

Inflammation in the Healing Wound: Friend or Foe

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Stages of wound healing

Resolution/ Remodeling

Vessel regression, Collagen remodeling

Proliferation

Reepithelialization, Angiogenesis, Fibrogenesis

Hemostasis / Inflammation

Fibrin clot, platelet deposition, leukocyte infiltration



Time after injury

Major events in wound inflammation

- ◆ Increased blood flow
- ◆ Increased vascular permeability
- ◆ Cellular activation and infiltration

Mediated by soluble molecules and factors released or produced at the site

Increased
blood flow -
Vasodilatation

Increased
vascular
permeability

Endothelial cell
contraction

Direct injury

Net result of vascular changes:

Edema (swelling)

Activation of endothelium

Enhanced entry of mediators from serum

Major events in wound inflammation

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Mediated by soluble molecules and factors released or produced at the site

◆ Alexis Carrel, MD

“Possibly the white cells bring the substances which adult tissues require in order to cicatrize or regenerate. They would have the function of storing away the growth-promoting substances characteristic of embryonic tissues, and bringing them to the regions of the organism where they are needed.” 1922

An introduction to the key cell types involved in wound inflammation

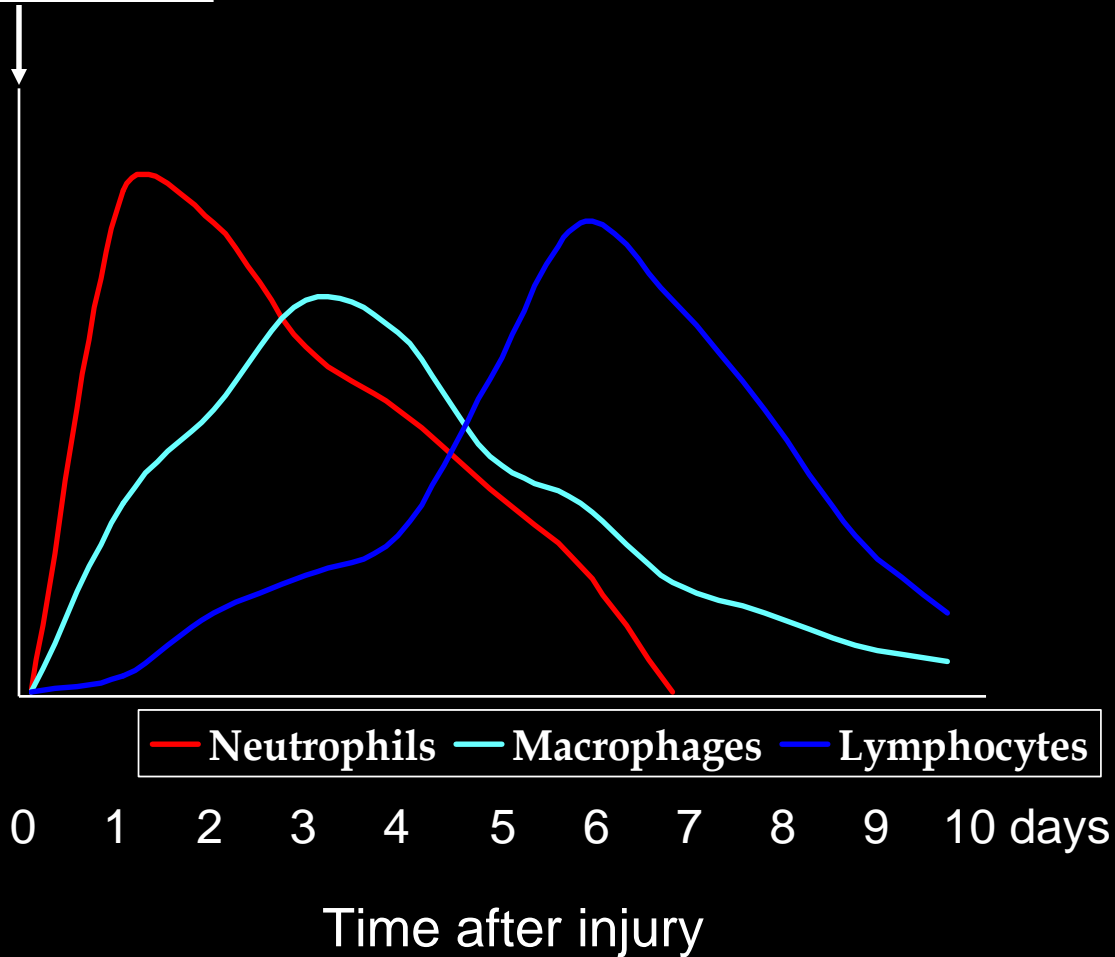
- ◆ Platelets
- ◆ Mast cells
- ◆ Neutrophils
- ◆ Macrophages
 - ◆ Derived from monocytes
- ◆ Lymphocytes (primarily T)
- ◆ Epithelial cells

