

A close-up photograph of pink cherry blossoms with yellow stamens, serving as the background for the slide.

# Anatomy of Skin and Wound Healing

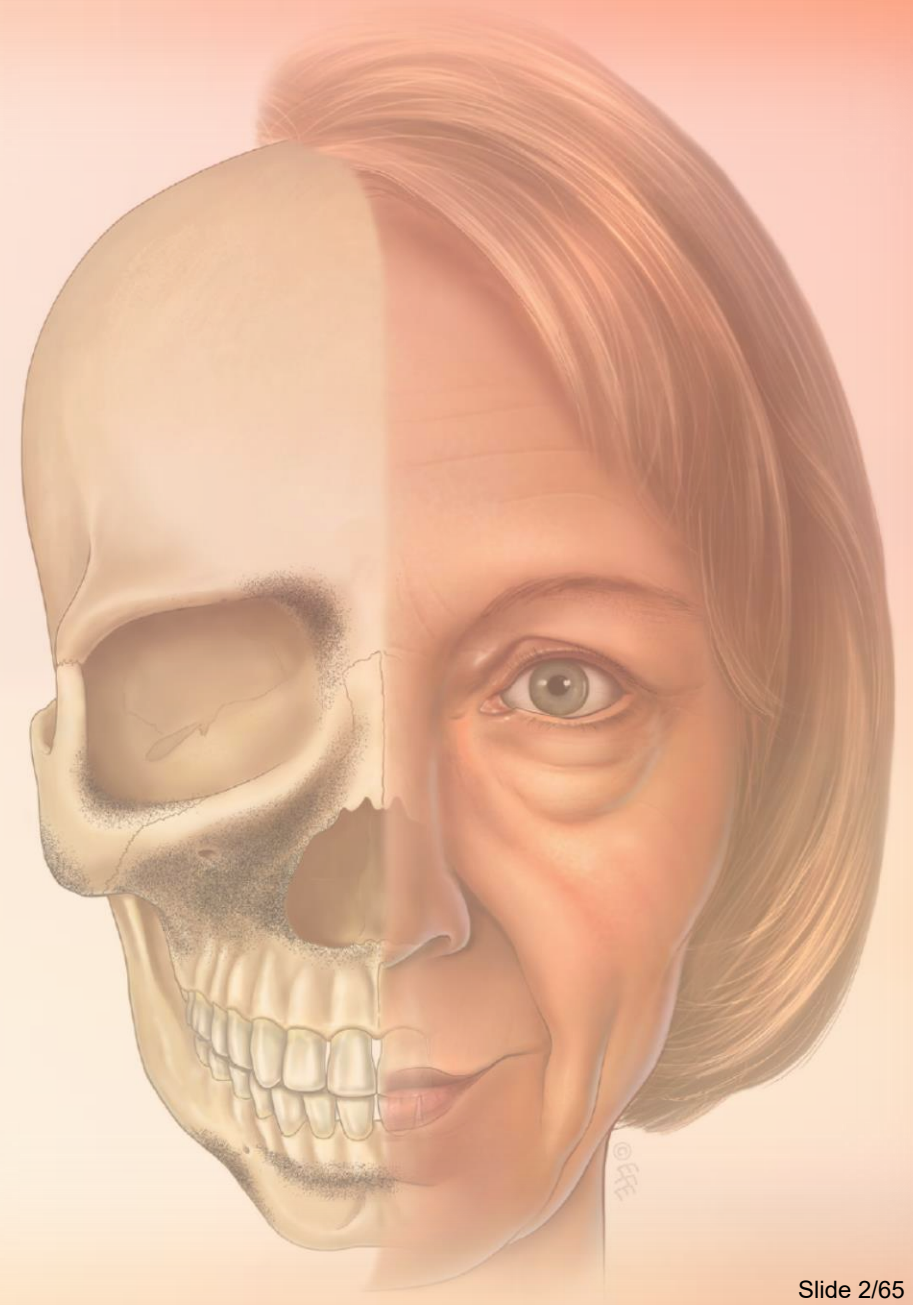
Suparerk Laohapitakworn, Ph.D., M.D.

*Burn unit & Wound clinic*

*Ramathibodi Hospital*

s\_laohapitakworn@hotmail.com

# Anatomy of skin



# Anatomy and physiology

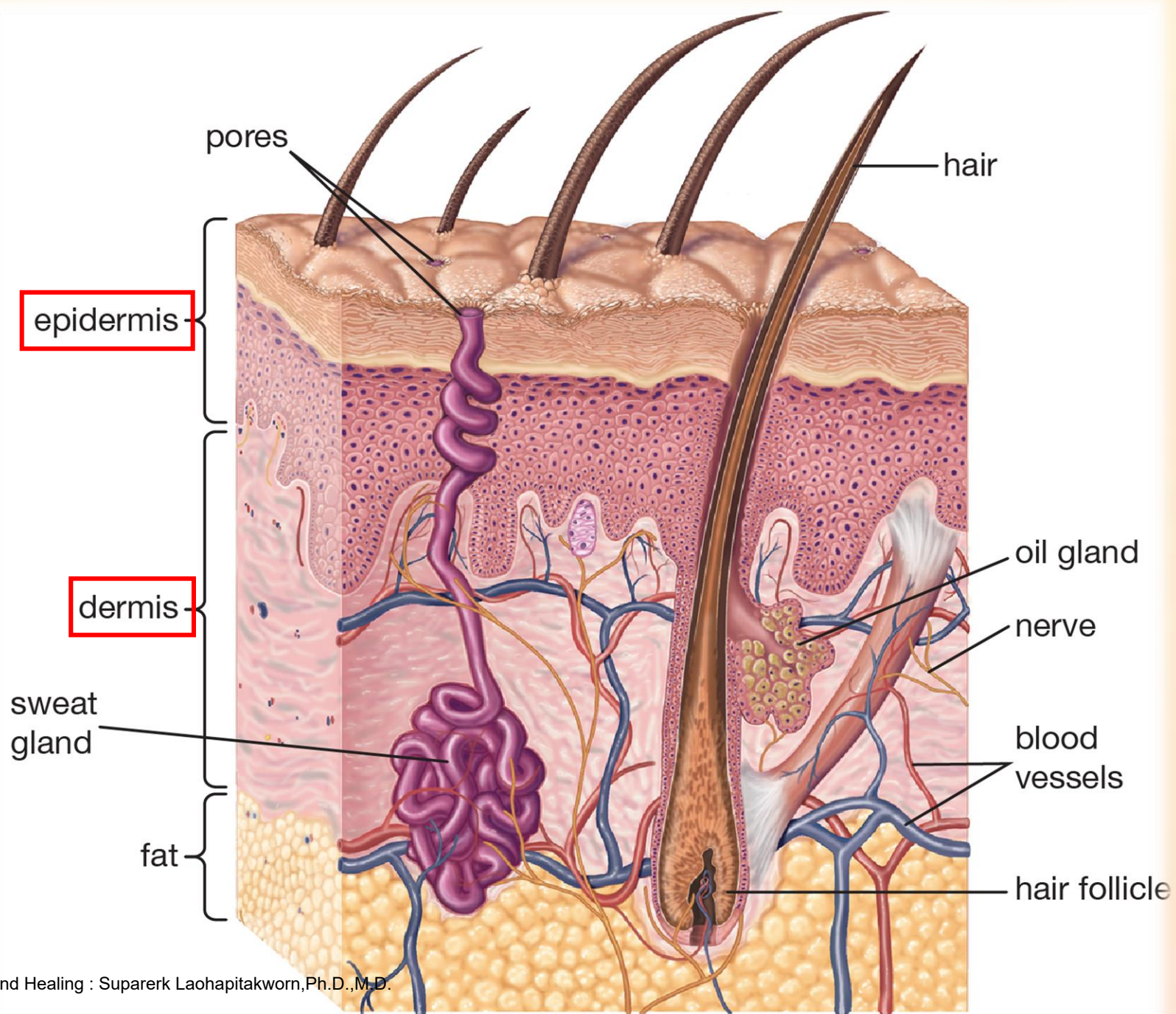
- Surface area of 1.2-2.2 m<sup>2</sup>
- Thickness of 0.5-4 mm



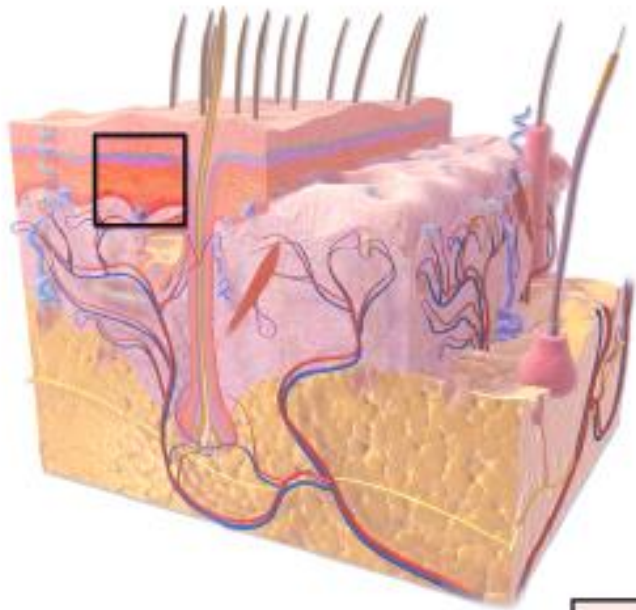
# Anatomy and physiology

- Function
  - Insulation
  - Temperature regulation
  - Sensation
  - Immune function
  - Synthesis of vitamin D



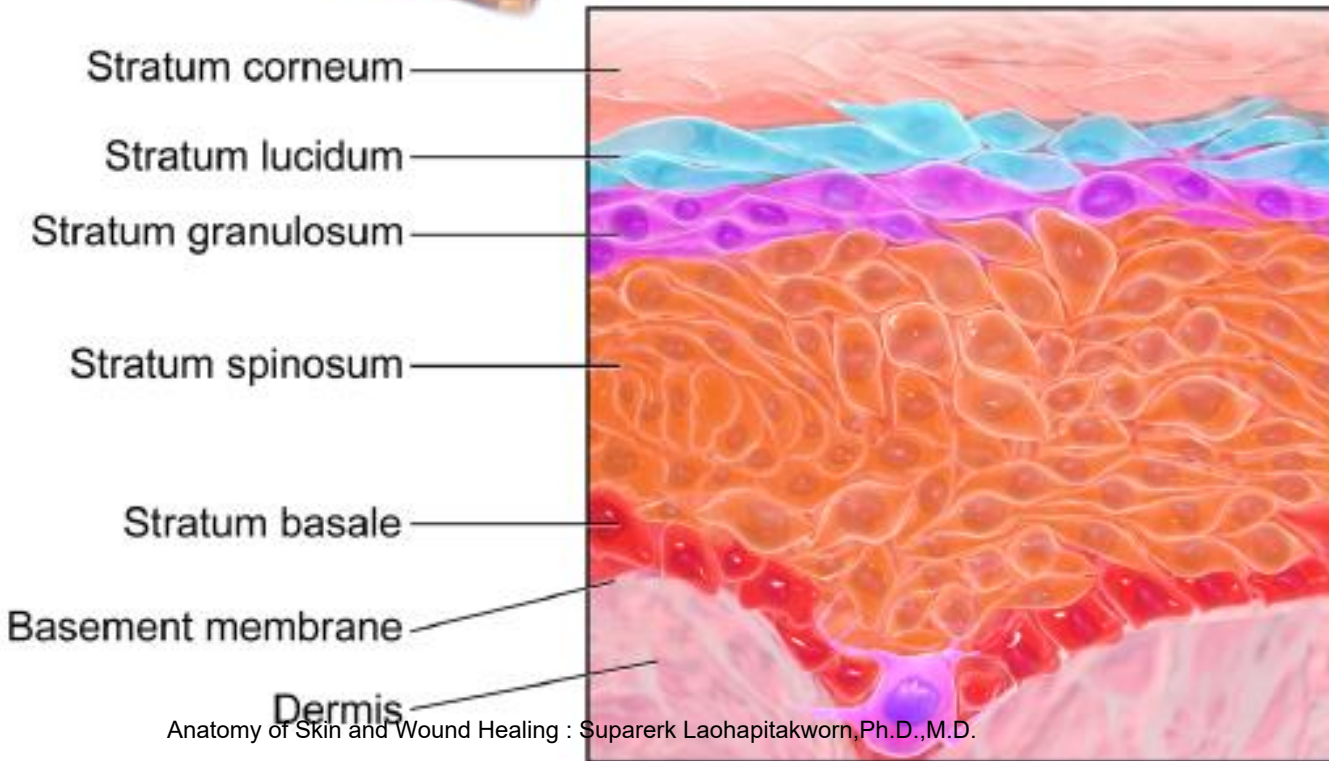




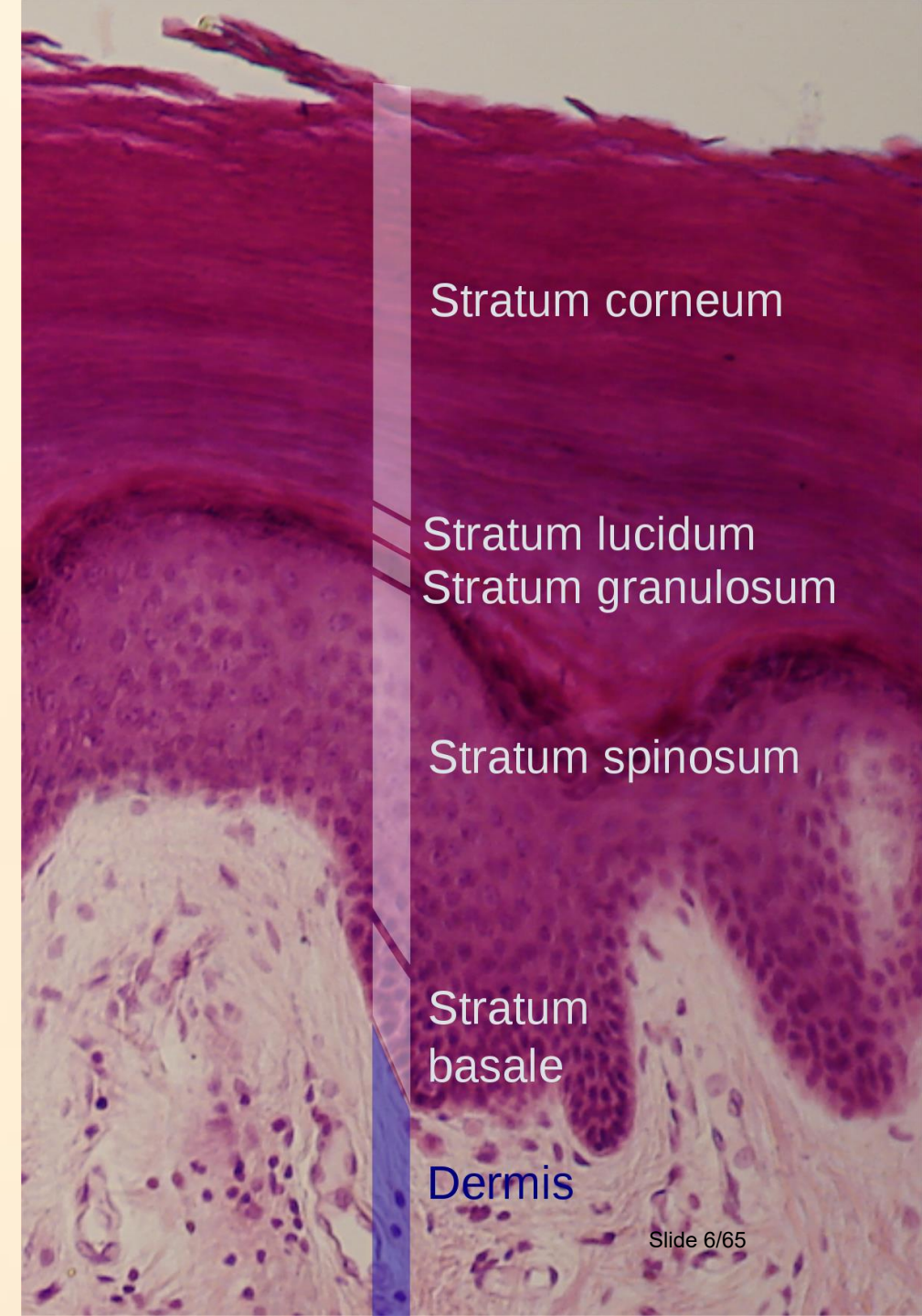


## The Structure of the Epidermis

### Keratinocytes



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Stratum corneum

Stratum lucidum  
Stratum granulosum

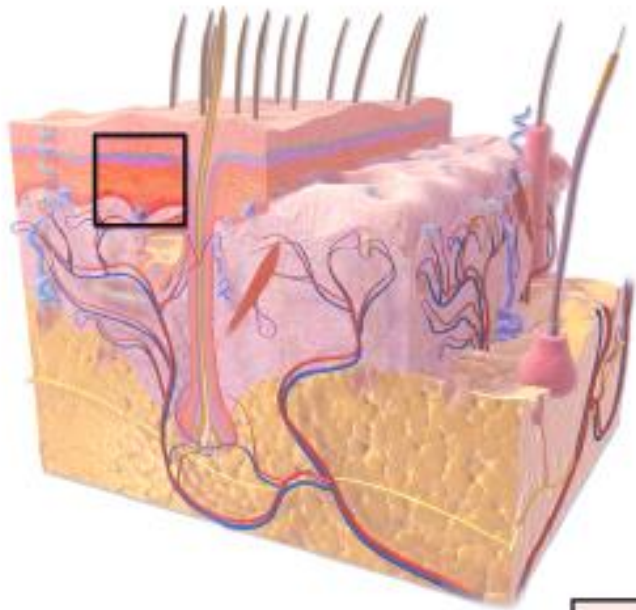
Stratum spinosum

Stratum  
basale

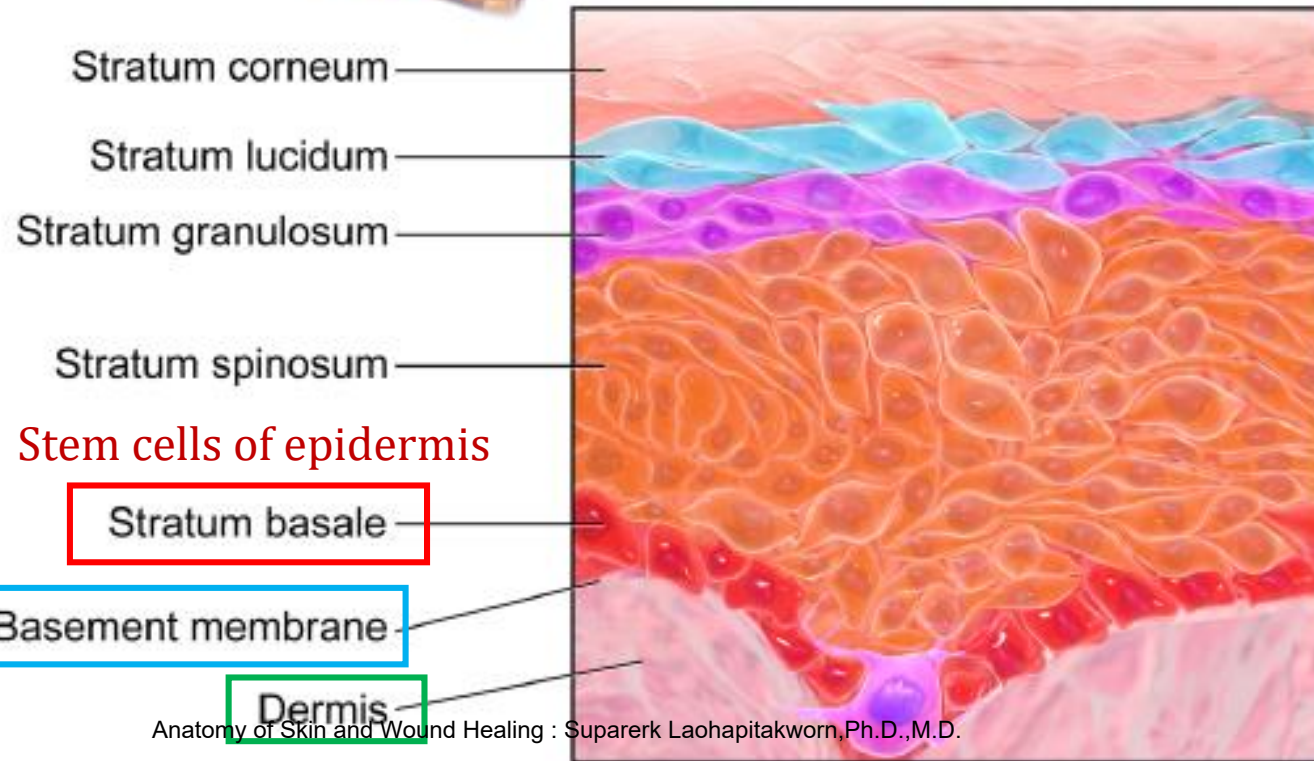
Dermis

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## The Structure of the Epidermis Keratinocytes



