WOUND HEALING

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Objectives:
After this lecture, the student should:

- have an understanding of the importance of wound healing
- recognize that wound healing requires the orderly progression through three phases
- understand the factors that affect wound healing
- be able to design a rational clinical approach to the treatment of wounds

Wound healing "NEED-TO-KNOWS":

- **goals:** - speedy restoration of anatomy
  - avoidance of "over-healing"
- **general wound physiology**
- **a clinical approach to optimizing wound healing**

Definition of a wound is broad:

- **ANY** disruption of normal anatomy

etiology:
- trauma
- radiation
- infection
- iatrogenic etc.

Wound Healing Is the Physiologic Response to Tissue Trauma:
It is related to tissue reconstitution (e.g. skin, GI-tract) which is the process by which the body replenishes cells that are being lost by normal physiologic events. In both processes similar events occur in varying degrees. The same basic molecular mechanisms governing growth and differentiation are active to a different extent. Without these being properly in place, even old wounds may be become subject to "re-opening" (e.g. scurvy-Vitamin C deficiency in sailors).

Wound heals in 3 phases that partially overlap:

- inflammatory phase
- fibroplasia phase
- remodelling phase

INFLAMMATORY PHASE
(aka. lag phase or substrate phase) 0-48 hrs

- clinical signs: Rubor (redness), Calor (warmth), Tumor (swelling), Dolor (pain), Functiona Laesa (dysfunction)
- key elements - wound cleaned of bacteria, foreign material, necrotic debris, chemo-attraction of inflammatory cells and fibroblasts, development of a structural framework to allow for cell migration.

Vascular Response:

primary phase - haemostasis

- vasoconstriction
- platelet aggregation and degranulation
- fibrinous clot

- ➢ fibrinolysis

secondary phase

- ➢ venular vasodilatation - 10 x increase in blood flow
- ➢ increased vascular permeability - aids in flow of chemical and cellular mediators (PDGF) in inflammation to site of injury
- ➢ lymphatic obstruction leads to tissue edema

Cellular Response:

increased vascular permeability during inflammatory phase facilitates margination, extravasation and migration of cellular mediators (PMN's and macrophages)

PMN's

- ➢ *function short lived* (0-48 hours)
- ➢ *attacks bacteria, then leaves*
- ➢ *not essential for wound healing process*

Macrophages

- ➢ *resident and monocyte–derived*
- ➢ *central cell during inflammatory phase of wound repair*
- ➢ *activated by lymphokines, immune complexes and C3b*
- ➢ *release angiogenesis factor*
- ➢ *phagocytosis of wound debris*
- ➢ *essential for wound healing*

Active Entities:

Chemical Mediators:

Prostanoids

- ➢ *prostaglandins and leucotriens*
- ➢ *affect platelet adherence and vasoconstriction*
- ➢ *polyunsaturated, hydroxylated long chain fatty acids*

Growth factors

- ➢ PDGF, EGF, bFGF

Other mediators:

- ➢ *ADP, serotonin, epinephrine, IL-1, monokines, angiogenesis factors etc*

Fibronectin

- ➢ *glycoproteins which are a major constituent of granulation tissue*
- ➢ *seen in cell surfaces, basement membrane, pericellular matrices*
- ➢ *produced by fibroblasts, endothelial cells, monocytes*
- ➢ *in wound healing act as growth factor, mediate migration of epithelium through integrin receptors, chemotactic for monocytes and fibroblasts, stimulate endothelial migration and organization, releases fibroblast growth factor from monocyte*

Factors Affecting Inflammatory Phase:

- ➢ pus contains both proteolytic and collagenolytic enzymes; prolongs inflammatory phase and retards epithelialization and fibroplasia
- ➢ necrotic tissue, foreign body, haematoma - prolongation of inflammatory phase
- ➢ ASA - inhibition of cyclo-oxygenase mechanism
- ➢ steroids - inhibit macrophage function

PROLIFERATIVE PHASE

(aka. fibroplasia phase) day 2 to ~ 6 weeks

- ➢ clinical signs: disappearance of inflammatory signs, reduction of swelling, reduction of wound size (contraction), itching
- ➢ key-elements: net collagen synthesis, increase in wound tensile strength, scar formation

Epithelialization

- is a requirement for orderly progression into the proliferative phase. It starts in the inflammatory phase, it requires de-differentiation, mitosis, migration and then re-differentiation by basal cells of epidermis
- *epithelial cells have capacity to digest through eschar and clot by secreting matrix metalloproteinases*
- basal cells from epithelial remnants (sebaceous glands, hair follicles and sweat glands) in partial thickness wounds, or from wound edge in full thickness wound will:
- *dedifferentiate and lose contact inhibition*
- undergo rapid mitosis and migrate across wound surface
- *once basal cells have moved across and covered entire wound they regain contact inhibition, re-differentiate, and begin to mature with increasing skin thickness as upward displacement and differentiation of cells occurs, restoring the normal layers of epidermis*