Yellow – circulating, White - resident

Cell activation and infiltration

Platelet
deposition

Mast cell
degranulation



Release initiating factors

Platelet deposition

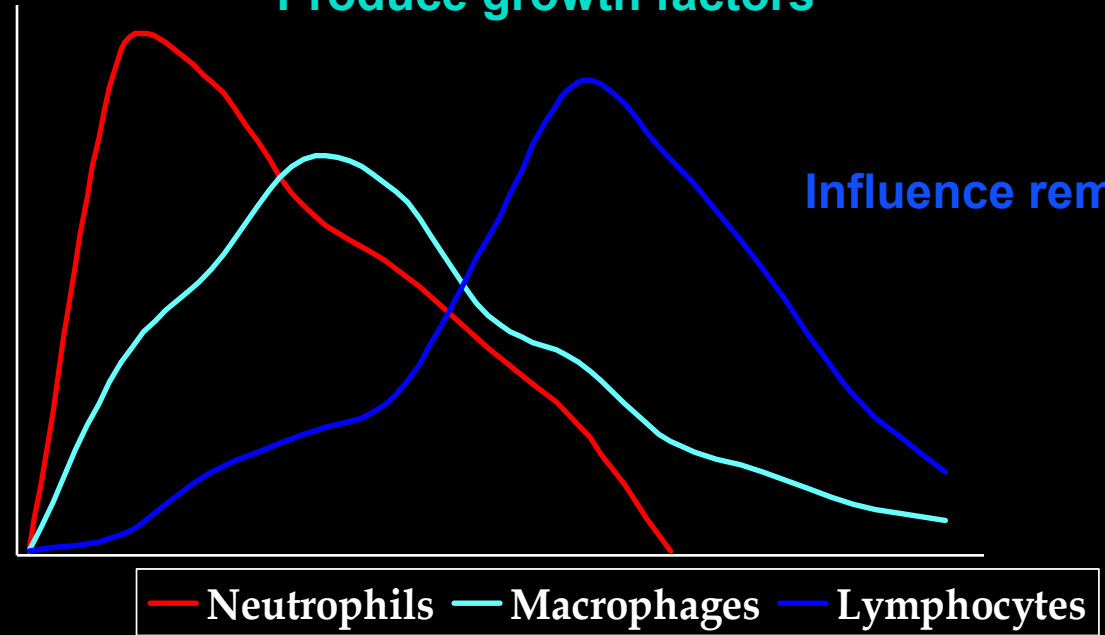
Mast cell degranulation

Major functions

Clear microbes

Remove spent neutrophils,
Produce growth factors

Influence remodeling




0 1 2 3 4 5 6 7 8 9 10 days

Time after injury

How do we evaluate the functional importance of specific inflammatory cells in wounds?

- ◆ In situ evaluation
- ◆ Depletion of specific cell types
- ◆ Strains that are deficient in particular cell types
- ◆ Add more of the cell type

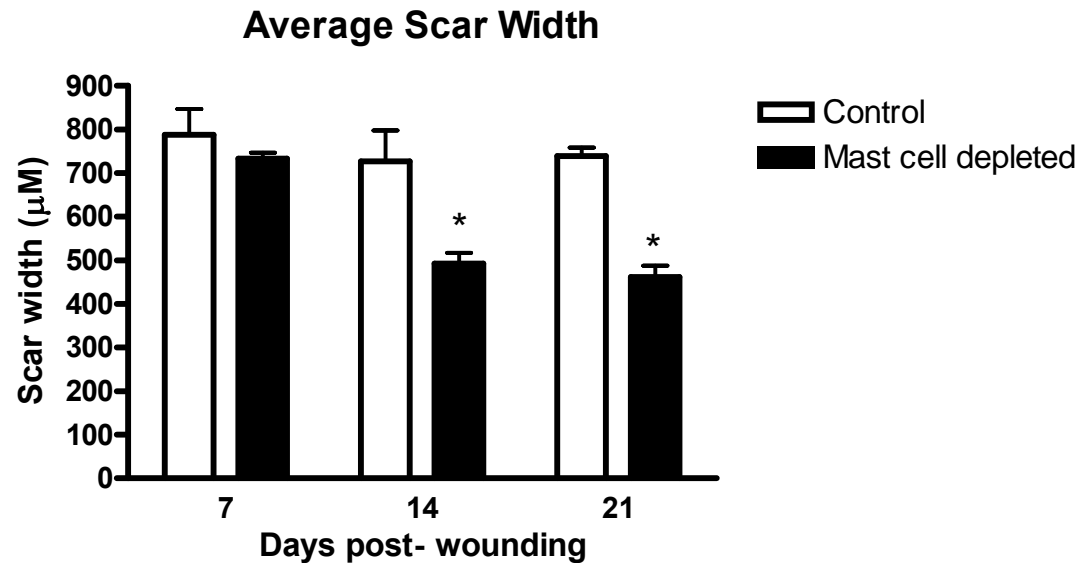
Key cell types involved in wound inflammation

- ◆ Platelets
 - ◆ Mast cells
 - ◆ Neutrophils
 - ◆ Macrophages
 - ◆ Lymphocytes
 - ◆ Epithelial cells
- 

Mast cells

Egozi EI, Ferreira AM, Burns AL, Gamelli RL,
DiPietro LA: Mast cells modulate the inflammatory
but not the proliferative response in healing
wounds. *Wound Repair Regen* 2003, 11:46-54

Inhibition of mast cells leads to reduced scar width in skin wounds.



