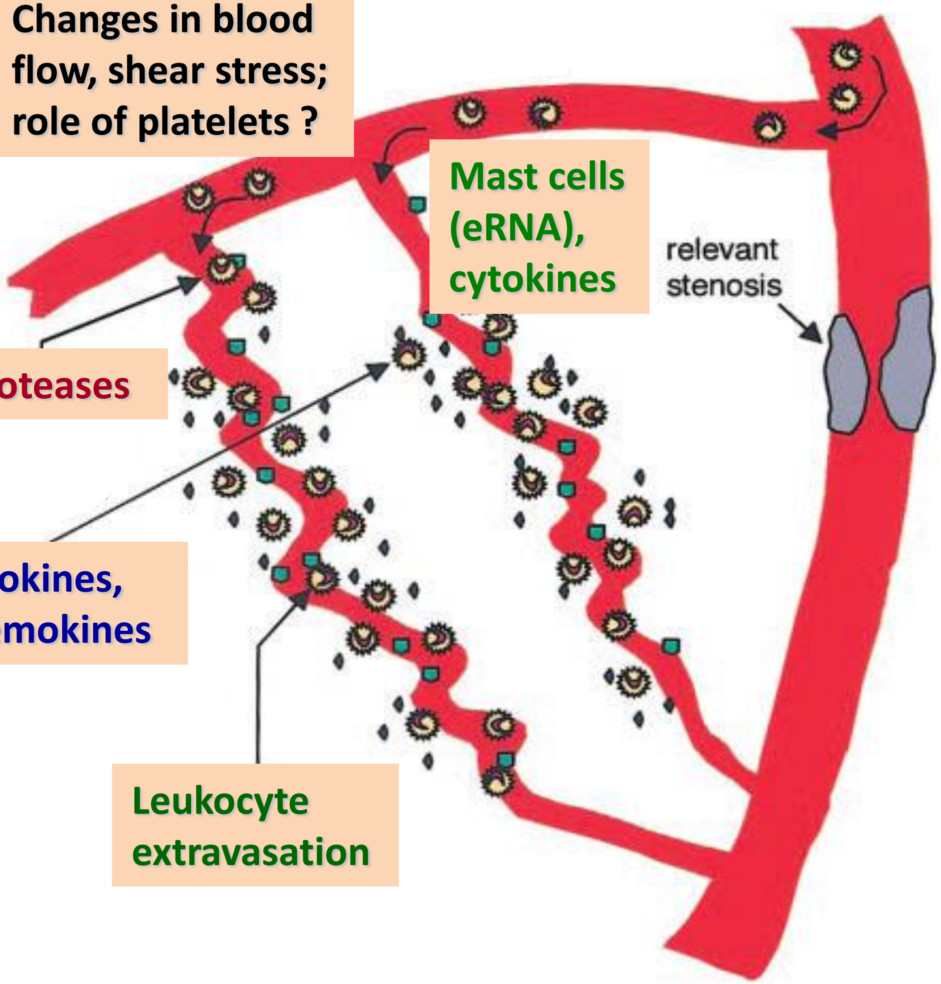
Changes in blood flow, shear stress; role of platelets ?

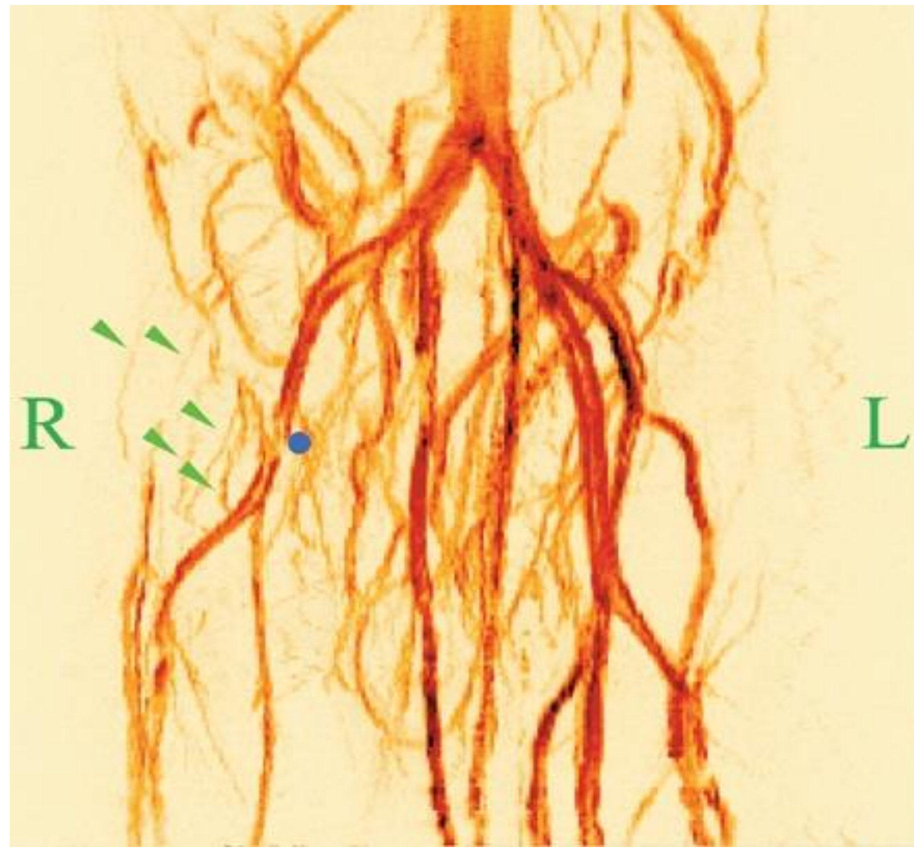
Mast cells (eRNA), cytokines

Proteases

Cytokines, chemokines

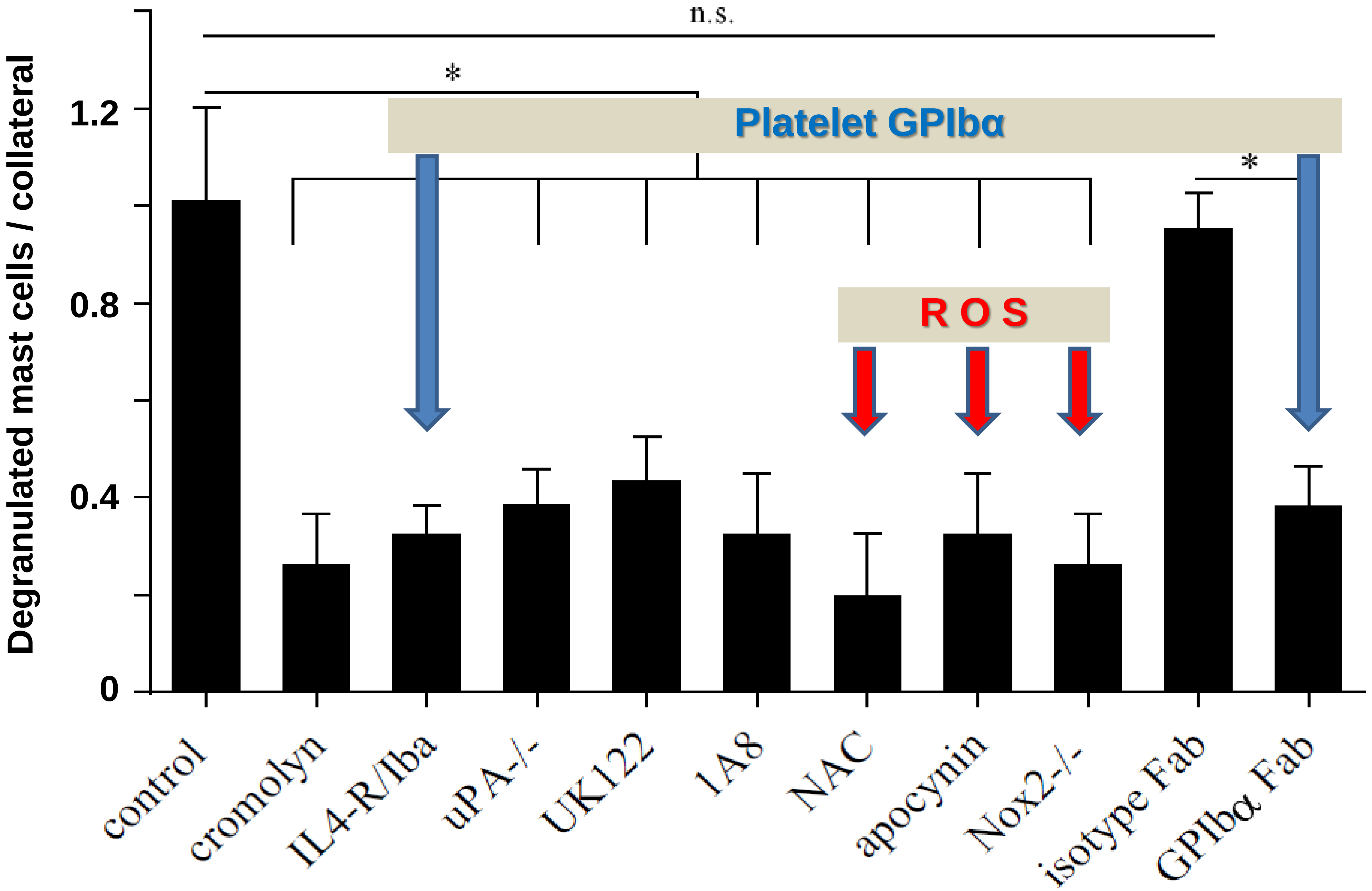
Leukocyte extravasation



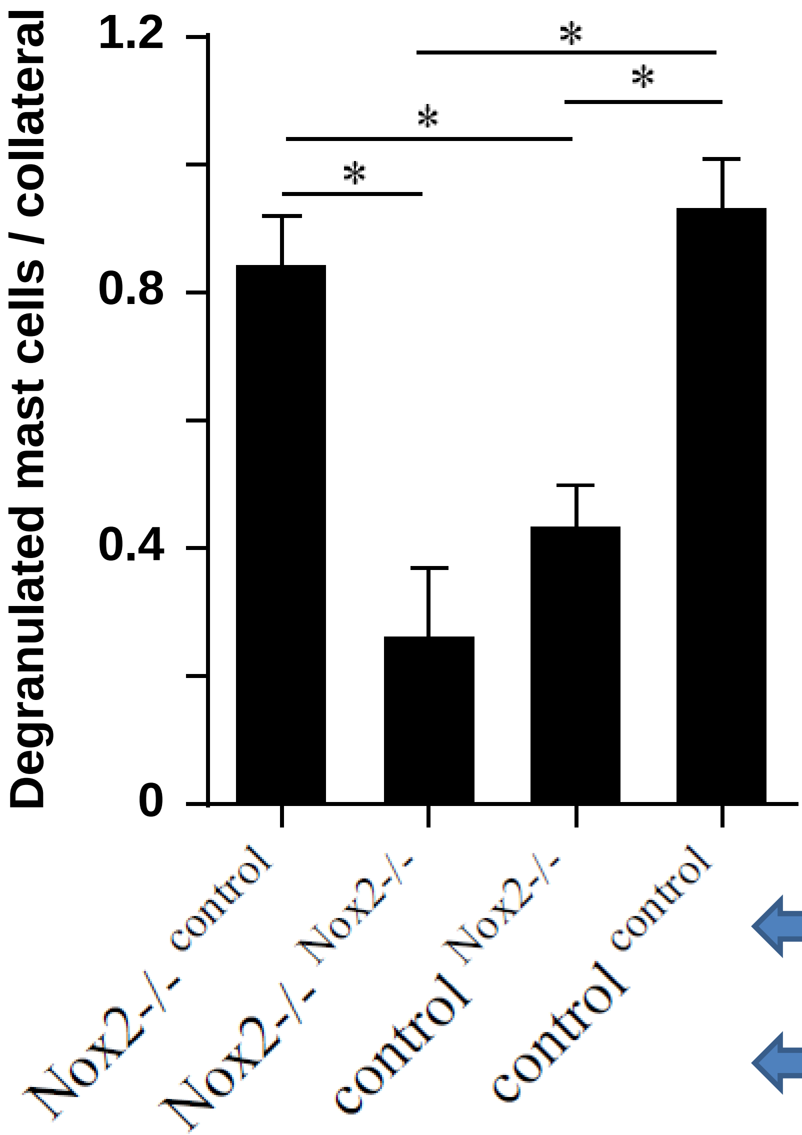


Ligation of *A. femoralis (R)*; sham-operation (L); quantification by Laser-Doppler analysis

Platelets and Reactive Oxygen Species (ROS) Contribute to Mast Cell Stimulation and Arteriogenesis



Reactive Oxygen Species (ROS)-dependent Mast Cell Stimulation: The Role of Nox2-deficiency



Transplanted bone marrow

Mouse genotype

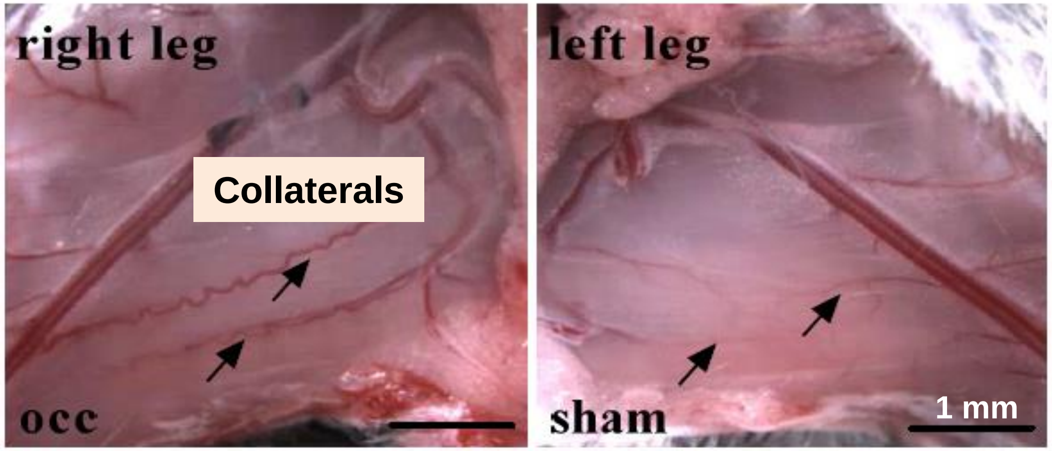
Day 1 following *femoral artery ligation*:

