EXTRACELLULAR MATRIX (ECM) AND CELL-MATRIX INTERACTIONS

Tissue repair depends not only on growth factor activity but also on interactions between cells and ECM components. *The ECM is a dynamic, constantly remodeling macromolecular complex synthesized locally, which assembles into a network that surrounds cells.* It constitutes a significant proportion of any tissue. Synthesis and degradation of ECM accompanies wound healing & chronic fibrotic processes.

ECM occurs in two basic forms:

1. Interstitial matrix, which is present in the spaces between mesenchymal (connective tissue) cells, and between epithelium and supportive vascular and smooth muscle structures; it is synthesized by the mesenchymal cells (e.g., fibroblasts). Its major constituents are fibrillar and nonfibrillar collagens, as well as fibronectin, elastin, proteoglycans, hyaluronate, and other elements.

2. Basement membrane, which lies beneath the epithelium and is synthesized by overlying epithelium and underlying mesenchymal cells; it tends to form a platelike "chicken wire" mesh. Its major constituents are amorphous nonfibrillar type IV collagen and laminin.

Functions of the ECM

1. Mechanical support **for cell anchorage and migration, and maintenance of cell polarity**

2. Control of cell growth by signaling through cellular receptors of the integrin family.

3. Maintenance of cell differentiation through the type of ECM proteins, also acting largely via cell surface integrins.

4. Scaffolding for tissue renewal: the maintenance of normal tissue structure requires a basement membrane or stromal scaffold.

5. Establishment of tissue microenvironments*:* basement membrane acts as a boundary between epithelium and underlying connective tissue and also forms part of the filtration apparatus in the kidney.

6. Storage and presentation of regulatory molecules*.* For example, growth factors like FGF is excreted and stored in the ECM in some tissues. This allows the rapid deployment of growth factors after local injury, or during regeneration.

Components of the Extracellular Matrix

There are three basic components of ECM:

1. Fibrous structural proteins (collagens and elastins).

2. Water-hydrated gels (proteoglycans and hyaluronan).

3. Adhesive glycoproteins (that include *fibronectin,* and *laminin)*

and ***adhesive receptors*** eg. (selectins, integrins and cadherins).

CELL AND TISSUE 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.
- Tissue regeneration can occur in parenchymal organs with **stable cell populations,** but with the exception of the liver, this is usually a limited process.
- *EGFR* (epidermal growth factor receptor) with intrinsic tyrosine kinase activity, is mitogenic for hepatocytes and most epithelial cells, including keratinocytes. In cutaneous wound healing EGF is produced by keratinocytes, macrophages, and other inflammatory cells
- It should be emphasized that 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.

REPAIR BY CONNECTIVE TISSUE

Healing or repair by connective tissue is encountered if

1. A severe or persistent (chronic) tissue injury that result in damage to parenchymal cells as well as the stromal framework

2. Injury affects non-dividing cells

Under these conditions, repair occurs by replacement of the non-regenerated cells with connective tissue, or by a combination of regeneration of some cells and scar formation.

Repair begins within 24 hours of injury by the emigration of fibroblasts and the induction of fibroblast and endothelial cell proliferation. By 3 to 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 the scab of a skin wound. Its microscopic appearance is characterized by proliferation of fibroblasts and new thin-walled, delicate capillaries, in a loose ECM. Granulation tissue then progressively accumulates connective tissue matrix, eventually resulting in the formation of a scar, which may remodel over time.

Repair by connective tissue deposition consists of four sequential processes:

1. *Formation of new blood vessels (angiogenesis)*

2. *Migration and proliferation of fibroblasts*
3. *Deposition of ECM (scar formation)*
4. *Maturation and reorganization of the fibrous tissue (remodeling)*

Angiogenesis (neo-vascularization)

The preexisting vessels send out capillary sprouts to produce new vessels. New vessels formed during angiogenesis are leaky. This leakiness explains why granulation tissue is often edematous, and accounts in part for the edema that may persist in healing wounds long after the acute inflammatory response has resolved. Several factors induce angiogenesis, but the most important are *VEGF* and *basic fibroblast growth factor*

Angiogenesis is also encountered in development of collateral circulation in ischemic tissues and in growing tumors.

Migration of Fibroblasts and ECM Deposition (Scar Formation)

Scar formation builds on the granulation tissue framework of new vessels and loose ECM that develop early at the repair site. It occurs in two steps:

1. Migration and proliferation of fibroblasts into the site of injury and

2. Deposition of ECM by these cells.

CUTANEOUS WOUND HEALING

This is a process that involves both epithelial regeneration and the formation of connective tissue scar and is thus illustrative of the general principles that apply to wound healing in all tissues. The events are orchestrated by interplay of growth factors and ECM.

Cutaneous wound healing has three main phases:

1. *inflammation*
2. *formation of granulation tissue*
3. *ECM deposition and remodeling*

Larger wounds also contract during the healing process. Events in wound healing overlap to a great extent and cannot be completely separated from each other.

Based on the nature of the wound, the healing of cutaneous wounds can occur by first or second intention.

Healing by First Intention

One of the simplest examples of wound repair is the healing of a clean, uninfected surgical incision approximated by surgical sutures. This is referred to as *primary union* or *healing by first intention.* The incision causes only focal disruption of epithelial basement membrane continuity and death of a relatively few epithelial and connective tissue cells. As a result, epithelial regeneration predominates over fibrosis. A small scar is formed, but there is minimal wound contraction.

Healing by Second Intention (healing by secondary union)

This mode of healing occurs in

1. Large wounds 3- Abscesses
2. Ulcerations 4- After infarction in parenchymal organs.

Secondary healing differs from primary healing in several respects:

1. A larger clot or scab rich in fibrin and fibronectin 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. Much larger amounts of granulation tissue are formed that generally results in a greater mass of scar tissue.
4. 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 process has been ascribed to the presence of myofibroblasts, which are modified fibroblasts exhibiting many of the ultrastructural and functional features of contractile smooth muscle cells.

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.

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. This may lead to **Pseudoarthosis** (formation of a false joint).

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

Healing in the Nervous System

Mature neurons are permanent cells i.e. they cannot divide. Any damage to the brain or spinal cord is followed by capillary ingrowth and astrocytic and microgial proliferation (Gliosis). Gliosis in the CNS is the equivalent of scar formation elsewhere. Once gliosis is established, it remains permanently.

Neurons in the peripheral nervous system can regenerate their axons.

Under ideal circumstances section of a peripheral nerve results in complete functional recovery. However if the cut ends are not in perfect alignment, granulation tissue grows between them resulting in a traumatic neuroma.