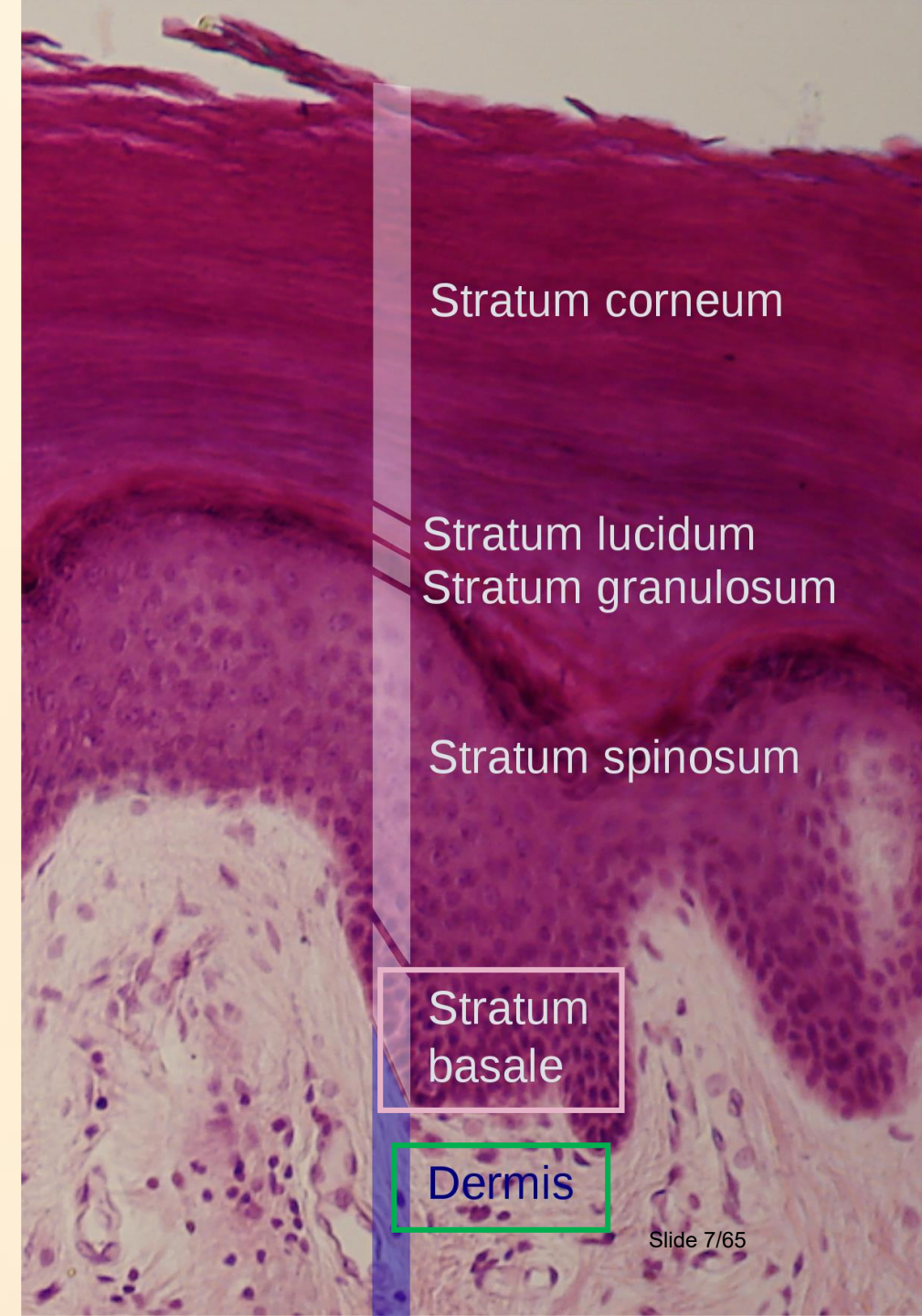
Stem cells of epidermis

Stratum basale

Basement membrane

Dermis

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Stratum corneum

Stratum lucidum  
Stratum granulosum

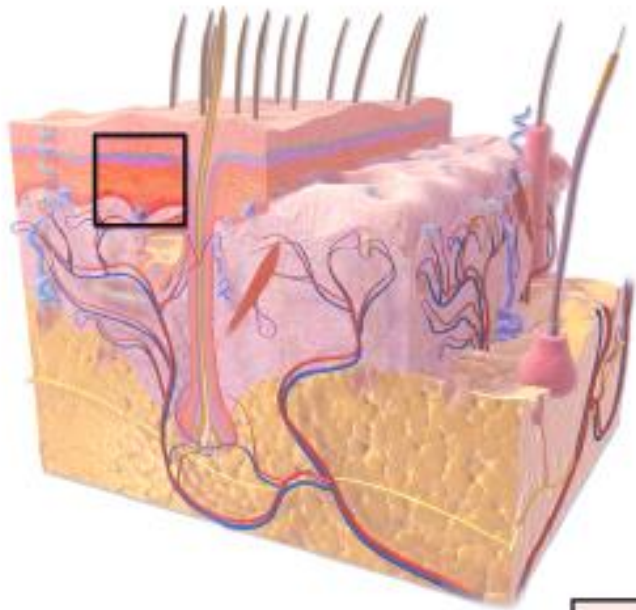
Stratum spinosum

Stratum  
basale

Dermis

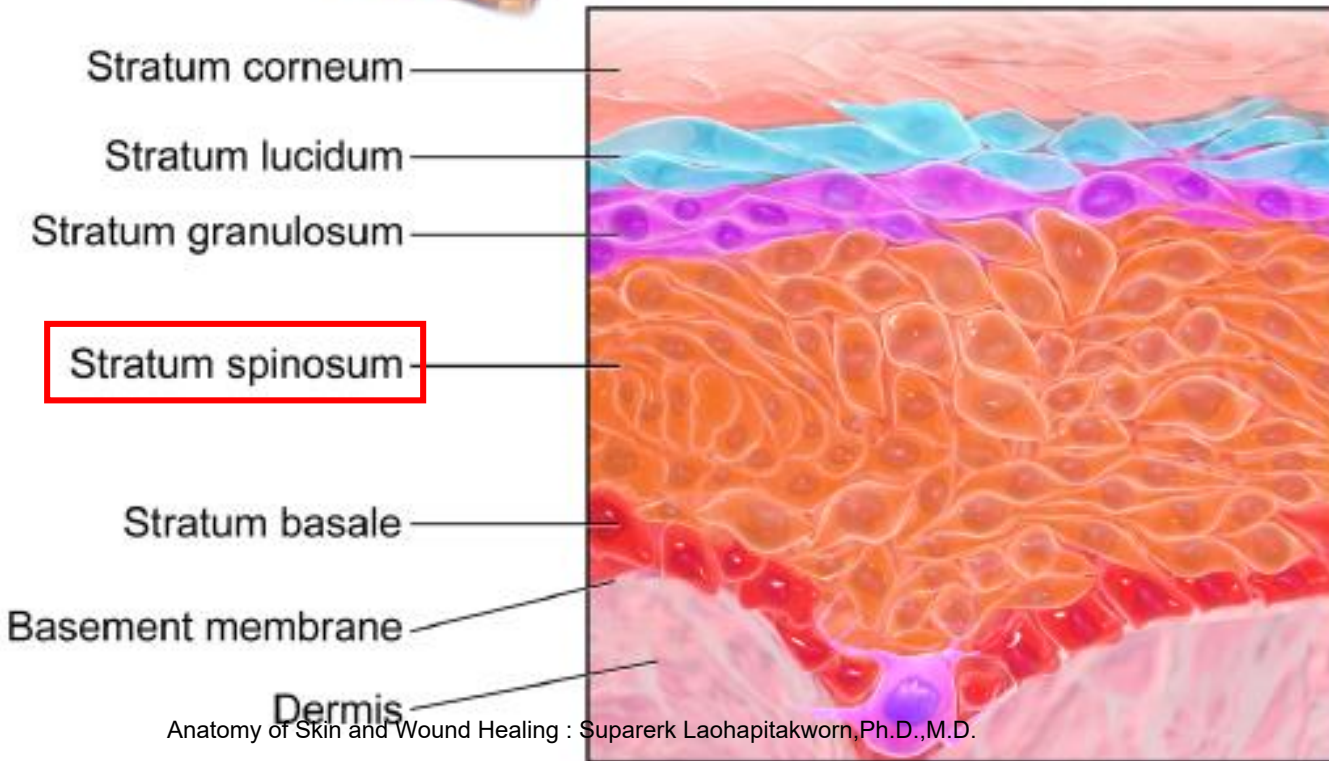
Slide 7/65



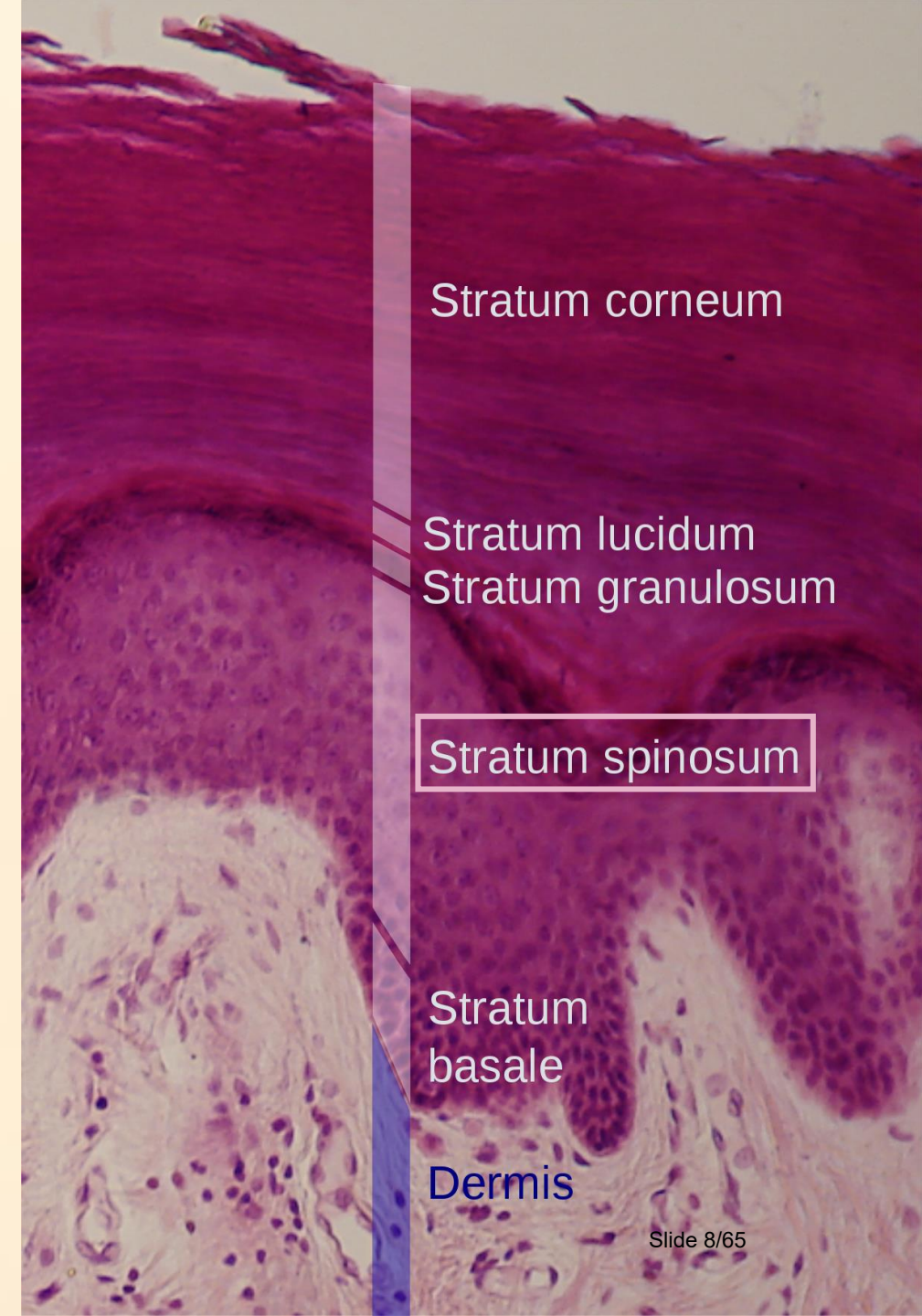


## The Structure of the Epidermis

### Keratinocytes

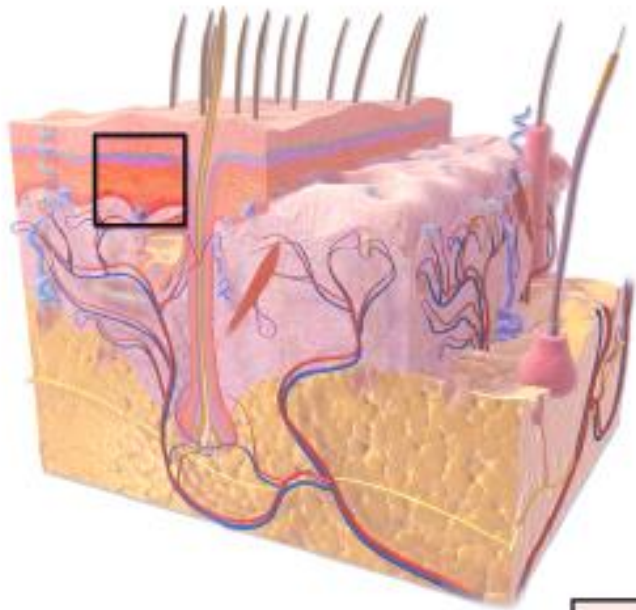


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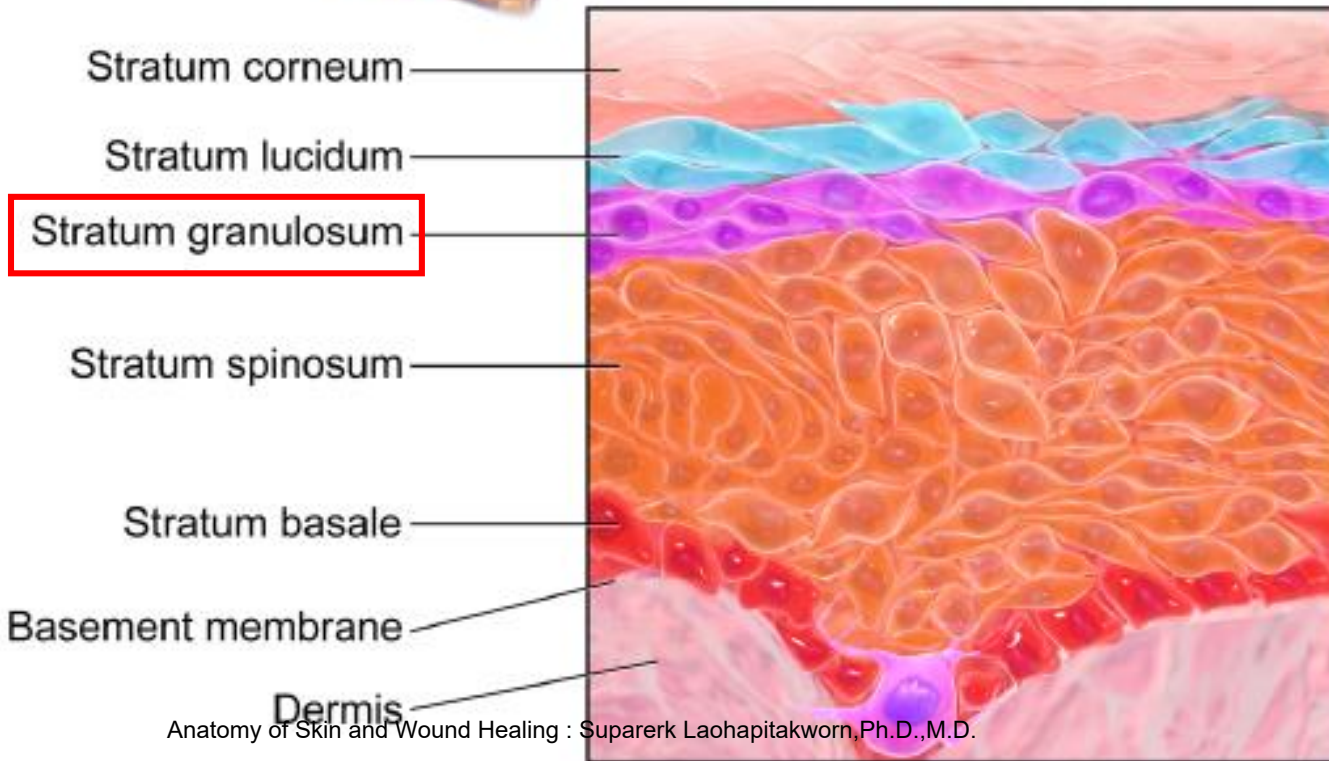
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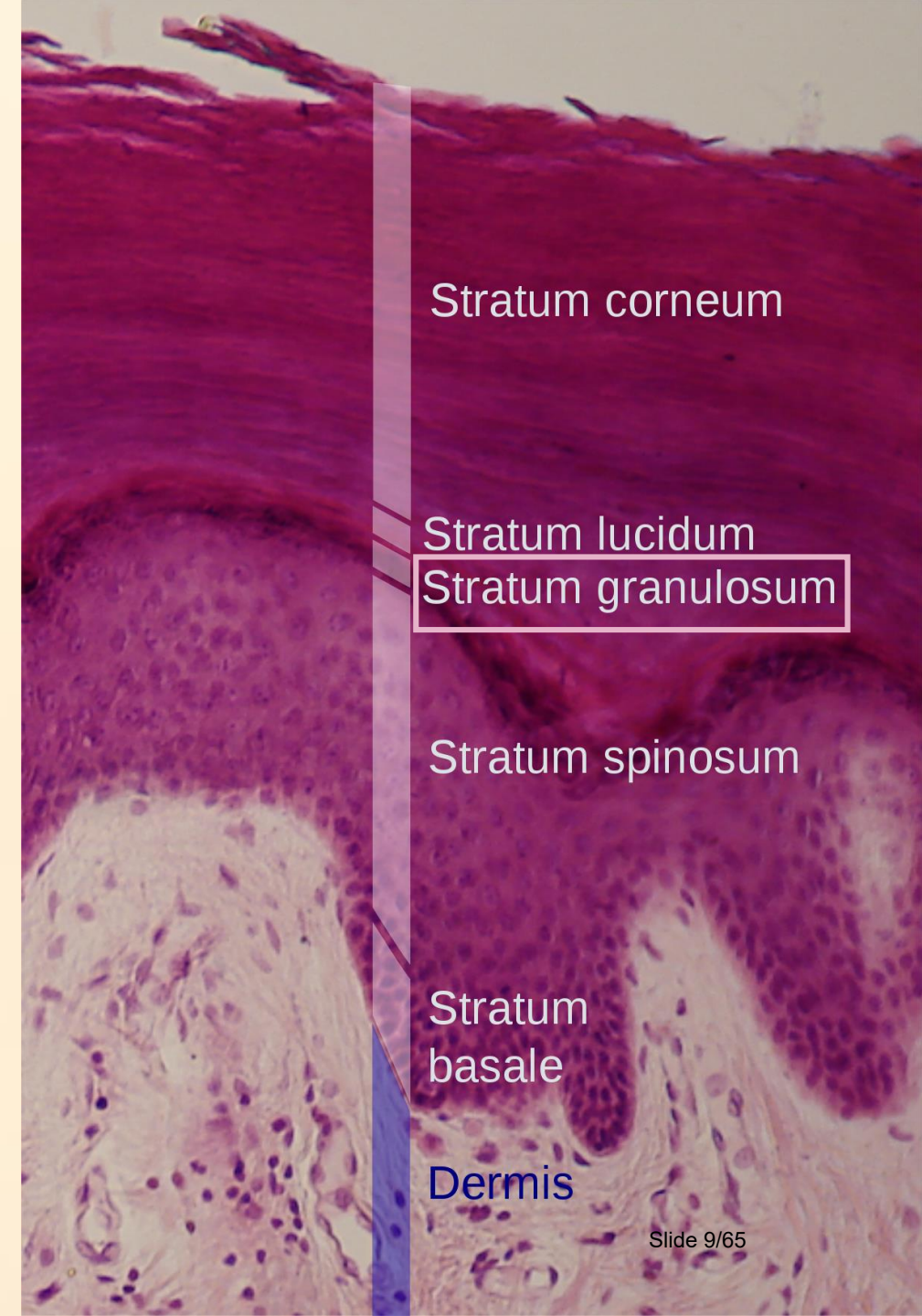


## The Structure of the Epidermis

### Keratinocytes

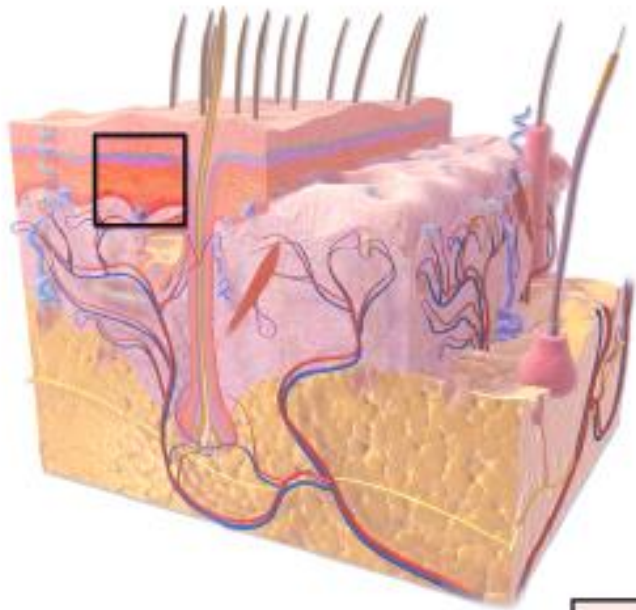


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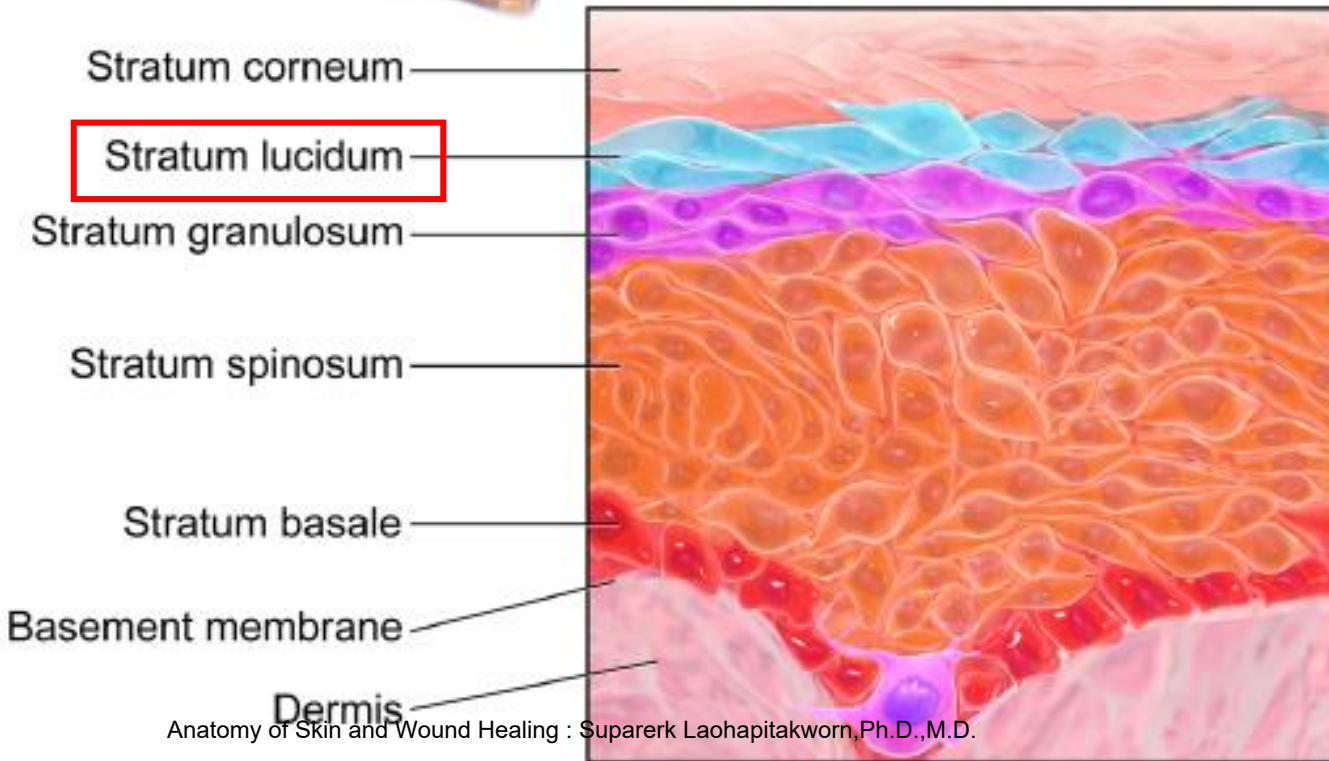
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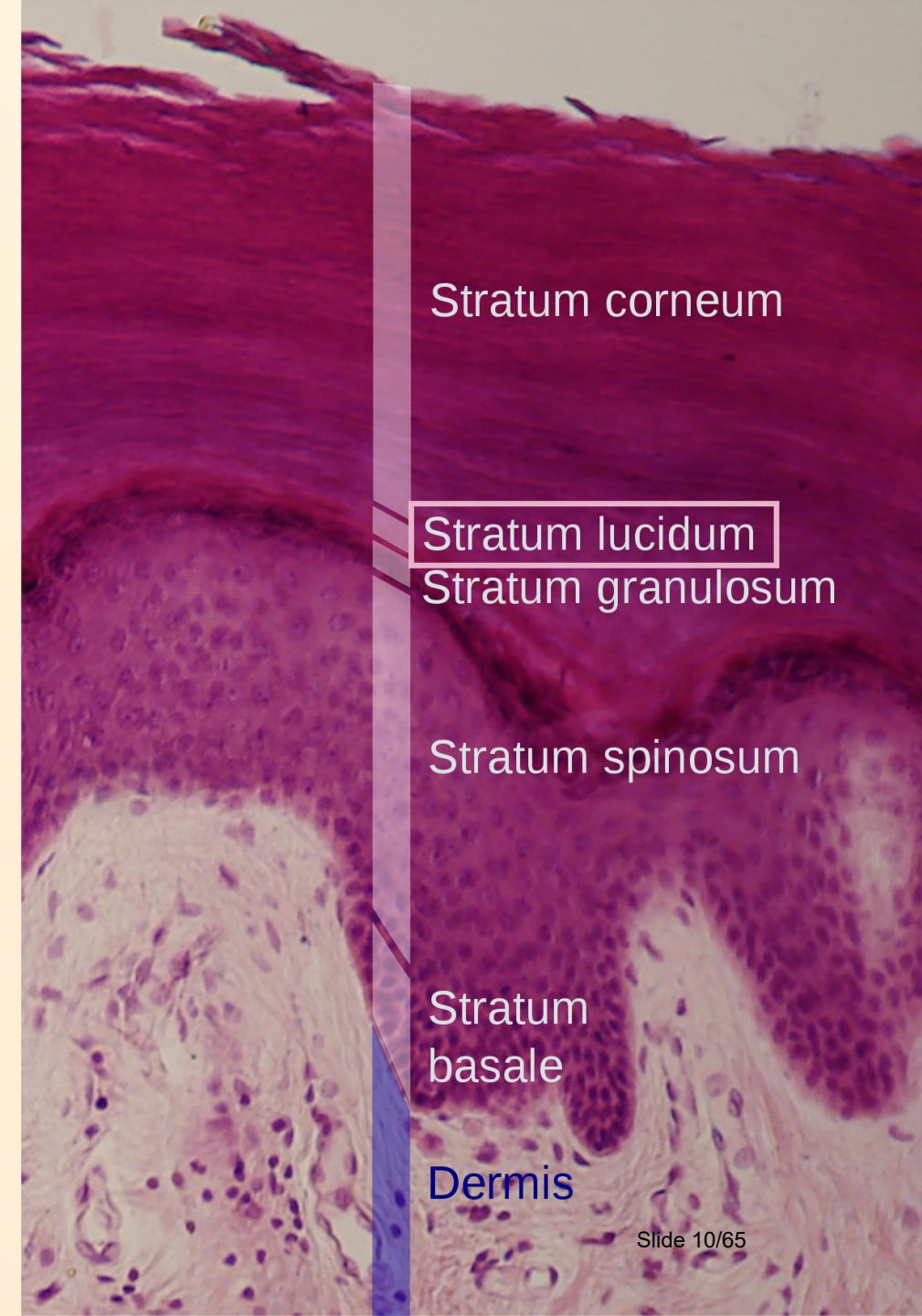


## The Structure of the Epidermis

### Keratinocytes

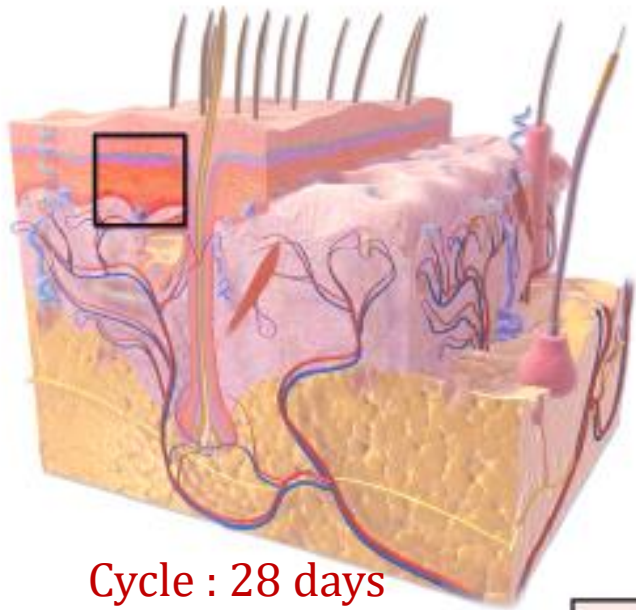


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## The Structure of the Epidermis