Wound Contraction

- "wounds heal from side to side but contract from end to end"
- thought to be mediated by myofibroblast - can produce collagen but also contains smooth muscle filaments
- *"lock-step" mechanism for contraction - myofibroblasts produce contraction and deposit collagen at shortened length*
- *new evidence points to intracellular actin contraction of the cytoskeleton that is transferred through other cells through so-called "focal adhesions" and integrin receptors*
- highest rate of contraction from days 10-21

Collagen Deposition

- prior to collagen deposition fibroblasts deposit "ground substance" composed mainly of glycosaminoglycans (which combine with protein to form proteoglycan - so called "wire-brush" which provides a large surface area for water accumulation and interdigitations for scaffold)
- function of ground substance is to create scaffold onto which collagen can be deposited, aggregated, and oriented in appropriate fashion
- starting at day 3 or 4 collagen is deposited, net collagen deposition is positive until day 21. After this day, collagen deposition is in balance with collagen resorption and no further net collagen deposition occurs. Tensile strength of wound begins to increase shortly after collagen deposition starts, it continues to increase after net collagen deposition has ceased. It reaches a maximum at 60 days post-injury (80% of tensile strength of normal skin)

Collagen:

- *protein composed of 3 polypeptide chains arranged in helix*
- *intracellular helical procollagen cleaved into tropocollagen which is secreted, tropocollagen helices (15 Angstrom) form collagen filaments (200 Angstrom) and through further intra and inter-molecular bonds form collagen fibrils (2,000 Angstrom) and fibers (20,000 Angstrom), which in turn form collagen fiber bundles (100-200,000 Angstrom)*
- *collagen fibers maintained with covalent bonds and disulfide linkages*
- *limiting step is hydroxylation of proline which requires 3 cofactors -oxygen, iron, vitamin C*

- *collagen types:*

  Type I collagen - 80% of collagen in normal adult skin, comprises majority of collagen in bone, tendon, ligaments, also found in fascia, arteries, uterus, dentin

  Type II collagen - remaining 20% of collagen, more major constituent in early

wound, fetal scan, hypertrophic scar or keloid, also found in cartilage and eye
Type III collagen - main type of collagen found in granulation tissue and healing wound, also found in skin, arteries, uterus and bowel wall
Type IV collagen – main component of basement membranes
Type V collagen – also found in basement membranes

MATURATION AND RE-MODELLING PHASE

3 weeks to 1-2 years

- represents time during which type III collagen is replaced by type I collagen, and is re-oriented across lines of tension with the creation of more stable bonds between fibers - net results decreases the amount of collagen required to maintain wound integrity
- duration of phase dependent upon patient age (decreased age - increased duration), racial differences, type of wound, body location and duration of inflammatory phase
- *collagenases act to resorb necessary fibers that have been deposited randomly - initially collagen deposition = collagen resorption, but eventually resorption is greater than deposition*

Collagenase

- *many different types - all require calcium as co-factor*
- *activity*
  - *increased by adrenocorticoids, colchicine, PTH*
  - *decreased by progesterone, cysteine*

PATIENT FACTORS AFFECTING WOUND HEALING

SYSTEMIC FACTORS:

Drugs:

steroids

- inhibit macrophage function, decrease inflammatory response
- inhibit prolyl/lysyl hydroxylase - unstable collagen bonds
  inhibitory effects reversed by Vit A, anabolic steroids
- inactivates complement
- leads to T and B cell dysfunction
- decreases leucocyte bactericidal activity

anti-neoplastic agents

- decreased WBC's, decreased fibroblast proliferation, decreased wound contraction, decreased protein synthesis
- if given greater than 14 days post-op - no apparent long-term effects on wound healing (noted early decreased wound strength)
- colchicine - decreased collagen precursors, decreased collagen secretion (cellular constipation), increases activity of collagenases
- penicillamine - Ca chelator (Ca required for collagen x-linking)

NSAIDs

- decrease collagen synthesis by 45% even at normal levels

Vitamins: (supra-normal levels of Vitamins are not beneficial)

vitamin A

- co-factor collagen synthesis and x-linking - increases wound strength
- capable of reversing inhibitory effects of steroids and Vit E
- does not increase wound healing in absence of steroids
  may reactivate disease for which steroids are given

vitamin C

- co-factor for proline hydroxylation
- absence (scurvy) - causes capillary fragility and wound healing

problems

vitamin E
- ➢ decreases collagen synthesis, inhibits wound healing inhibited and decreases tensile strength