Gallant-Behm et al. The mast cell stabilizer ketotifen prevents development of excessive skin wound contraction and fibrosis in red Duroc pigs. *Wound Repair Regen*, 2008. 16:226-33.

Inhibition of mast cell function did not affect the production of granulation tissue within the wounds, nor did it affect the rate of reepithelialization.

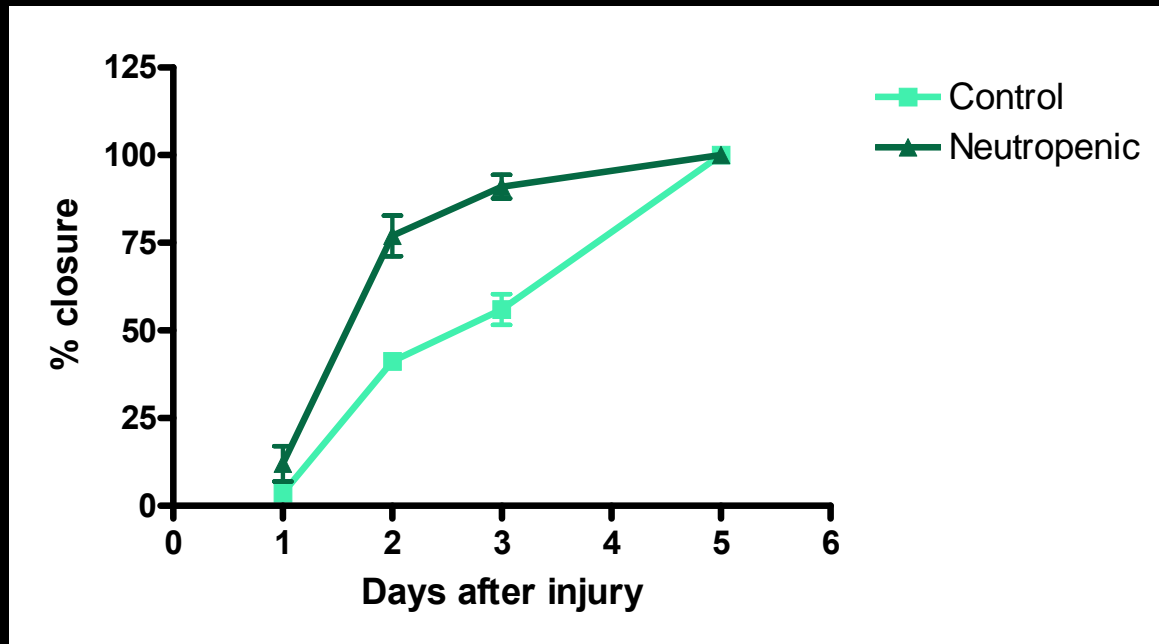
Ketotifen treatment significantly reduced contraction and collagen deposition in red Duroc wounds, effectively reducing scar formation.

Neutrophils

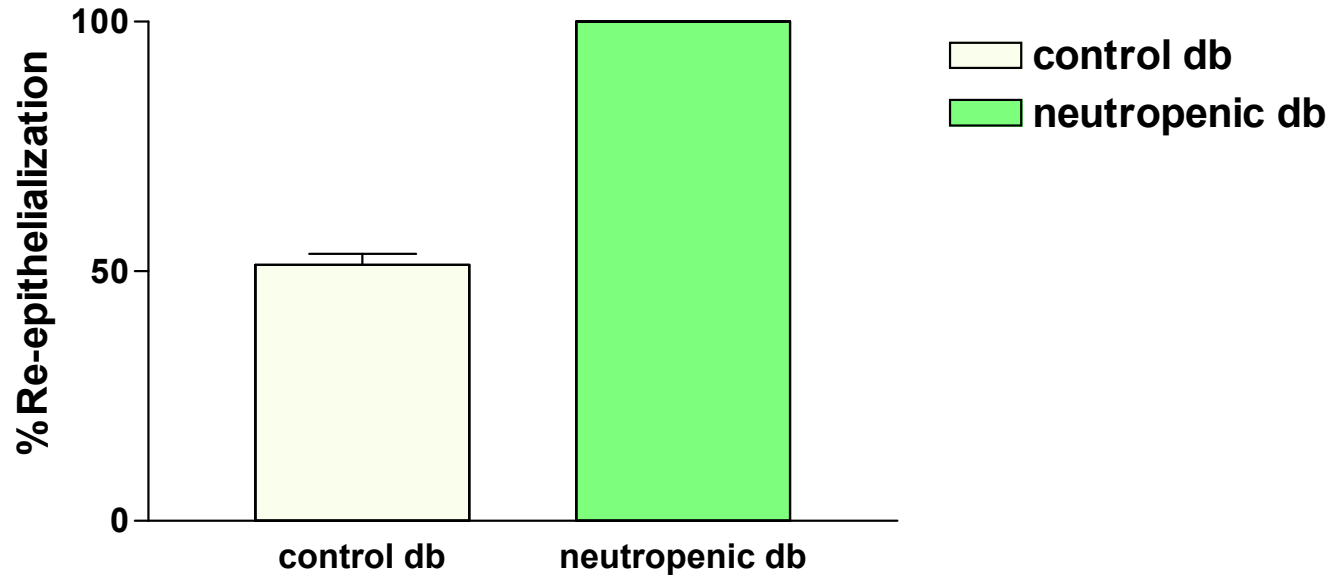
- masters of microbial clearance*
- produce toxic products*
- bystander injury*