### Keratinocytes

Cycle : 28 days

Stratum corneum

Stratum lucidum

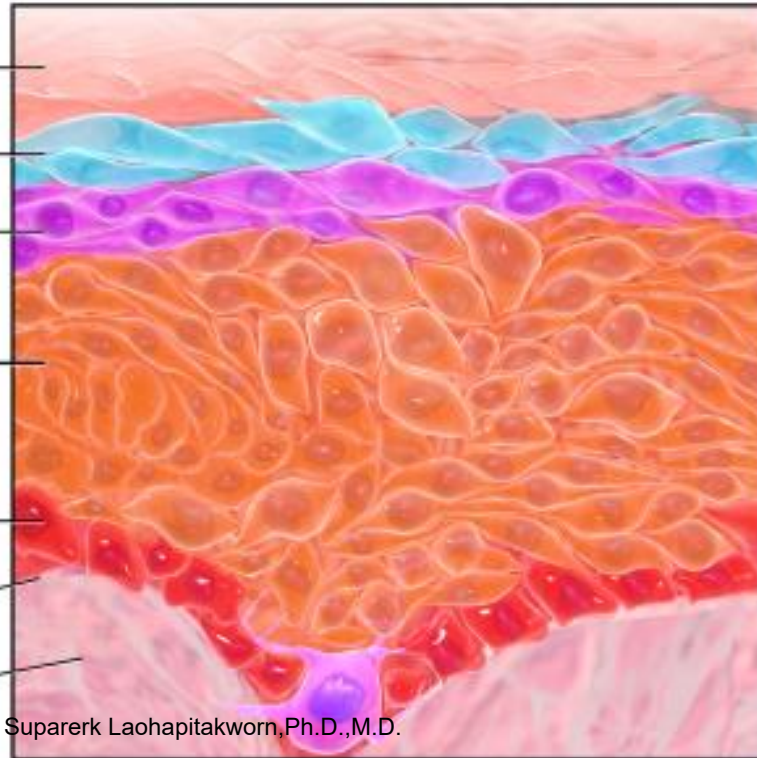
Stratum granulosum

Stratum spinosum

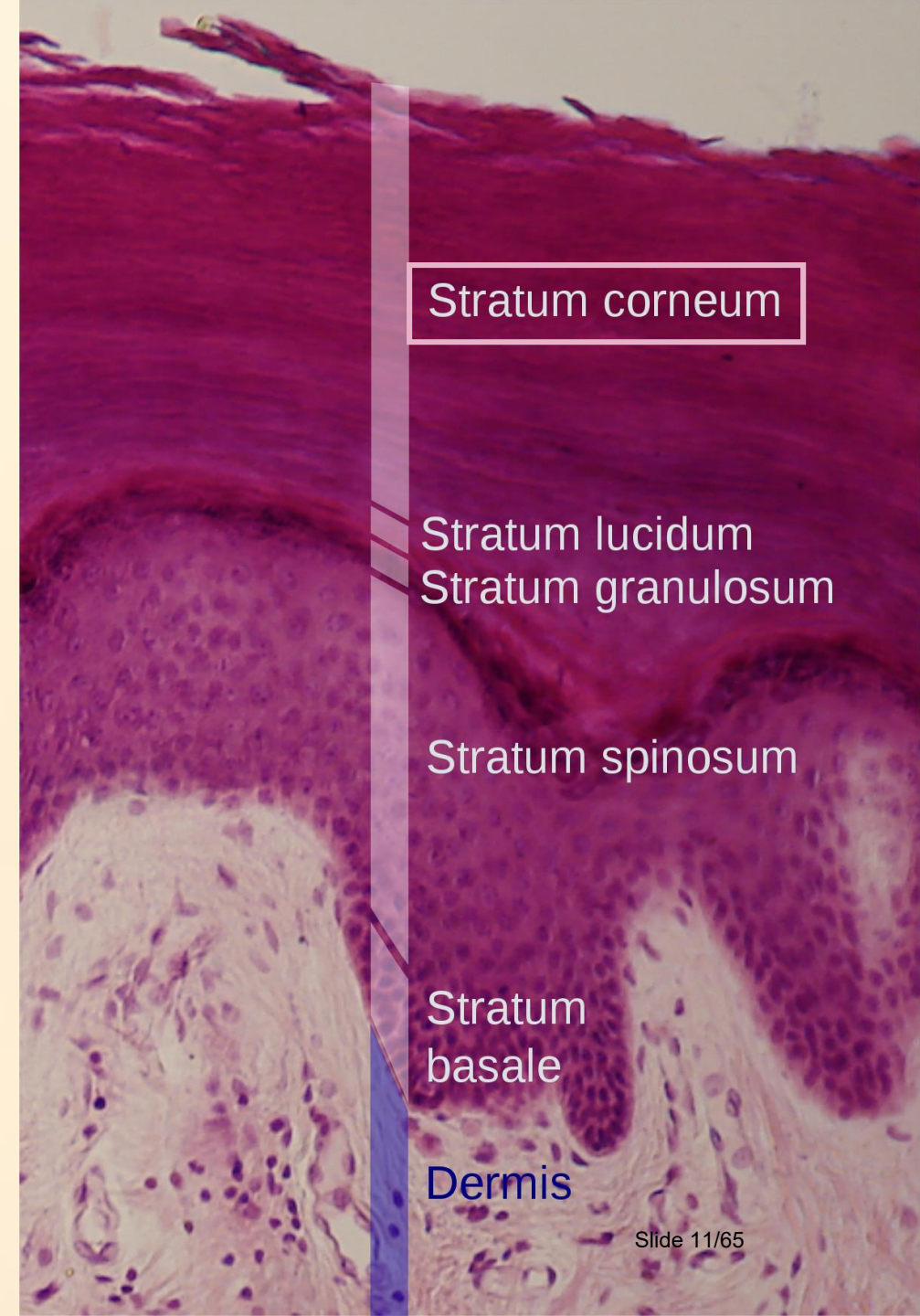
Stratum basale

Basement membrane

Dermis



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Stratum corneum

Stratum lucidum  
Stratum granulosum

Stratum spinosum

Stratum  
basale

Dermis

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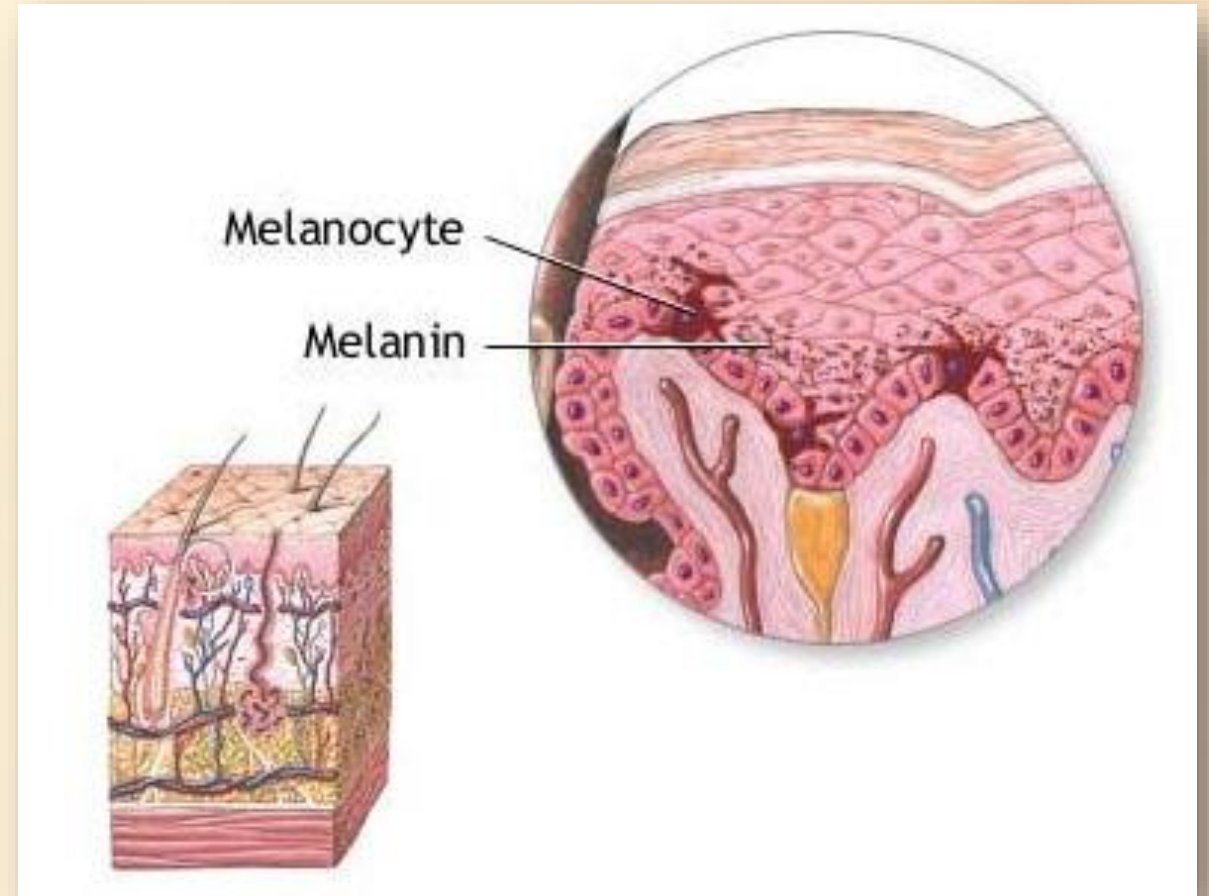
# Epidermis

- Keratinocytes



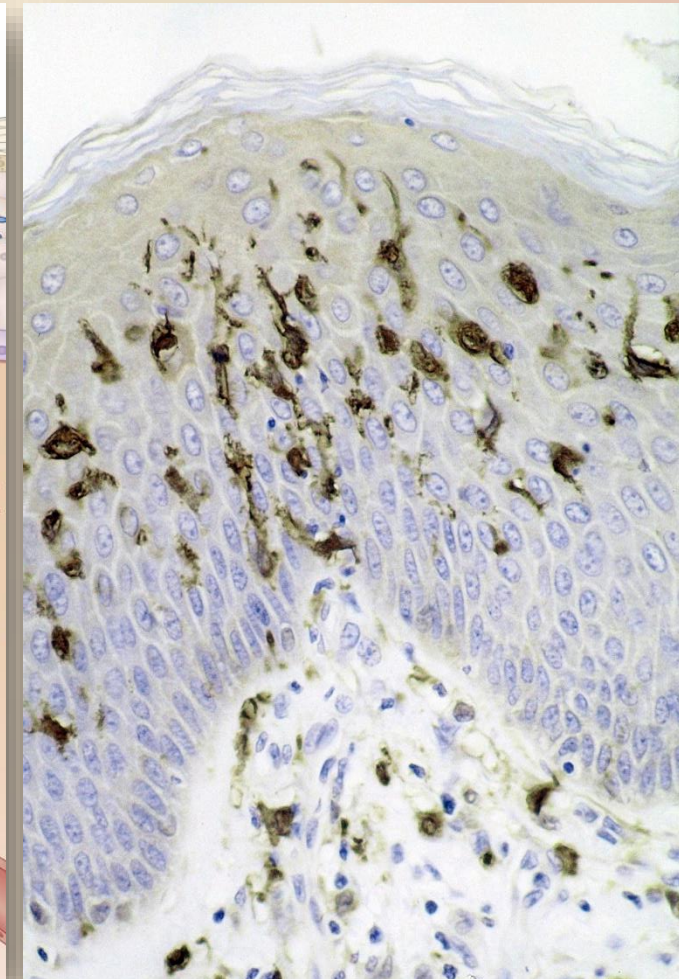
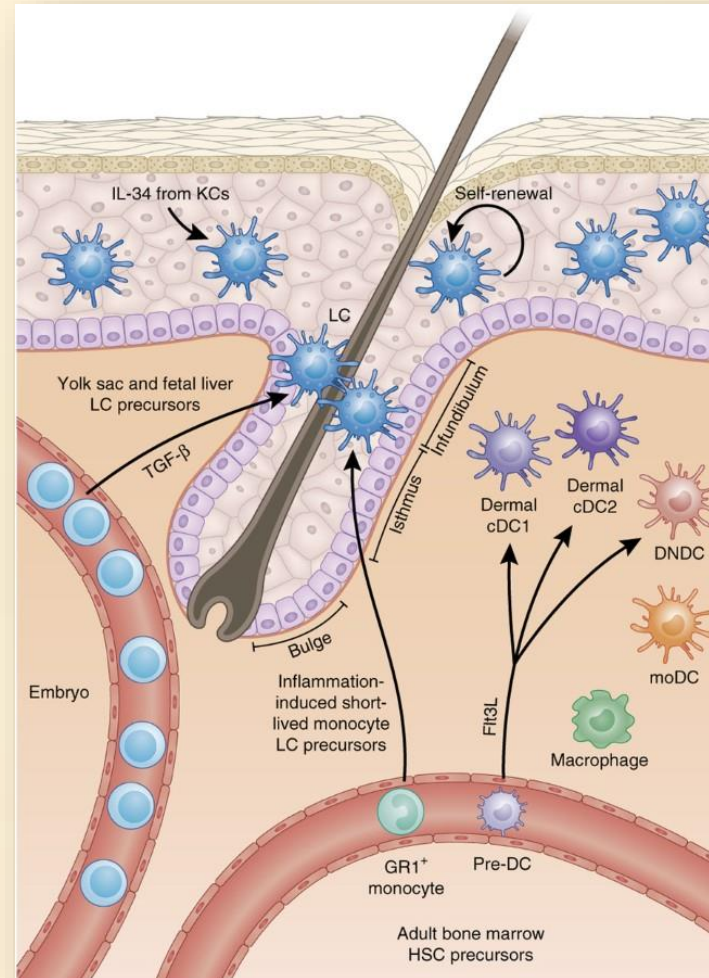
# Epidermis

- Keratinocytes
- Melanocytes : melanin production



# Epidermis

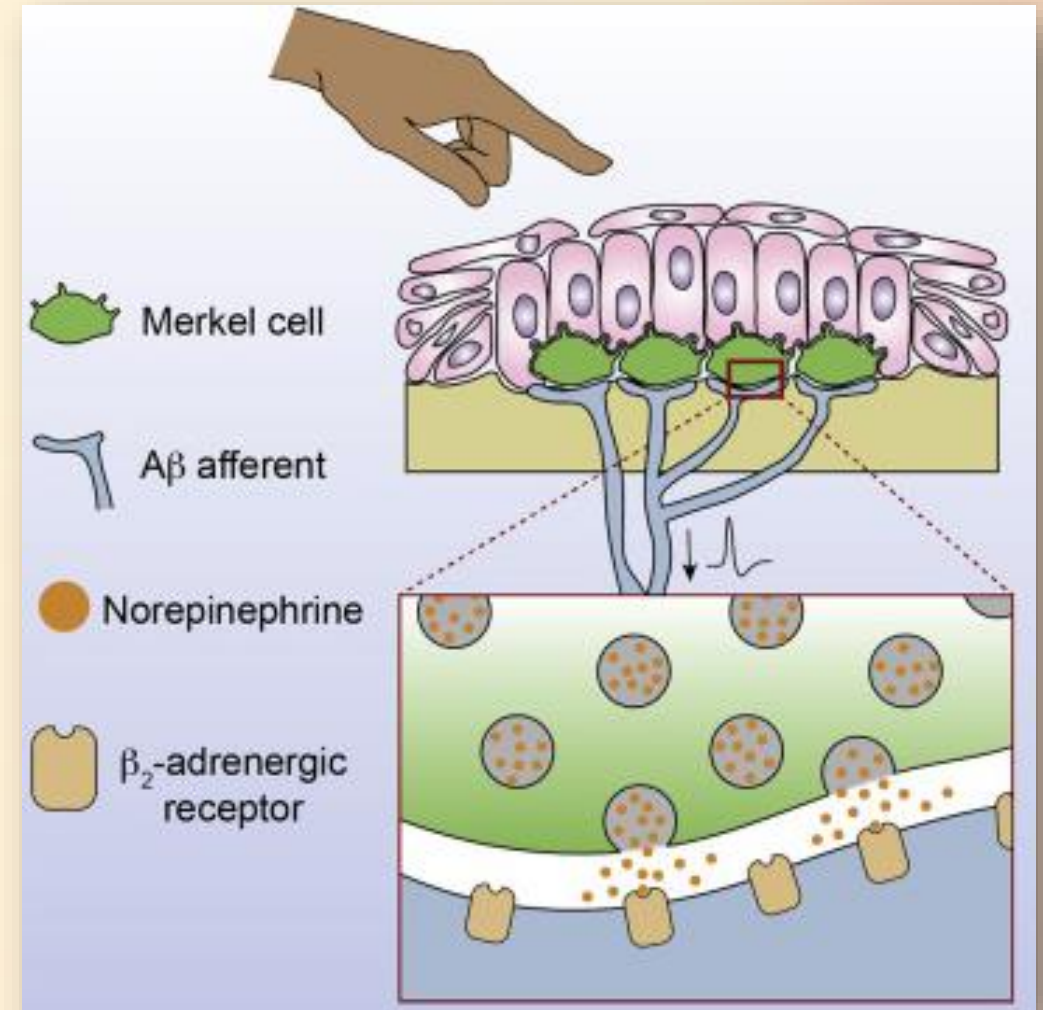
- Keratinocytes
- Melanocytes : melanin production
- Langerhans cells : immune cells (like macrophages)

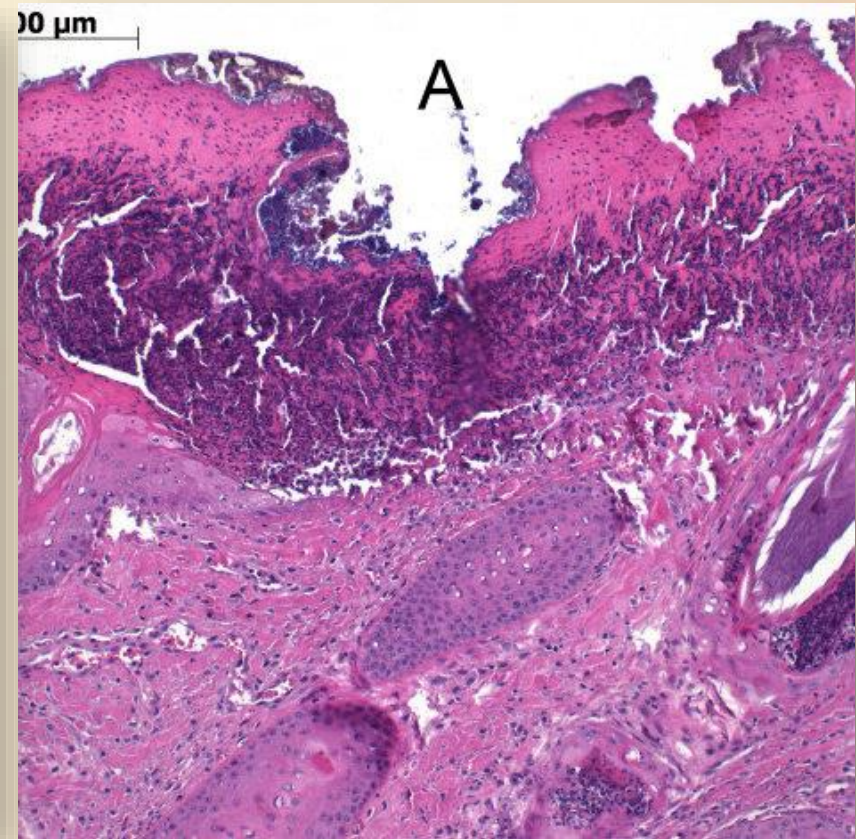




# Epidermis

- Keratinocytes
- Melanocytes : melanin production
- Langerhans cells : immune cells (like macrophages)
- Merkel cells : mechanoreceptors for palms, soles, nailbeds, oral-genital areas
- Sensitive nerves



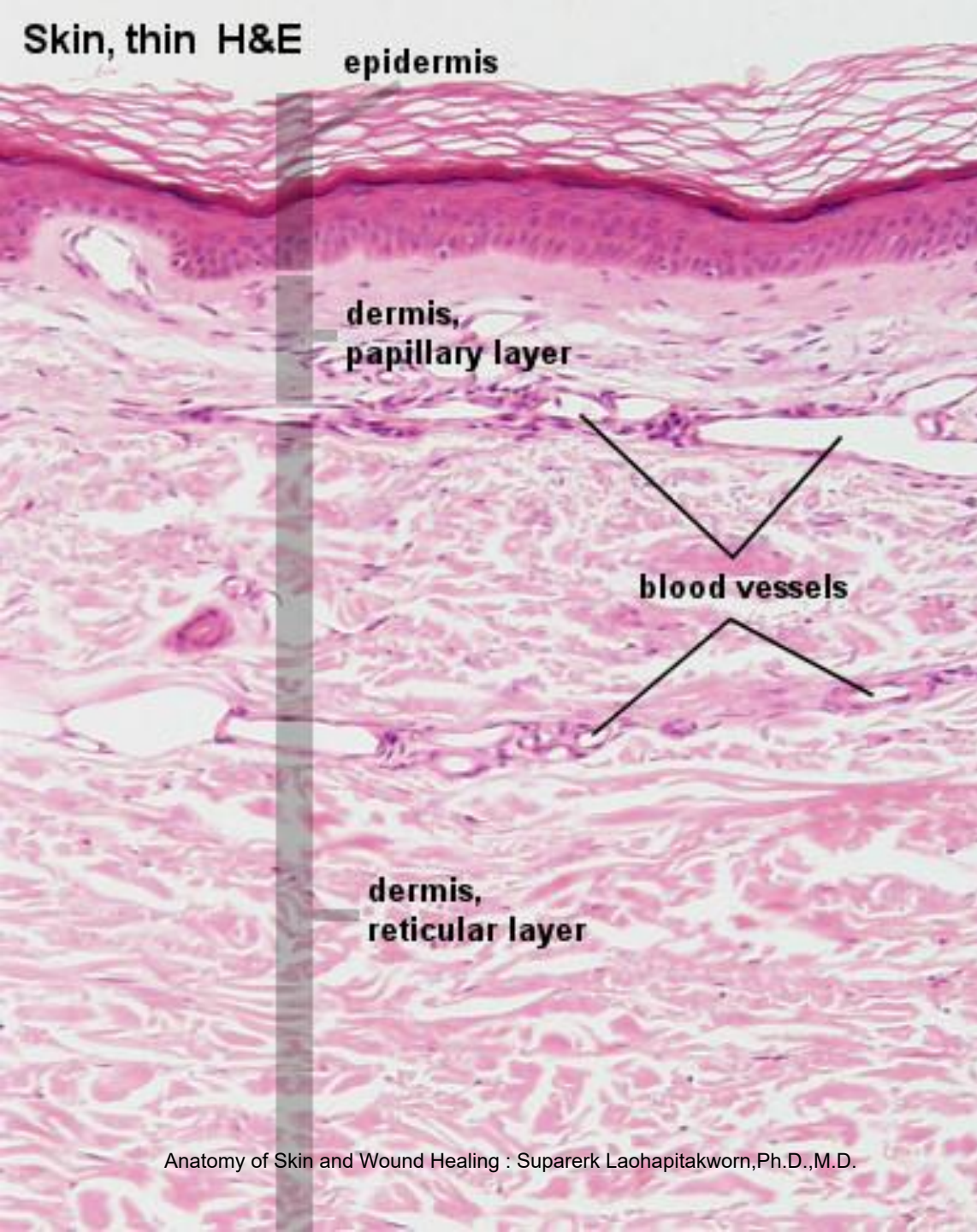


**Abrasion wound**





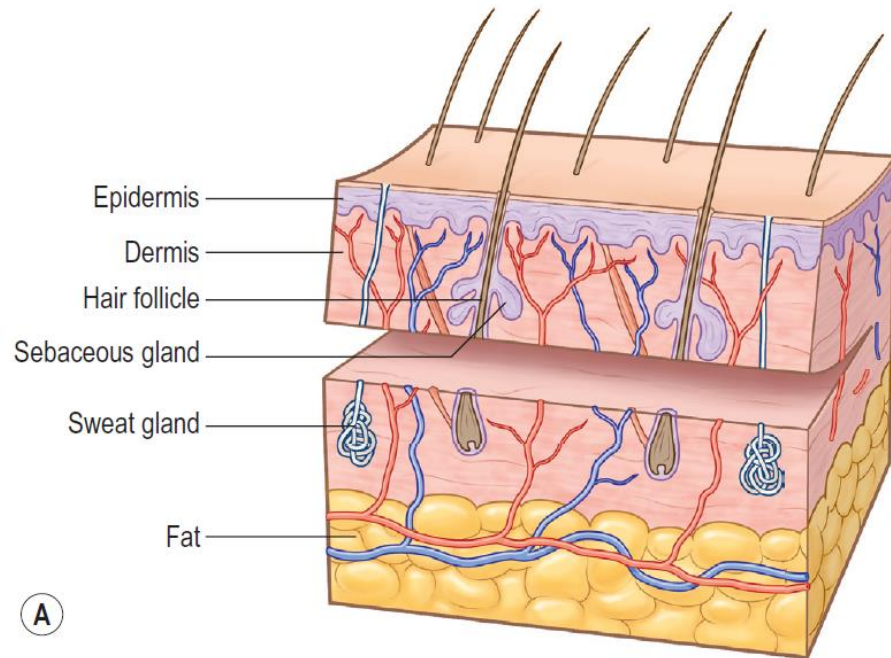
**First degree burn : sunburn**



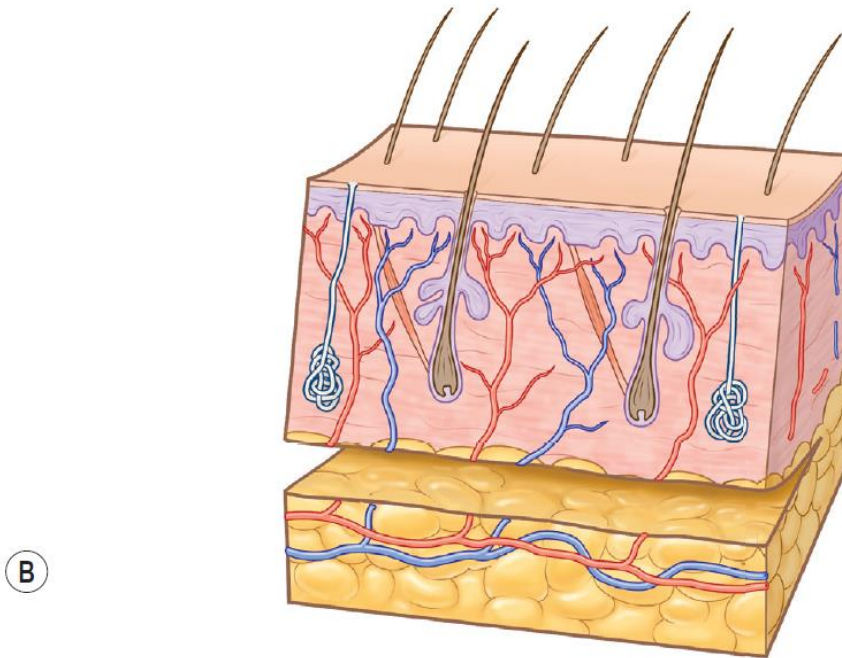
# Dermis

- Main components (from fibroblasts)
  - Collagens
  - Glycosaminoglycans
  - Elastins
- Mechanical features of the skin





**Split-thickness skin grafting**



**Full-thickness skin grafting**



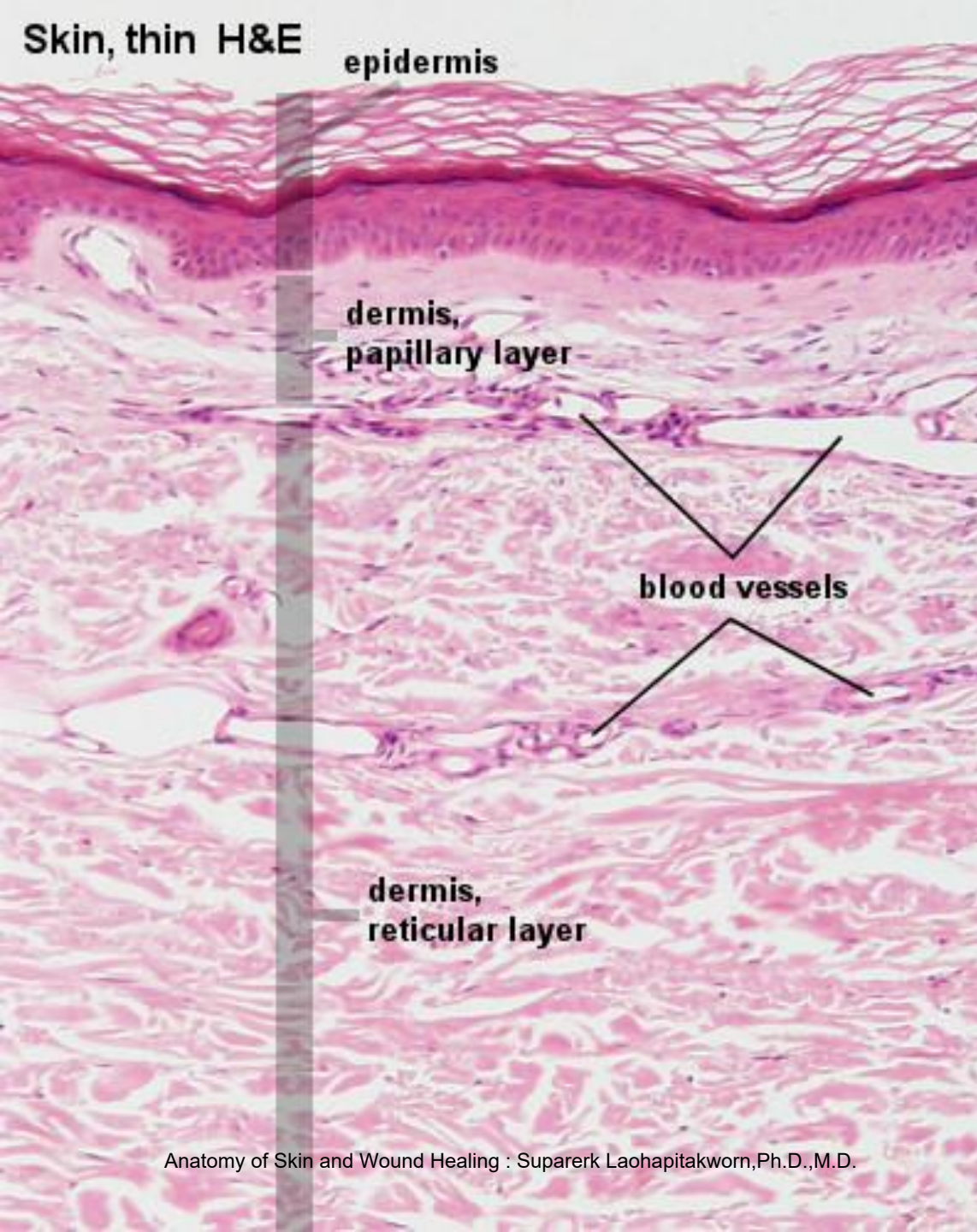
**Split-thickness skin grafting**



**Full-thickness skin grafting**

- Less secondary contraction
- Excellent skin quality and stability
- Hair regrowth and skin appendage function