Number of perivascular, degranulated mast cells

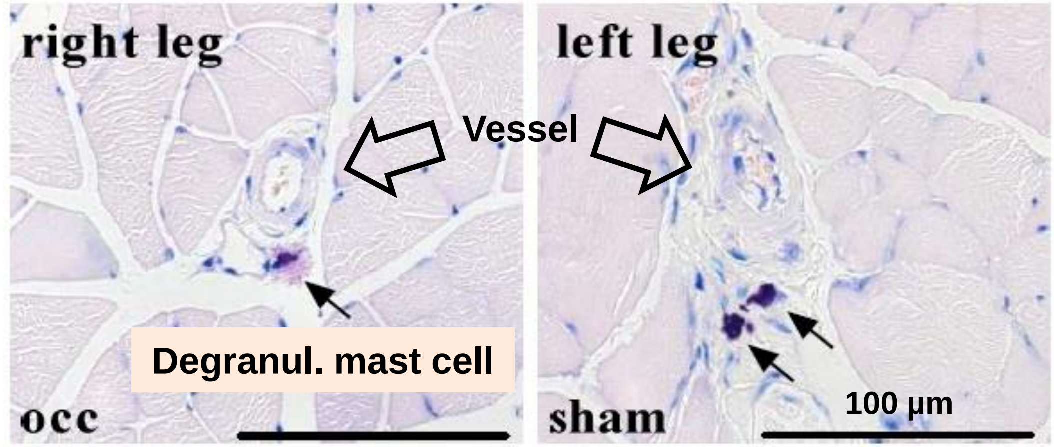
Chandraratne et al., ATVB 2015

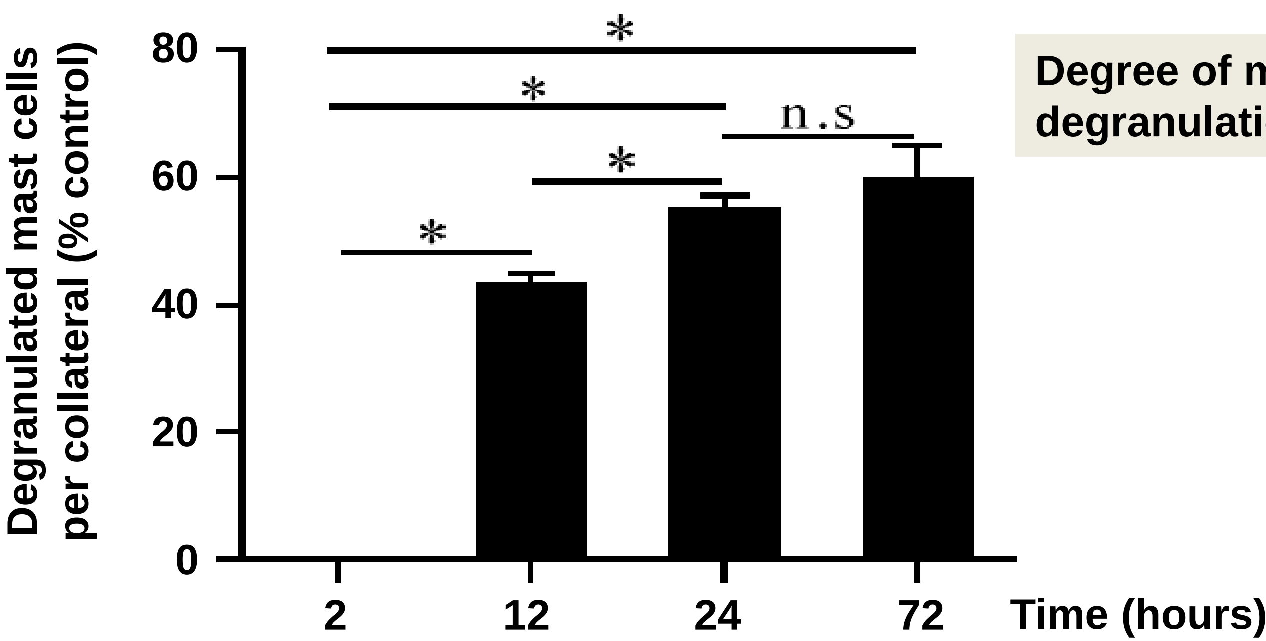
Mast Cell Degranulation Promotes Arteriogenesis (I)

Collateral vessel formation



Pericellular mast cell degranulation



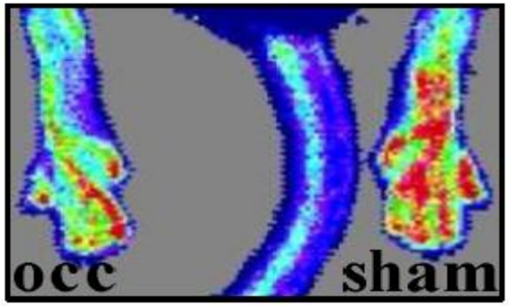


Degree of mast cell degranulation

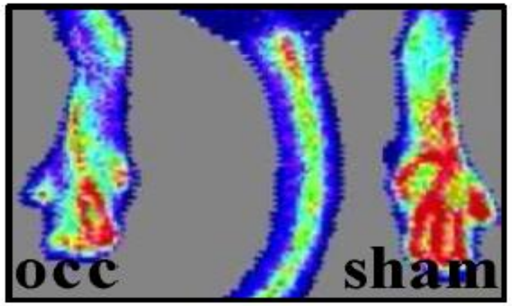
Chillo et al., 2014 (unpublished data)

Mast Cell Degranulation Promotes Arteriogenesis (II)

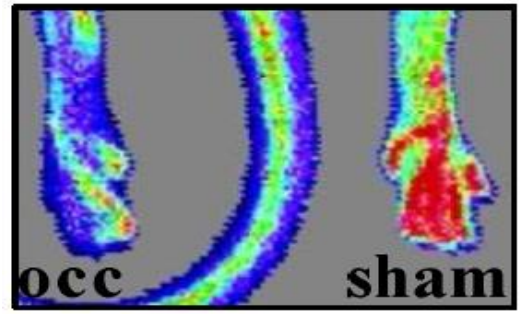
Control



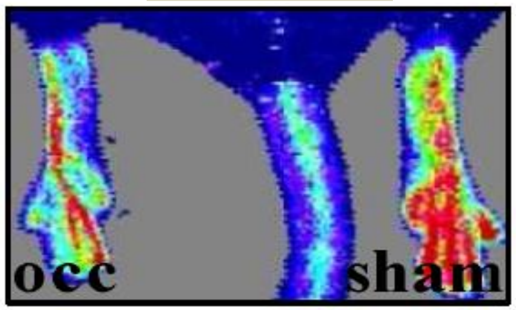
Compound 48/80



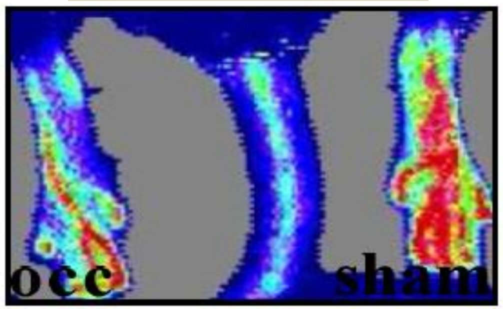
Cromolyn



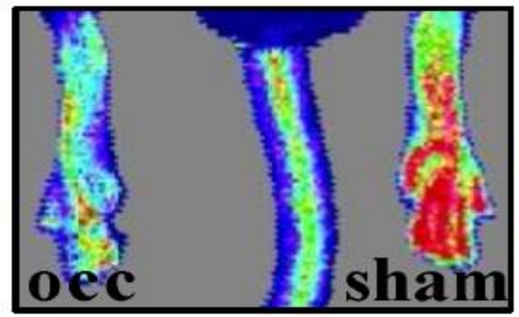
Diprotin A



Stem cell factor



Cromolyn + Diprotin A

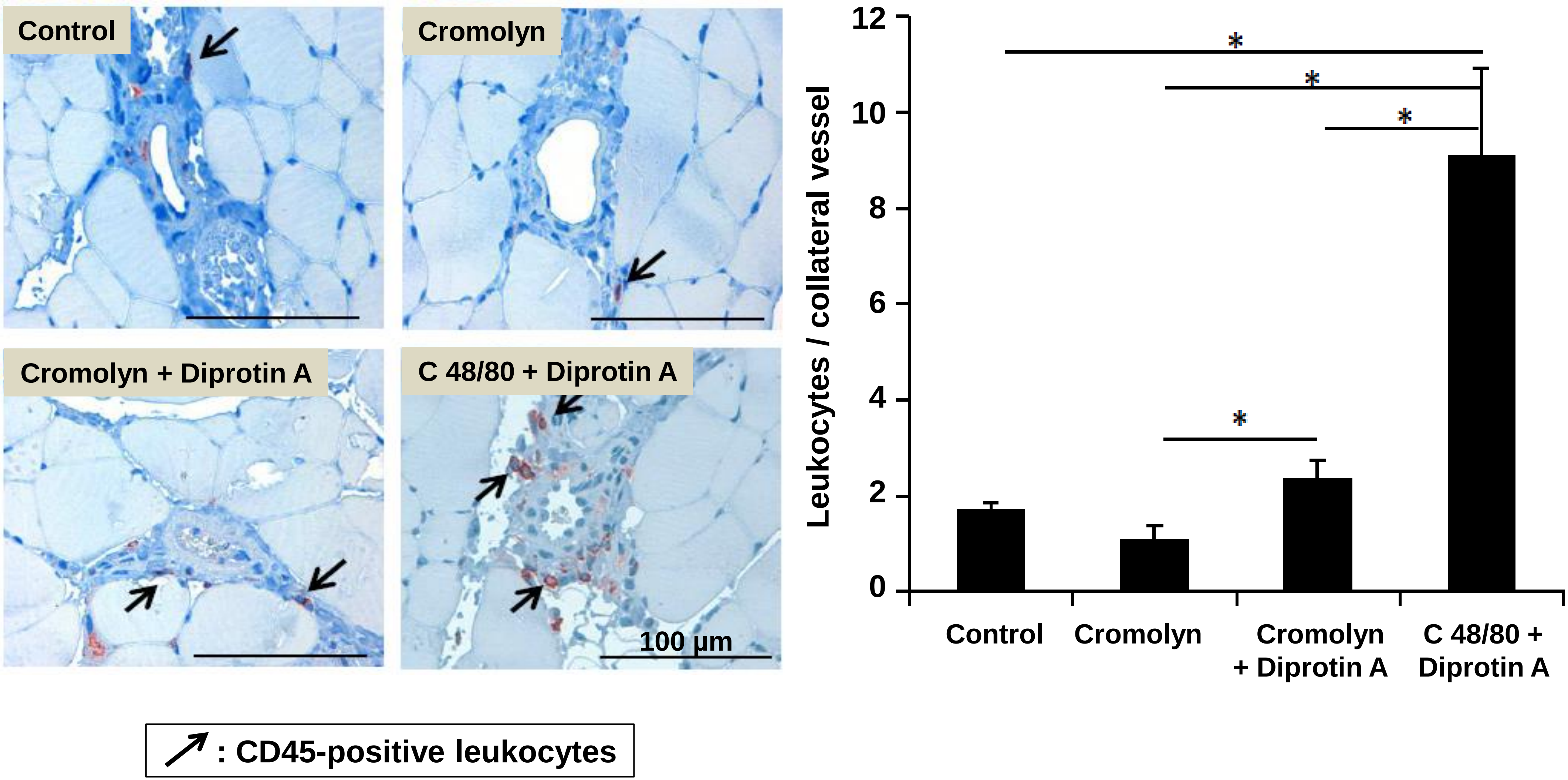


- Control
- Compound 48/80
- Diprotin A
- Stem cell factor
- Comp. 48/80 + Diprotin A

- Control
- Diprotin A
- Cromolyn
- Cromolyn + Diprotin A

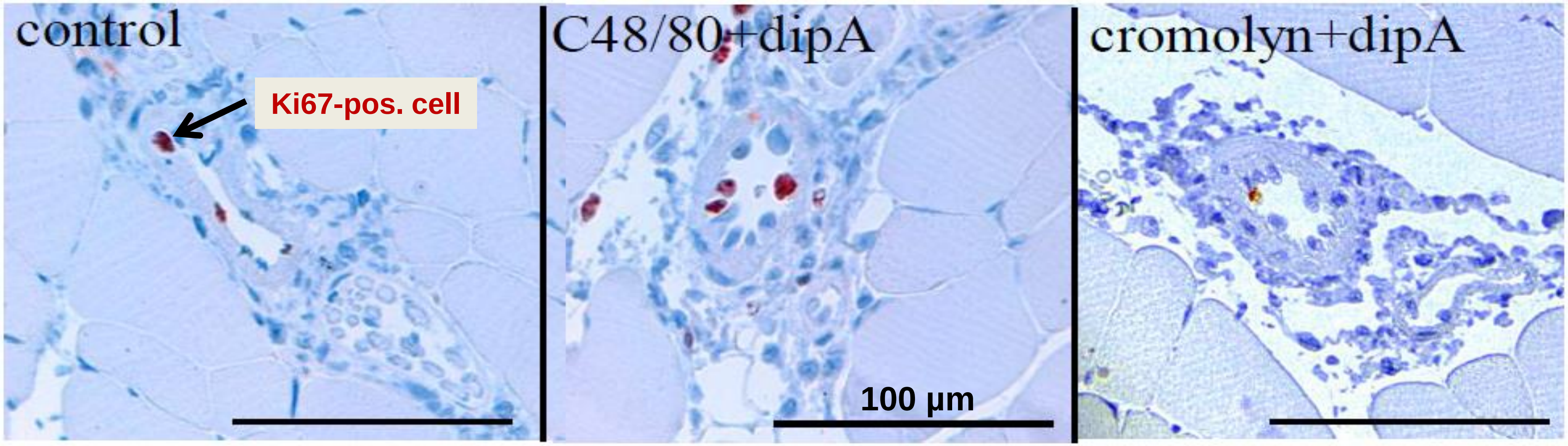
Chillo et al., 2014 (unpublished data)

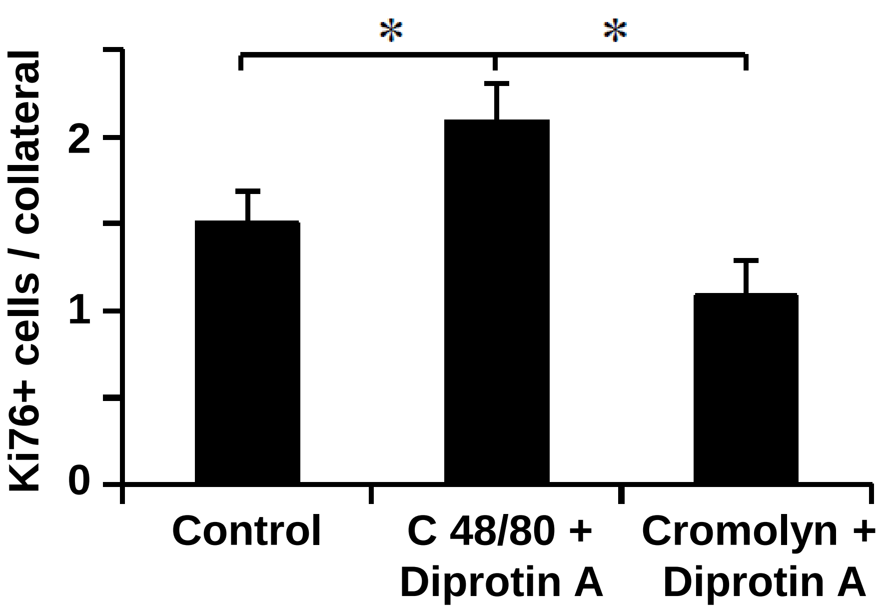
Mast Cell Products Recruit Leukocytes in Arteriogenesis



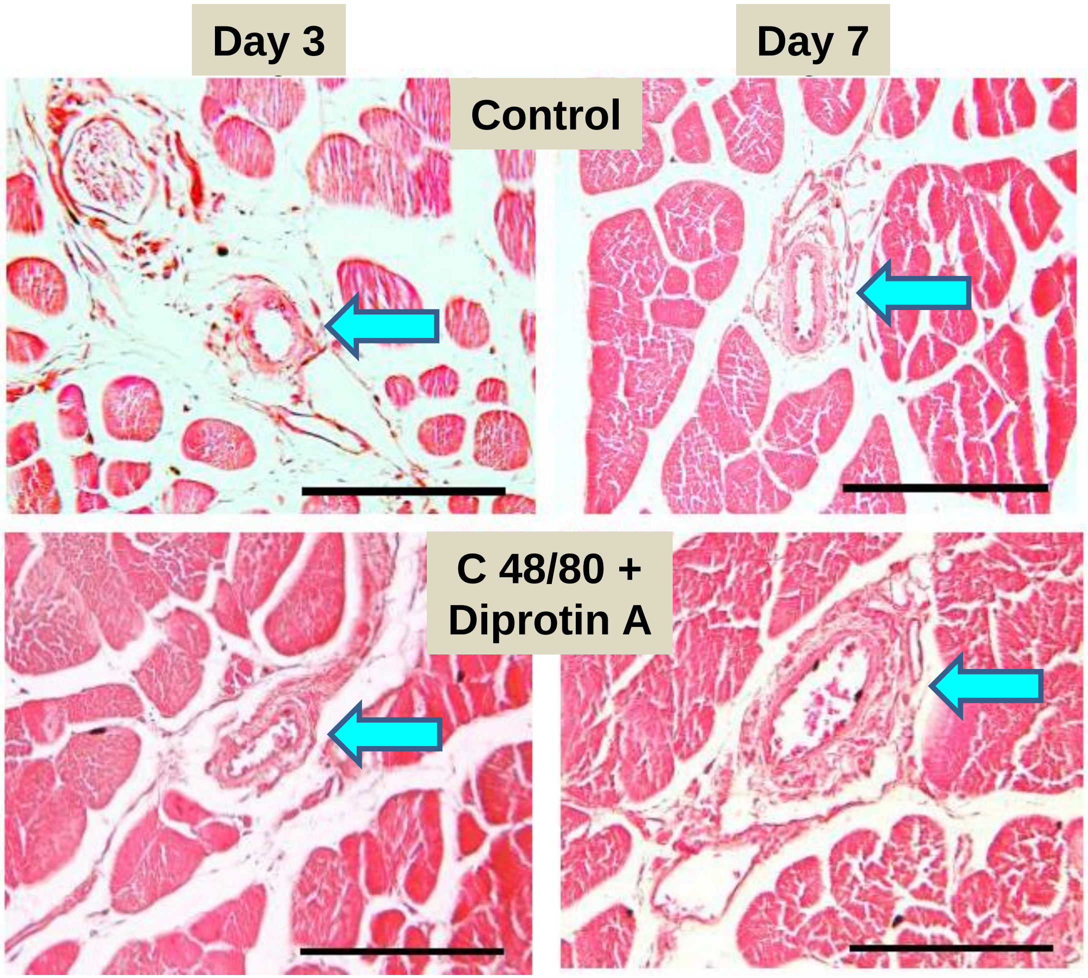
Chillo et al., 2014 (unpublished data)