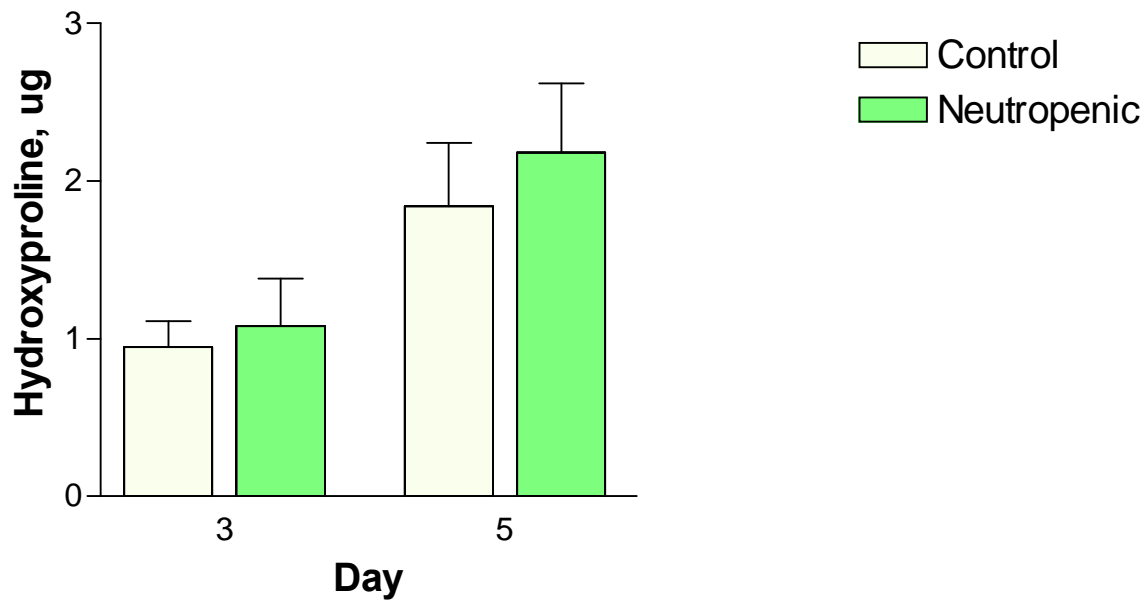
Neutrophil depletion enhances wound closure.



Neutrophil depletion improves wound closure in diabetic mice



Do neutrophils influence collagen synthesis?



Activated Macrophage

Inflammation

Cytokines

Reactive O₂ species

Proteases

AA metabolites

Tissue Repair

**Growth Factors for
angiogenesis,
fibroblast proliferation,
& collagen synthesis**

“Remodeling” proteases

Studies of the function of individual cell types suggest....

Mast cells – may promote scar formation but are dispensable for wound closure

Neutrophils – inhibit closure but are needed to control infection

Macrophages – phenotype dependent, but can be beneficial and promote the proliferative phase

T lymphocytes – may be dispensable, or in some cases may enhance fibrotic scar formation

Leukocyte infiltration requires
specific interaction with endothelium

Major events in wound inflammation

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- ◆ Increased vascular permeability
- ◆ Cellular activation and infiltration

Mediated by soluble molecules and factors released or produced at the site

Mediators of Inflammation

Type	Examples (source)
Plasma derived	Complement, acute phase proteins (serum)
Preformed intracellular	Histamine (mast cells) PDGF, TGF-β (platelets)
Locally produced	IL-1, IL-8, IL-10, TNF-α, NO (multiple cell types)

**Measurable and levels change during the healing process.
Highly complex - Large number (>50).**

How do we evaluate the role of specific mediators in wound healing?

- ◆ Depletion or neutralization of individual mediators
- ◆ Specific inhibition of synthesis
- ◆ Genetic knockouts or knock-ins

Eming SA, et al. Accelerated wound closure in mice deficient for interleukin-10. *Am J Pathol*, 2007. 170:188-202.

**How informative is depletion
of a single mediator?**

Ferreira AM, Rollins BJ, Faunce DE, Burns AL, Zhu X, **DiPietro LA**: The effect of MCP-1 depletion on chemokine and chemokine-related gene expression: Evidence for a complex network in acute inflammation. *Cytokine* 2005, 30:64-71.

Roy S et al. Characterization of the acute temporal changes in excisional murine cutaneous wound inflammation by screening of the wound-edge transcriptome. *Physiol Genomics* 2008. 34:162-184

SPECIAL CIRCUMSTANCES I

Some model systems strongly suggest that inflammation is either entirely or substantially dispensable for healing.