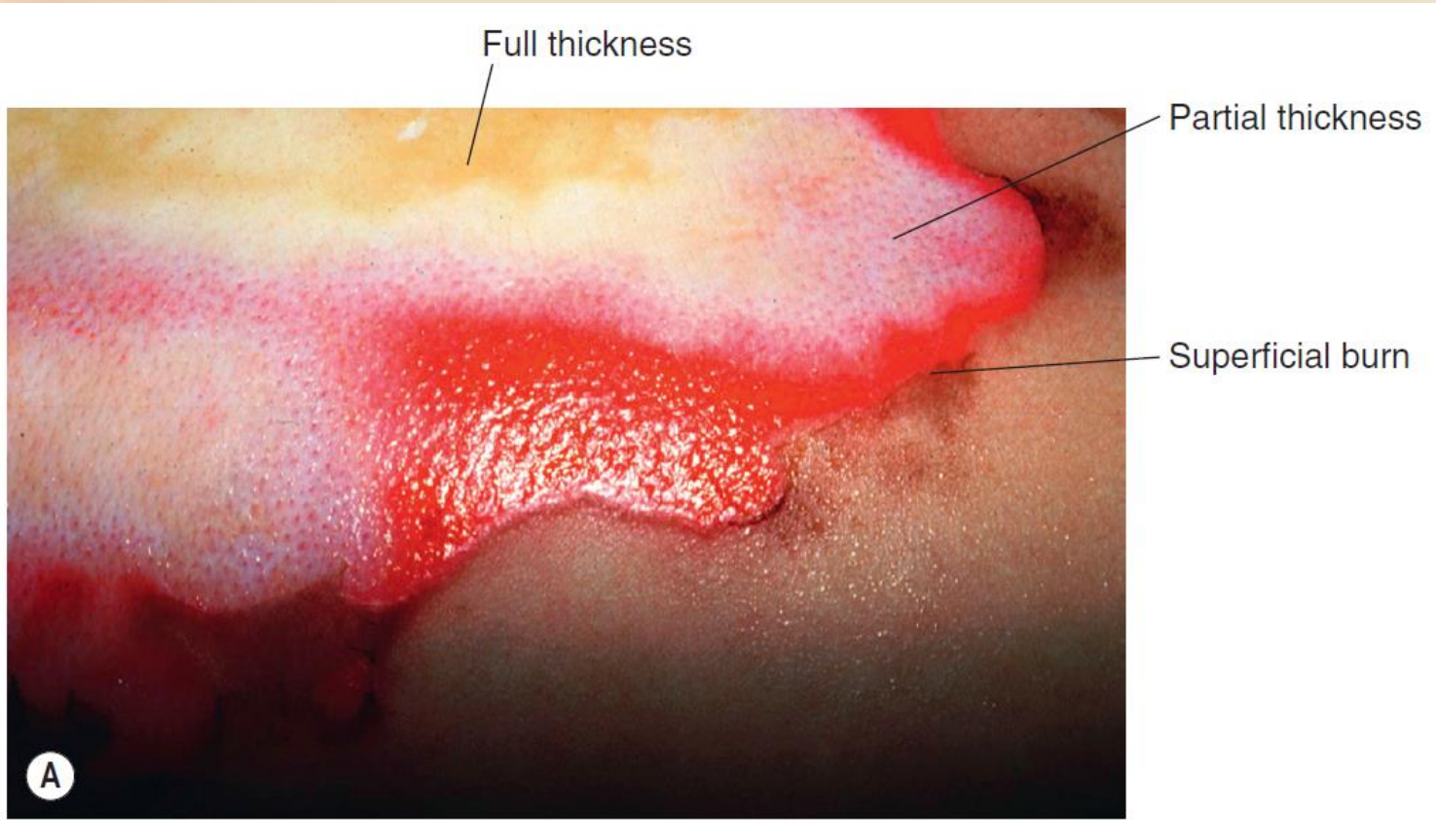




# Dermis

- Papillary dermis
  - Fine collagenous fibers
  - Undulating interface : mechanical stability
  - Blood vessels : rich papillary plexus and nerve fibers
- Reticular dermis
  - Thicker collagenous fibers
  - Blood vessels : reticular plexus

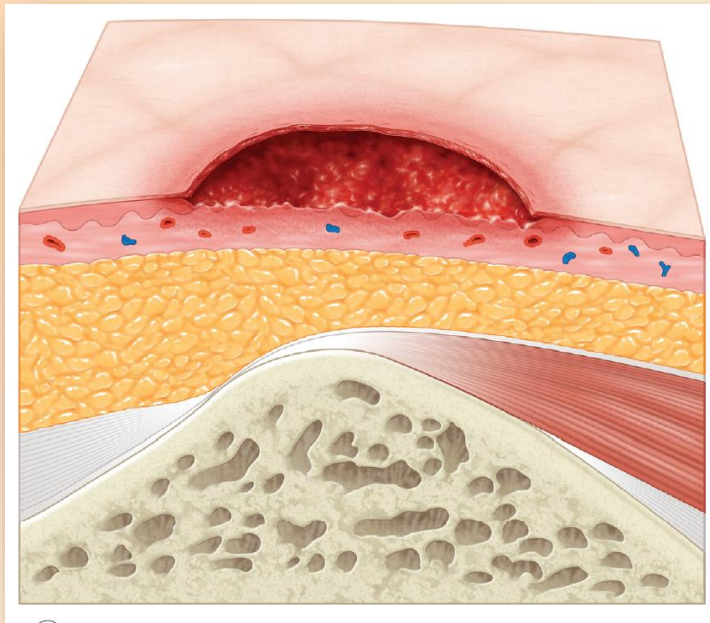
## 3<sup>rd</sup> degree burn (full-thickness loss of dermis)



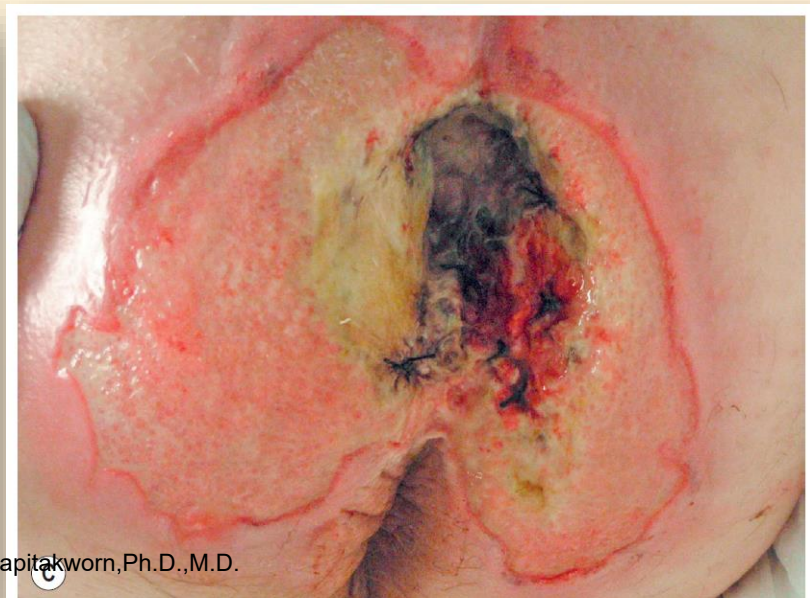
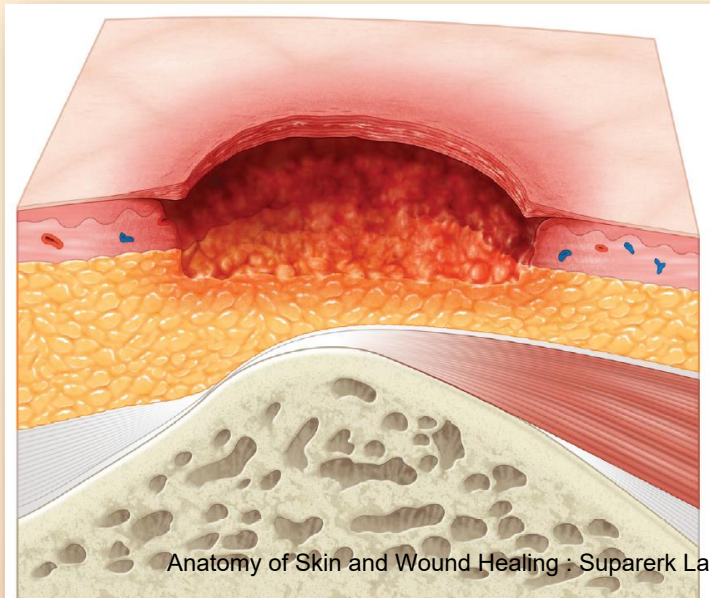
## 2<sup>nd</sup> degree burn

- Superficial (papillary dermis)
- Deep (reticular dermis)





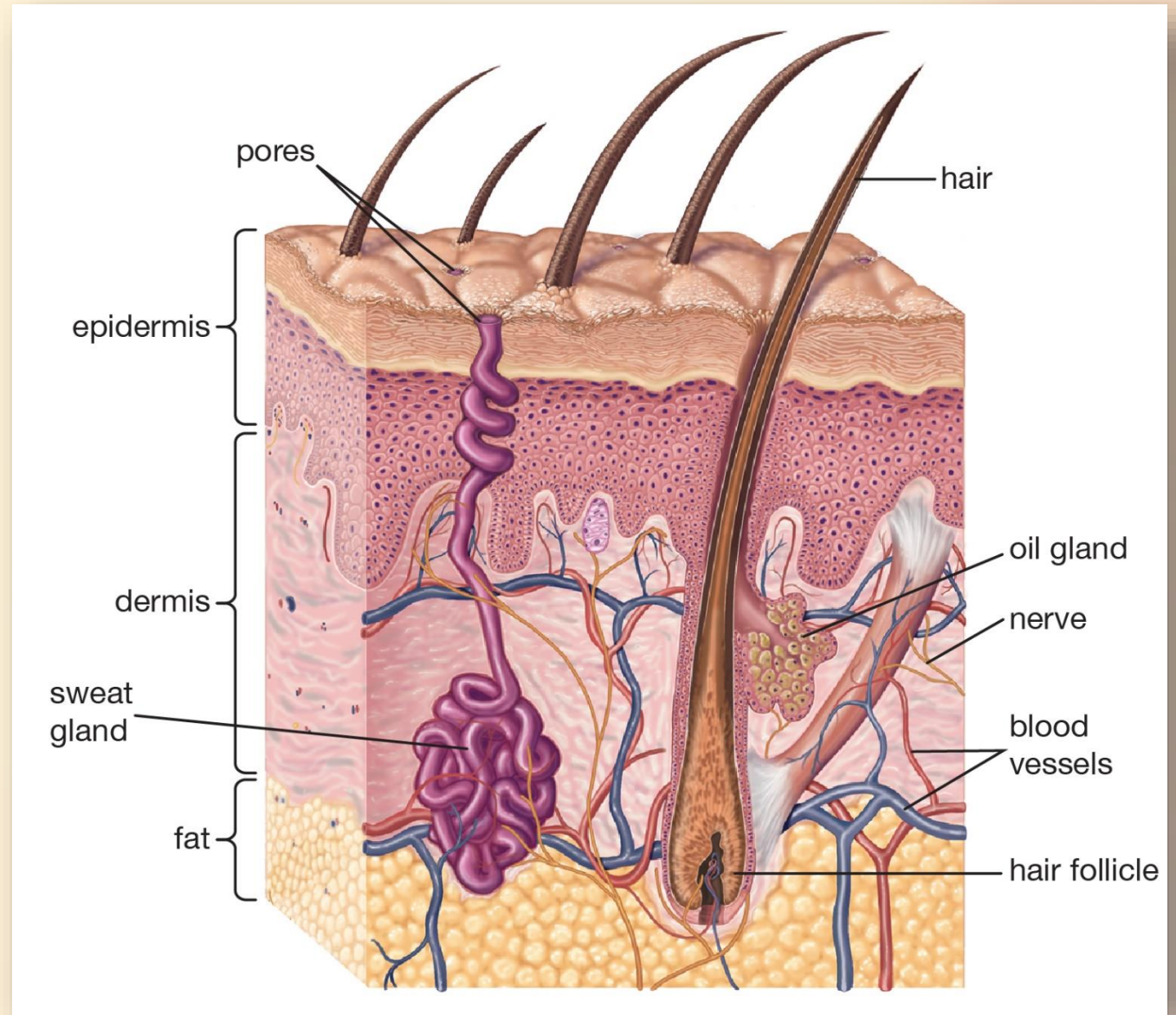
**Pressure ulcer grade 2**  
(partial-thickness loss of dermis)



**Pressure ulcer grade 3**  
(full-thickness loss of dermis)

# Adnexal structures

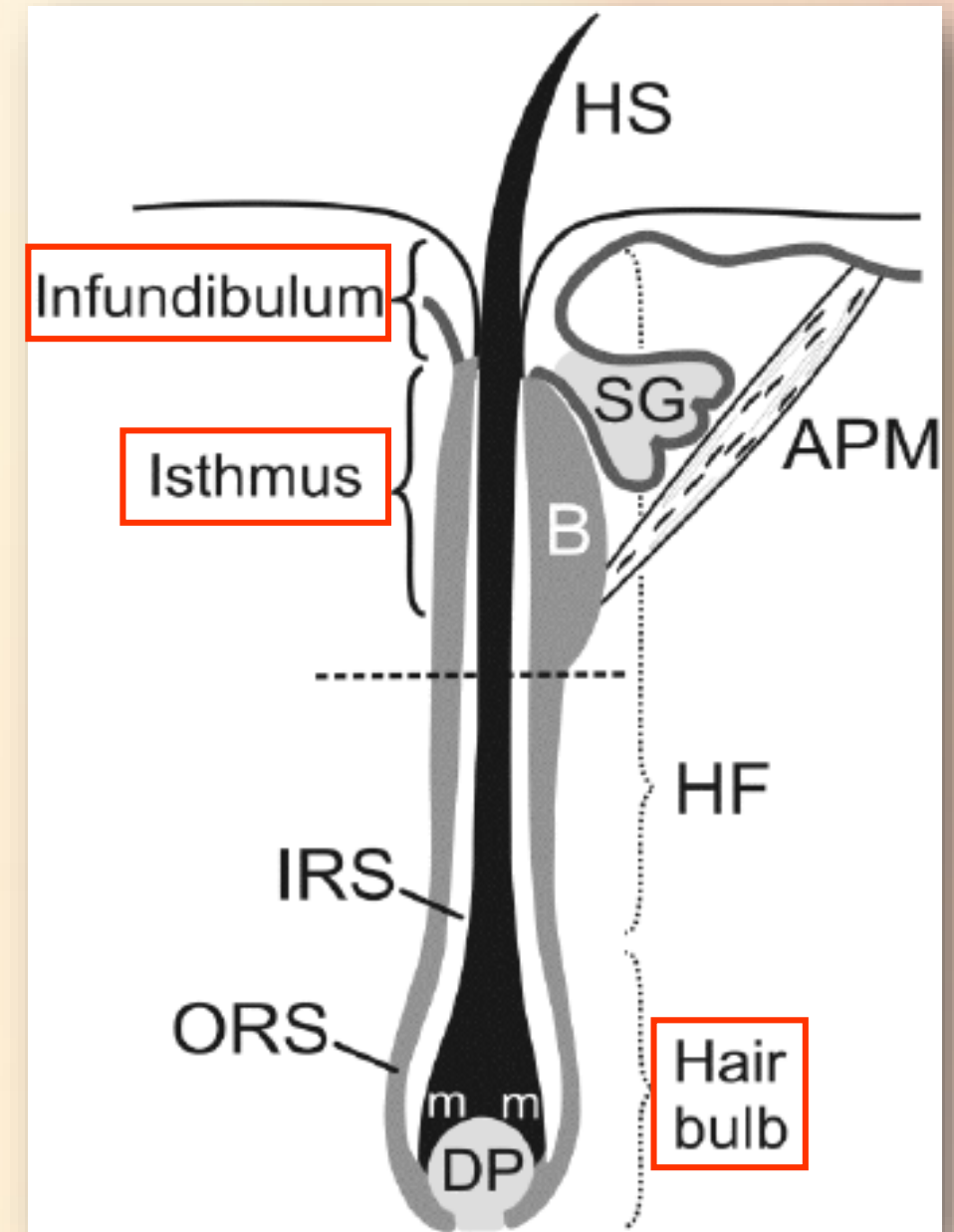
- Hair follicles
- Sebaceous glands
- Sweat glands





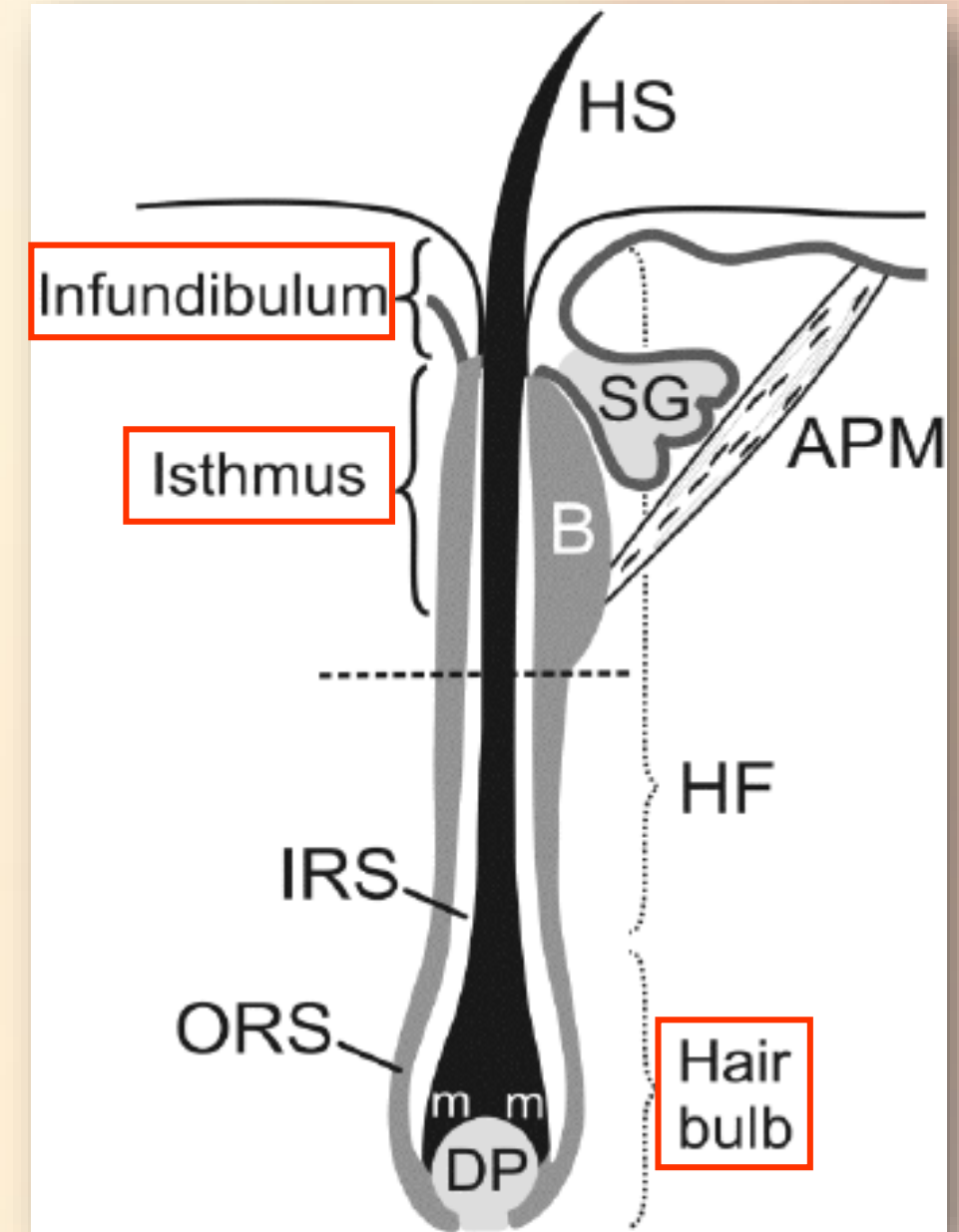
# Hair follicles

- Infundibulum
- Isthmus
- Suprabulbar
- Bulb



# Hair follicles

- Infundibulum
- Isthmus : bulge (multipotent stem cells)
  - Hair follicles
  - Sebaceous glands
  - Epidermis
- Suprabulbar
- Bulb





# Transplantation of the LGR6<sup>+</sup> Epithelial Stem Cell into Full-Thickness Cutaneous Wounds Results in Enhanced Healing, Nascent Hair Follicle Development, and Augmentation of Angiogenic Analytes

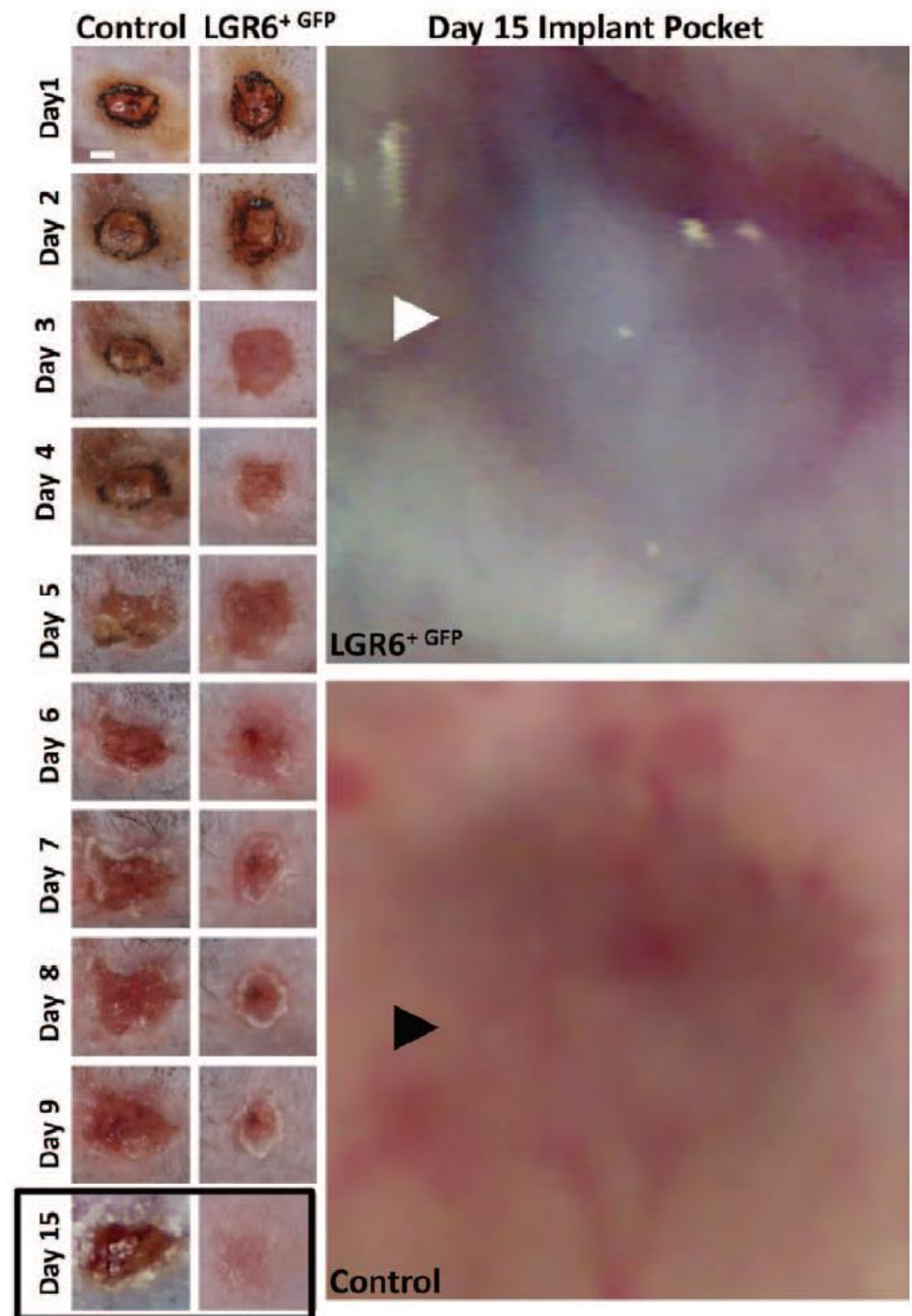
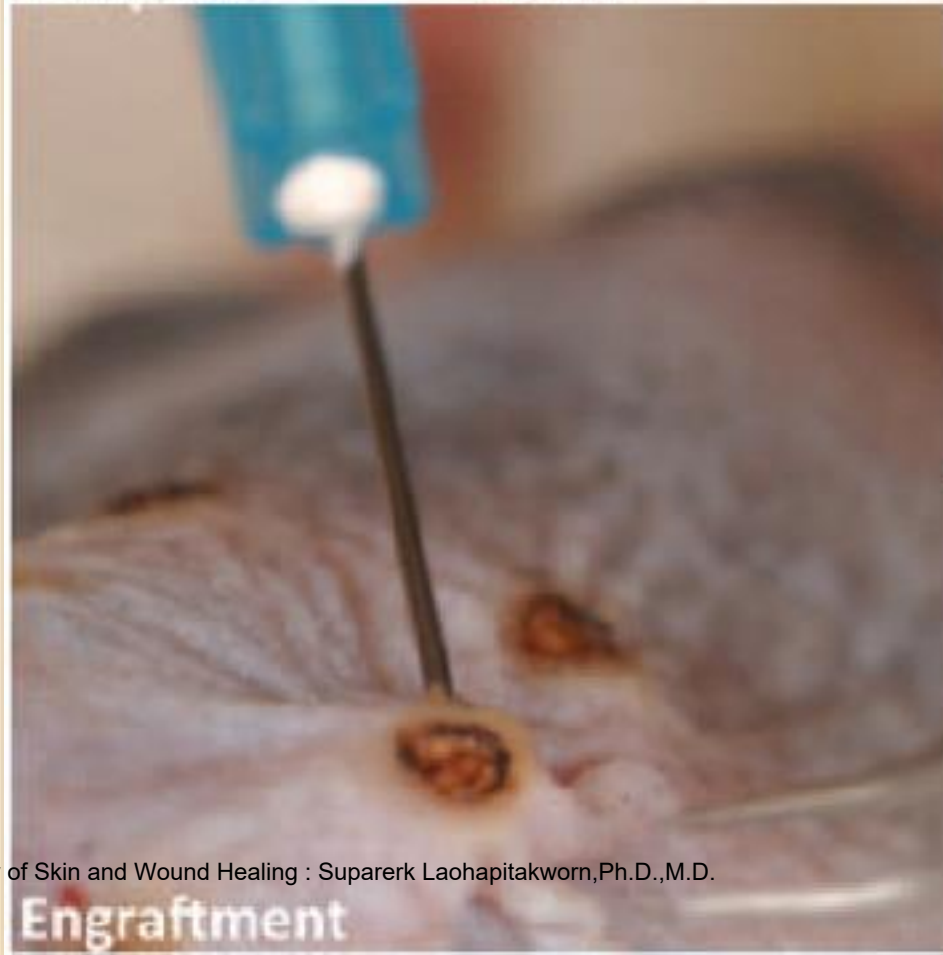
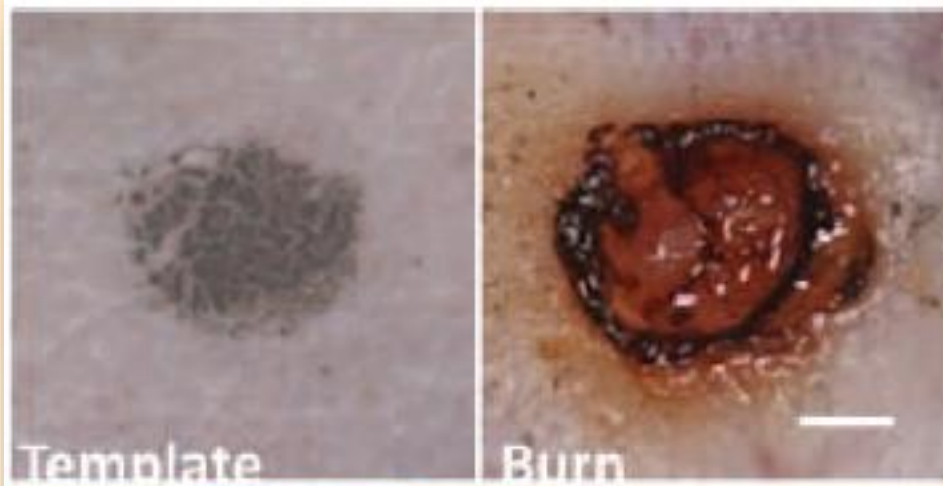
Denver M. Lough, M.D.,  
Ph.D.  
Mei Yang, M.D., Ph.D.  
Anthony Blum, B.S.  
Joel D. Reichensperger, B.S.  
Nicole M. Cosenza, M.S.  
Nathan Wetter, M.D.  
Lisa A. Cox, B.S.  
Carrie E. Harrison, B.S.  
Michael W. Neumeister, M.D.  
*Springfield, Ill.; and Baltimore, Md.*

**Background:** The recently discovered leucine-rich repeat-containing G-protein coupled receptor 6 (LGR6<sup>+</sup>) epithelial stem cell located within the follicular bulge of the adnexal compartment is capable of producing all cellular lineages of the skin. In this study, the authors sought to determine whether these cells can be transplanted for use as a type of cellular therapy for the repair of full-thickness wounds in which the native stem cell niche has been obliterated.