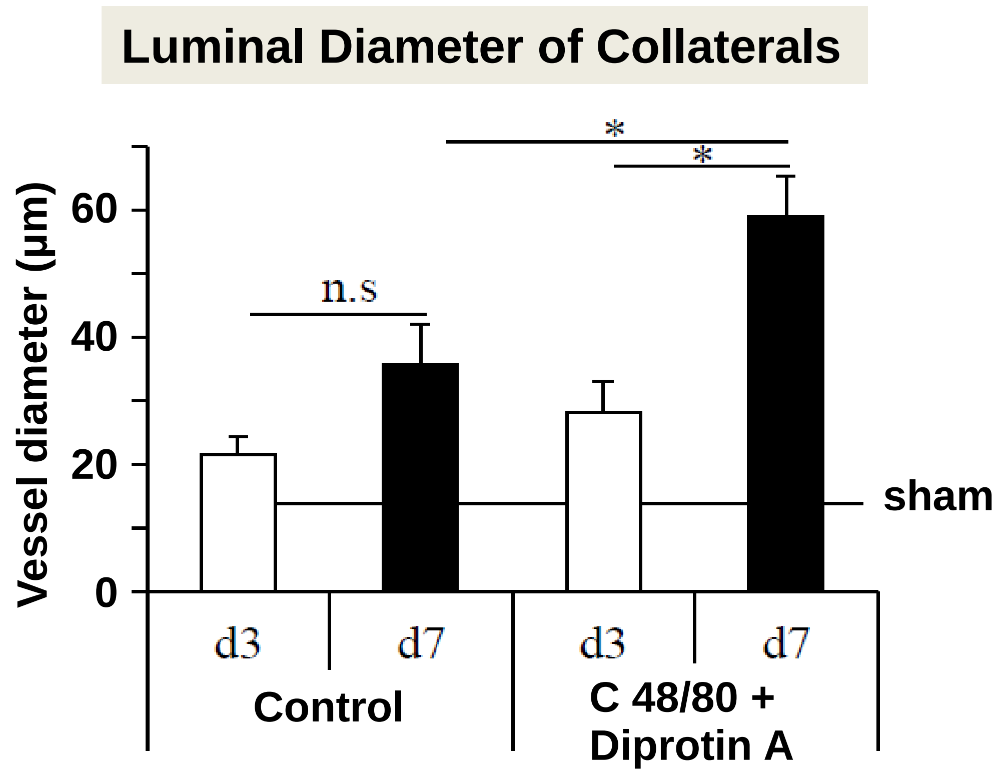
Mast Cell Degranulation Promotes Collateral Vessel Growth





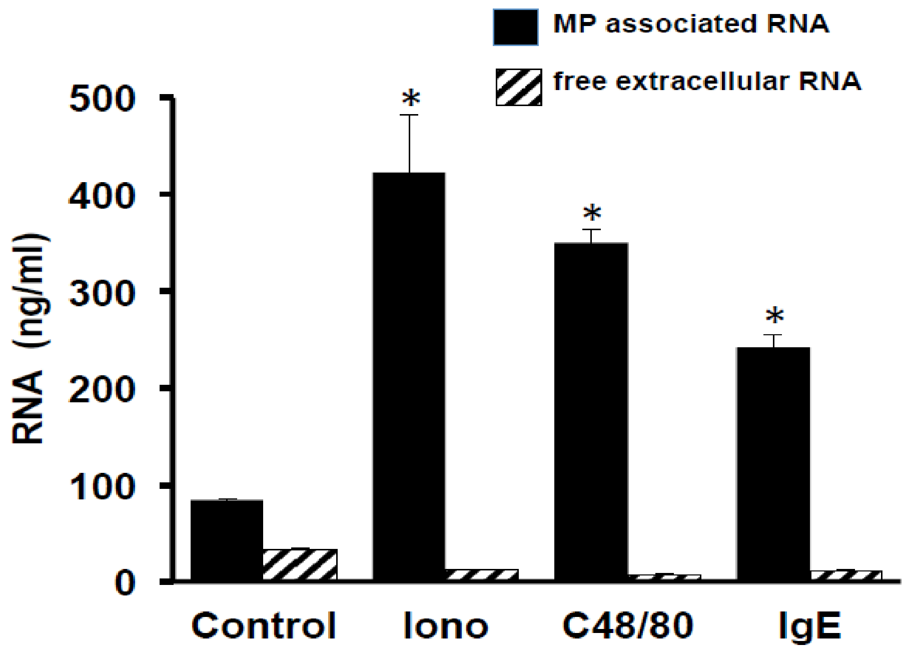
Mast Cell Degranulation Promotes Collateral Vessel Enlargement

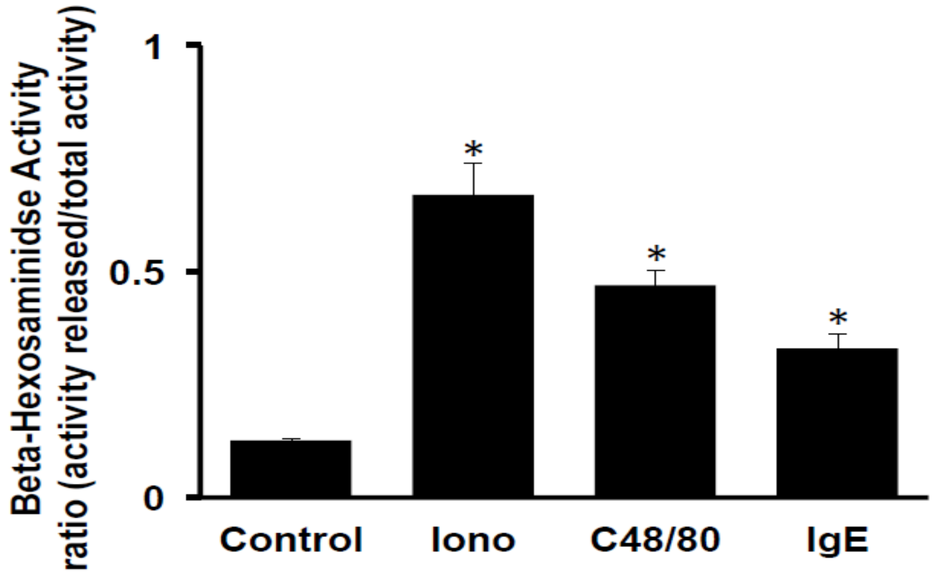


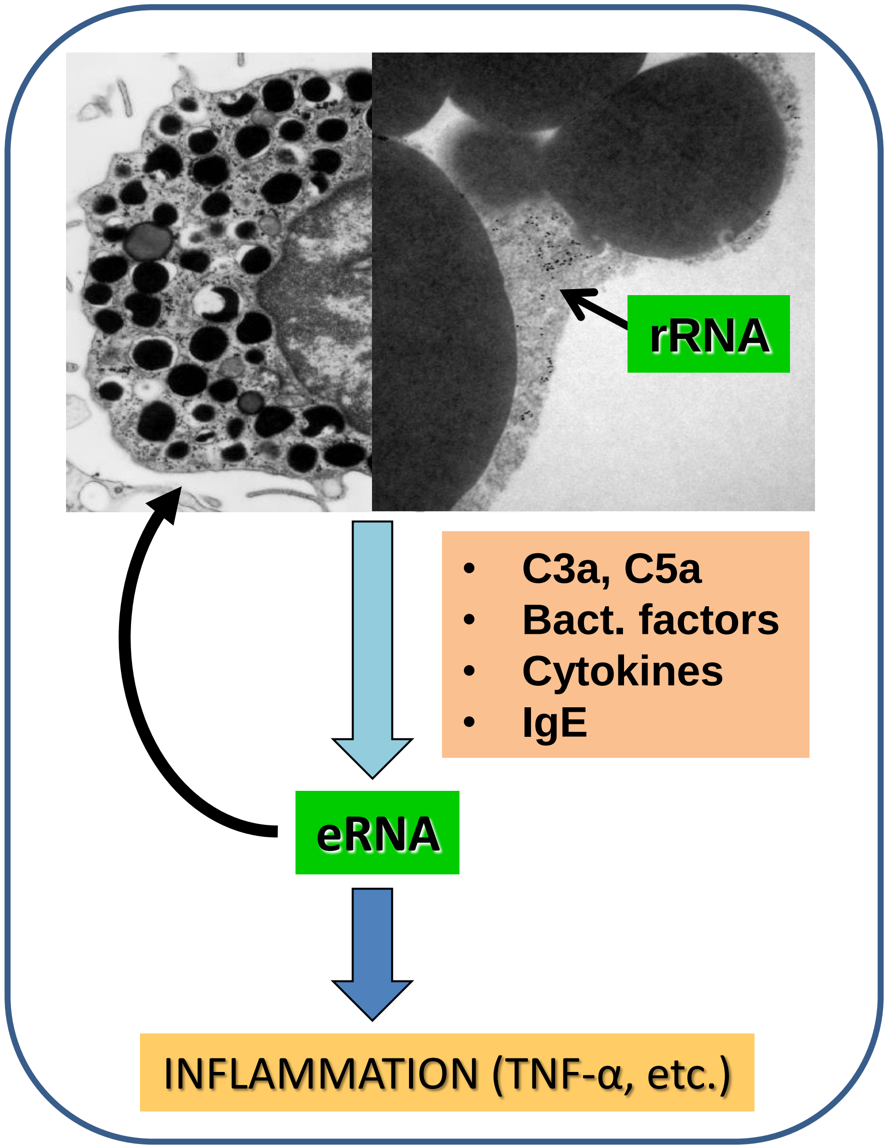


Chillo et al., 2014 (unpublished data)

Mast cells: Major Source of Extracellular RNA

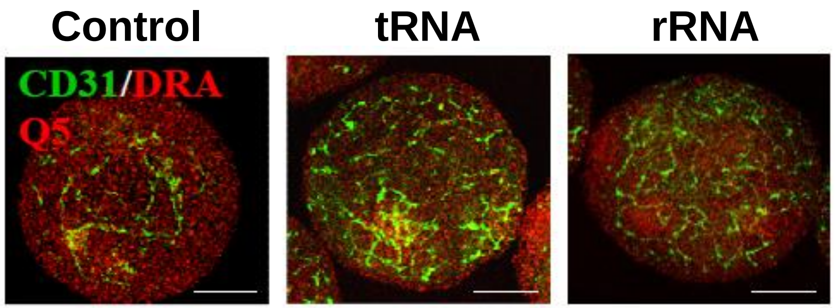


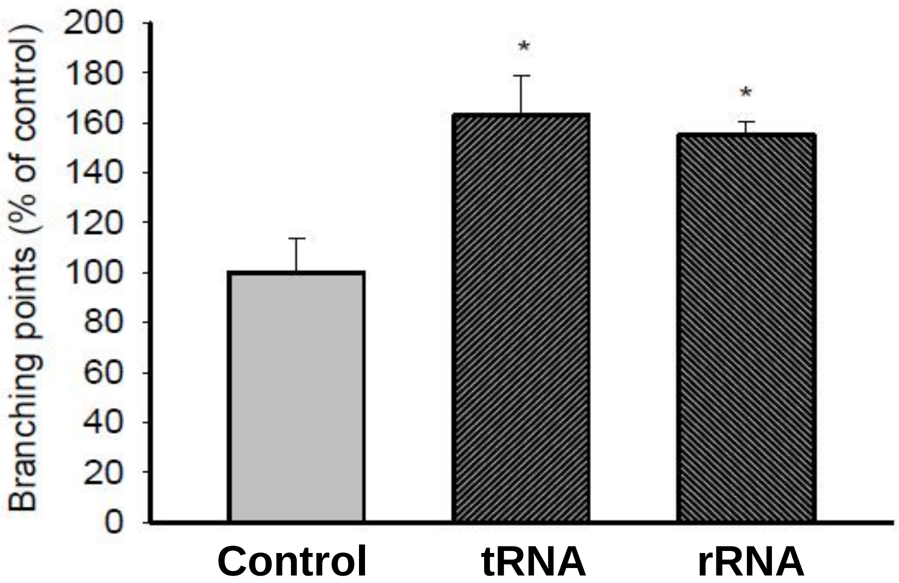


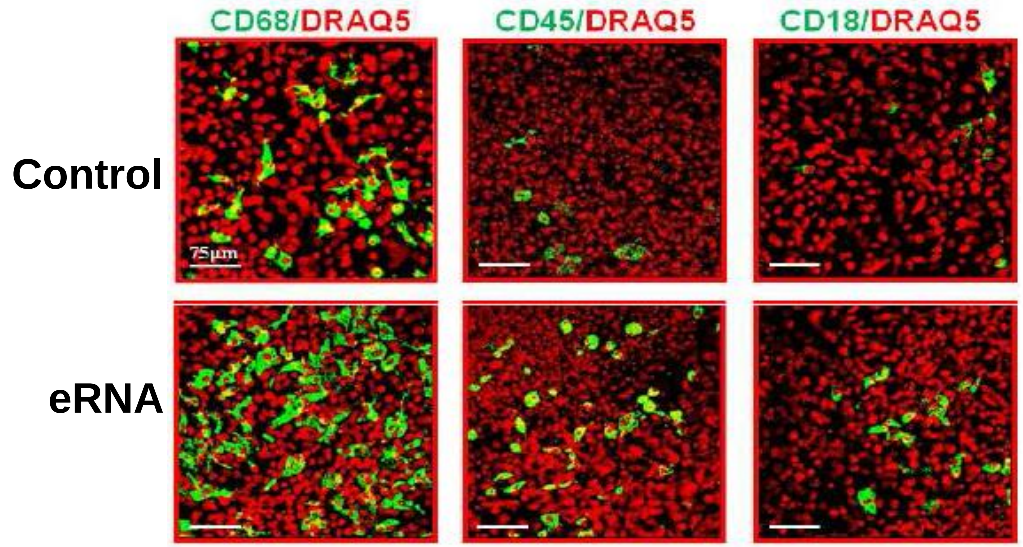


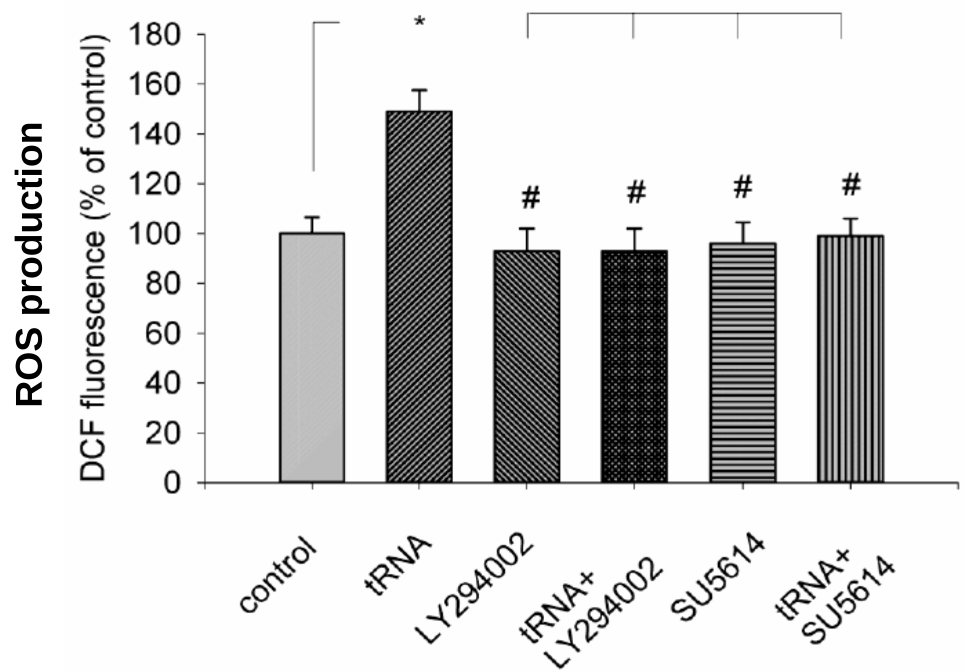
Fischer et al., 2014 (unpublished data)