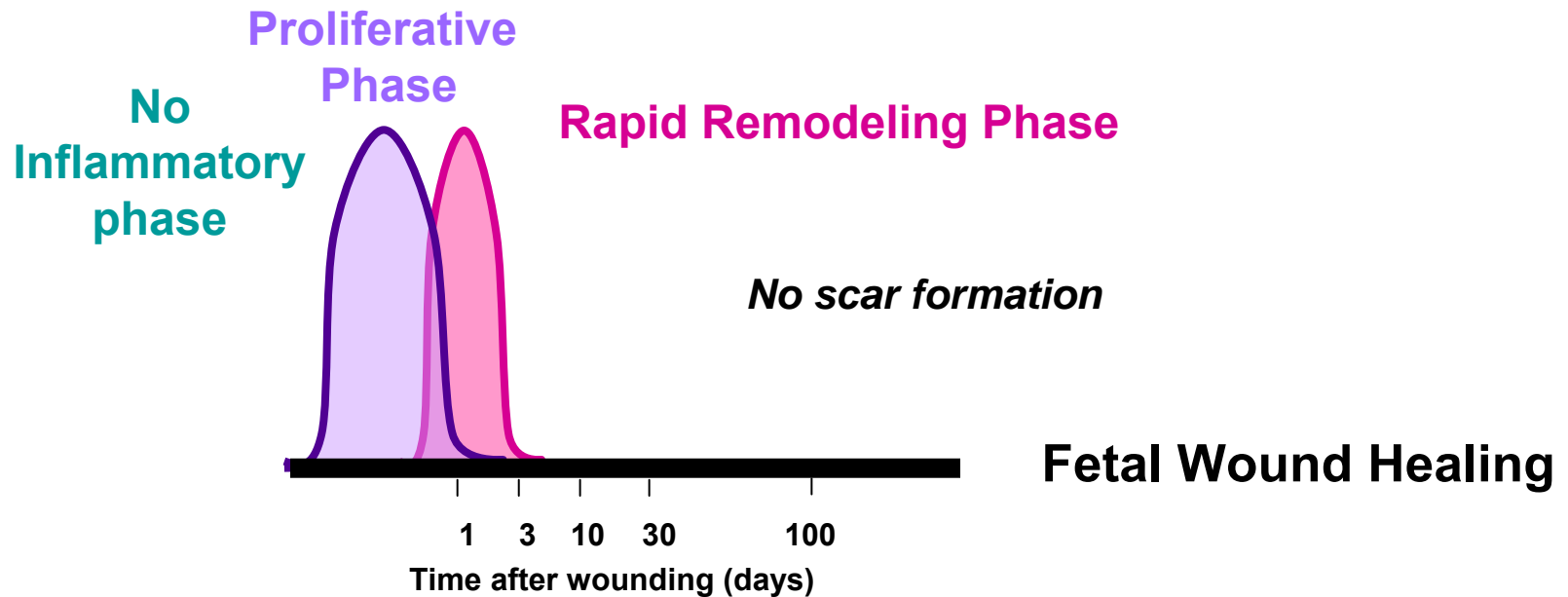
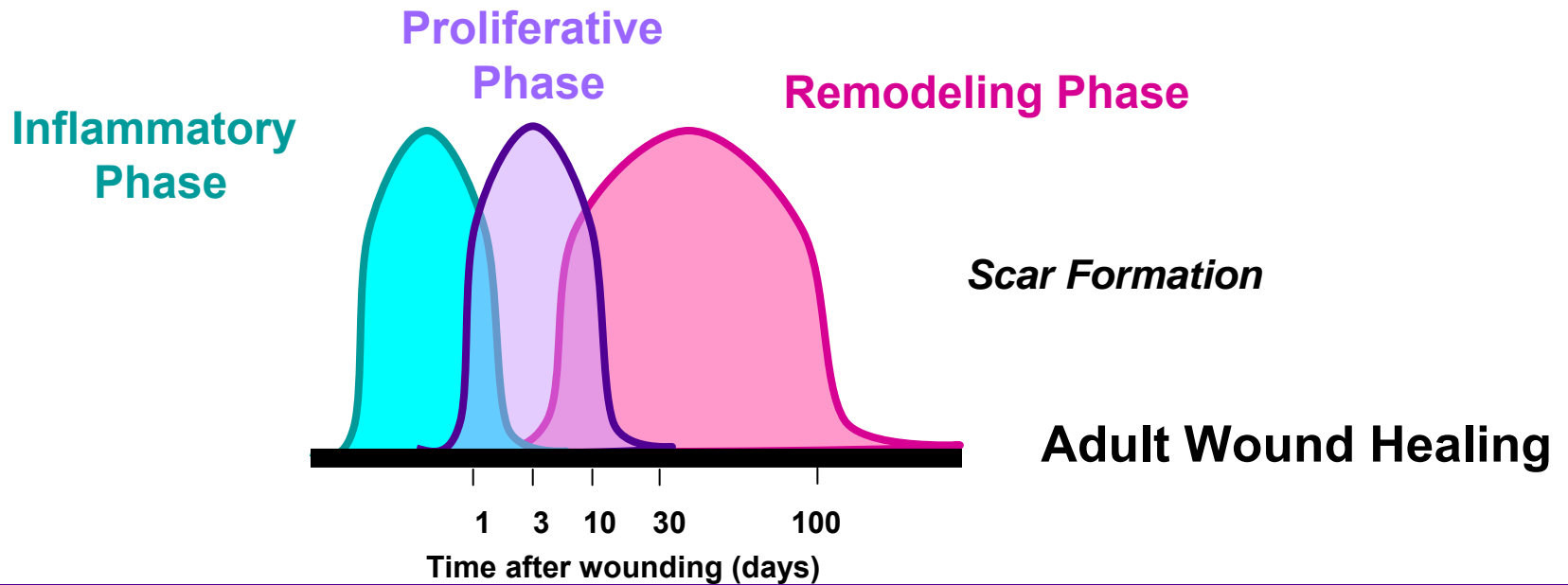
Differential healing of fetal wounds