**Methods:** Full-thickness murine skin was harvested and LGR6<sup>+</sup>GFP epithelial stem cells were isolated using fluorescence-activated cell sorting. This enriched epithelial stem cell population was then transplanted by means of local injection into wound beds on the dorsum of nude mice. Viability, migration, healing, the development of nascent hair follicles, and gene and proteomic expression studies were performed to determine whether the engraftment of LGR6<sup>+</sup>GFP epithelial stem cells enhanced healing when compared with controls.

**Results:** Wound beds receiving LGR6<sup>+</sup>GFP epithelial stem cells showed enhanced healing; nascent follicle growth; and augmentation of the Wnt, vascular endothelial growth factor, epidermal growth factor, and platelet-derived growth factor pathways when compared with controls.

**Conclusions:** The LGR6<sup>+</sup> epithelial stem cells appear to hold great promise for the development of a clinically useful stem cell-based therapy for the repair of full-thickness wounds and hair regeneration. These results indicate that transplantation of LGR6<sup>+</sup> epithelial stem cells promotes epithelialization, hair growth, and angiogenesis in tissues destined for scar formation. (*Plast. Reconstr. Surg.* 133: 579, 2014.)





# Reepithelialization from Stem Cells of Hair Follicles of Dermal Graft of the Scalp in Acute Treatment of Third-Degree Burns: First Clinical and Histologic Study

Gilbert Zakine, M.D., Ph.D.

Maurice Mimoun, M.D.

Julien Pham, M.D.

Marc Chaouat, M.D., Ph.D.

*Tours and Paris, France*

**Background:** The scalp, an excellent donor site for thin skin grafts, presents a limited surface but is rich in keratinocyte stem cells. The purpose of this study was to double scalp harvesting in one procedure and to evaluate the capacity of the dermal layer to spontaneously reepithelialize from hair follicle stem cells.

**Methods:** Two layers of 0.2-mm split-thickness skin graft, a dermoepidermal graft and a dermal graft, were harvested from scalp during the same procedure. Fifteen burn patients were included in this study. Healing of the scalp donor site and percentage of graft taken were evaluated. The Vancouver Scar Scale was used at 3 months and 1 year. Histologic studies were performed at day 0 and 3 months on grafts, and on the scalp at day 28.

**Results:** Nine patients were treated on the limbs with meshed dermal graft. Six were treated on the hands with unmeshed dermal graft. Graft take was good for both types of grafts. The mean time for scalp healing was 9.3 days. Histologic study confirmed that the second layer was a dermal graft with numerous annexes and that, at 3 months, the dermis had normal thickness but with rarer and smaller epidermal crests than dermal graft. The difference between the mean Vancouver Scar Scale score of dermal graft and dermoepidermal graft was not significant.

**Conclusion:** The authors' study shows the efficacy of dermal graft from the scalp and good scalp healing. (*Plast. Reconstr. Surg.* 130: 42e, 2012.)

**CLINICAL QUESTION/LEVEL OF EVIDENCE:** Therapeutic, II.





Dermoepidermal graft

Dermal graft

0.008 inch



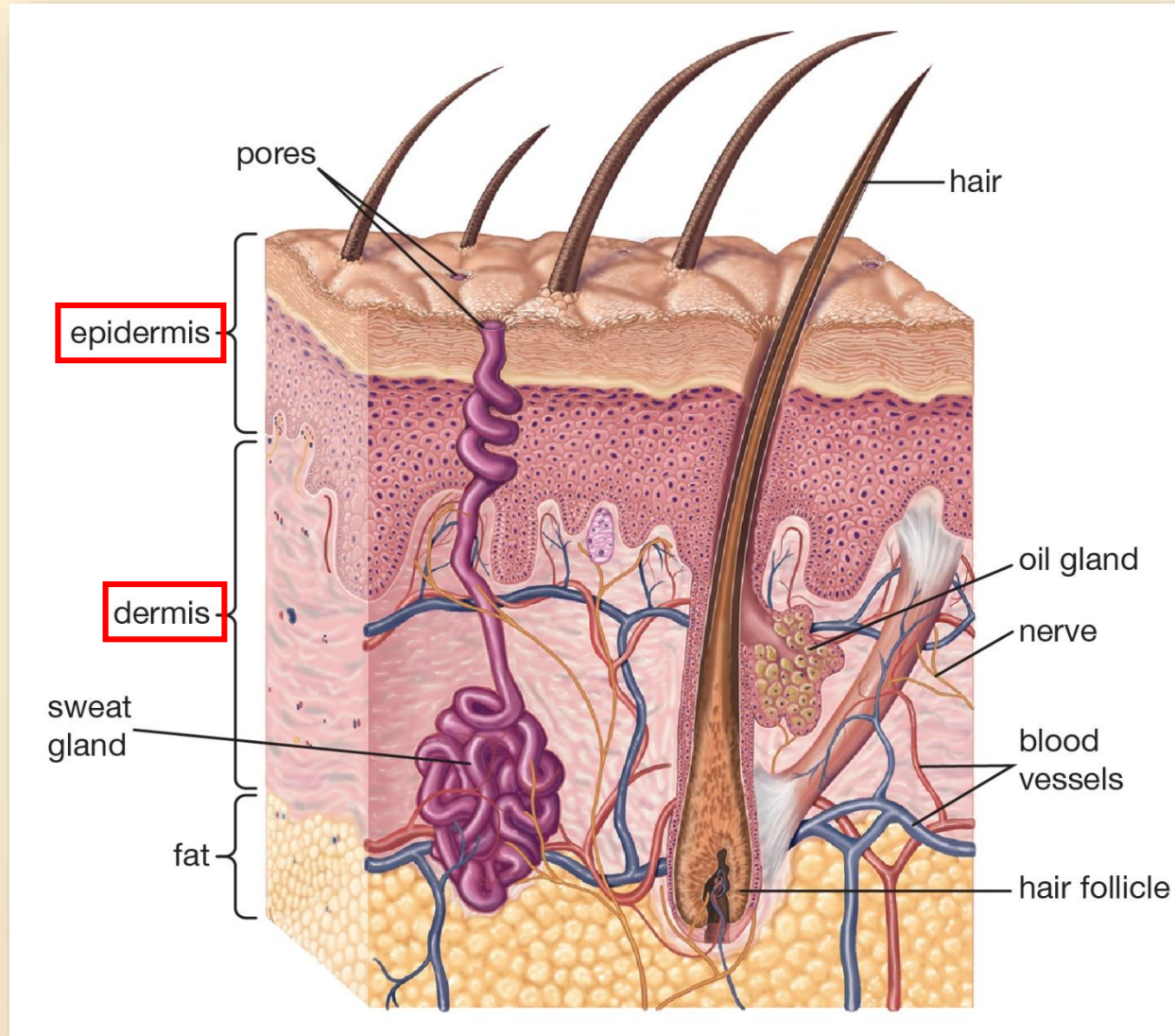


**Fig. 4.** (Left) Left hand covered by unmeshed dermal graft and (right) the right hand covered by unmeshed dermoepidermal graft on (above) day 0 and (below) day 9.



## Epidermis

- Keratinocytes
- Melanocytes
- Langerhans cells
- Merkel cells
- Sensitive nerves



## Dermis

- Fibroblasts
  - Collagens
  - Glycosaminoglycans
  - Elastins
- Adnexal structures
  - Hair follicles
  - Sebaceous glands
  - Sweat glands



# Wound healing

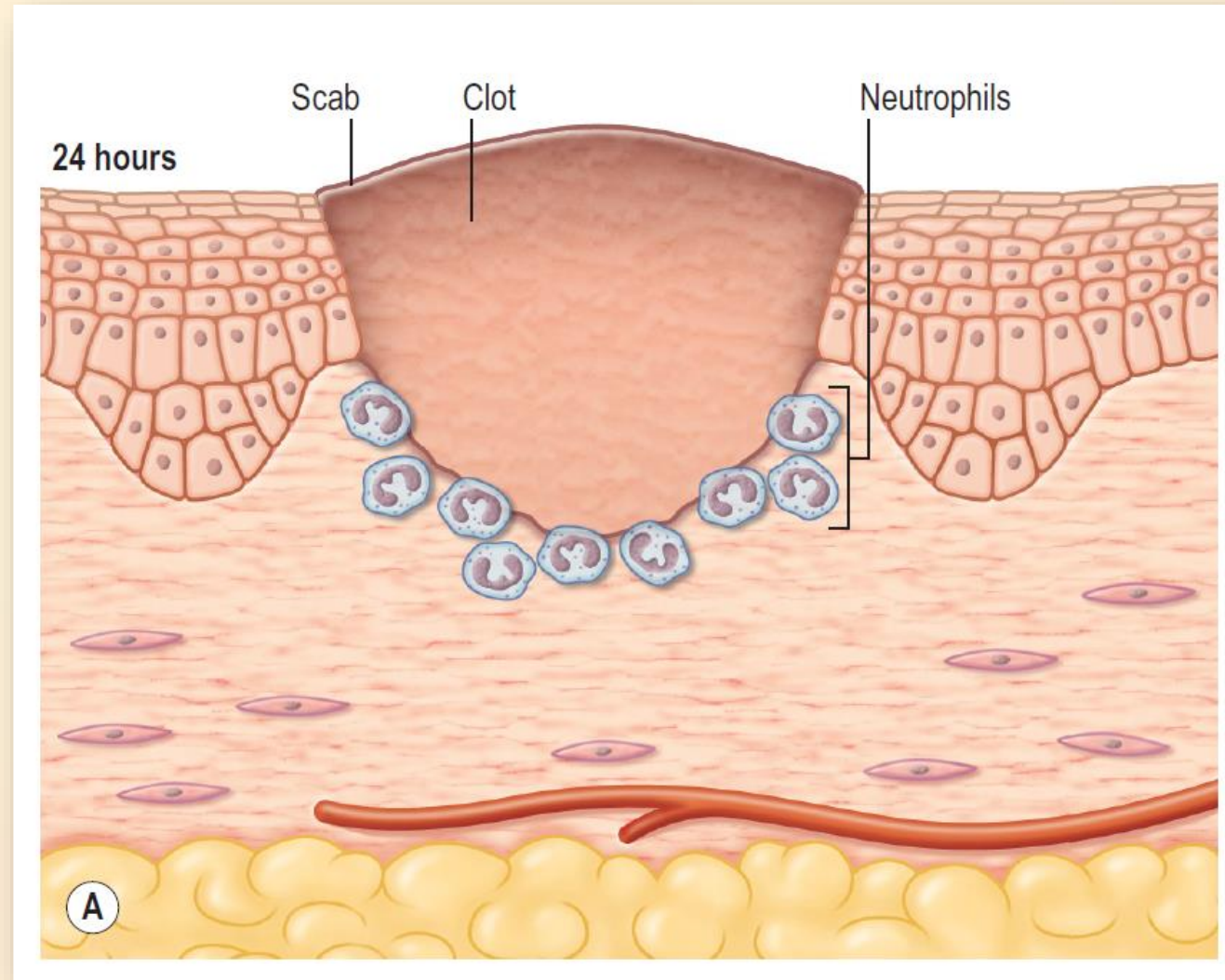


# Wound healing process

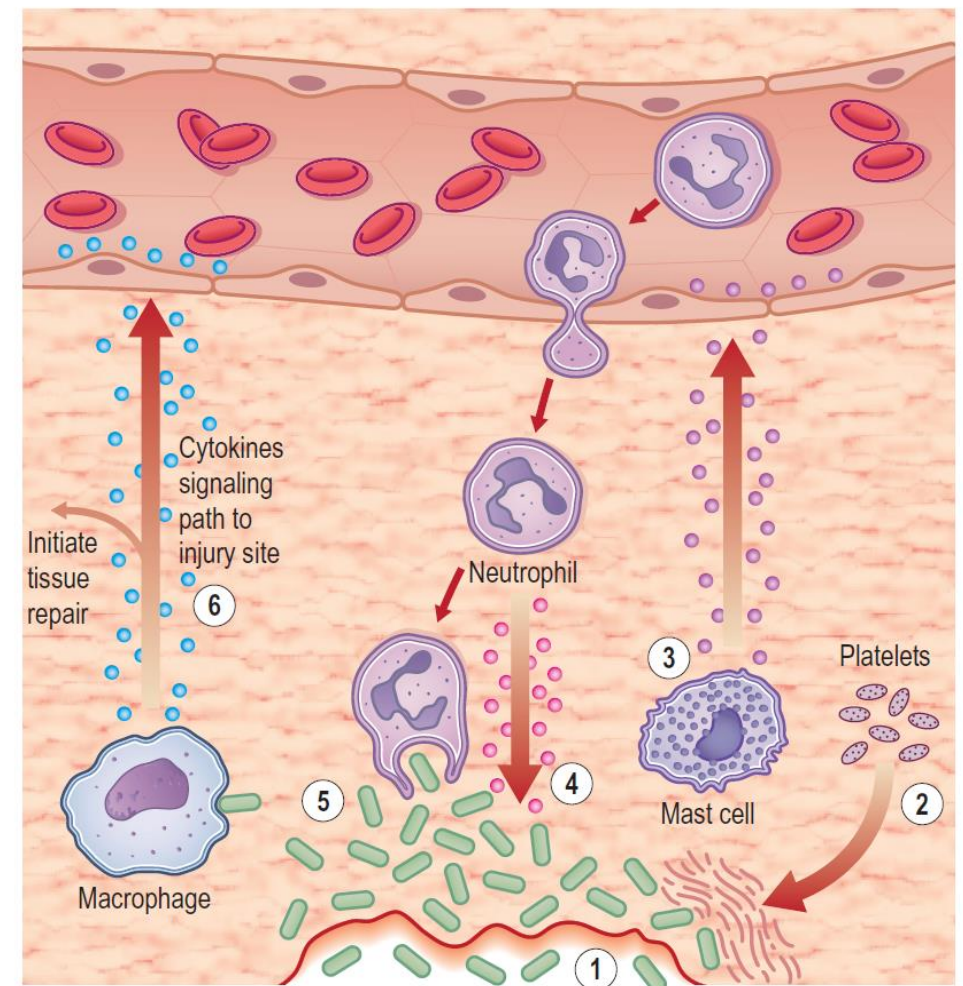
- Hemostasis and inflammation
- Proliferation
- Remodeling



# Hemostasis and inflammation



# Hemostasis and inflammation

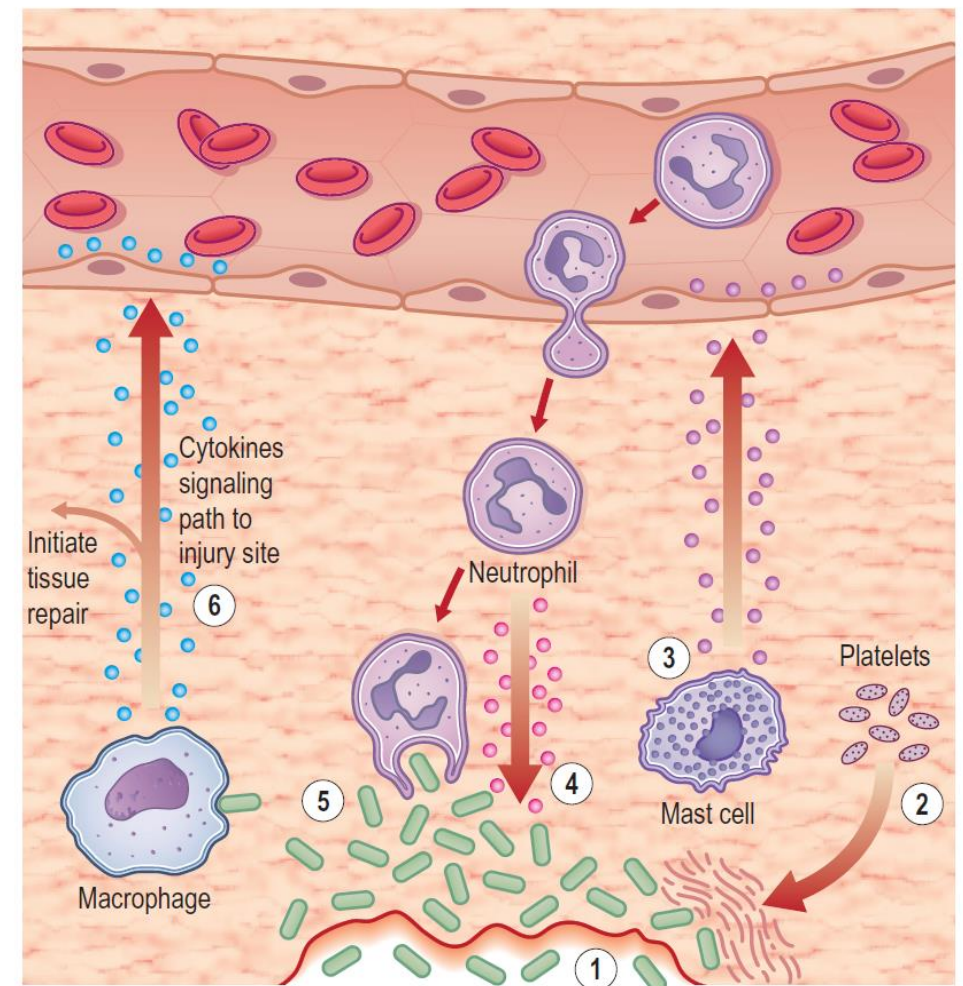


- 1 Bacteria and other pathogens enter wound.
- 2 Platelets from blood release blood-clotting proteins at wound site.
- 3 Mast cells secrete factors that mediate vasodilation and vascular constriction. Delivery of blood, plasma and cells to injured area.
- 4 Neutrophils and macrophages remove pathogens by phagocytosis.
- 5 Macrophages secrete hormones called cytokines that attract immune system cells to the site and activate cells involved in tissue repair.
- 6 Inflammatory response continues until the foreign material is eliminated and the wound is repaired.

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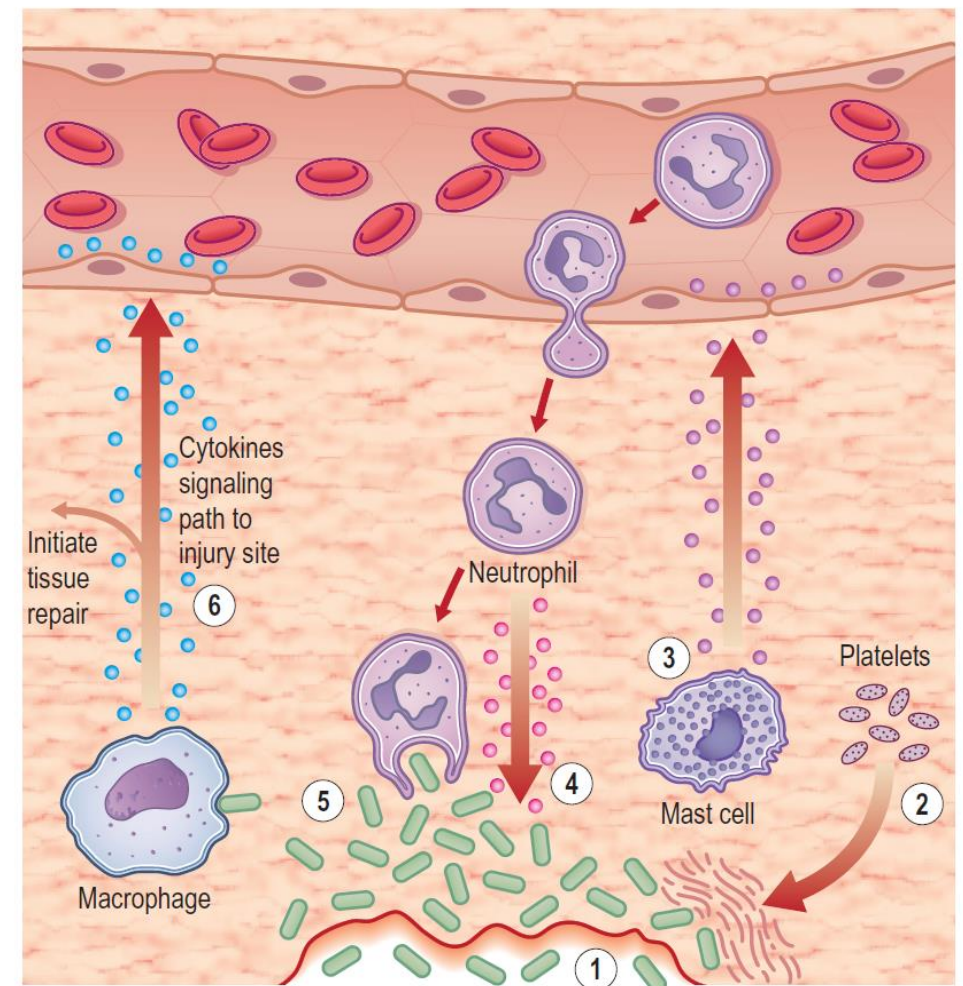
# Hemostasis and inflammation



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Slide 37/65

# Hemostasis and inflammation

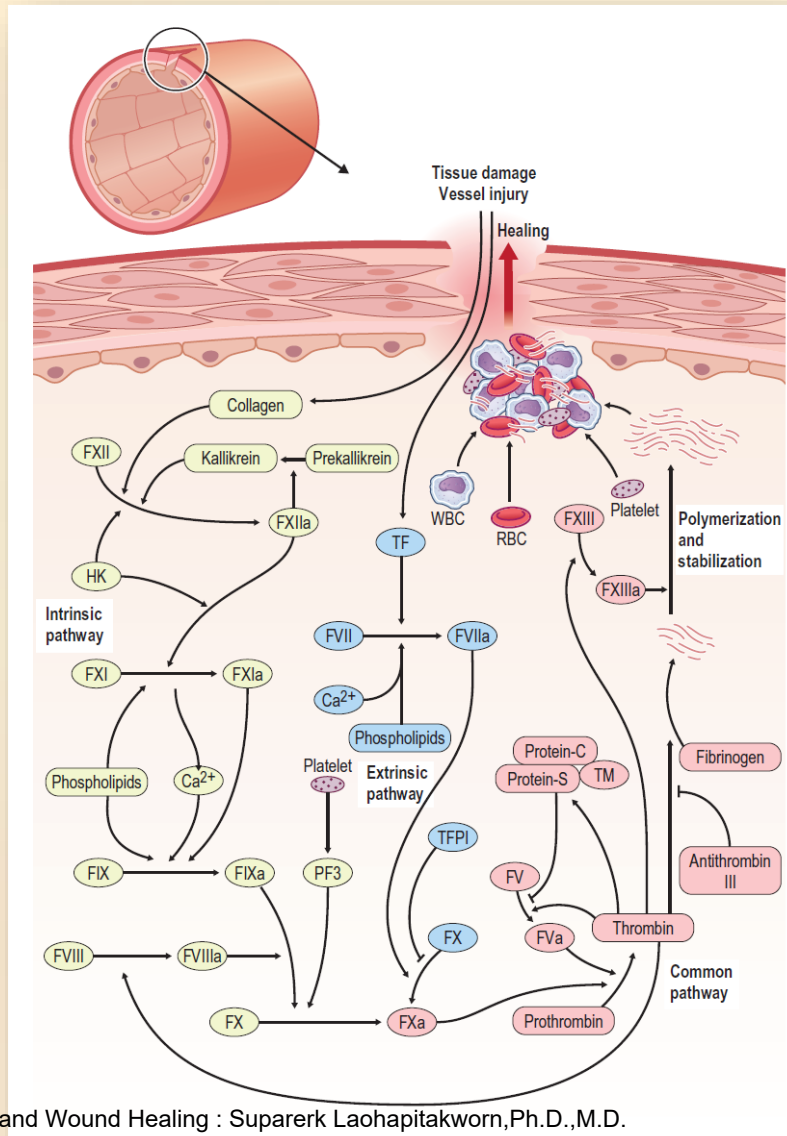


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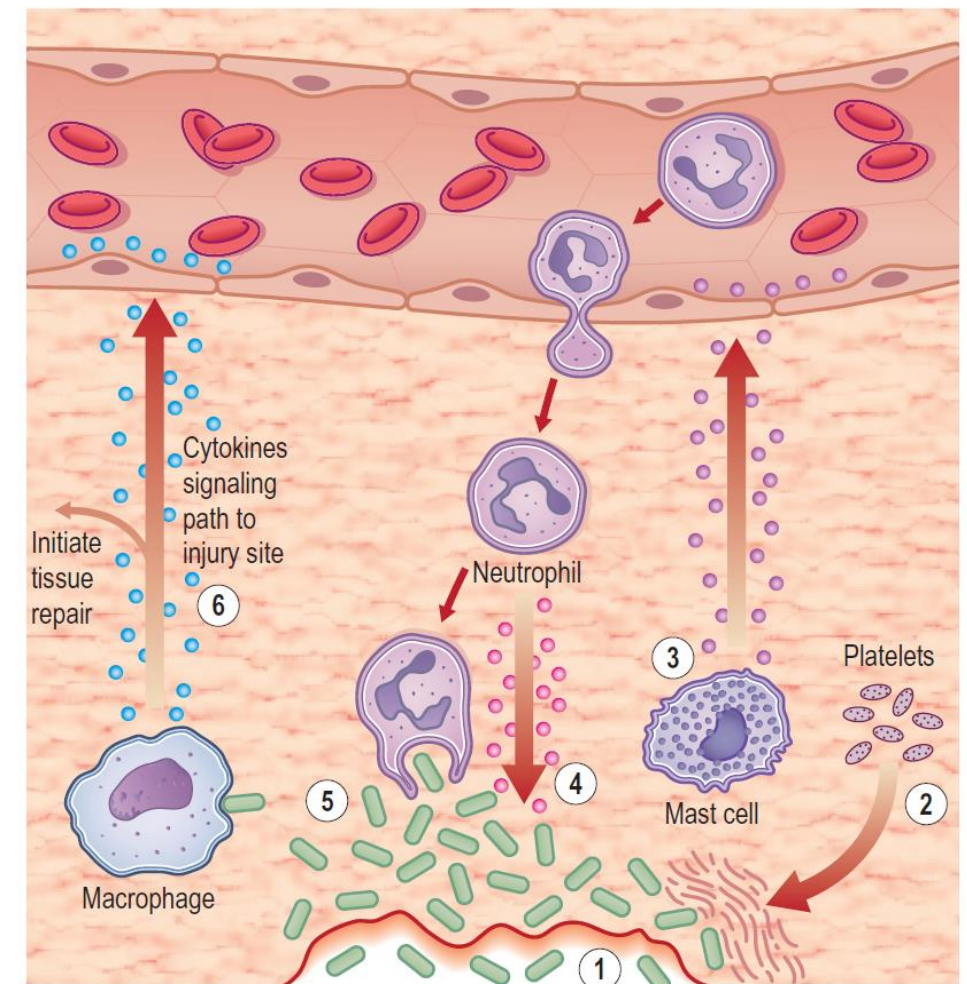
Slide 38/65