Induction of Vasculogenesis/Angiogenesis by Extracellular RNA



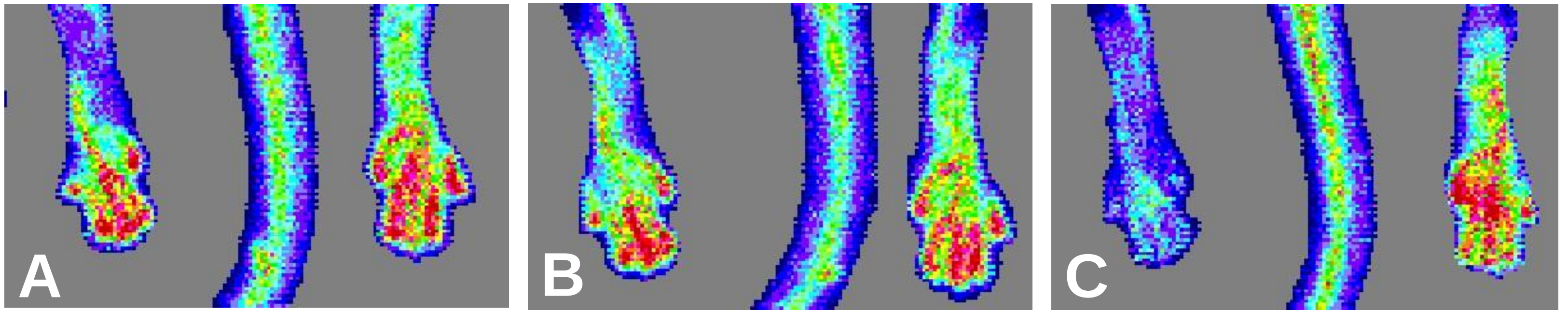


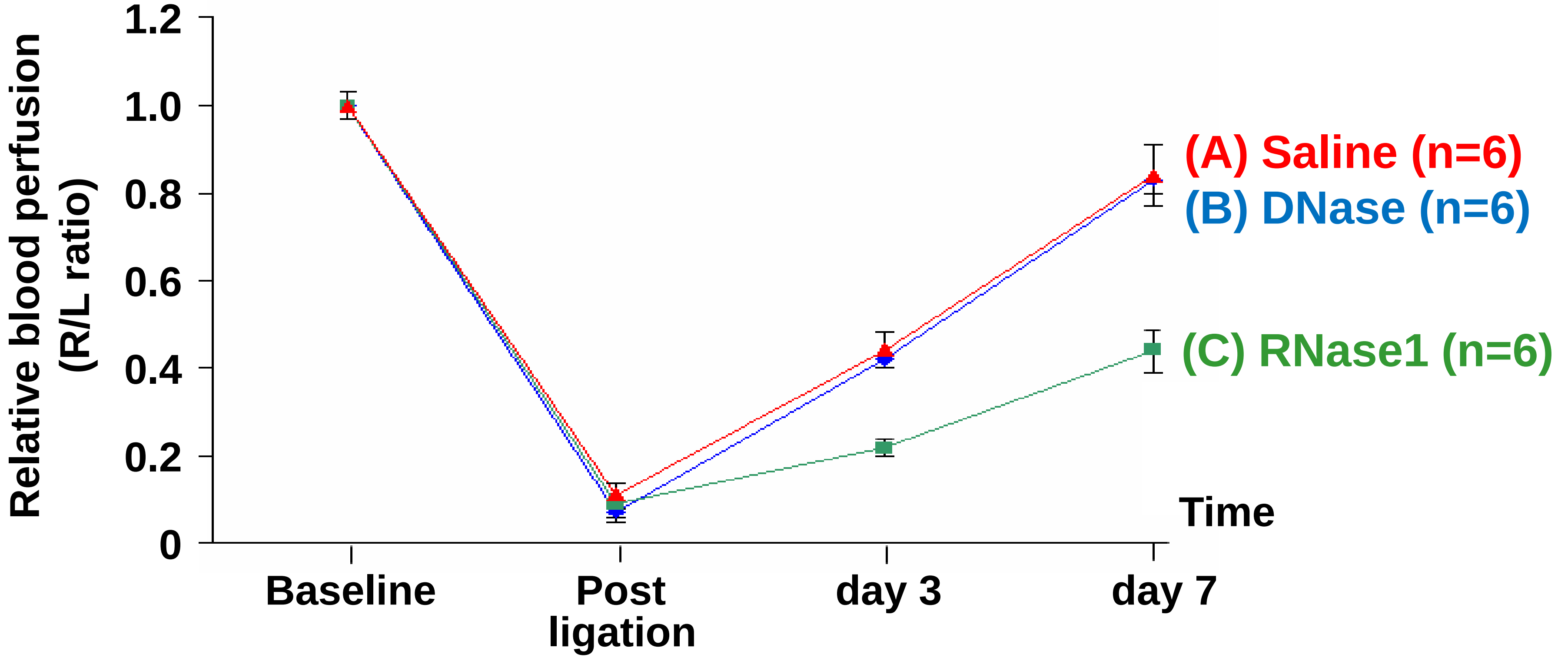




Sharifpanah et al., Free Rad Biol Med 2015

The Extracellular RNA – RNase1 System and Arteriogenesis

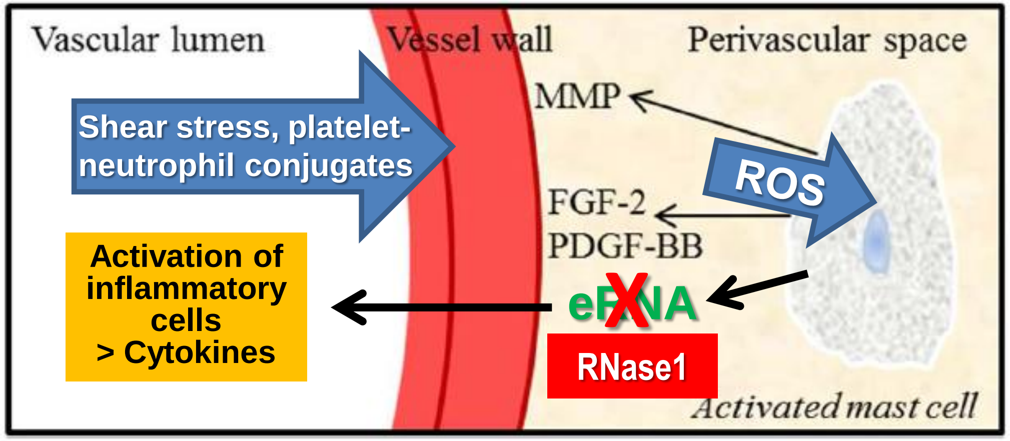




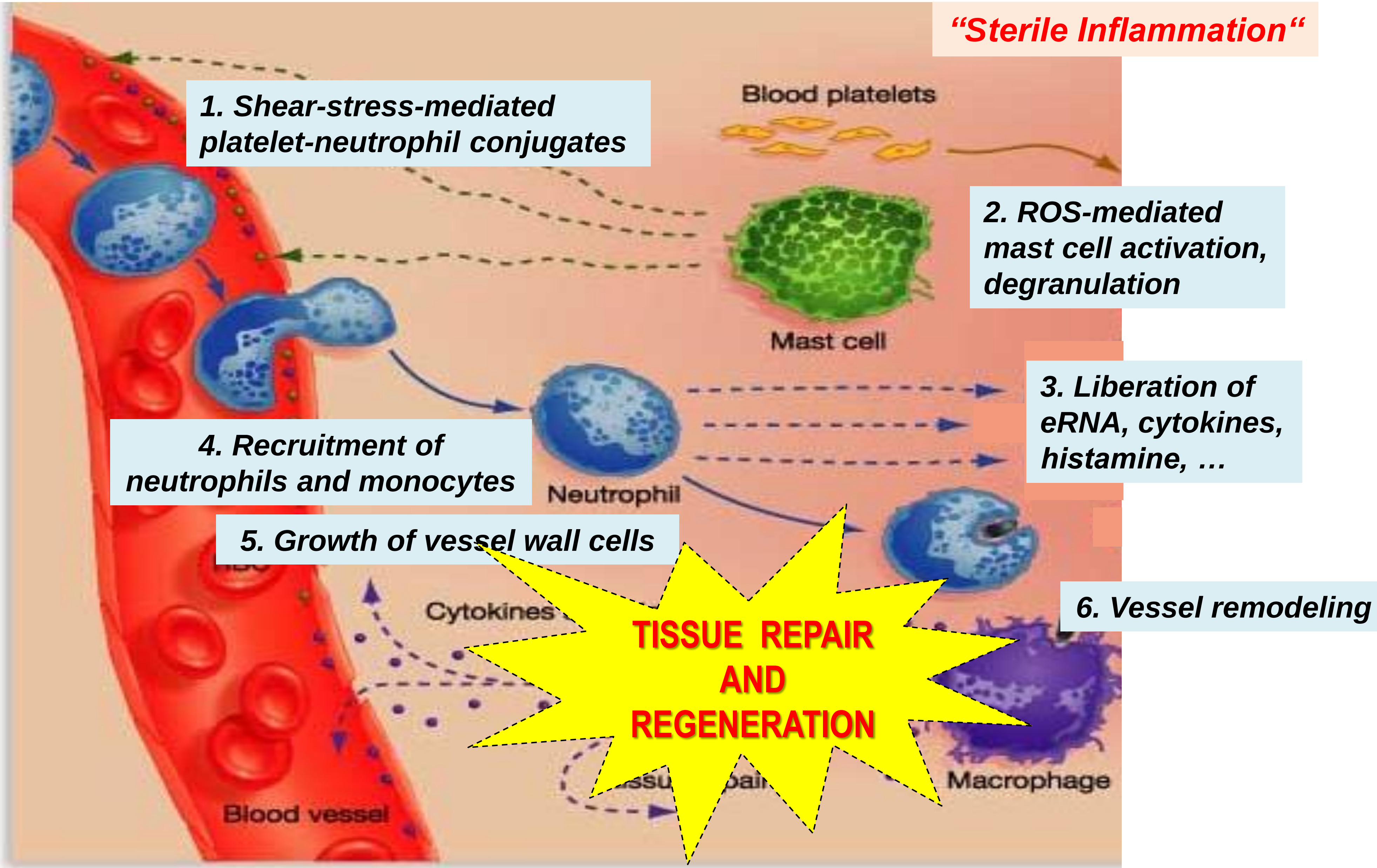
Novel molecular relations between extracellular RNA, mast cells and arteriogenesis

Mast Cells Orchestrate Arteriogenesis

**1. Mast cell degranulation >>>
Vascular remodeling & cell proliferation**



Innate Immunity Reactions as Template for Tissue Regeneration



Acknowledgements ....

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Omary Chillo

Elisabeth Deindl

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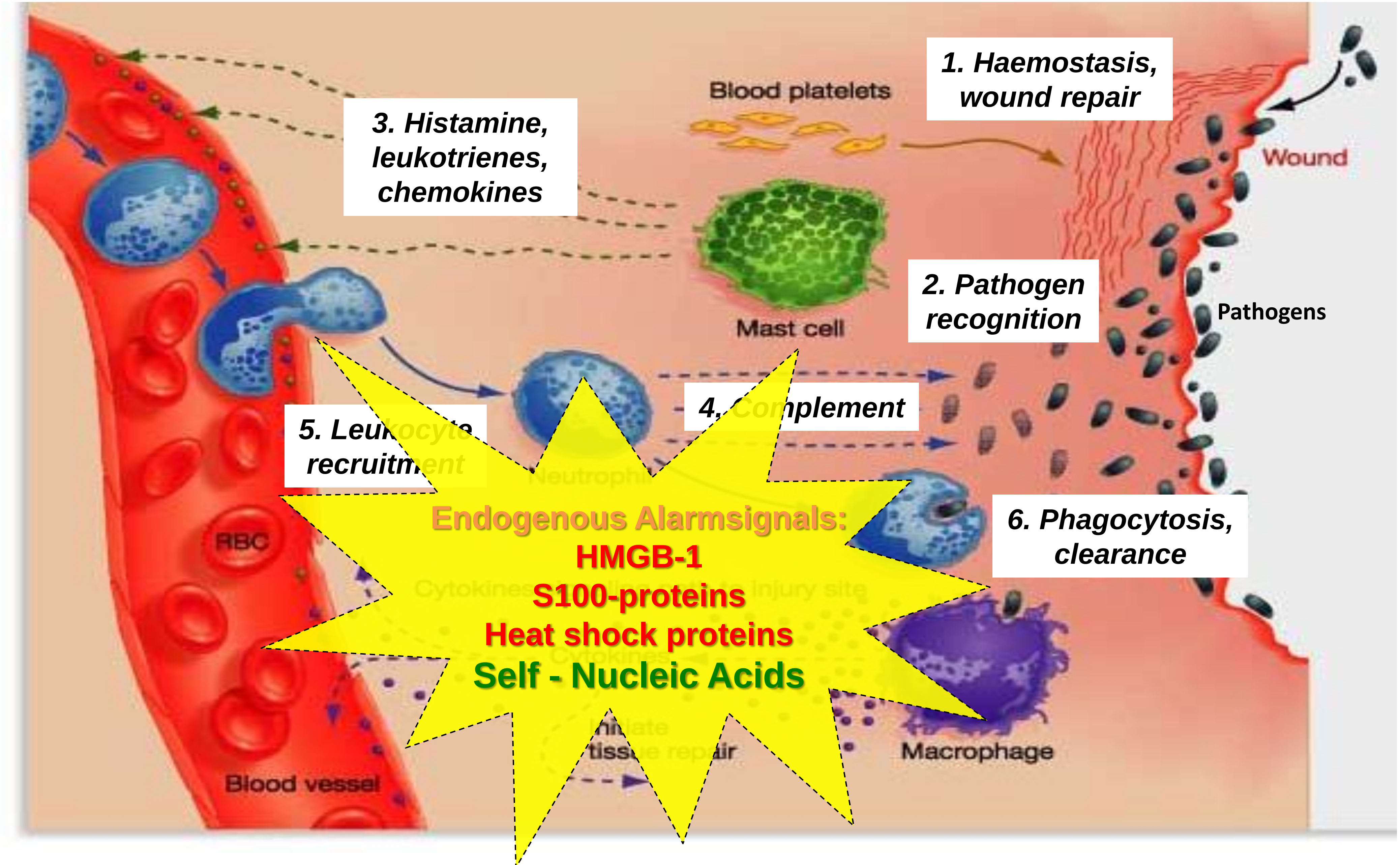
SFB-TR 84 Innate Immunity of the Lung