Mouse, gestation ~21 days

Scarless (E15)

Scar-forming (E18)

Wilgus et al, 2008



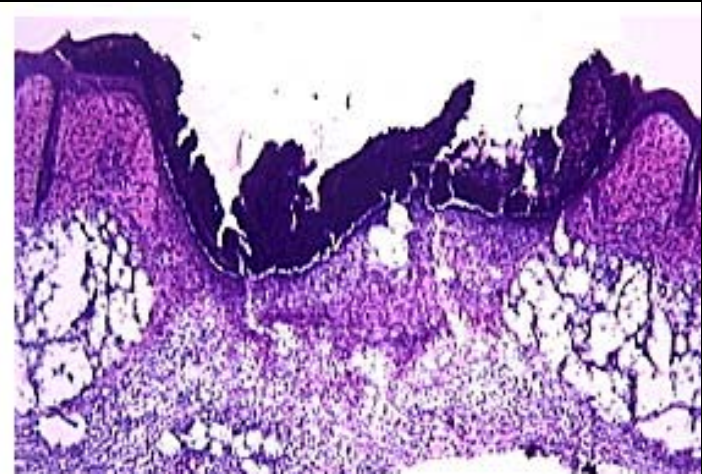
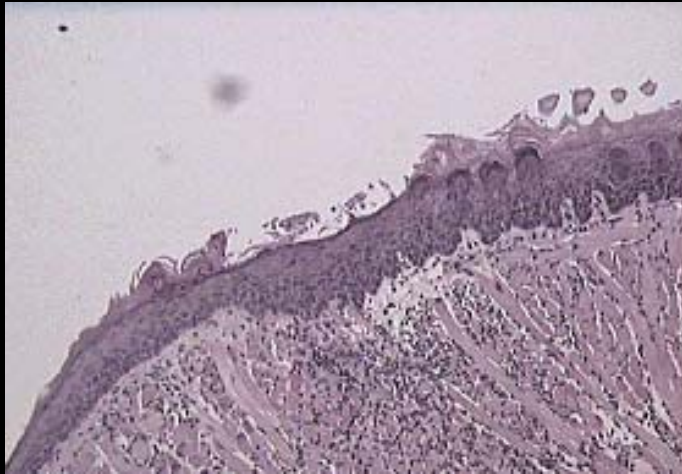
Adapted from TA Wilgus

Differential healing of oral mucosal versus cutaneous wounds.