# Hemostasis and inflammation



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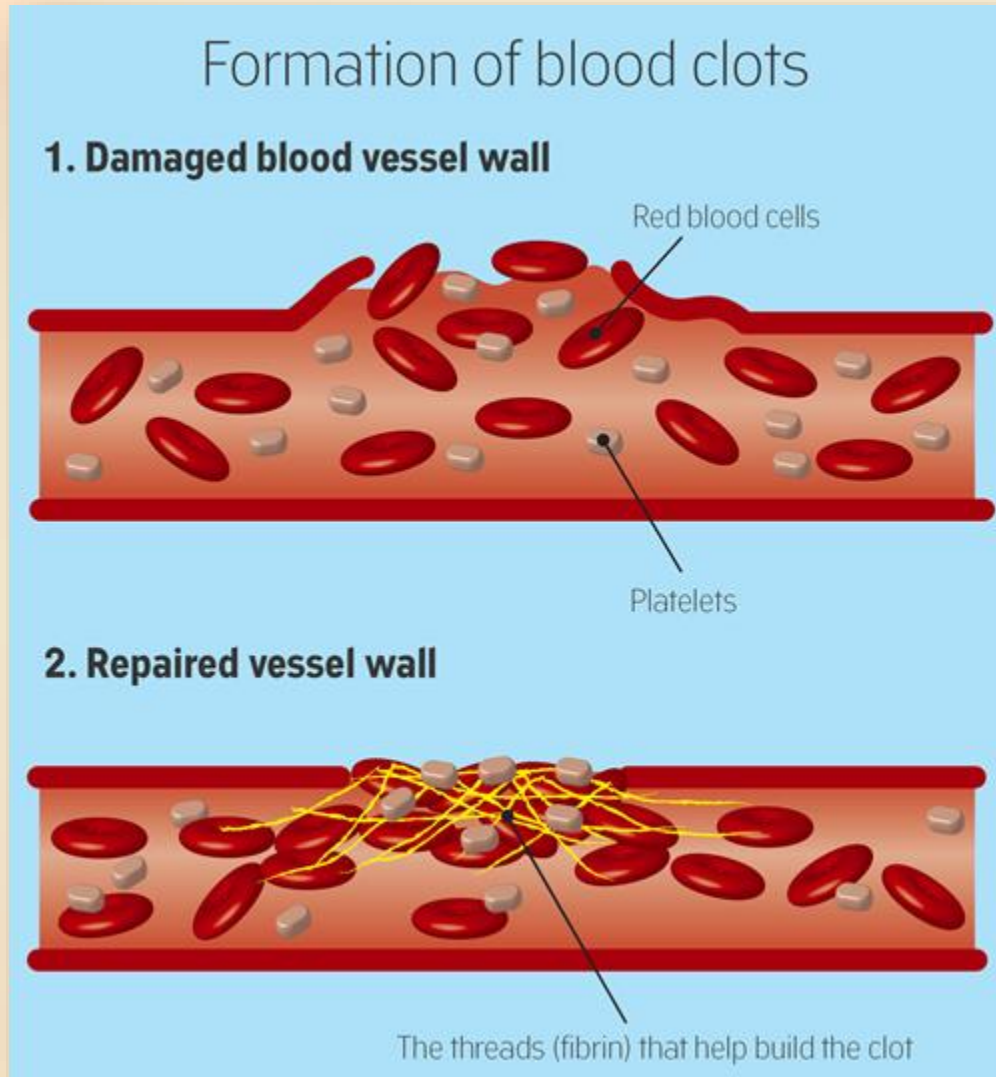


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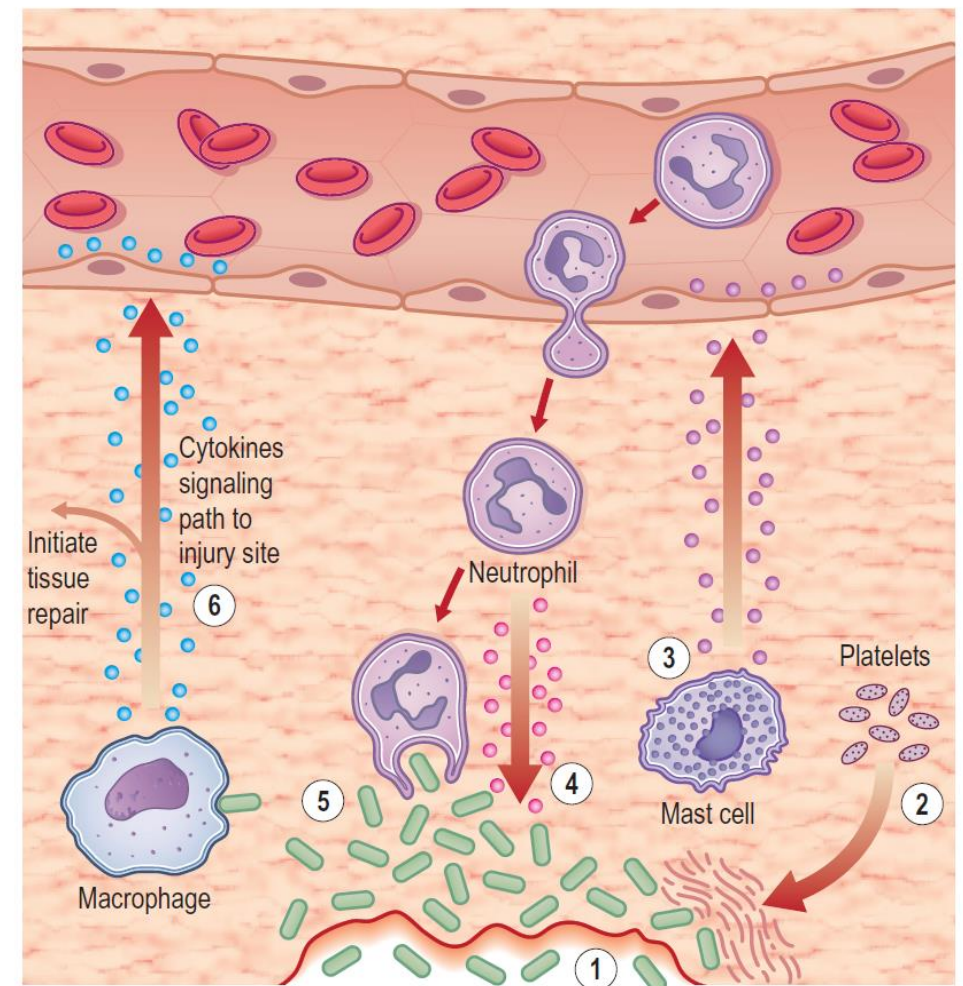
Slide 39/65



# Hemostasis and inflammation



Anatomy of Skin and Wound Healing : Suparerk Laohapitakworn, Ph.D., M.D.



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Slide 40/65

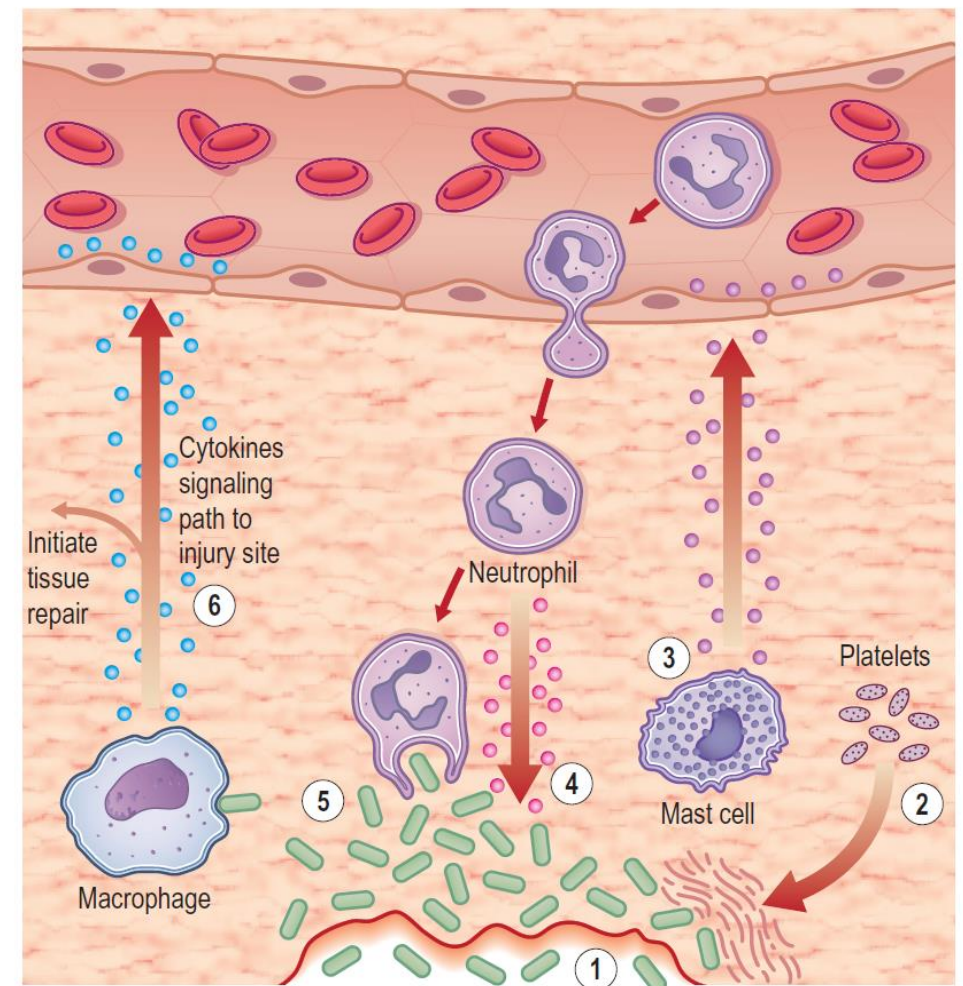


# Hemostasis and inflammation

- **Fibrin glue** : fibrinogen and thrombin
- **Tranexamic acid** : antifibrinolytic
- **Aspirin/Clopidogrel** : antiplatelet
- **Warfarin** : inhibit clotting factors
- **Calcium alginate** : sodium alginate and calcium ion



# Hemostasis and inflammation

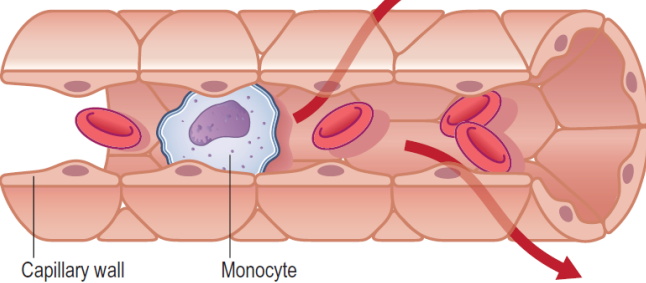


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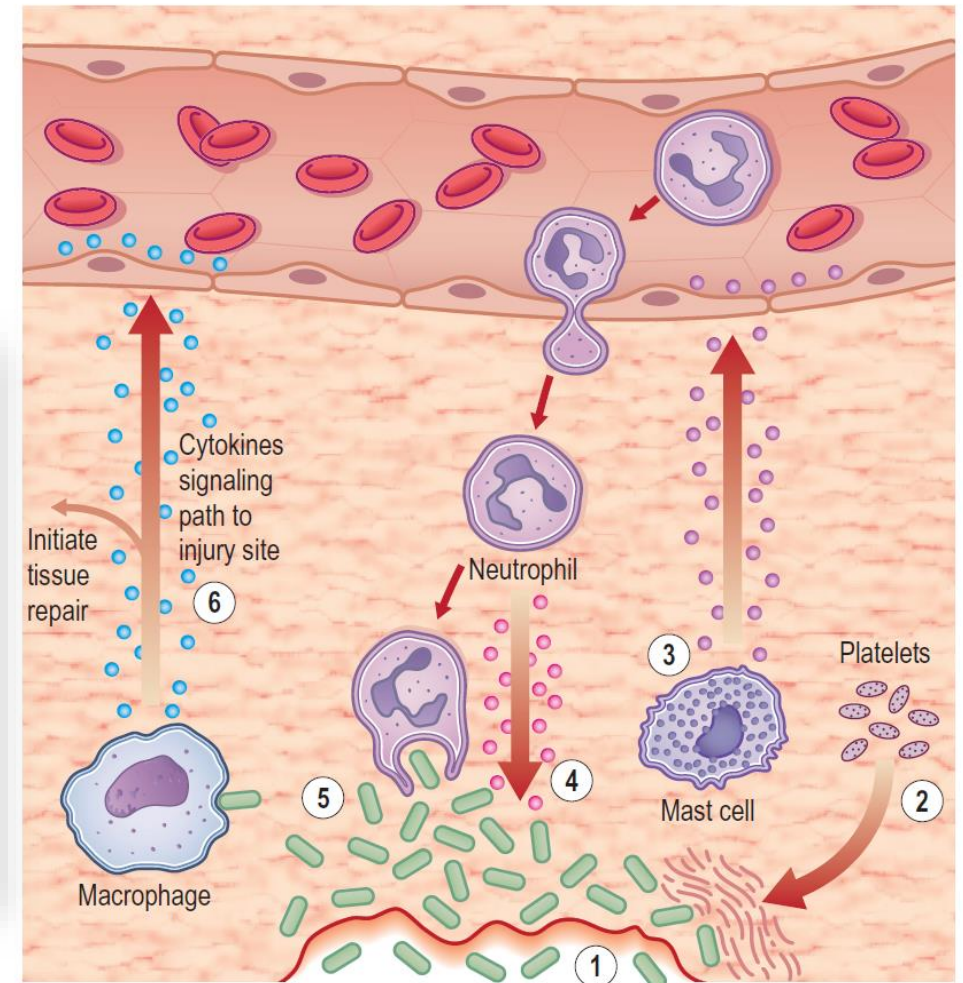
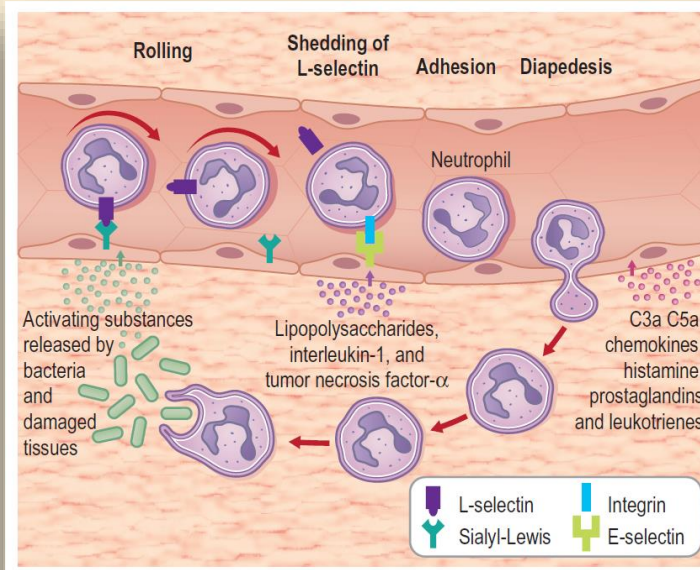
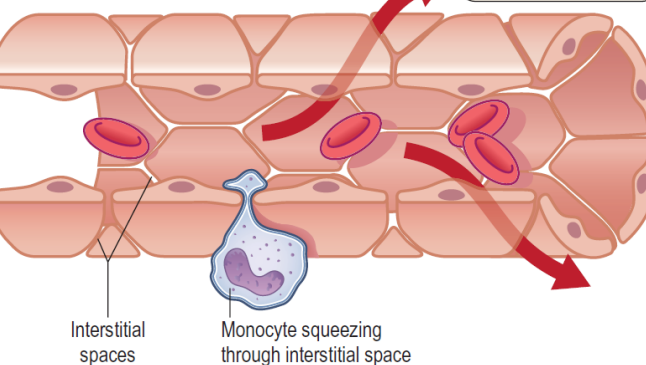


# Hemostasis and inflammation

Normal permeability of capillary



Increased permeability of capillary during inflammation

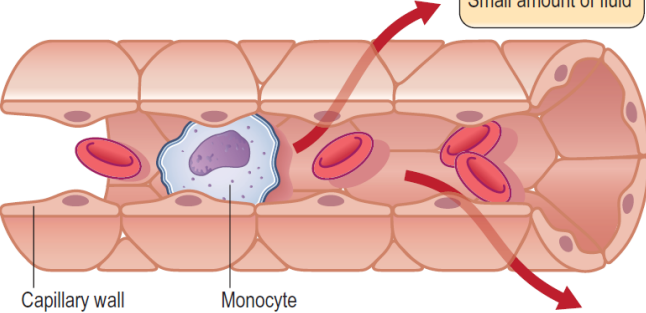


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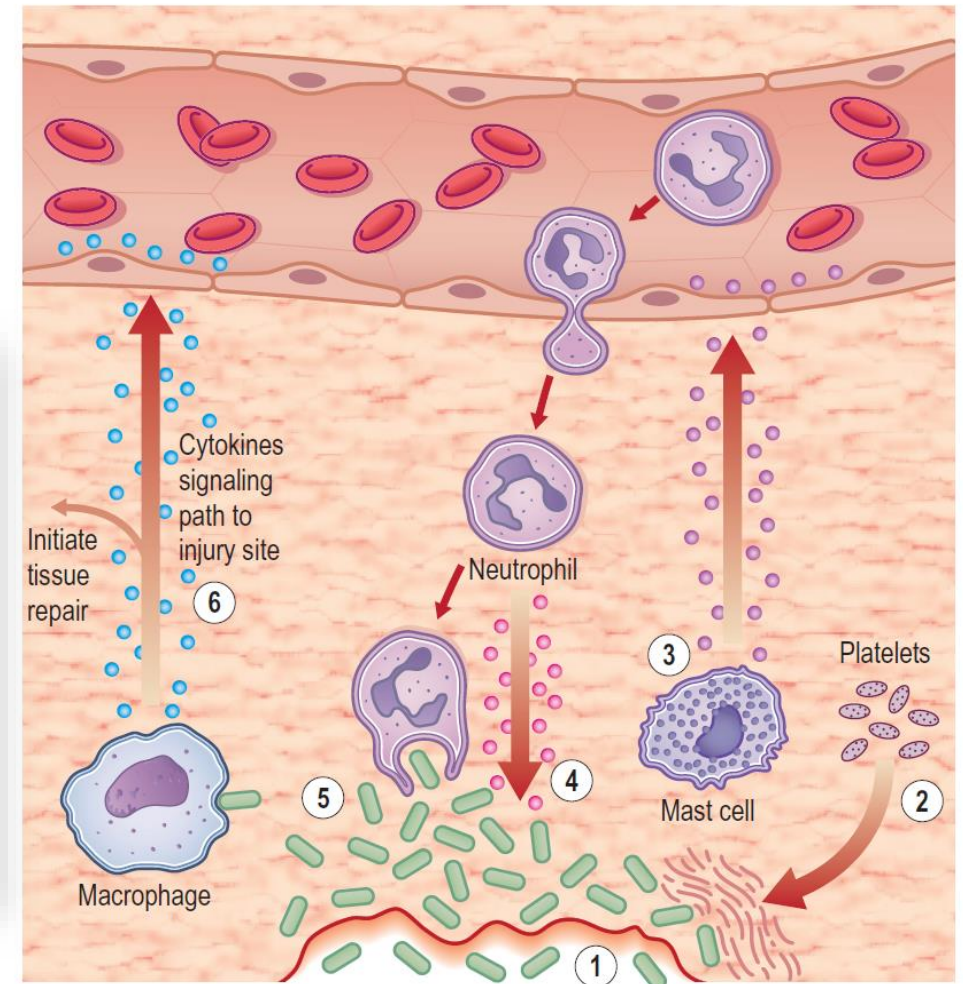
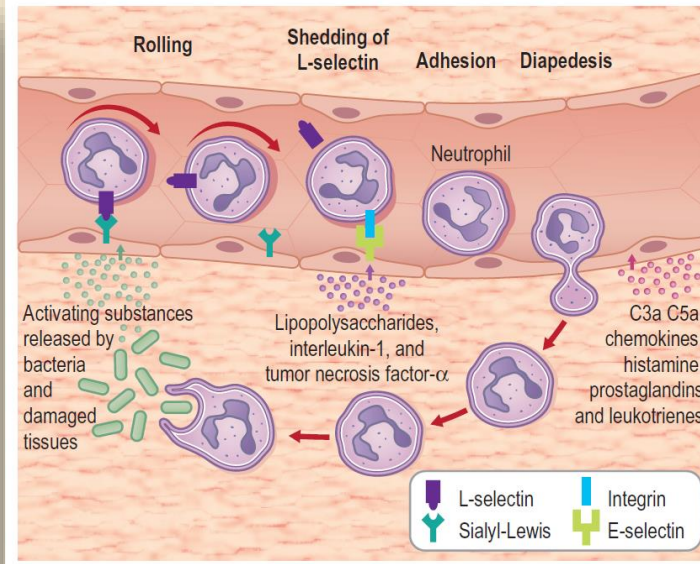
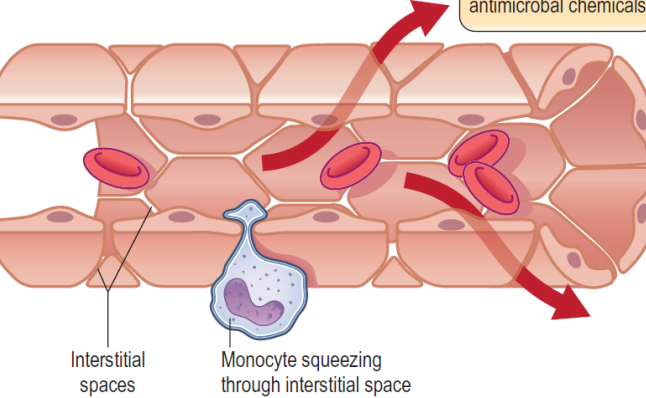


# Hemostasis and inflammation

Normal permeability of capillary



Increased permeability of capillary during inflammation



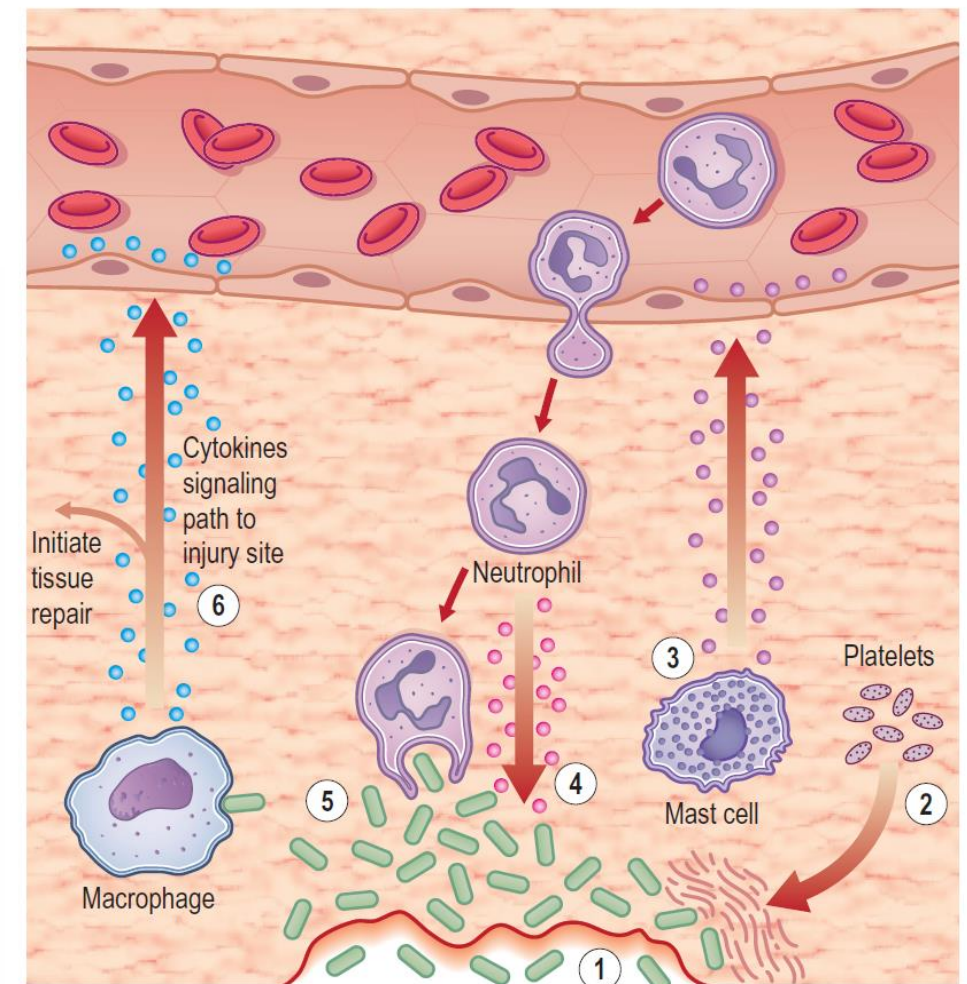
- ① Bacteria and other pathogens enter wound.
- ② Platelets from blood release blood-clotting proteins at wound site.
- ③ Mast cells secrete factors that mediate vasodilation and vascular constriction. Delivery of blood, plasma and cells to injured area.
- ④ Neutrophils and macrophages remove pathogens by phagocytosis.
- ⑤ Macrophages secrete hormones called cytokines that attract immune system cells to the site and activate cells involved in tissue repair.
- ⑥ Inflammatory response continues until the foreign material is eliminated and the wound is repaired.