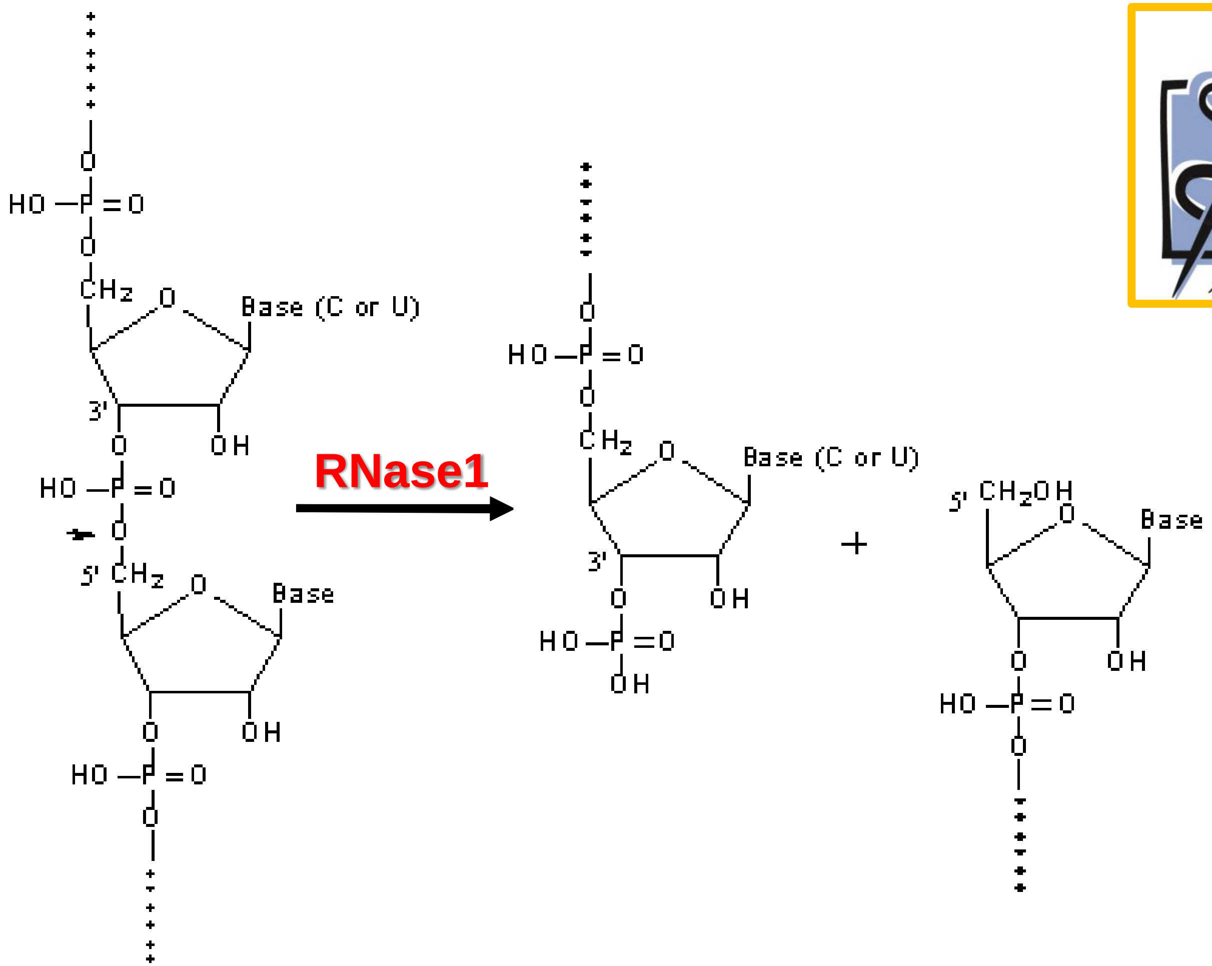
von behring | röntgen | stiftung

DFG

Players and Reactions in Innate Immunity and Defense



RNase1-mediated Hydrolysis of RNA: Generation of Vaso- and Tissue-protective Products?



Nucleotidases

(CD73)

Nucleosides (e.g. Adenosine)

RNA → **ss-Oligonucleotides** + **3`Nucleotides** / **5`Nucleotides**

Damaging Nature of Extracellular RNA and Induction of Endogenous Inflammatory Pathways

- Atherosclerosis
- Acute inflammation
- Ischemia/reperfusion injury
- Stroke, edema formation
- Tumor development
- Transplantation (?)

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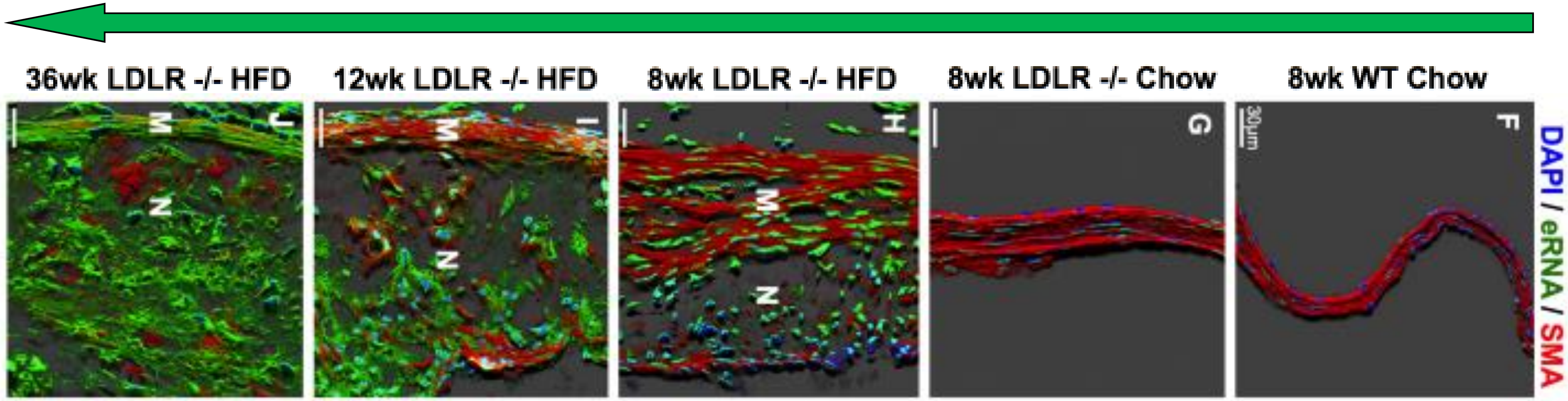
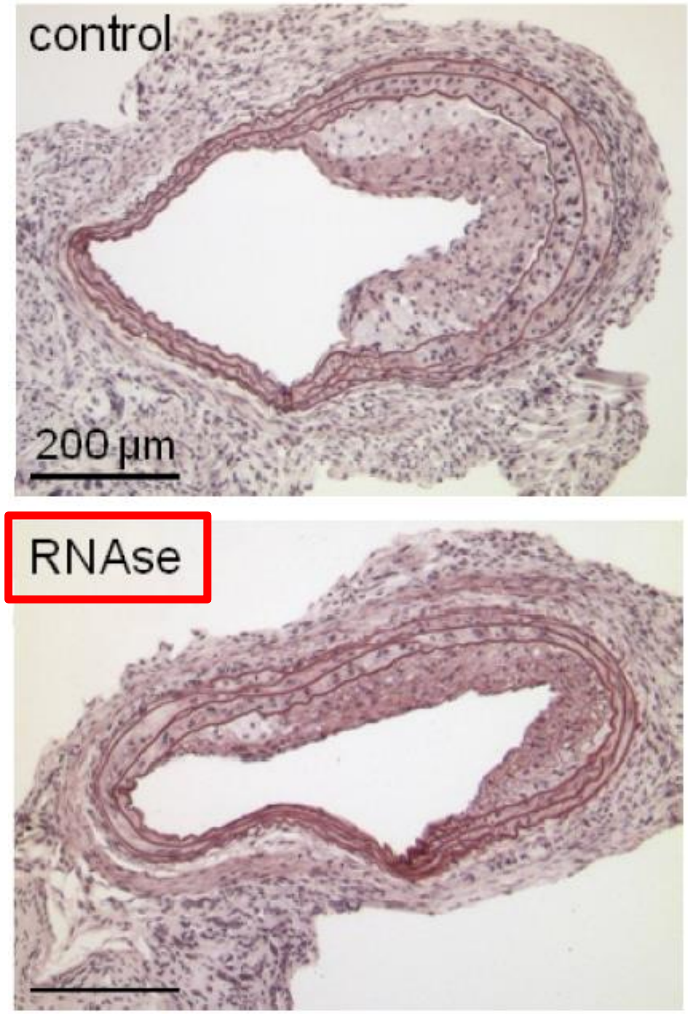
Development of atherosclerotic lesions (LDLR-/- mice) in an eRNA-dependent manner



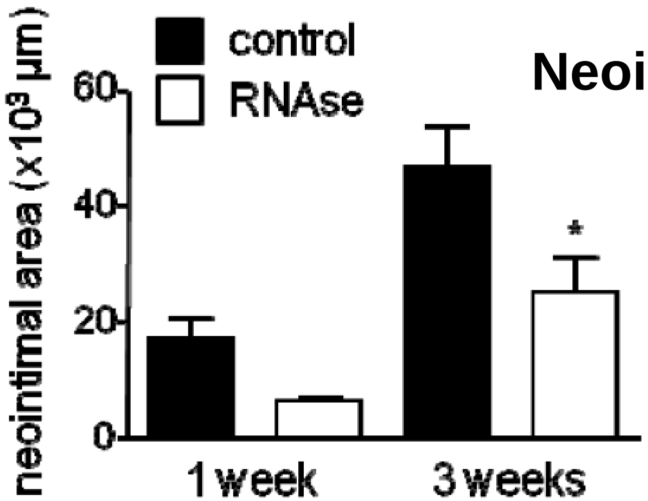
Simsekyilmaz et al., Circulation 2014

RNase1 Treatment of Atherosclerosis-prone Apo-E$^{-/-}$ Mice

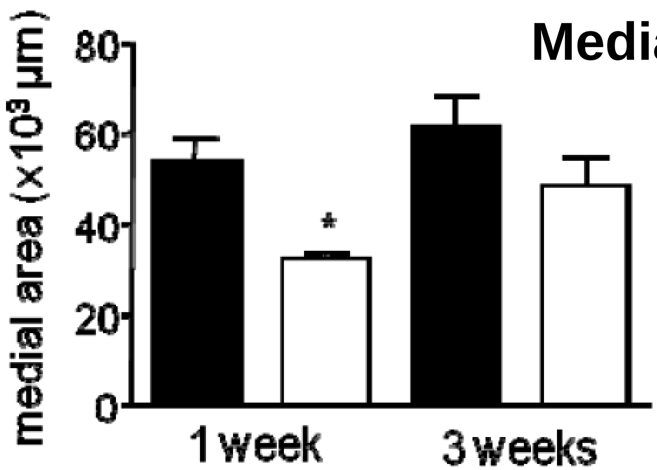
Atherosclerosis-prone apo-E$^{-/-}$ mice: Wire-induced vessel injury
(Alma Zernecke, Aachen/Würzburg)