Oral mucosa

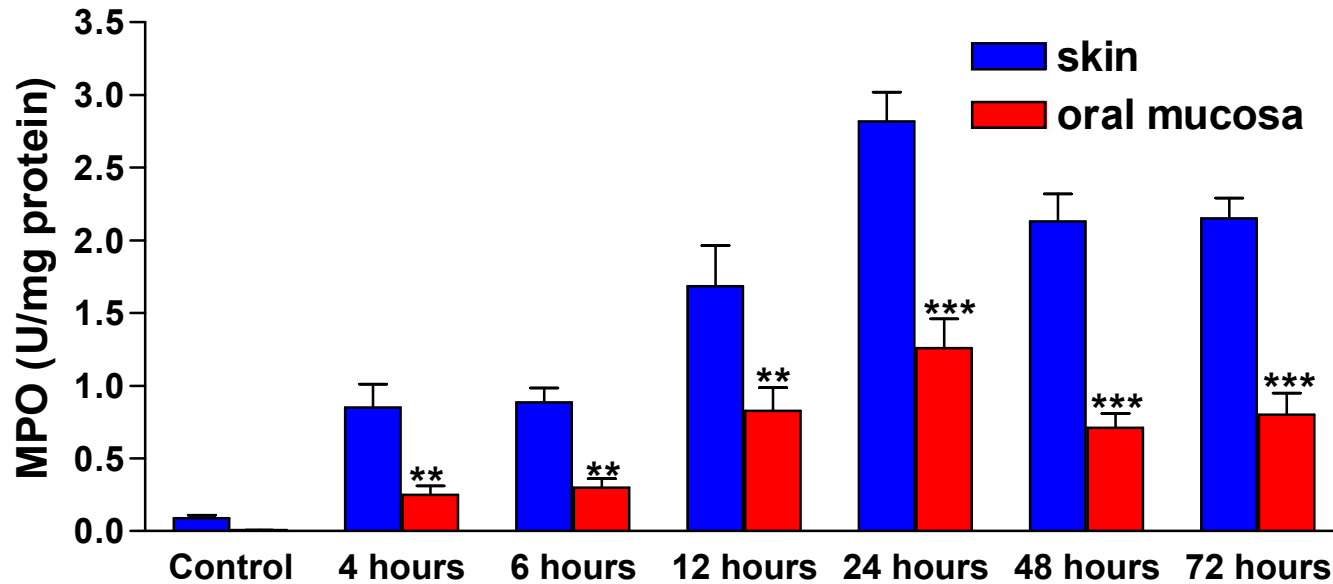
Skin

24h

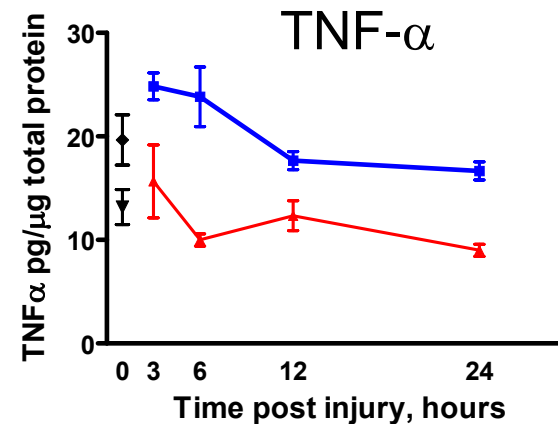
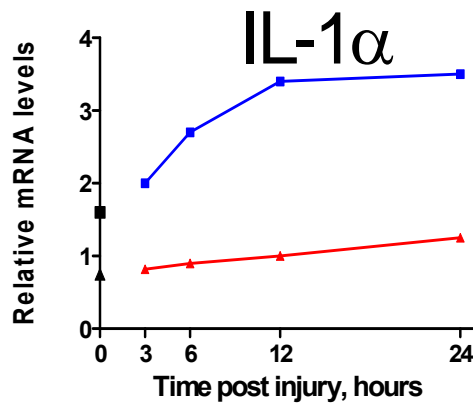
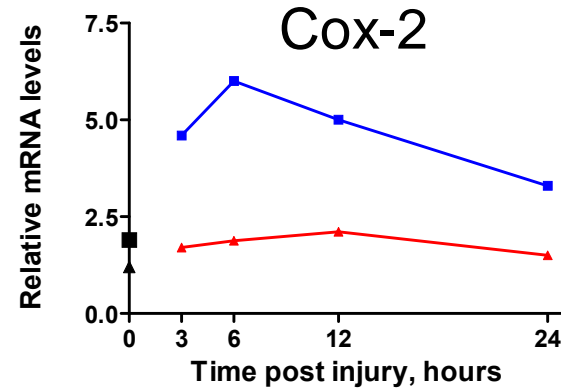
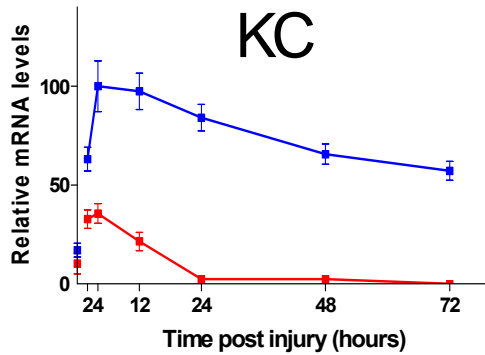