# Hemostasis and inflammation

Table 15.1 A partial list of growth factors present at the wound site\*

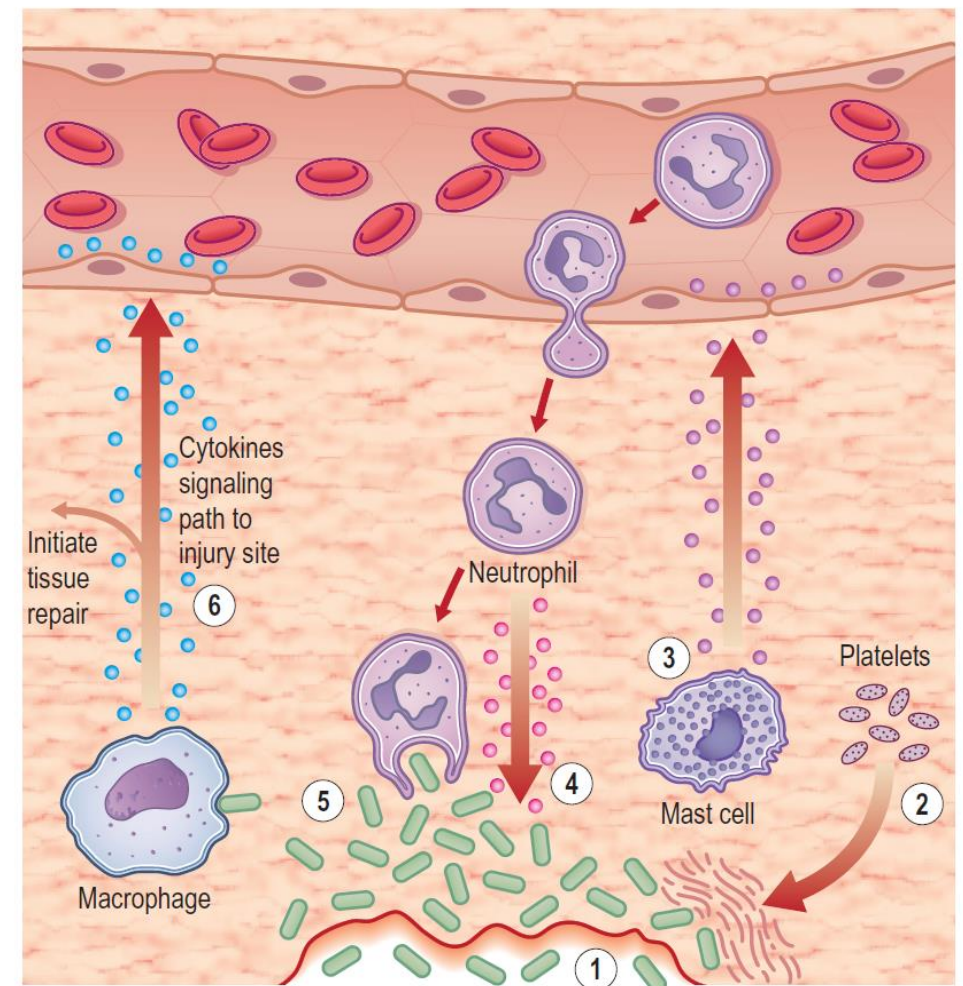
Growth factor	Cellular source	Target cells	Biologic activity
Activin	Fibroblasts, keratinocytes	Stromal cells	Granulation tissue formation, scarring
TGF- $\beta_1$ and TGF- $\beta_2$	Macrophages, platelets, fibroblasts, keratinocytes	Inflammatory cells, keratinocytes, fibroblasts	Chemotaxis, proliferation, matrix production (fibrosis)
TGF- $\beta_3$	Macrophages	Fibroblasts	Antiscarring?
TGF- $\alpha$	Macrophages, platelets, keratinocytes	Keratinocytes, fibroblasts, endothelial cells	Proliferation
TNF- $\alpha$	Neutrophils, mast cells	Macrophages, keratinocytes, fibroblasts	Activation of growth factor expression
PDGF	Macrophages, platelets, keratinocytes, fibroblasts, endothelial cells, vascular smooth-muscle cells	Neutrophils, macrophages, fibroblasts, endothelial cells, vascular smooth-muscle cells	Chemotaxis, proliferation, matrix production
FGF-1, FGF-2, FGF-4	Macrophages, fibroblasts, endothelial cells	Keratinocytes, fibroblasts, endothelial cells, chondrocytes	Angiogenesis, proliferation, chemotaxis
FGF-7 (KGF-1), FGF-10 (KGF-2)	Fibroblasts	Keratinocytes	Proliferation, chemotaxis
EGF	Platelets, macrophages, keratinocytes	Keratinocytes, fibroblasts, endothelial cells	Proliferation, chemotaxis
HB-EGF	Macrophages, keratinocytes	Keratinocytes, fibroblasts	Proliferation, epithelial migration, synergistic with IGF
IGF-1/Sm-C	Fibroblasts, macrophages, platelets	Fibroblasts, endothelial cells	Proliferation, collagen synthesis
IL-1 $\alpha$ and IL-1 $\beta$	Macrophages, neutrophils	Macrophages, fibroblasts, keratinocytes	Proliferation, collagenase synthesis, chemotaxis
CTGF/CCN2	Fibroblasts, endothelial cells	Fibroblasts	Downstream of TGF- $\beta_1$
VEGF	Macrophages, keratinocytes, fibroblasts	Endothelial cells	Angiogenesis



- 1 Bacteria and other pathogens enter wound.
- 2 Platelets from blood release blood-clotting proteins at wound site.
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- 6 Inflammatory response continues until the foreign material is eliminated and the wound is repaired.

Slide 45/65

# Hemostasis and inflammation



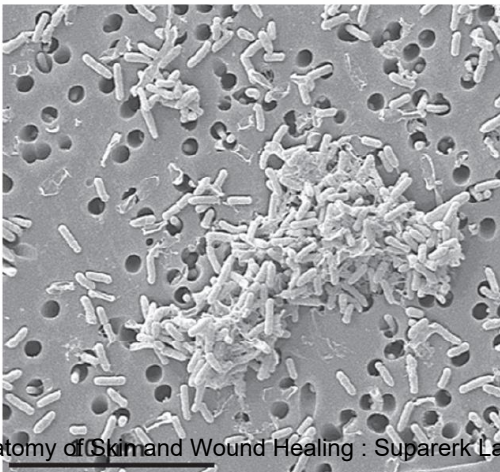
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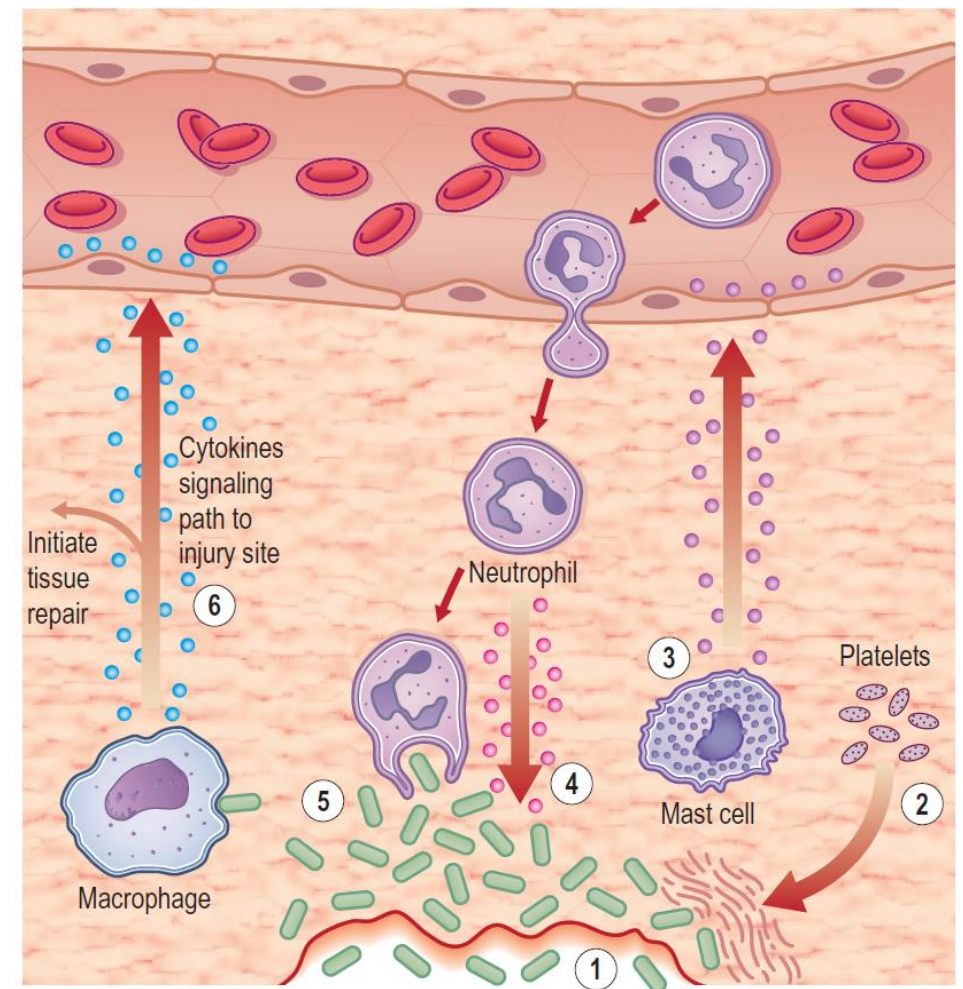
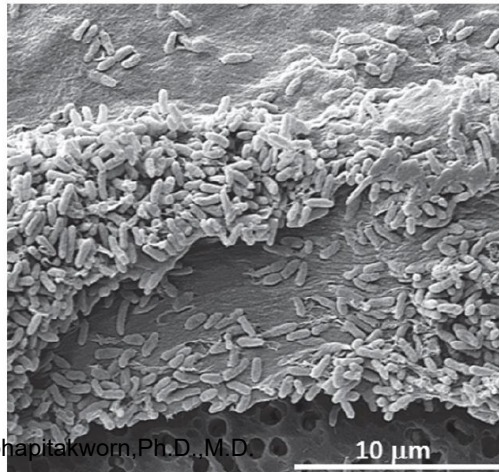
# Hemostasis and inflammation



PLANKTONIC

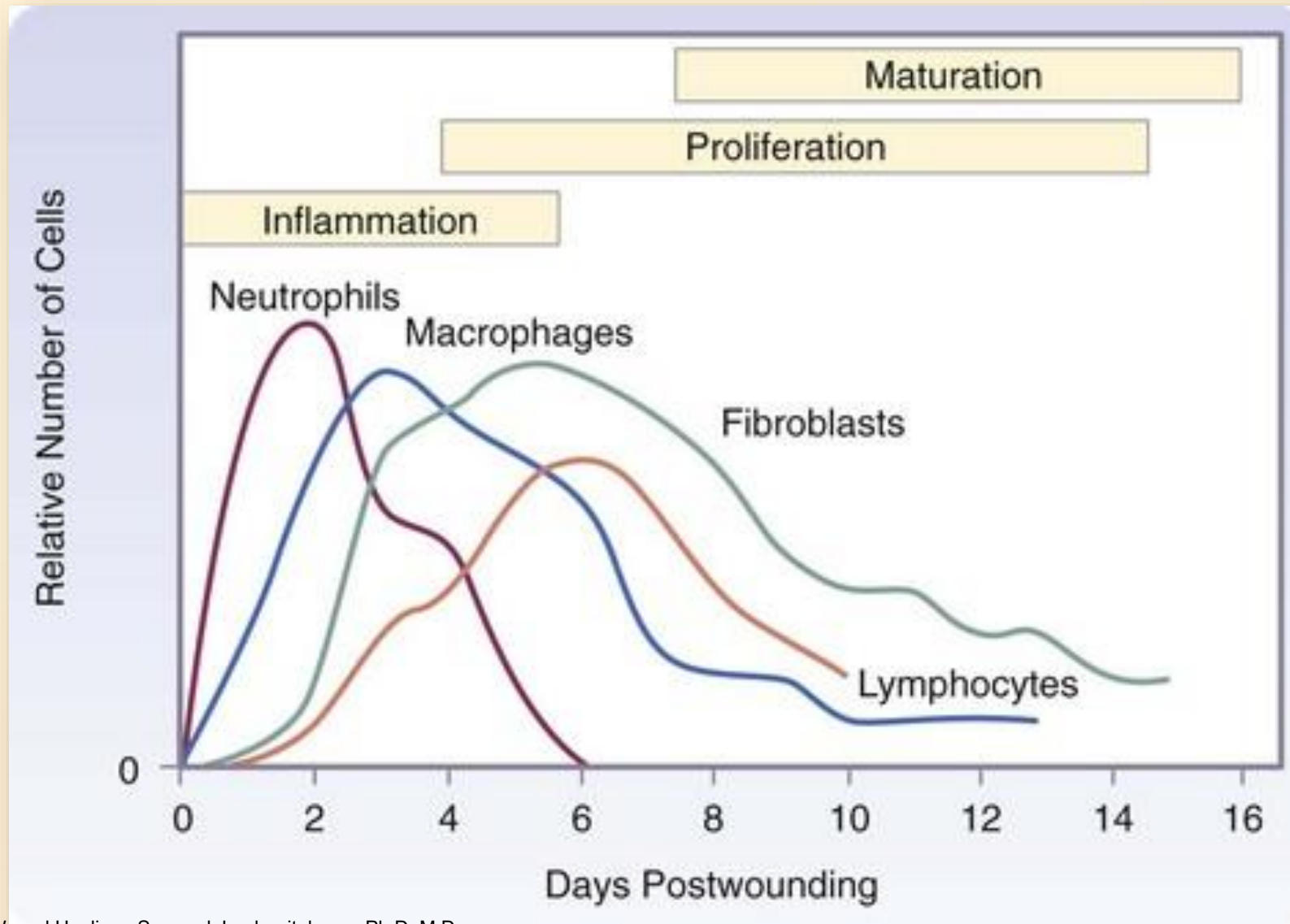


BIOFILM



- ① Bacteria and other pathogens enter wound.
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# Hemostasis and inflammation



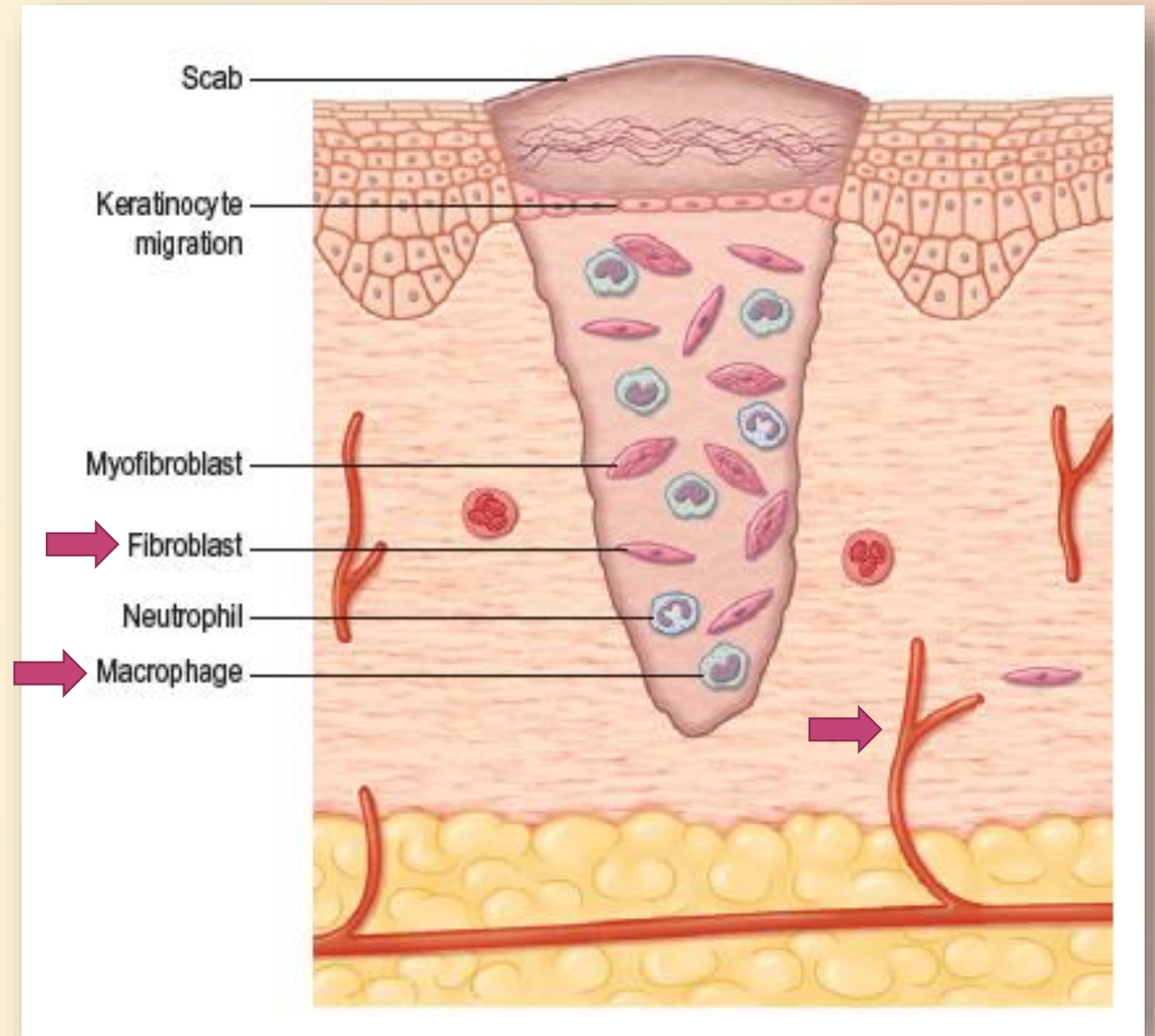


# Proliferation

- Granulation and vascularization
- Wound closure

# Proliferation

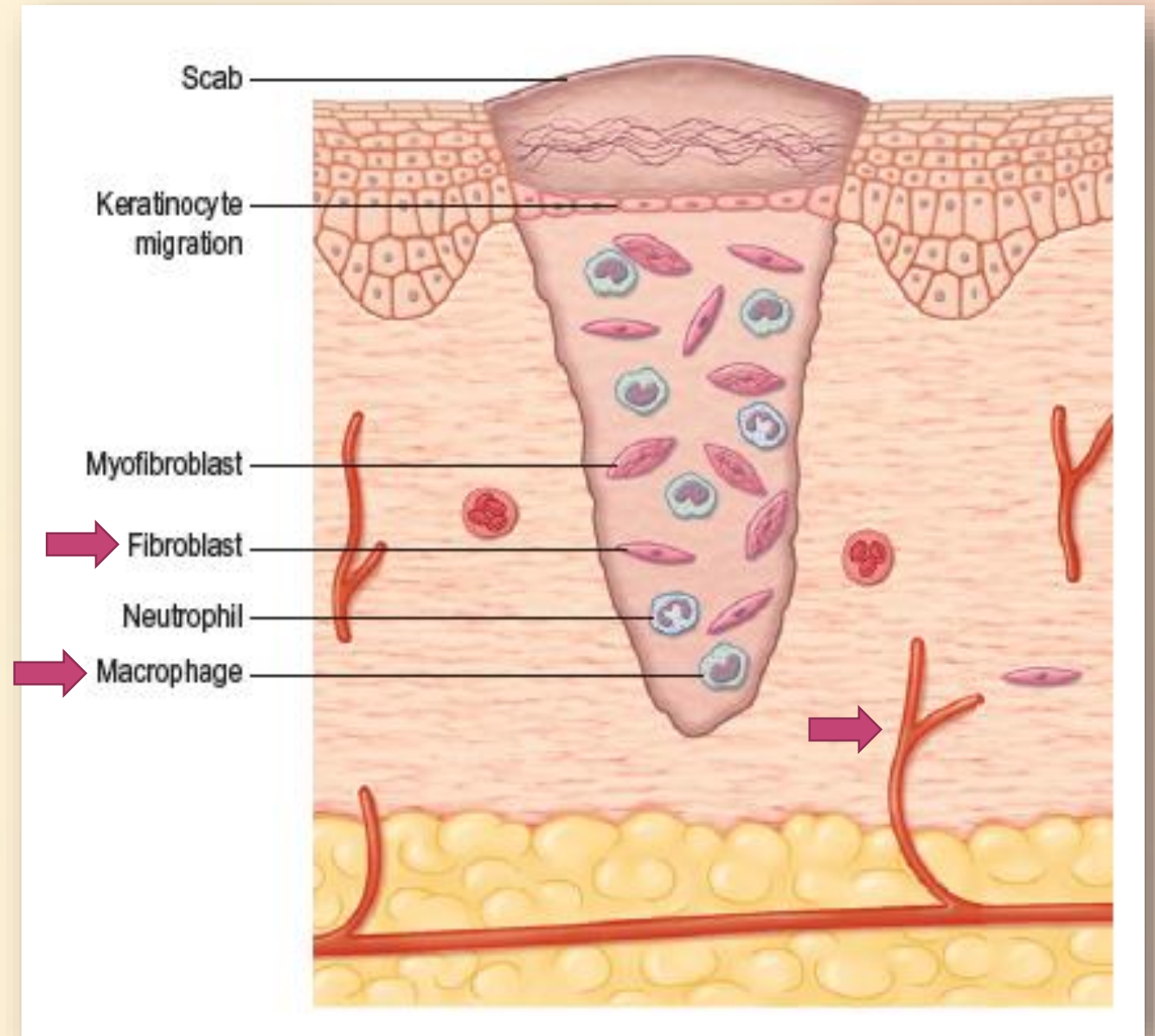
- Granulation and vascularization
  - Fibroblasts
  - Macrophages
  - Blood vessels



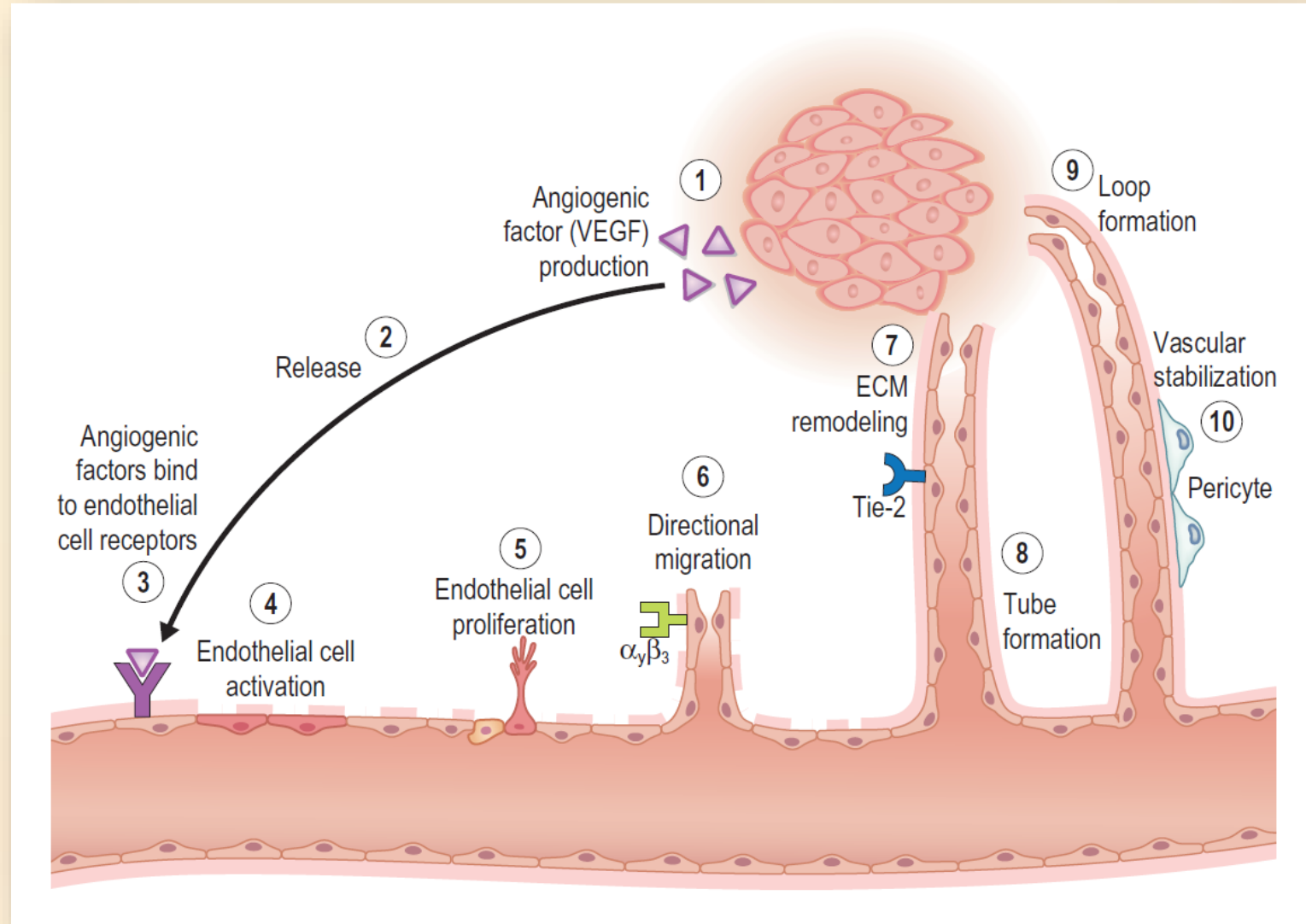


# Proliferation

- Granulation and vascularization
  - Fibroblasts : extracellular matrix
  - Macrophages : growth factors
  - Blood vessels