Neointimal area

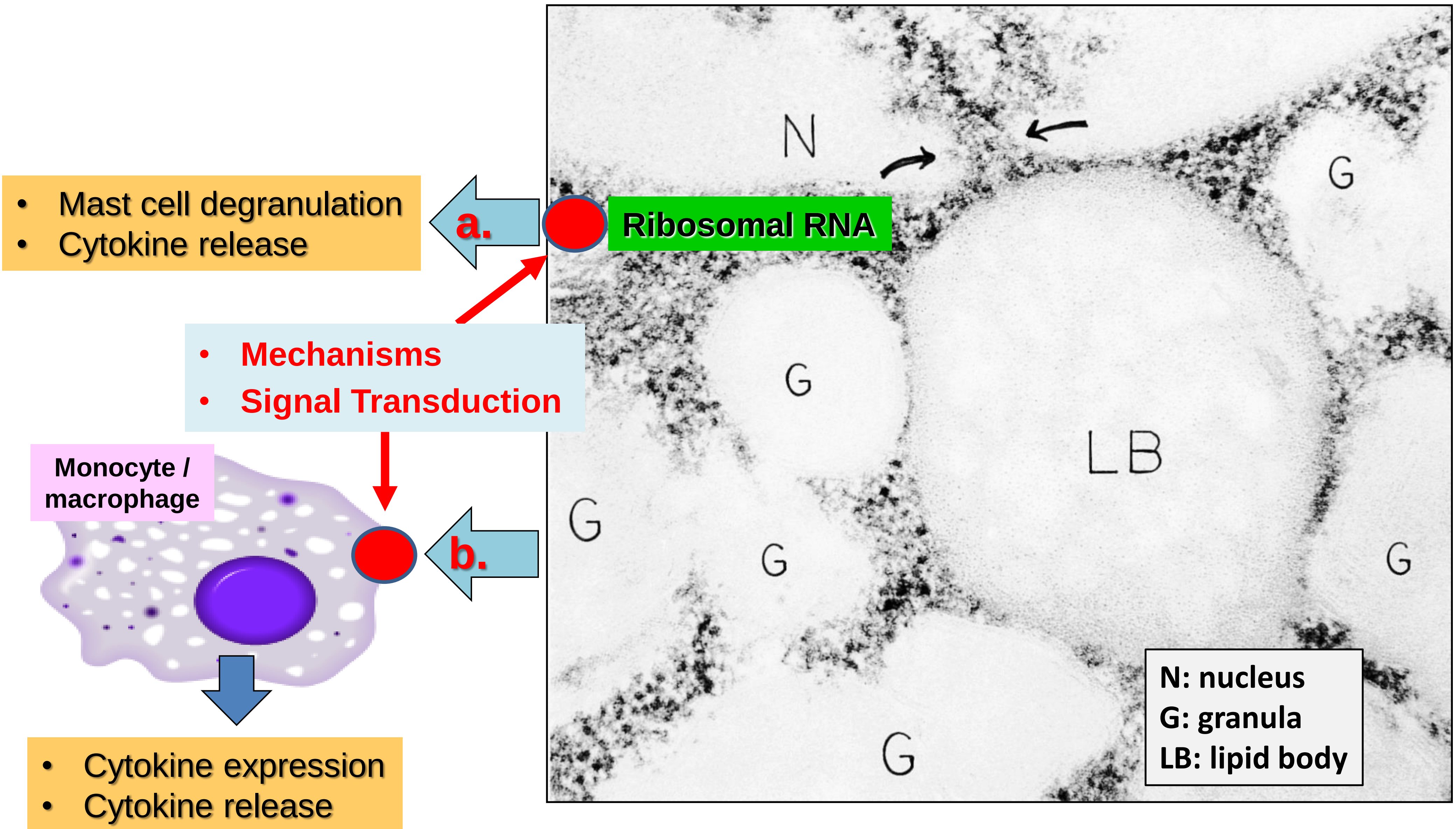


Medial area



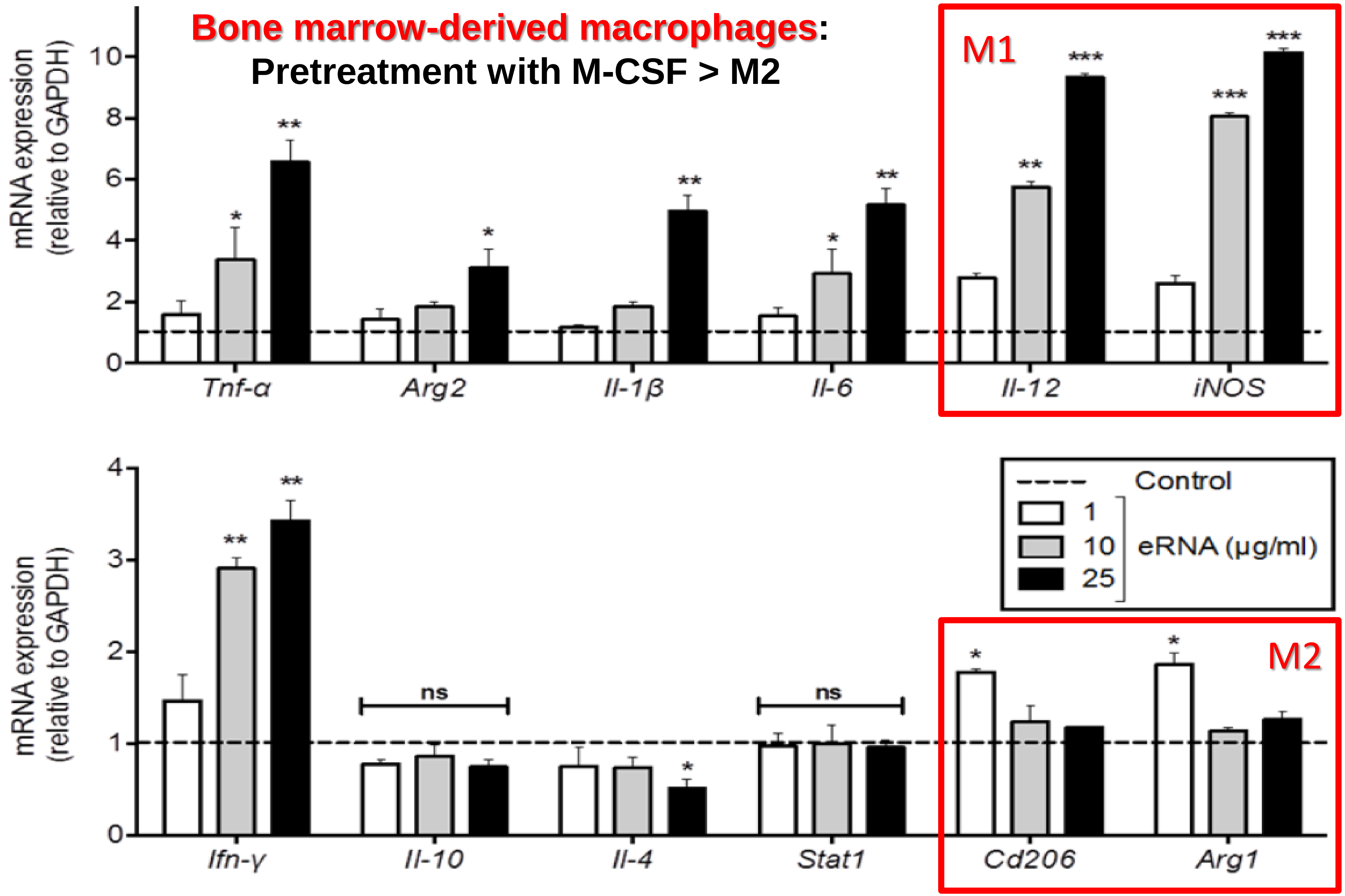
Simsekyilmaz et al., Circulation 2014

Mast Cell Degranulation, eRNA Release and Cytokine Storm



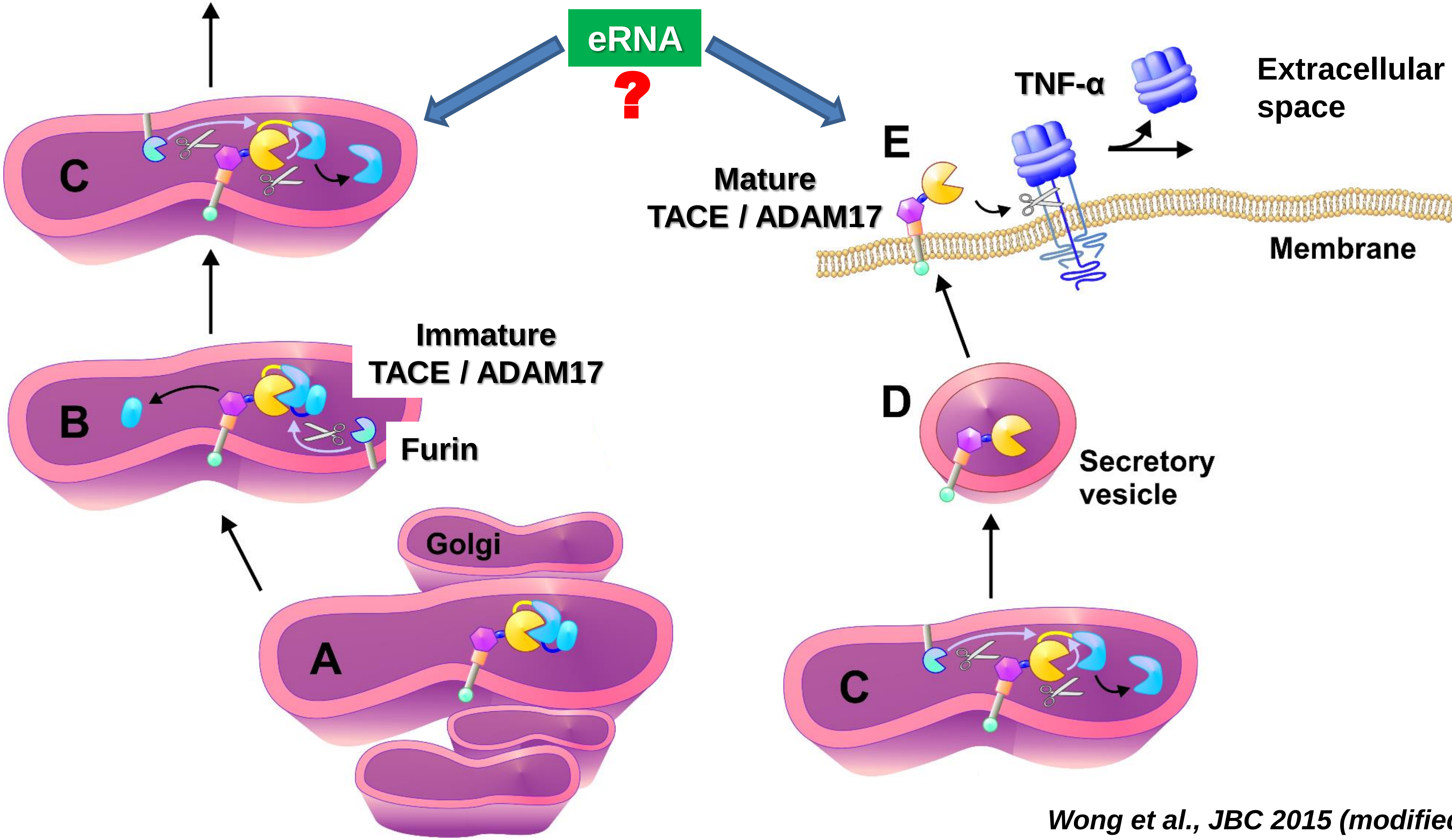
Dvorak et al., Histol. Histopathol. 2003

Extracellular RNA Drives Macrophages Towards M1-Polarization



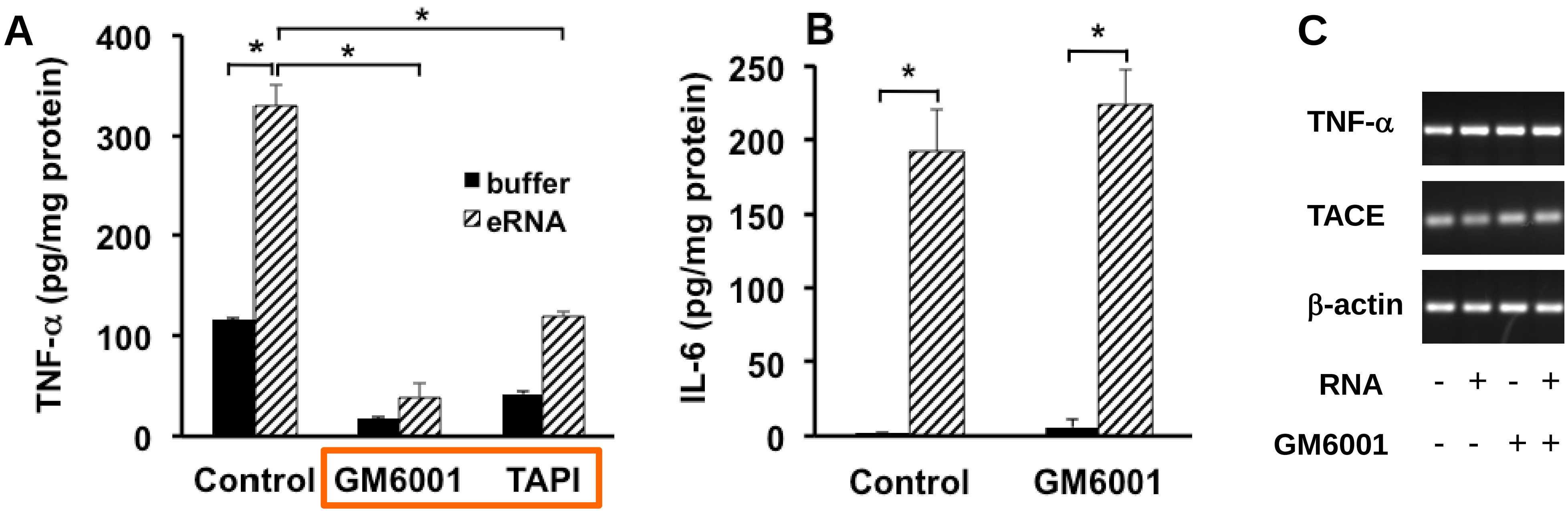
Cabrera-Fuentes et al., T&H 2015

Intracellular Processing of Immature TACE / ADAM17 by Furin-mediated Removal of Auto-inhibitory Prodomain

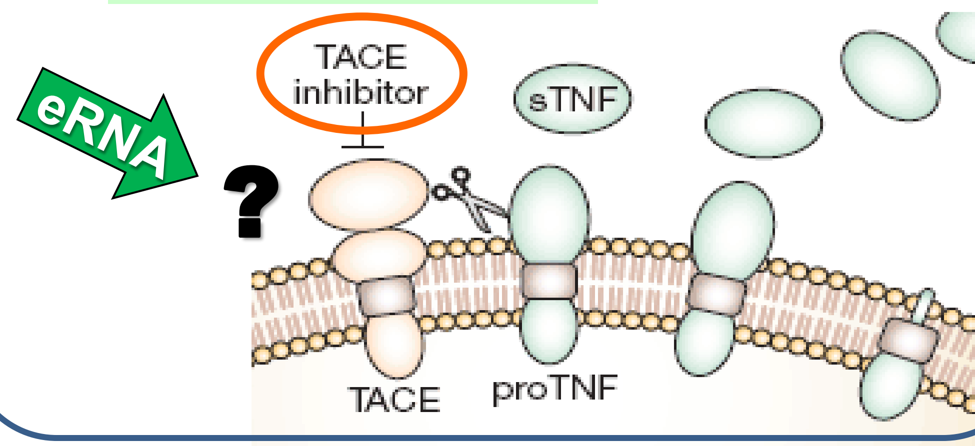


Wong et al., JBC 2015 (modified)

Extracellular RNA-induced TNF-α Release: Contribution of the Sheddase TACE / ADAM-17



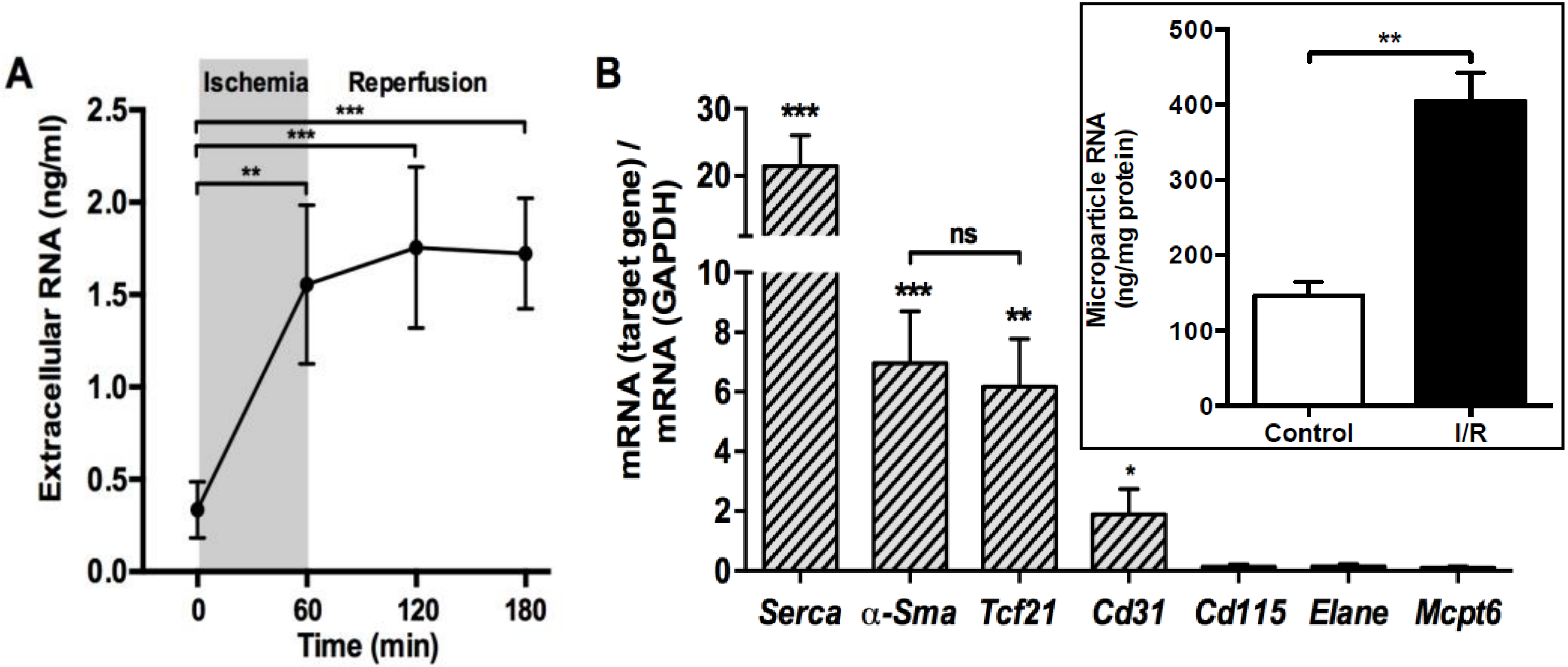
Proposed mechanism



Extracellular RNA promotes TACE-induced cleavage of membrane-bound pro-TNF.