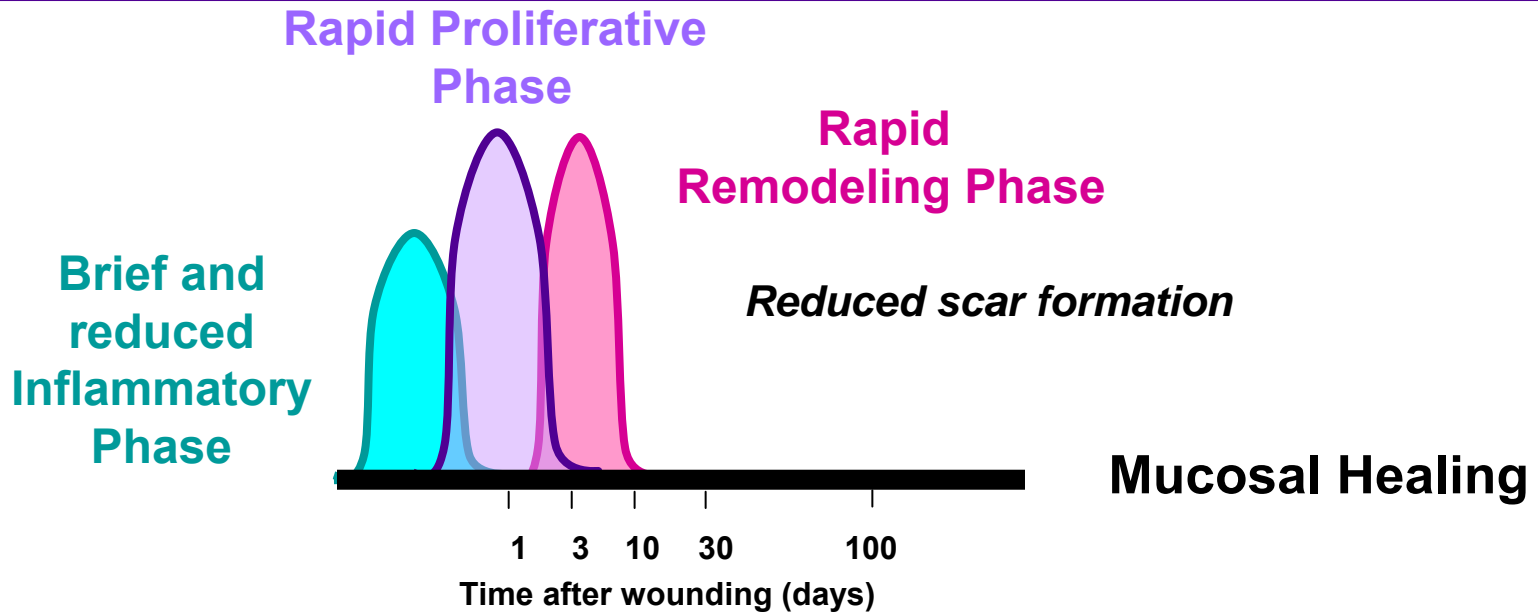
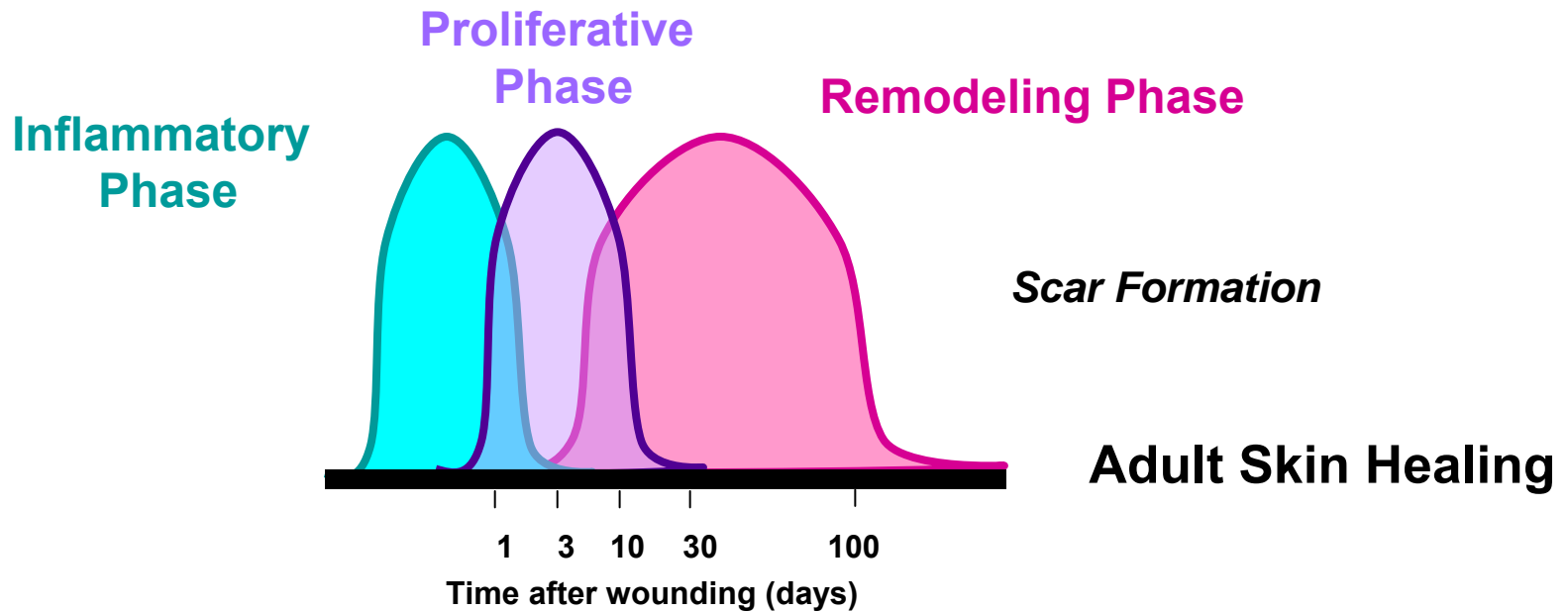
Inflammatory cells in mucosal and skin wounds

Neutrophils



Inflammatory cytokines in mucosal and skin wounds





Fetal and mucosal wounds heal very well despite significantly reduced inflammation.

SPECIAL CIRCUMSTANCES II

Altered inflammation is frequently observed in poorly healing wounds.

Impaired healing circumstances that exhibit aberrant inflammation.

- ◆ Diabetes
- ◆ Steroids (long term use)
- ◆ Chemotherapy
- ◆ Smoking/Drug/Alcohol Use
- ◆ Aging
- ◆ Stress

Summary - Inflammation and Healing

1. The complexity of the inflammatory response in wounds is extreme.
2. The importance of understanding wound inflammation is underscored by specific observations.
 - Certain rapidly healing regenerative wounds exhibit reduced inflammation.
 - Alterations in inflammation are frequently observed in poorly healing wounds.
3. The development of immunomodulatory therapeutics will require a more complete understanding of the roles and interactions of all components, and must consider the risk of infection.

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