Tissue Repair part II

1. Healing by Second Intention (secondary union):

When tissue loss is more extensive, such as in large wounds, abscess formation, and ulceration, the repair process is more complex. In second intention healing, the inflammatory reaction is more intense, there is abundant development of granulation tissue, and the wound contracts by the action of myofibroblasts. This is followed by accumulation of ECM and formation of a large scar.

Repair begins within 24 hours of injury by the emigration of fibroblasts and the induction of fibroblast and endothelial cell proliferation.

By 3 - 5 days, a specialized type of tissue that is characteristic of healing, called **granulation tissue** is apparent.

The term granulation tissue derives from the pink, soft, granular gross appearance, such as that seen beneath skin wound.

Its **histologic appearance** is characterized by

1- proliferation of fibroblasts.

2- new thin-walled, delicate capillaries (angiogenesis).

3- with a loose ECM.

Granulation tissue then progressively accumulates connective tissue matrix, eventually resulting in the formation of a scar which may remodel over time.

Steps of repair by connective tissue deposition:

1- Formation of new blood vessels (angiogenesis).

2- Migration and proliferation of fibroblasts and deposition of ECM (scar formation).

3-Maturation and reorganization of the fibrous tissue (remodelling).

4-Wound contraction.

1. Formation of new blood vessels (Angiogenesis):

Angiogenesis, or neovascularization, is a process of formation of new blood vessels in which pre-existing vessels send out capillary sprouts to produce new vessels. Endothelial precursor cells may migrate from the bone marrow to areas of injury and participate in angiogenesis at these sites. Several growth factors induce angiogenesis, but the most important are vascular endothelial growth factor VEGF and fibroblast growth factor (FGF).

2. Migration of Fibroblasts and ECM Deposition (Scar Formation)

Scar formation occurs in two steps:

(1)-Migration and proliferation of fibroblasts into the site of injury.

** The granulation tissue is started by formation of a framework of

New vessels and loose ECM that develop early at the repair site.

** Then the recruitment and stimulation of **fibroblasts i**s driven by many growth factors, including PDGF, FGF-2 and TGF- β . (Derived from activated endothelium and inflammatory cells).

** **Macrophages**, in particular, are important cellular constituents of granulation tissue, and besides clearing extracellular debris and fibrin at the site of injury, they elaborate a host of mediators that induce fibroblast proliferation and ECM production.

** Sites of inflammation are also rich in **mast cells**, and **lymphocytes**. These can contribute directly or indirectly to fibroblast proliferation and activation.

(2)-Deposition of ECM by fibroblasts:

As healing progresses, the number of proliferating fibroblasts and new vessels decreases; however, the fibroblasts progressively assume a more synthetic phenotype, and hence there is increased deposition of ECM.

Collagen synthesis by fibroblasts begins early in wound healing (days 3 to 5) and continues for several weeks, depending on the size of the wound.

Many of the same growth factors that regulate fibroblast proliferation also participate in stimulating ECM synthesis and scar formation including **TGF-β, PDGF, and FGF**. These are potent fibrogenic agent stimulating the production of collagen, fibronectin and proteoglycans, causes migration and proliferation of fibroblasts, smooth muscle cells, and macrophages.

Cytokines as mediators of inflammation and may also function as growth factors and participate in ECM deposition and scar formation. **IL-1 and TNF**, for example, induce fibroblast proliferation and can have a fibrogenic effect. They are also chemotactic for fibroblasts and stimulate the synthesis of collagen by these cells.

The granulation tissue formed will evolves into a scar composed of largely **inactive,** spindle-shaped fibroblasts, dense collagen, fragments of elastic tissue, and other ECM components. Matured scar has vascular regression, which eventually transforms the highly vascularized granulation tissue into a **pale,** largely **avascular scar.**

3-Maturation and reorganization of the fibrous tissue (remodelling)

The transition from granulation tissue to scar involves shifts in the composition of the ECM; even after its synthesis and deposition, scar ECM continues to be modified and remodelled. The outcome of the repair process is, in part, a balance between ECM synthesis and degradation.