Trace Metals

Zinc
- ➢ *enzyme constituent*
- ➢ *cofactor for collagen synthesis*

Copper
- ➢ *extracellular cofactor*
- ➢ *required for collagen crosslinking*

Magnesium
- ➢ *cofactor in glycolization*

Nutritional Status
- ➢ catabolic state (net -ve nitrogen balance) inhibits wound healing
- ➢ protein depletion - prolongs inflammatory phase, impairs fibroplasia
- ➢ methionine required for forming disulfide bonds in collagen synthesis
- ➢ carbohydrates - energy source for WBC's
- ➢ fats - no significant effects, but fats are essential elements of all cell membranes

Associated Illnesses/Immunity
- ➢ cancer, infection, peripheral vascular disease, COPD/ hypoxia, obesity smoking - all negatively affect wound healing
- ➢ diabetes - affects wound healing in numerous ways - associated peripheral vascular disease, neuropathy, immuno-dysfunction (decreases chemotaxis, phagocytosis, intracellular killing), decreases collagen synthesis

Patient Age
- ➢ associated with delayed onset of healing, protraction of phases and an inability to reach same level of healing
- ➢ associated with decreased tensile strength and wound closure rate

LOCAL FACTORS:

pO_2
- ➢ in a person with normal nutritional status, oxygen level in healing wound is the rate limiting step (requires adequate inspired oxygen, Hb for transport, adequate tissue perfusion via large and small vessels)
- ➢ local ability to supply oxygen to healing wound process is inhibited by peripheral vascular disease, previous radiation, chronic inflammation, diabetes mellitus, infection;
- ➢ a paucity of vascularization limits the development of an adequate inflammatory reaction
- ➢ *required for collagen synthesis*
  - *proline hyroxylation*
  - *collagen secretion*
- ➢ required for leucocyte killing capacity (phagosomal peroxidation)
- ➢ *also needed for growth factor secretion and fibroblast proliferation*
- ➢ *hyperbaric O_2*
- ➢ *increases fibroblast proliferation (requires adequate perfusion)*

- ➢ *may induce neovacularization*

Localized Infection

- ➢ pus lowers local pO_2, has collagenolytic effect, prolongs inflammatory phase, inhibits re-epithelialization

Foreign Body

- ➢ clot, necrotic debris, dirt, suture, glass etc. prolong inflammatory phase, lead to increased susceptibility for infection and increased duration of re-epithelialization

Mechanical Stress

- ➢ affects quantity, aggregation and orientation of collagen fibers

Wound Hydration

- ➢ moist wound healing environment increases rate of re-epithelialization

Temperature

- ➢ environment temperature greater than 30^oC increases tensile strength

Previous Radiation Therapy

- ➢ acute radiation changes cause vascular stasis and associated decreased wound healing
- ➢ chronic radiation changes include irreversible damage to skin with progressive obliterative endarteritis and an inability for fibroblasts to replicate or contract resulting in progressive skin changes and inability to heal minor wounds, the wound is chronically hypoxic

Pressure

- ➢ undue pressure may lead to ischaemia of the wound, therefore, all wounds, particularly chronic wounds, should be off-loaded.

A CLINICAL APPROACH TO OPTIMALIZING WOUND HEALING:

The multitude of factors affecting wound healing should be examined for every individual. In general, to provide for optimal wound healing one should strive to eliminate underlying causative and/or contributory factors and stimulate positive physiologic factors required for the healing process. A systemic approach includes attention to the following:

SYSTEMIC FACTORS:

- optimize nutritional status
  - ➢ consult with dietician
  - ➢ ensure adequate protein and caloric intake, trace metals, vitamins
- examine psychosocial status
  - ➢ depression inhibits compliance and wound healing
  - ➢ pain control is extremely important
- optimize biochemical status
  - ➢ acid base balance
  - ➢ endocrinologic status, blood sugars, hypo-thyroidism
  - ➢ correct renal failure, liver failure
- optimize perfusion and oxygenation

- ➢ cardiac status
- ➢ pulmonary status
- ➢ anemia
- ➢ vascular bypass surgery if required

- correct spasticity and other physical factors contributing to abnormal mobility
- examine and reduce all medications

LOCAL FACTORS:

- optimize wound environment by
  - ➢ thorough wound debridement
  - ➢ eliminate foreign bodies, dead space and necrotic tissue
  - ➢ elimination of infection
    - systemic/topical antimicrobials
    - pH active agents
  - ➢ decrease edema
  - ➢ off-load pressure
  - ➢ provide moist wound environment