Fischer et al., T&H 2012

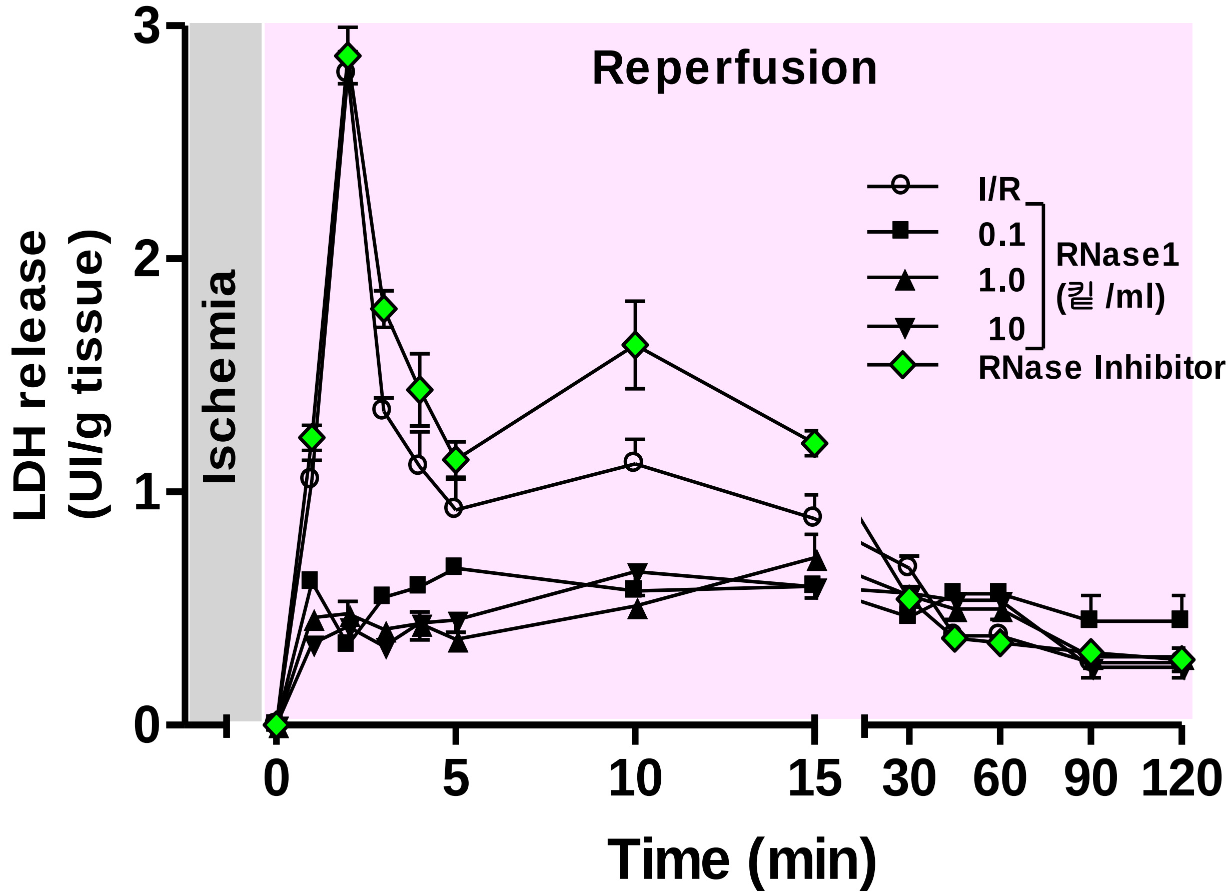
In vivo Release of eRNA Following Ischemia-Reperfusion in the Mouse Heart

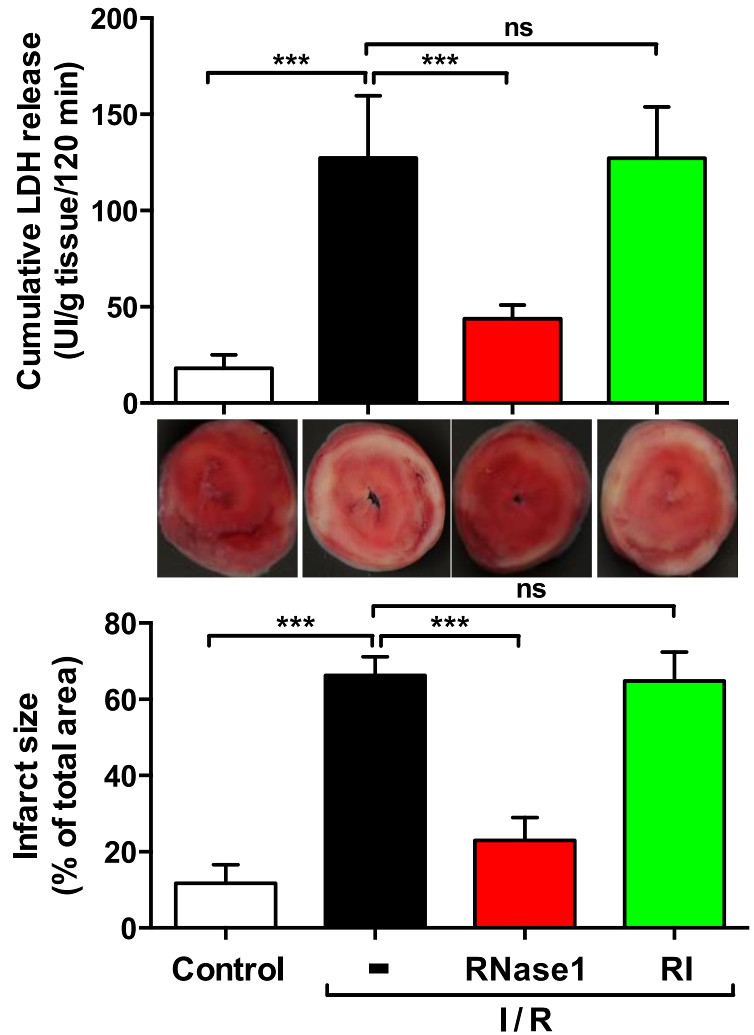


> eRNA is mainly derived from cardiomyocytes <

Cabrera-Fuentes et al., T&H 2014

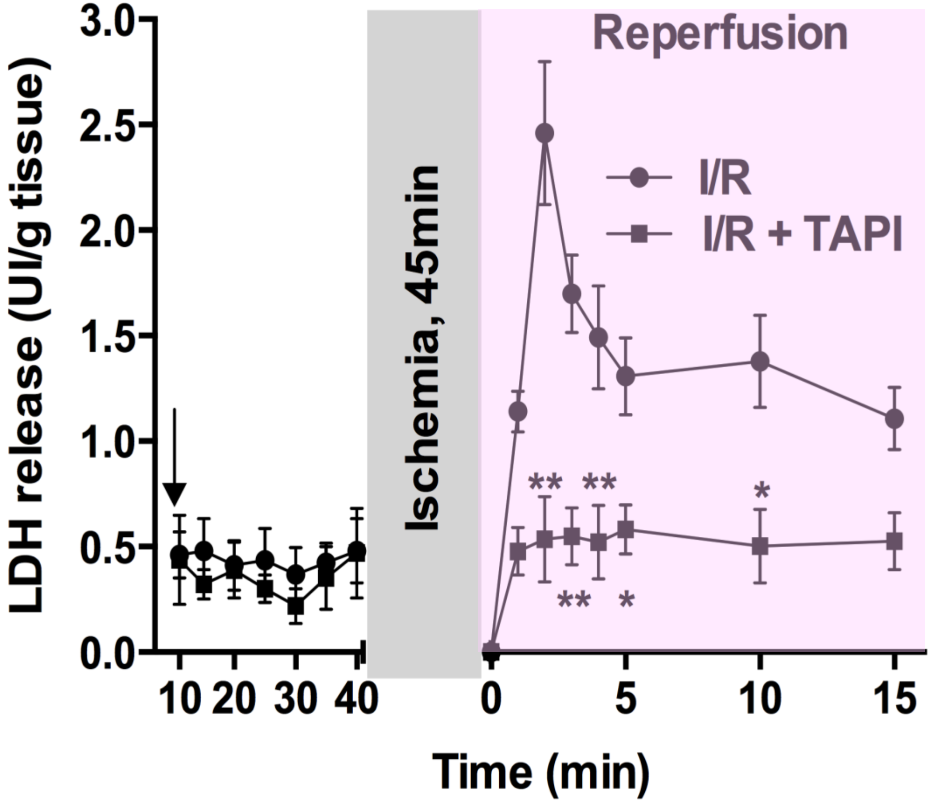
Ischemia/Reperfusion Injury in Isolated Rat Hearts: RNase1 Prevents Tissue Damage and Reduces Infarct Size





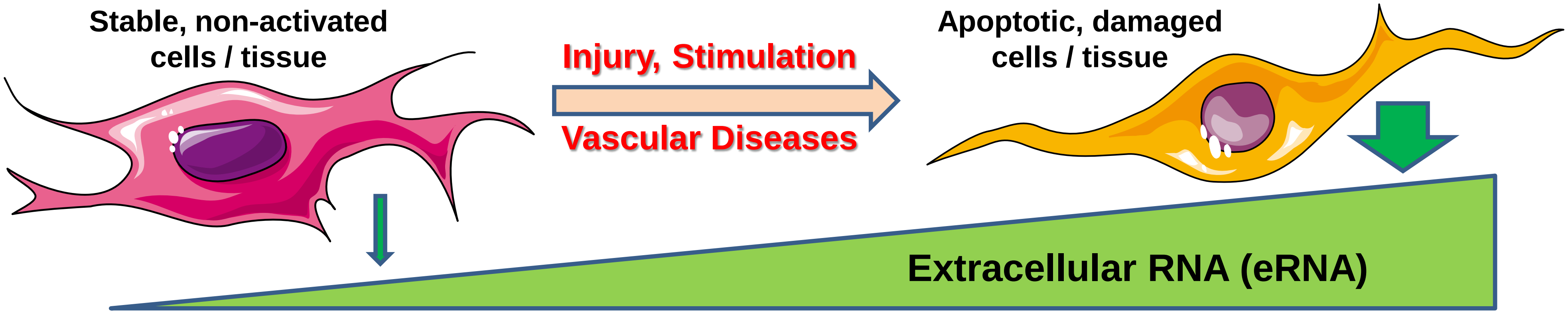
LDH and TNF-a Release in Isolated Rat Hearts Submitted to I/R:
Influence of TACE/ADAM17-Inhibition by TAPI

A. **LDH release**

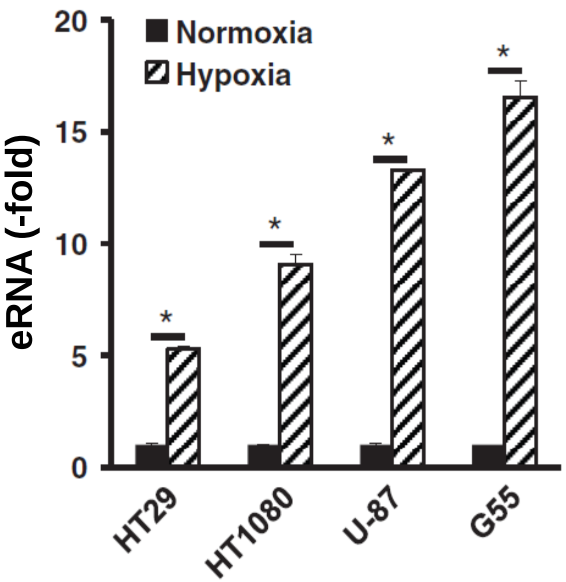


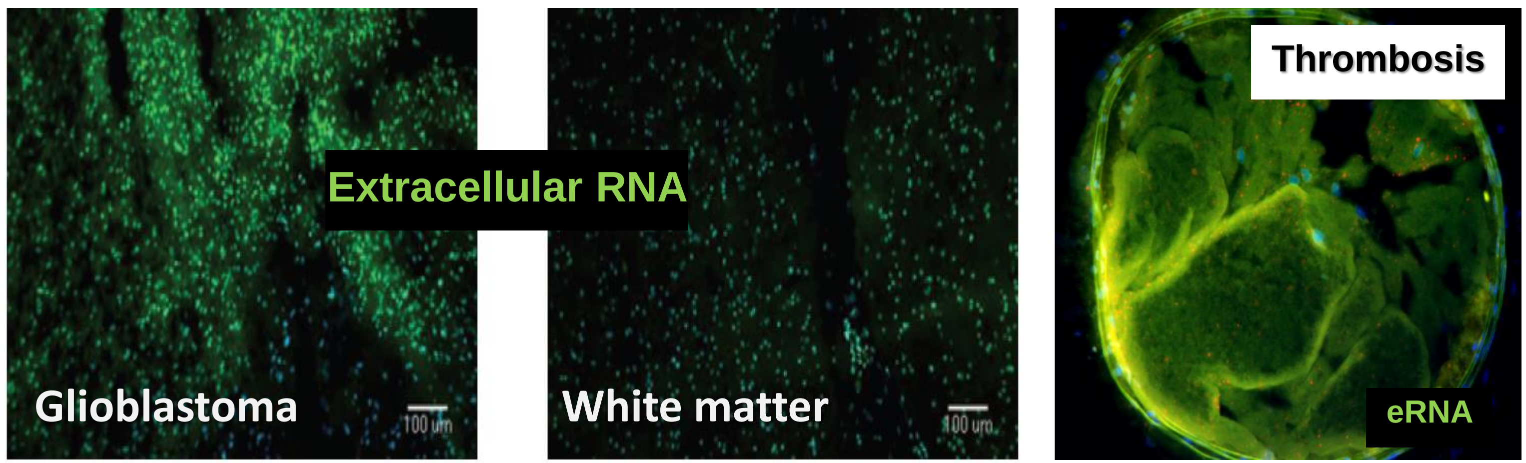
Administration of TAPI

Distribution and Cellular Release of Extracellular RNA

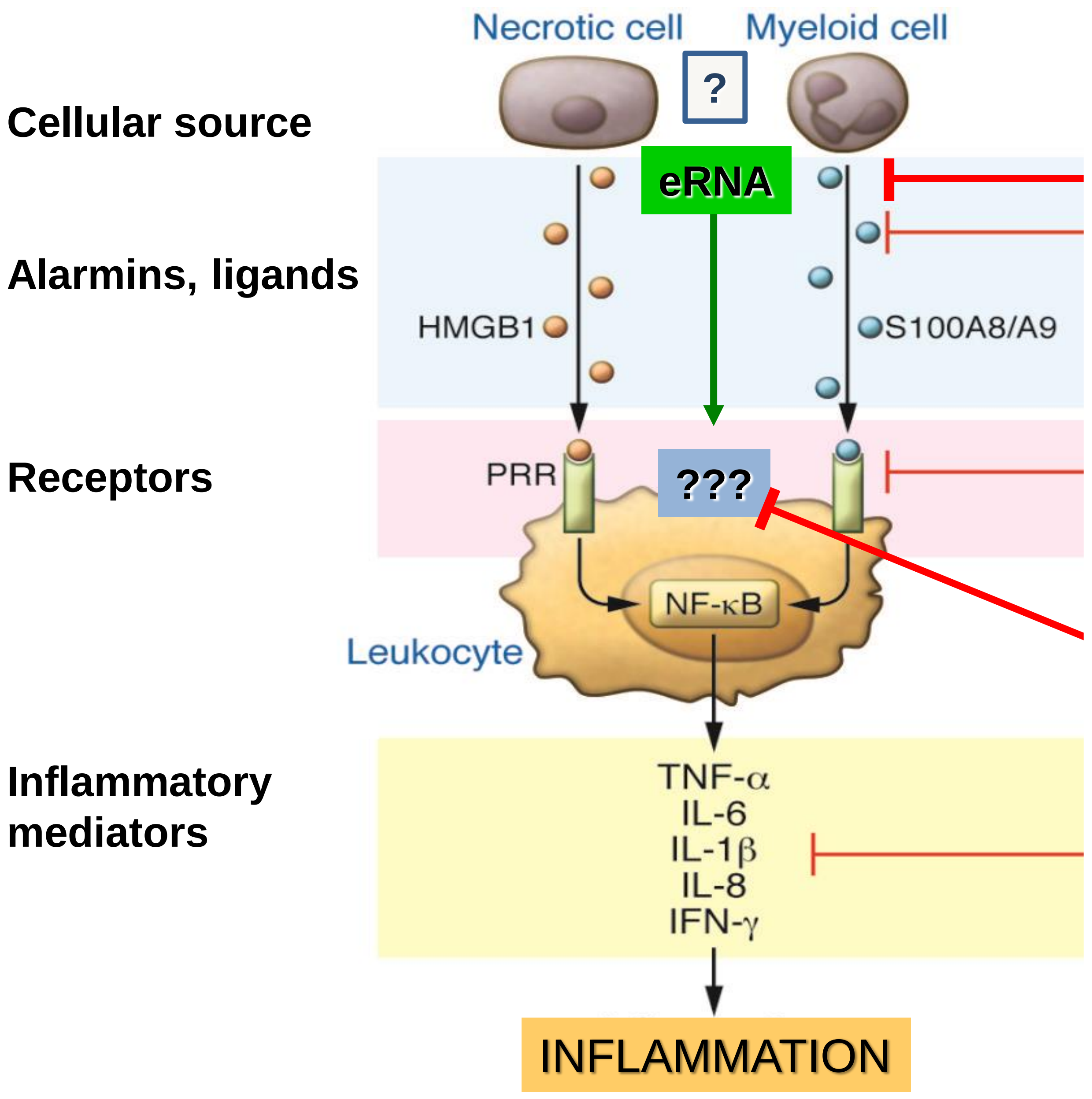


eRNA liberation from tumor cells (hypoxia)