The degradation of collagens and other ECM components is accomplished by a family of matrix metalloproteinases (MMPs). MMPs are produced by a variety of cell types (fibroblasts, macrophages, neutrophils, synovial cells, and some epithelial cells), and their synthesis and secretion are regulated by growth factors, cytokines, and other agents.

4-Wound contraction:

Secondary healing involves wound contraction. Within 6 weeks, for example, large skin defects may be reduced to 5% to 10% of their original size, largely by contraction. This occurs due to the presence of **myofibroblasts,** which are modified fibroblasts exhibiting many of the ultrastructural and functional features of contractile smooth muscle cells.

Differences between secondary healing and primary healing:

1- A larger clot forms at the surface of the wound.

2- Inflammation is more intense because large tissue defects have a greater volume of necrotic debris, exudate, and fibrin that must be removed.

3- Large defects have a greater potential for secondary infections.

4- Much larger amounts of granulation tissue are formed to fill the gaps and provide the underlying framework for the regrowth of epithelium.

5- A greater volume of granulation tissue results in a greater mass of scar tissue.

Types of repair according to the type of the tissue:

1- Repair by regeneration:

Cell renewal occurs continuously in **labile tissues,** such as the bone marrow, gut epithelium, and the skin. Damage to epithelia or an increased loss of blood cells can be corrected by the proliferation and differentiation of stem cells and, in the bone marrow, by proliferation of more differentiated progenitors.

The renewal of hematopoietic cells is driven by growth factors called colony-stimulative factors (CSFs), which are produced in response to increased consumption or loss of blood cells.

Tissue regeneration can occur in parenchymal organs with **stable cell** populations, but with the exception of the liver, this is usually a limited process. Pancreas, adrenal,

thyroid, and lung tissues have some regenerative capacity. Extensive regeneration or compensatory hyperplasia can occur only if the residual tissue is structurally and functionally intact, as after partial surgical resection. By contrast, if the tissue is damaged by infection or inflammation, regeneration is incomplete and is accompanied by scarring.

2- **Repair by connective tissue:**

Repair occurs by replacement of the non-regenerated cells with connective tissue, or by a combination of regeneration of some cells and scar formation:

1- If tissue injury is severe or chronic, and results in damage to parenchymal cells and epithelia as well as the stromal framework.

2 -If **nondividing cells** are injured, repair cannot be accomplished by regeneration alone.

Aberrations of cell growth and ECM production

This may occur even in what begins as normal wound healing.

1**. Keloid**: refers to the accumulation of exuberant amounts of collagen that give rise to prominent, raised scars. There appears to be a heritable predisposition to keloid formation, and the condition is more common in blacks.

2. **Exuberant granulation**: healing wounds may also generate excessive granulation tissue that protrudes above the level of the surrounding skin and hinders re-epithelialization. The restoration of epithelial continuity requires cautery or surgical resection of the granulation tissue.

3. **Disabling fibrosis** associated with chronic inflammatory diseases such as rheumatoid arthritis, pulmonary fibrosis, and cirrhosis have many similarities to those involved in normal wound healing. In these diseases, however, persistent stimulation of fibrogenesis results from chronic immune reactions that sustain the synthesis and secretion of growth factors, fibrogenic cytokines, and proteases.

<u>Delay healing:</u> Factors causes delay healing:

1- **Infection** is the single most important cause of delay in healing; it prolongs the Inflammatory phase of the process and increases the local tissue injury.