# Proliferation





# Proliferation



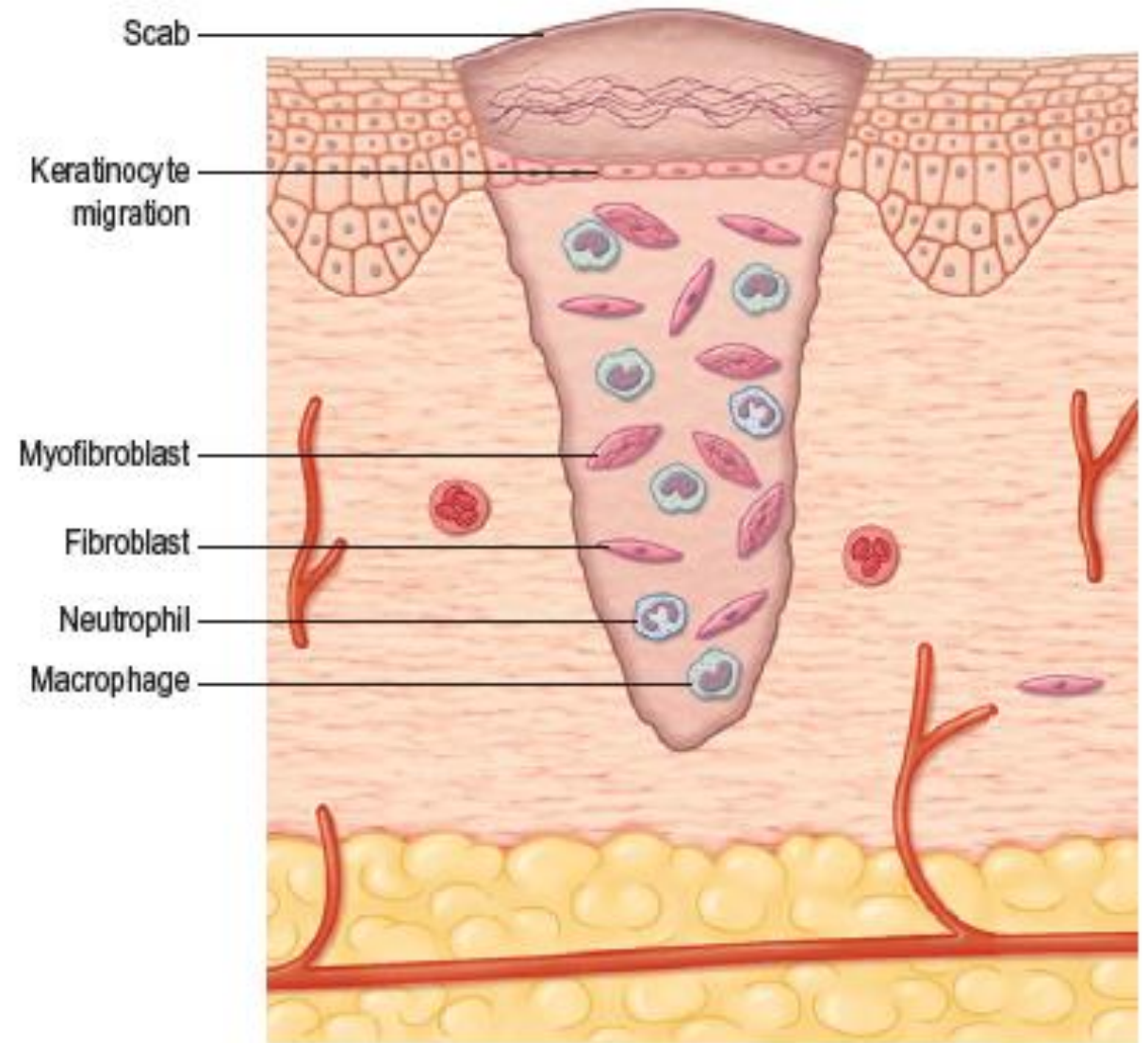
# Proliferation





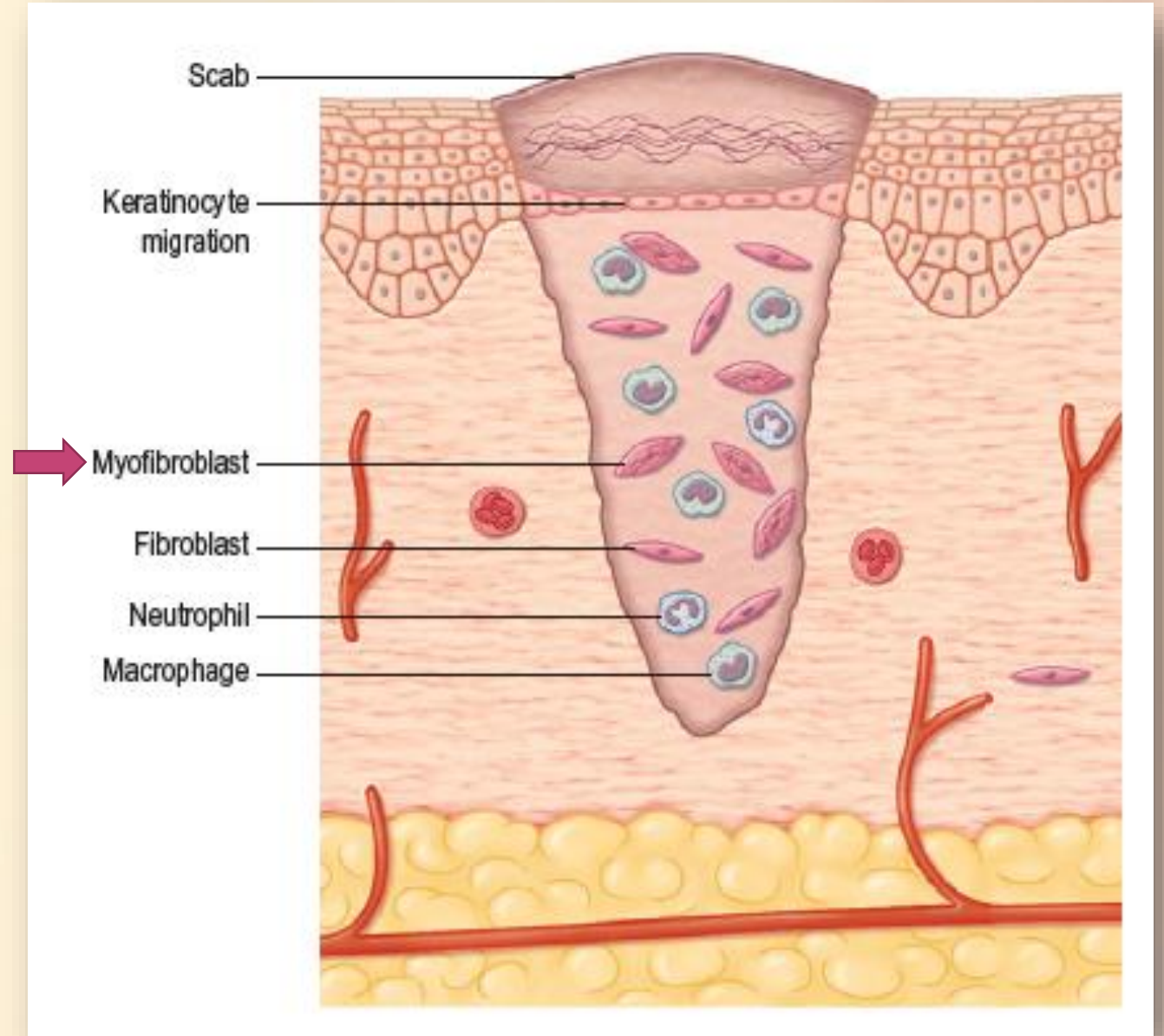
# Proliferation

- Wound closure
  - Wound contraction
  - Epithelialization



# Proliferation

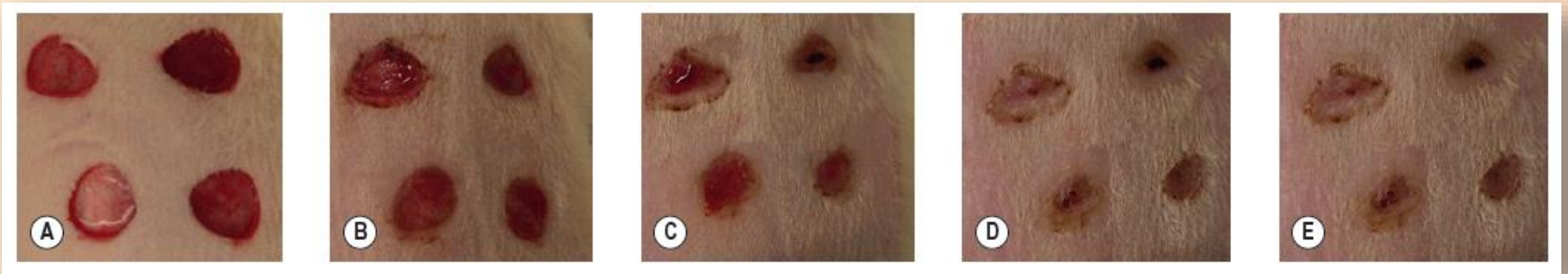
- Wound closure
  - Wound contraction : myofibroblasts
  - Epithelialization





# Proliferation

- Wound closure
  - Wound contraction : myofibroblasts
  - Epithelialization

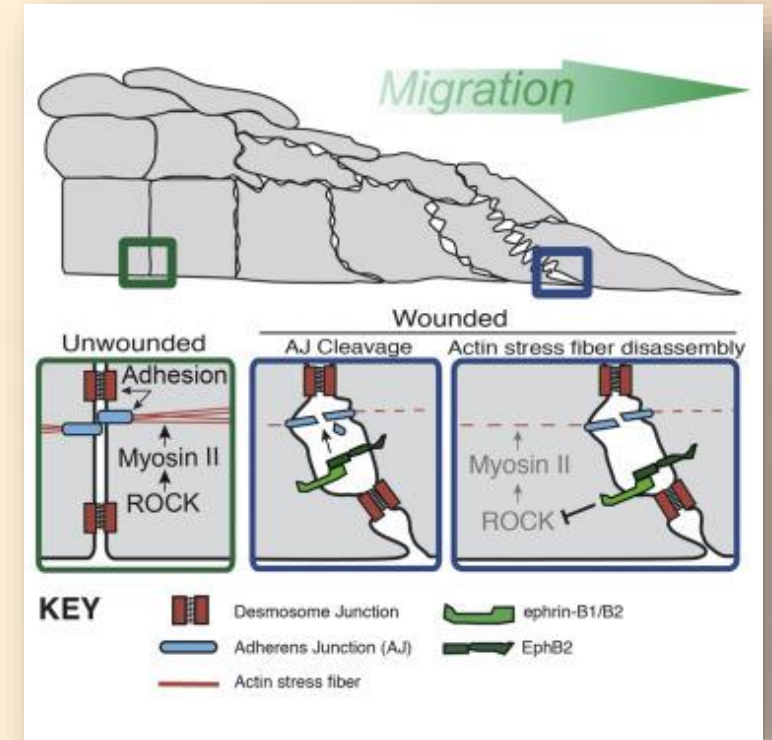
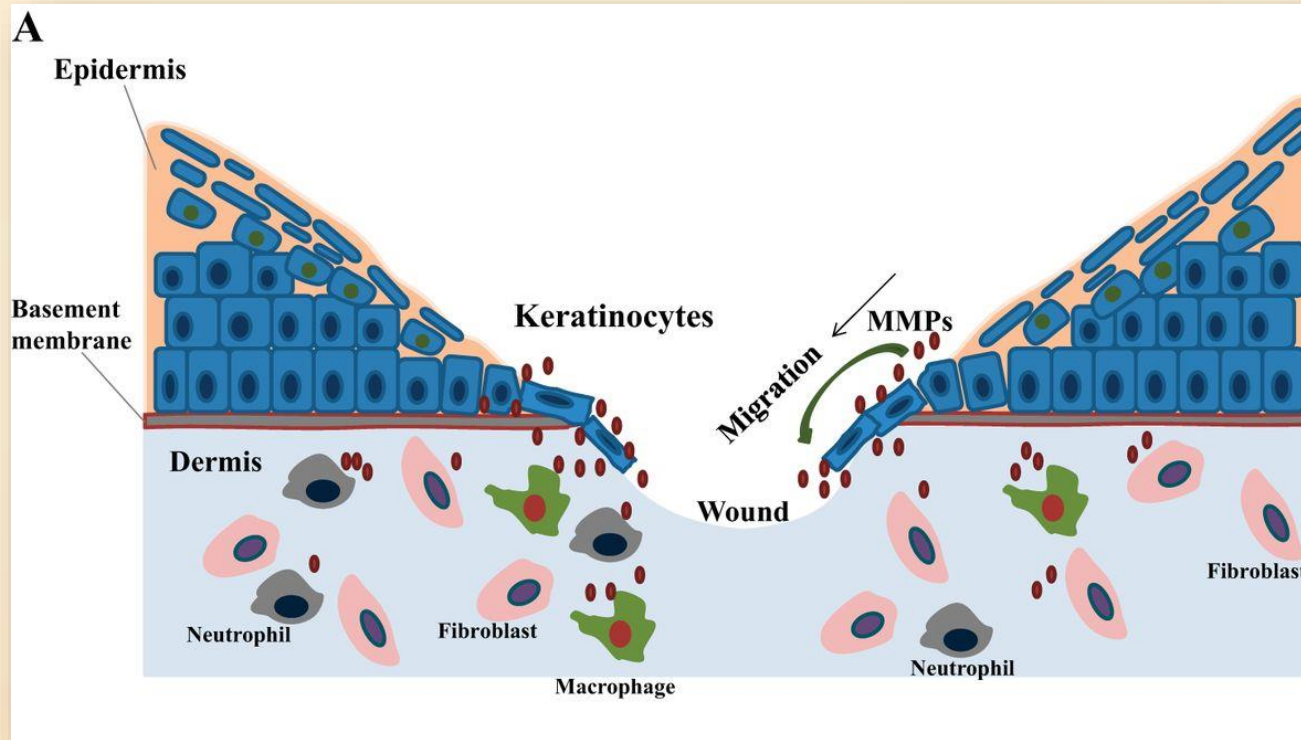
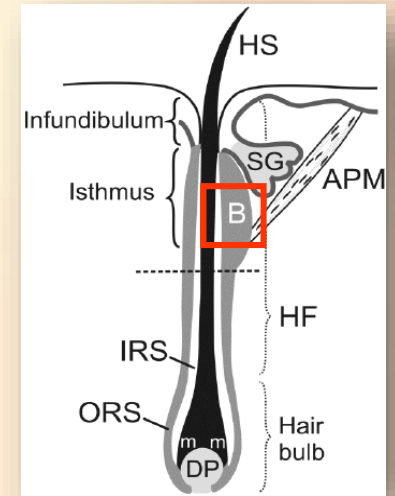






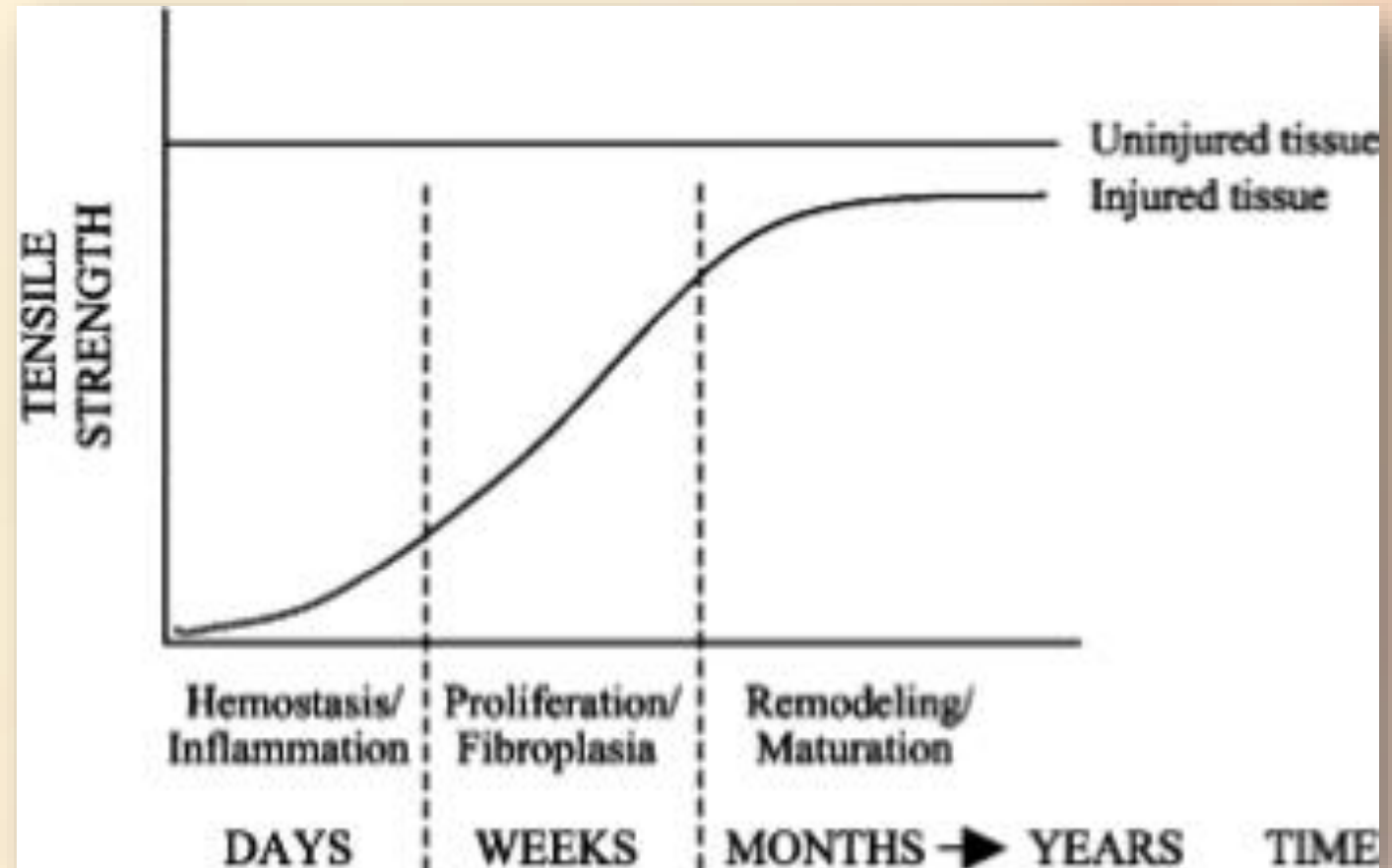
# Proliferation

- Wound closure
  - Wound contraction : myofibroblasts
  - Epithelialization



# Remodeling

- Increased tensile strength
  - Replace type III collagen with type I collagen
  - Collagen cross-linking by lysyl oxidase
- Maximum tensile strength approximately 80%



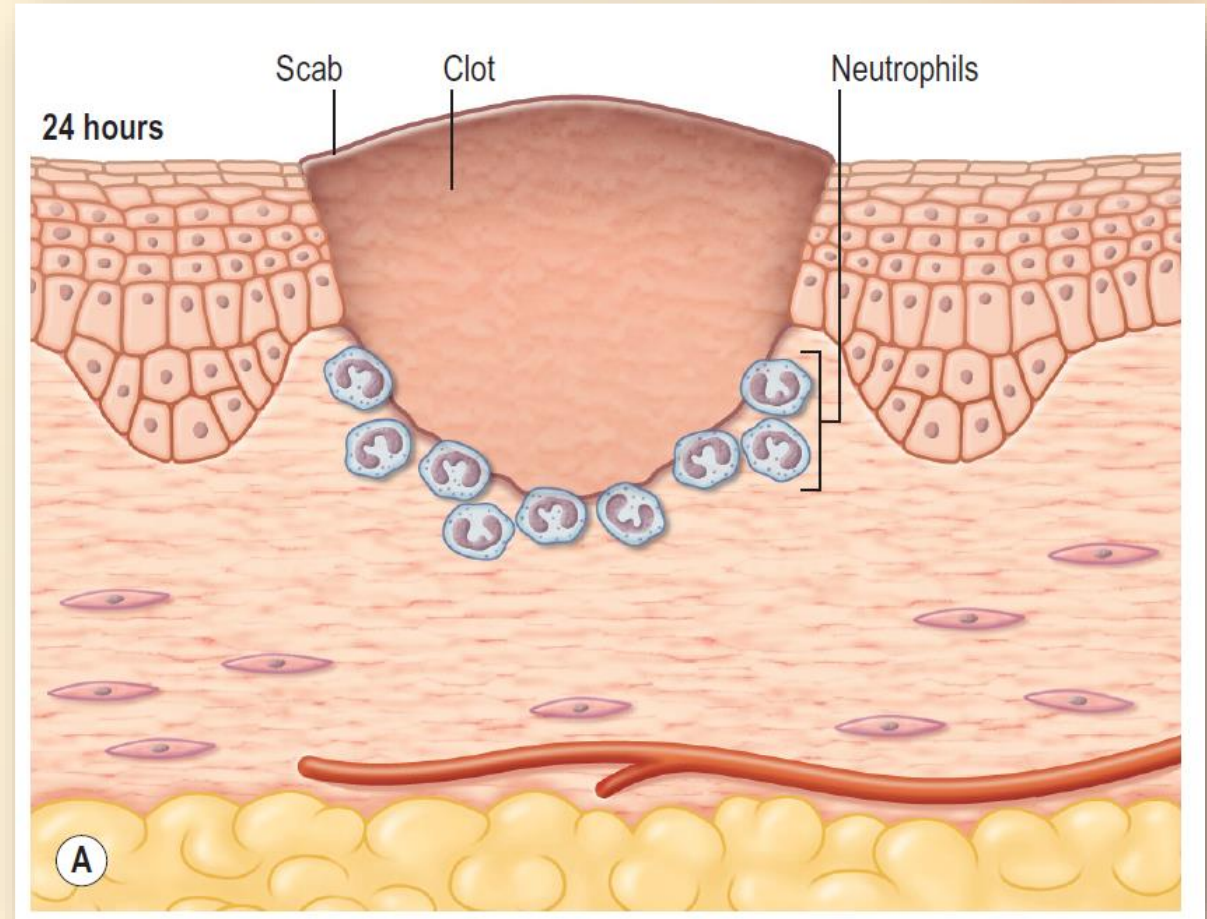


# Wound healing process

- Hemostasis and inflammation
- Proliferation
  - Granulation and vascularization
  - Wound closure
- Remodeling

# Wound healing process

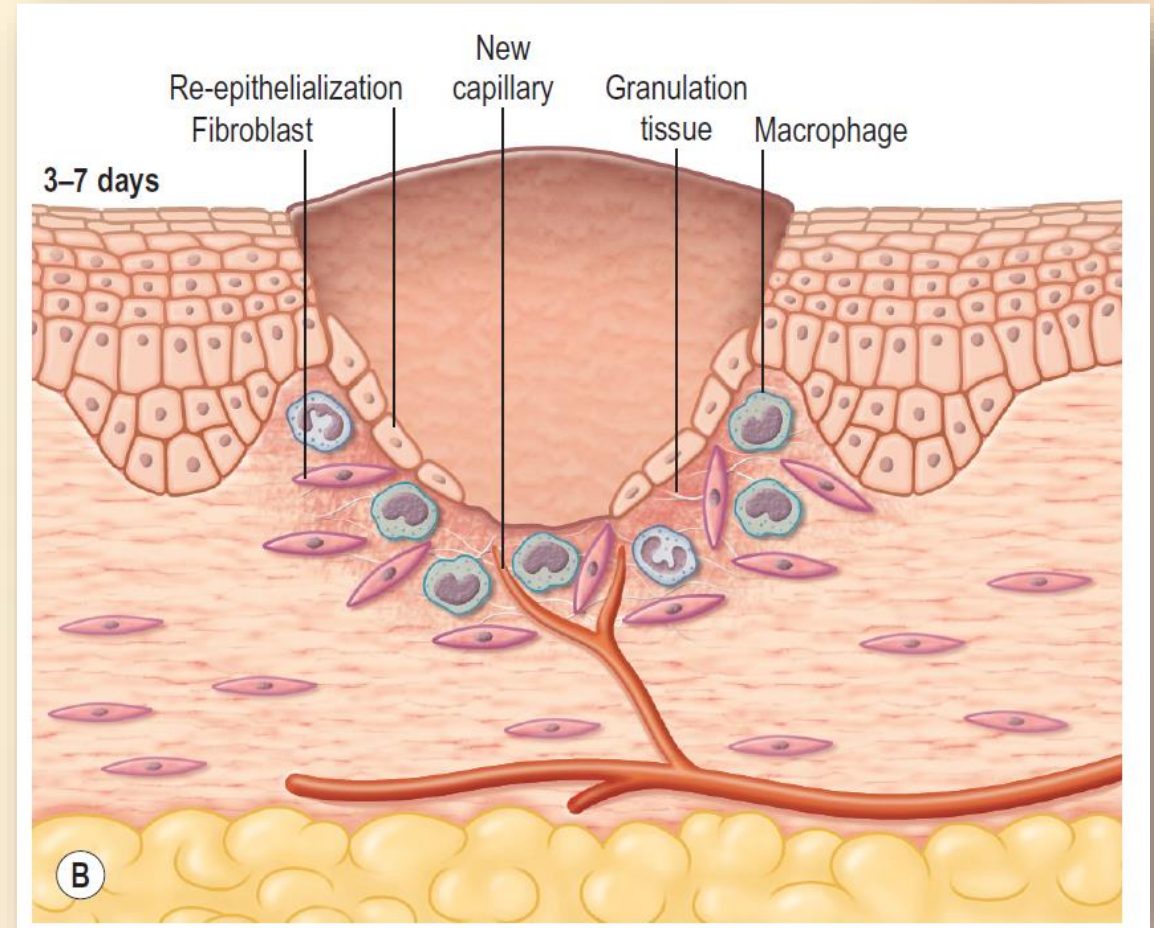
- Hemostasis and inflammation
- Proliferation
  - Granulation and vascularization
  - Wound closure
- Remodeling





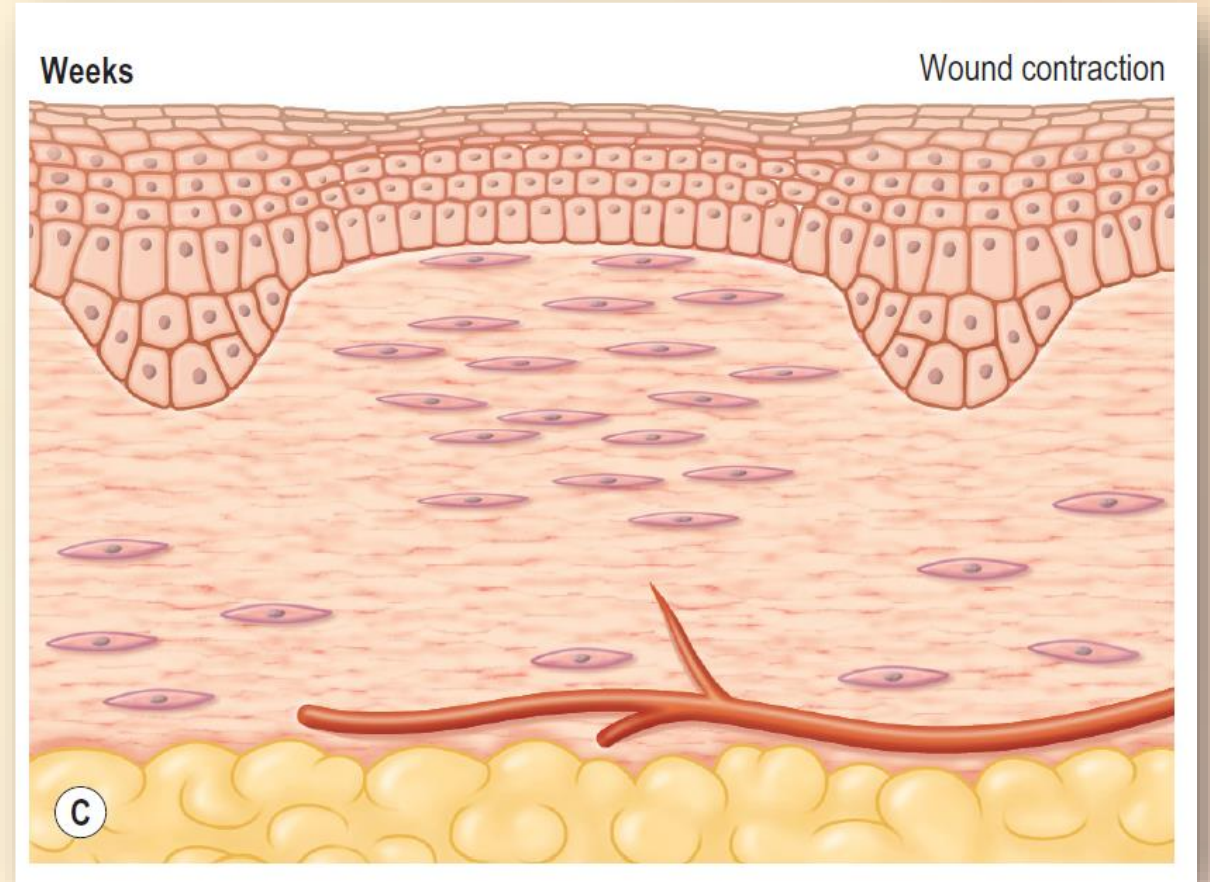
# Wound healing process

- Hemostasis and inflammation
- Proliferation
  - Granulation and vascularization
  - Wound closure
- Remodeling



# Wound healing process

- Hemostasis and inflammation
- Proliferation
  - Granulation and vascularization
  - Wound closure
- Remodeling







**Thank you**