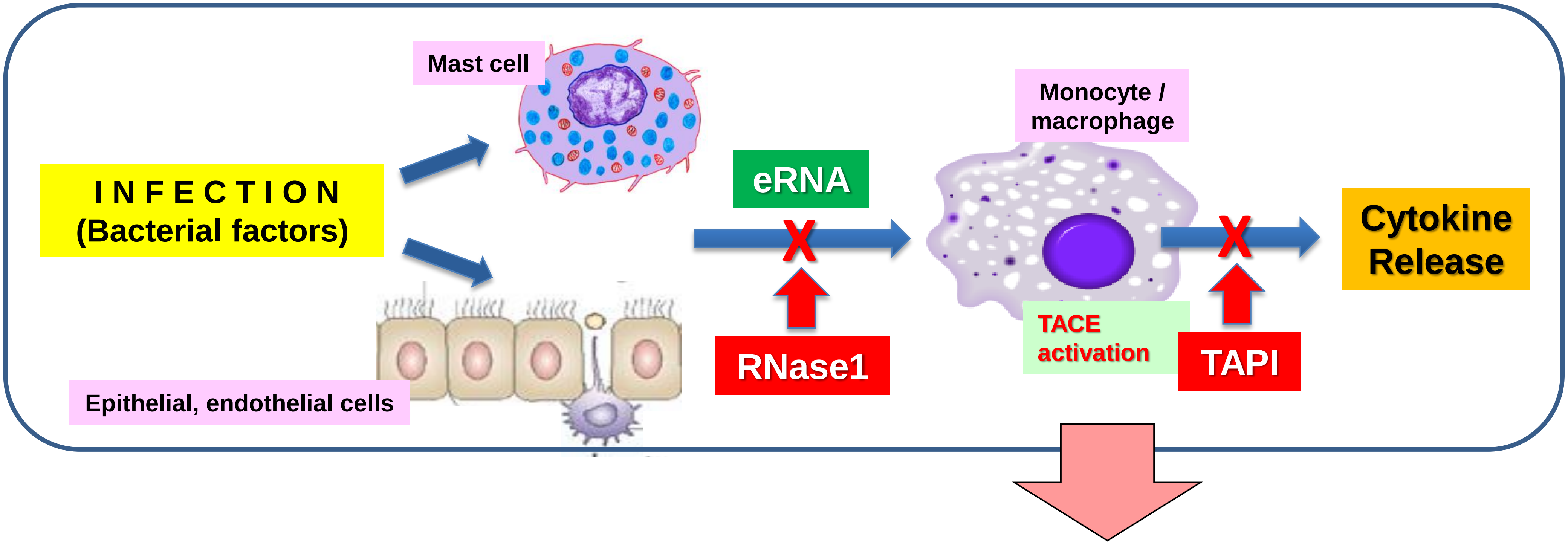


The Endogenous Inflammatory Cascade



Chan et al., JCI 2012 (modified)

Extracellular RNA-mediated Inflammatory Cascade Involving TACE /ADAM17

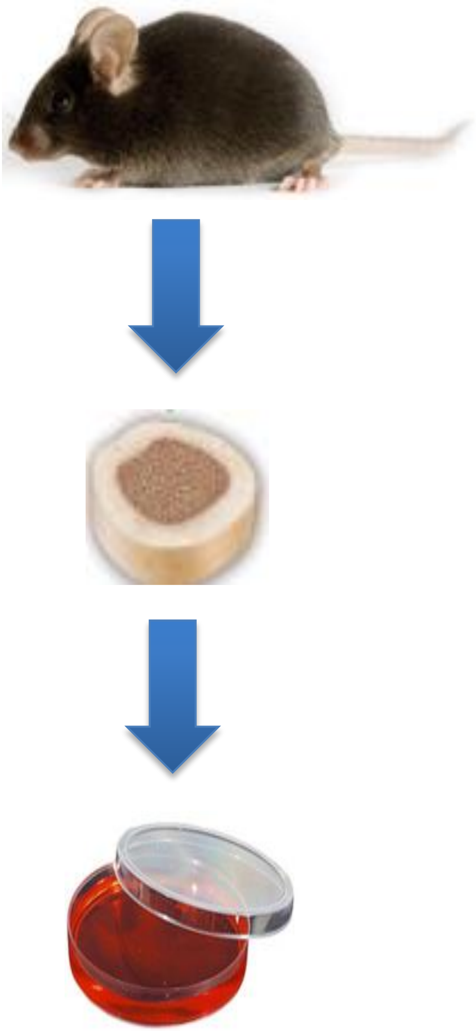


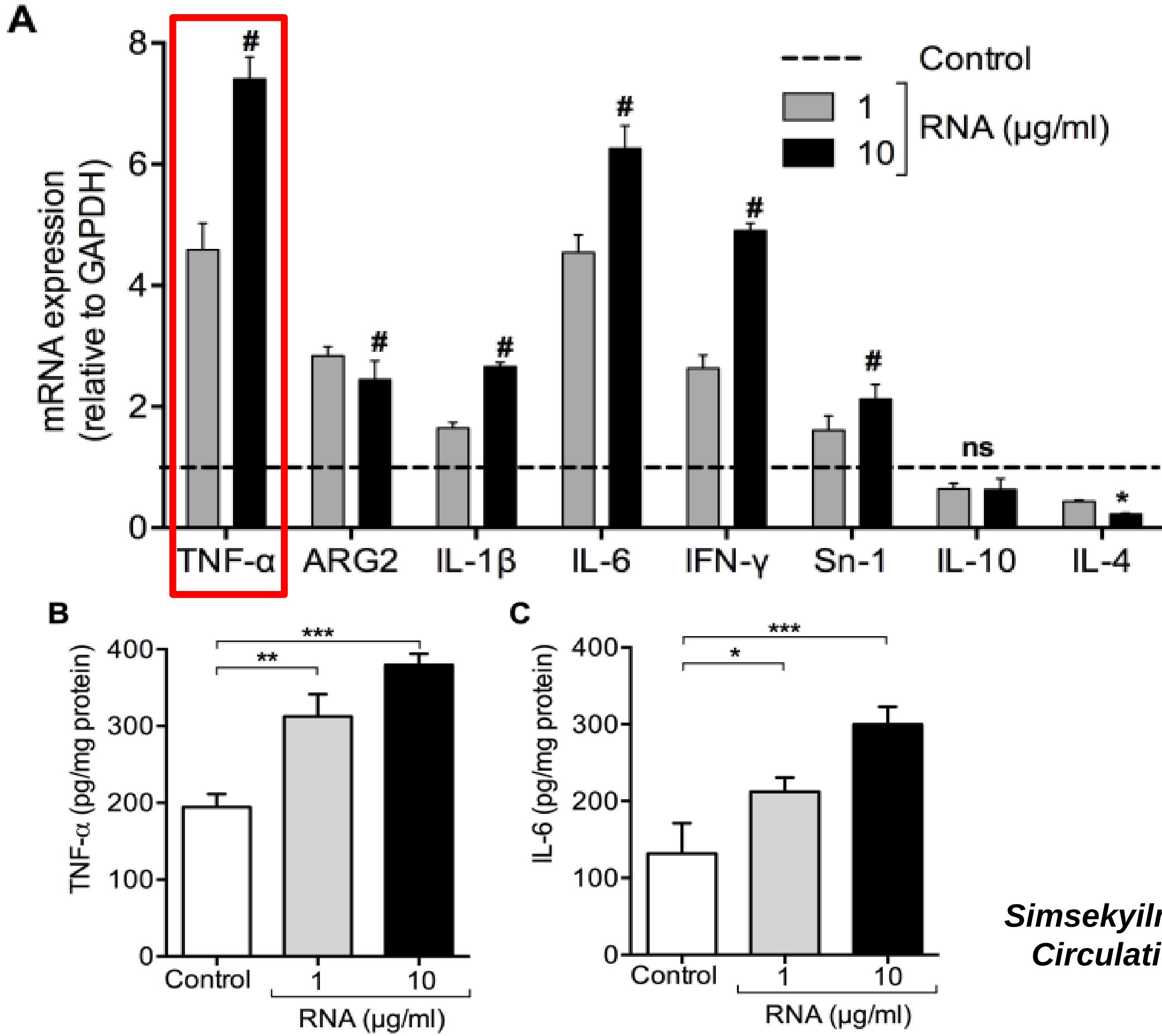
TACE / ADAM-17 cleaves more than 70 substrates. Examples are:

Pro-TNF-α ● Pro-EGF ● NOTCH ● ICAM-1 ● VEGF-R2 ● GPIbα

IL6-R ● L-selectin ● IGF2-R ● L-selectin ● ErB-ligand ● CD44

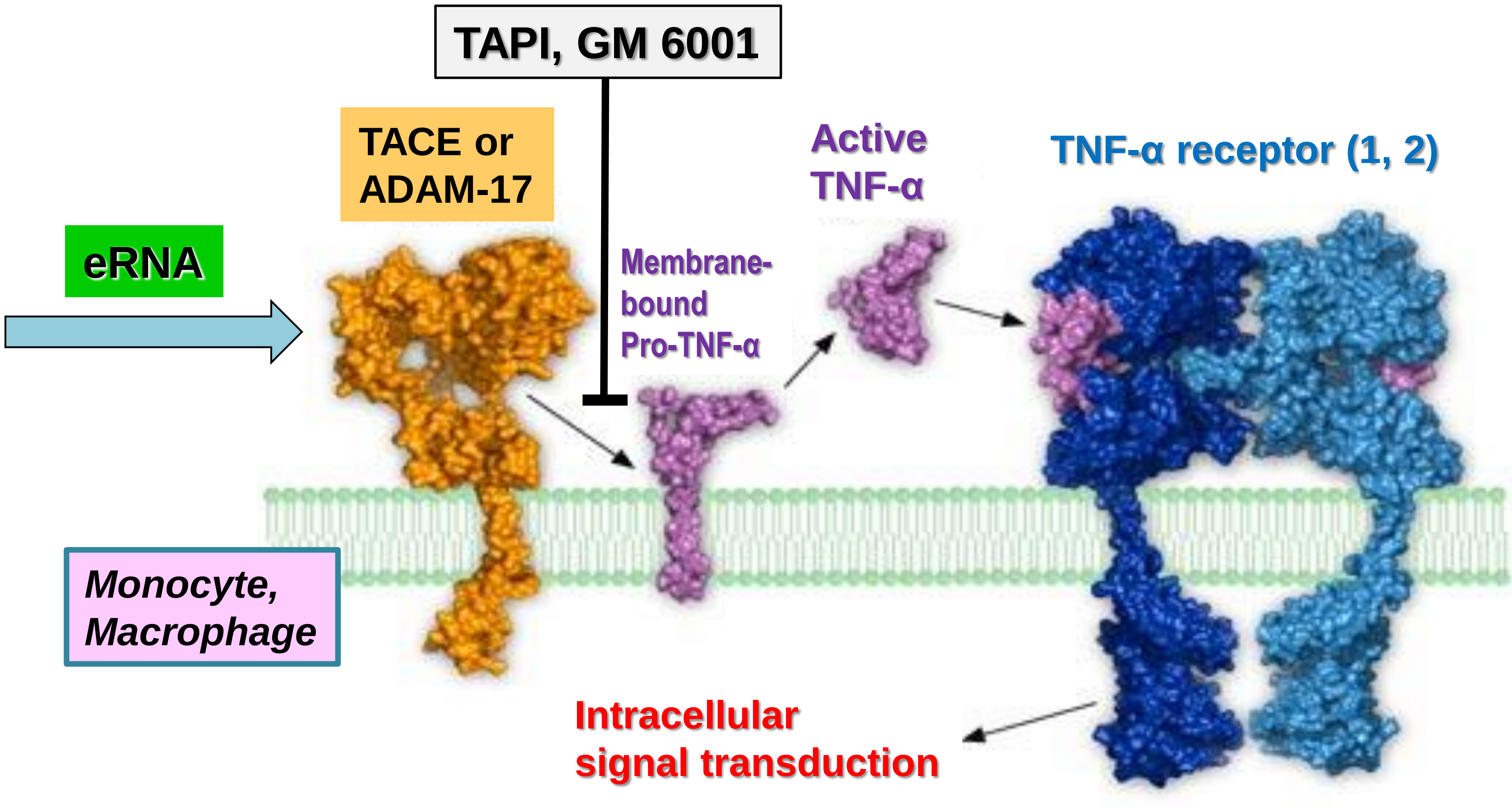
Extracellular RNA-mediated Cytokine Production in Bone Marrow-derived Macrophages (BMDM)





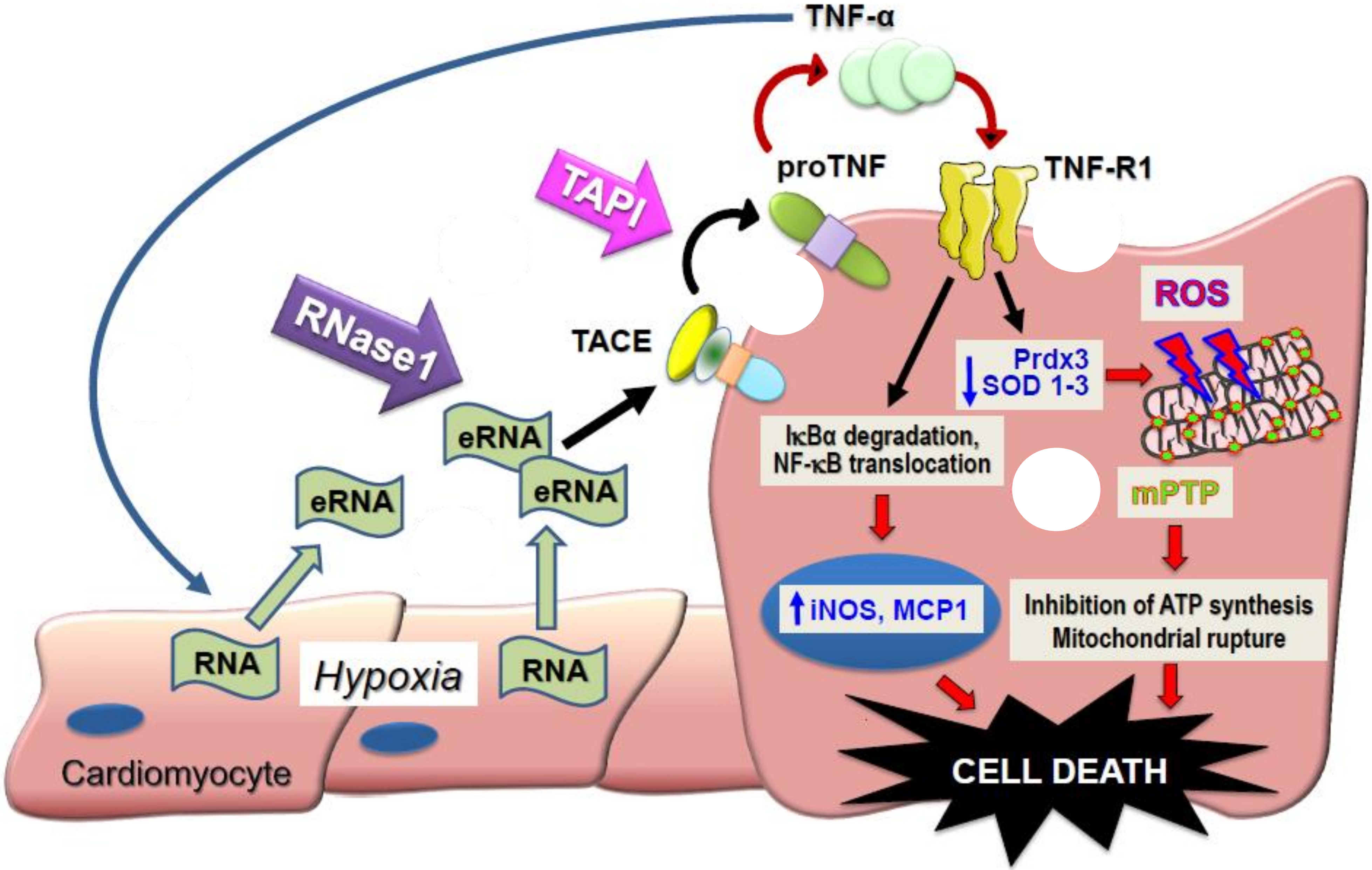
Simsekyilmaz et al., Circulation 2014

Mechanism of TNF-α Shedding: The Role of TACE / ADAM-17



TACE / ADAM-17: TNF-α Converting Enzyme
TAPI, GM 6001: TACE-inhibitors

Damaging Interplay Between eRNA and TNF-α in Cardiac Ischemia/Reperfusion Injury



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