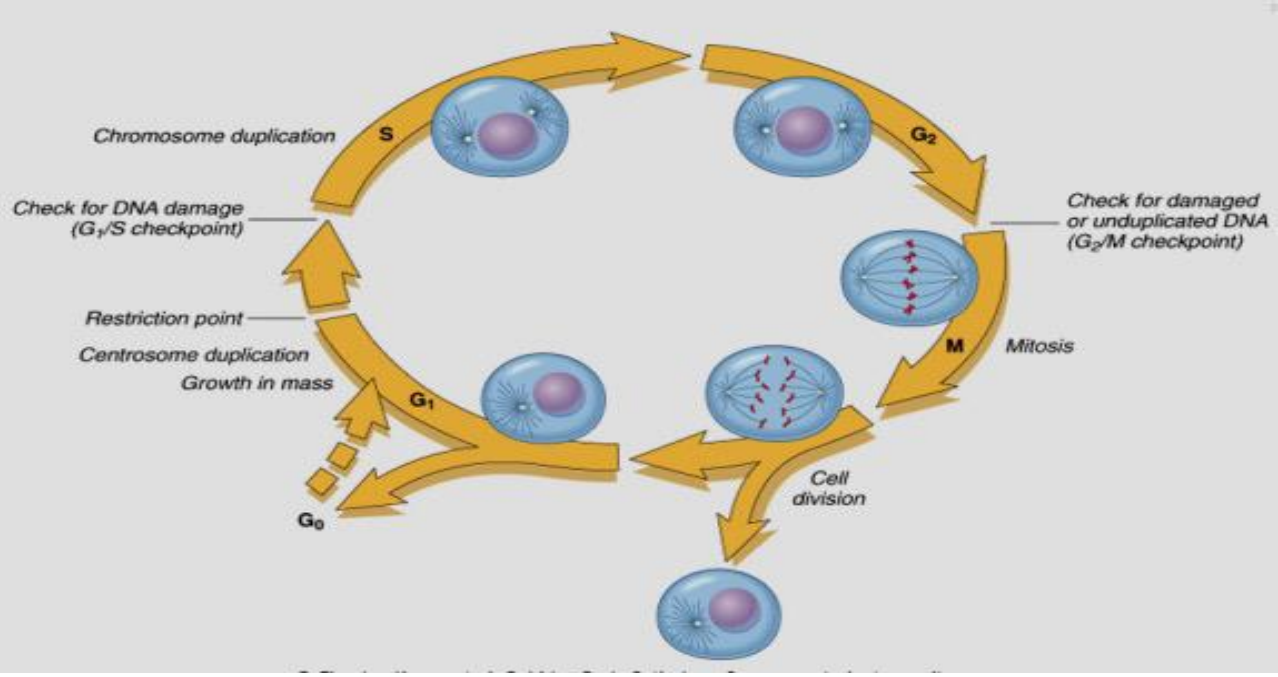
2- **Nutritional deficiency** like vitamin C deficiency, inhibits collagen synthesis and retards healing.

3- **Glucocorticoids** (steroids) have well-documented anti-inflammatory effects, and their administration may result in poor wound strength due to diminished fibrosis.

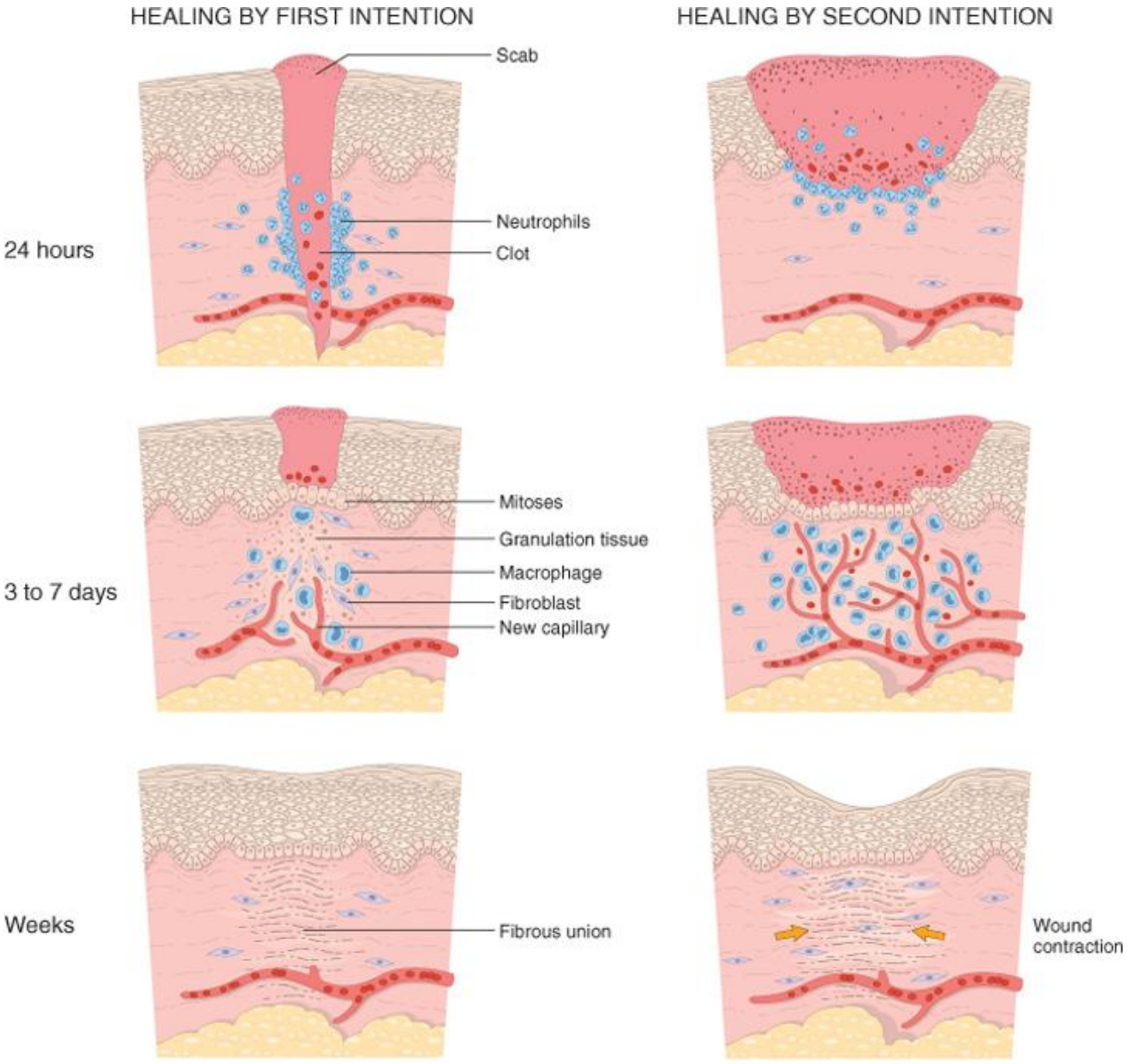
4- **Mechanical injuries** such as increased local pressure or torsion may cause wounds to pull apart, or opened.

5- **Diminished blood supply**, due either to arteriosclerosis and diabetes or to obstructed venous drainage (e.g. in varicose veins).

6- **Foreign bodies** such as fragments of steel, glass, or fractured bone.



THE CELL CYCLE: G0: quiescent stage, G1: growth presynthetic, S: DNA synthesis G2: growth premitotic, M: mitotic The G1 and S stages generally constitute the majority of the time of the cell cycle; the mitotic (M) phase is typically brief. Note the G1 restriction point, and the G1/S and G2/M checkpoints.



HEALING OF BONE FRACTURE

Bone fracture is caused by physical trauma, leading to discontinuity of the bone. The separation of fractured ends may be **complete** or **incomplete**. The latter is common in young children and called **greenstick fracture**. The fracture may be a **closed** one i.e. with an intact overlying skin or **open** i.e. the overlying skin is also injured so that the fractured bone is exposed through a gaping wound. A **comminuted** fracture is the one in which the bone is divided into multiple fragments.

Fracture healing

Due to tearing of blood vessels in the medullary cavity, cortex and periosteum, **a hematoma** forms at the site of fracture. The periosteum is stripped off form the bone surfaces. The bone with haemopoietic marrow around the fracture site undergoes **ischemic necrosis**. Bone death is recognized histologically by loss of osteocytes from lacunae (empty lacunae).

Organization of the hematoma is associated with a local inflammatory response, with hyperaemia, exudation of protein rich fluid, & migration of neutrophils & macrophages. These cells phagocytose the hematoma & necrotic debris. This is followed by in-growth of capillaries & fibroblasts, producing **fibrovascular granulation tissue.**

At the end of the 1st week, osteoblasts derived from osteoprogenitor cells of the inner layer of the periosteum will migrate into the granulation tissue and proceed to deposit larger quantities of osteoid in a haphazard way, producing a **woven bone** pattern. **External callus** is thus formed by the periosteum and tends to immobilize the bone fracture site. The two enlarging cuffs of callus advance towards each other until finally unite to bridge the fracture gap outside externally.

If there is a significant gap between the bone ends, it may induce cartilage formation.

The **internal callus derived from endosteal osteoprogenitor cells** bridges the fracture from within the medullary cavity, and rarely contains cartilage due to better vascularization.

The cartilaginous component of callus is converted to bone by endochondral ossification

Callus is usually formed by the 3rd week after the incident of fracture, but the **initial bony union is by woven bone, which is mechanically weak.**

The amount of periosteal callus formed (external callus) depends on the site of fracture & the degree of immobilization. It tends to be abundant in poorly immobilized fracture e.g. clavicle & ribs.

Remodelling of callus occurs once the defect between the two bone ends is bridged by bony callus, so the newly laid down bone is reconstructed to restore full mechanical strength. The newly formed woven bone is resorbed and gradually replaced by **lamellar bone** (compact).

The cortex is re-formed across the fracture gap & gradually the medullary callus is removed with restoration of the marrow cavity. Remodelling is done by the osteoblasts & osteoclasts.

The whole reparative process may take about a year, although the time varies from site to site. It is also more rapid and more complete in children.

Factors Affecting Fracture Healing

These are basically similar to those of affecting healing in general. However, mobility of fracture ends and mal-alignment play a detrimental role in interfering fracture healing. Vitamin D deficiency leads to abundant callus, which fails to calcify & remains soft.

Complications of fractures

1. **Delayed union,** after fibrous union, bony conversion is slow.

2. **Non-union,** in which the fractured bone ends do not join by bone. This occurs if the fibrous tissue becomes very dense. The latter is then is converted to fibrocartilage.

3. **Fat embolism,** which may follow damage to the bone marrow. In such cases globules of fat embolize to such sites as the lungs, brain, and kidneys with the ultimate result of ischemic necrosis (infarction).

4. **Osteonecrosis;** this refers to local bone necrosis after fracture. It may occur depending on local peculiarities of the blood supply, e.g. fracture of femoral neck is often followed by osteonecrosis of the femoral head.

5. **Osteoarthritis (osteoarthrosis)**; this degenerative joint disease may occur when the fracture line has involved the articular surface that result in the production of an in continuity of the articular cartilage.

Pathological Fracture

For fracture of normal bone to occur, the causative trauma has to be severe enough. In contrast, trivial trauma may cause fracture when the underlying bone is abnormal e.g**. presence of osteoporosis** (reduced bone mass) that occurs in the elderly may be associated with pathological fractures particularly in the femur & vertebral column. **Osteomalacia** (vitamin D deficiency lead to inadequate bone mineralization→ soft weak bone**), Primary or metastatic tumors** (from carcinoma of breast bronchus, thyroid & kidney) may be associated with pathological fractures.