Healing and Repair

Overview

The body attempts to heal damage induced by local injury very early in the process of inflammation. However, inflammation and repair are closely intertwined; but repair is discussed as a separate entity.

General considerations

Repair of injuries is intimately associated with the inflammatory response. The healing process begins early in the inflammatory process and results in repair of the injury by replacement of dead or damaged cells with healthy cells. The body uses two distinct processes to effect repairs:

1. **Regeneration**, which is the replacement of injured tissue with cells of the same type.
2. **Replacement** by connective tissue.

Most injuries are repaired by a combination of these processes. Obviously, it is most advantageous for repairs to occur by regeneration because this will restore the organ to normal functioning capabilities. Repair by regeneration is governed largely by several factors including the **regenerative capacity** of the cells involved and the **severity of the injury**.

Regeneration

Based on their regenerative capabilities, the cells of the body are divided into three groups: **labile cells**, **stable cells**, and **permanent cells**.

A. **Labile cells**. proliferate normally throughout life replacing cells that are continually being destroyed (continuously and rapidly replaced).
   Examples: Surface epithelium of skin, G.I. tract, G.U tract, hematopoietic cells.

B. **Stable cells**. These are continuously but slowly replaced. Their proliferation can be markedly accelerated during regeneration (stimulated to divide rapidly in response to various stimuli).
   Examples: Hepatocytes, renal tubular epithelial cells, endothelium, smooth muscle and mesenchymal cells such as fibroblasts, smooth muscle cells, osteoblasts, and chondroblasts.

C. **Permanent cells**: Although portions of these cells may be restored (e.g. neuron), the cells themselves are not replaced. Regeneration does not occur.
Examples: Skeletal and cardiac muscle, CNS neurons.

Injuries in organs or tissues composed largely of permanent cells will be repaired by connective tissue replacement. Injuries in organs composed largely of labile or stable cells are repaired either by regeneration or by a combination of regeneration and connective tissue replacement. The extent of the injury is a major factor in determining which of these occurs. Because the scaffolding provided by stroma and basement membranes is so critical, if the injury is such that these structures are preserved, it is more likely that injury by regeneration will occur. However, if these structures are also damaged, then repair by connective tissue replacement becomes more likely.

Repair by connective tissue replacement:

This type of repair predominates when injuries occur in tissues formed largely of permanent cells or when the injury results in extensive damage to stromal framework and supporting connective tissues. In these situations, the injured tissue is replaced by fibroblastic cells, usually in the form of granulation tissue, which eventually results in the formation of a scar.

Granulation tissue:

Early in the inflammatory process, fibroblasts and vascular endothelial cells start to proliferate. Sometimes this begins as early as 24 hours after injury. By 3 to 5 days, a specialized type of tissue appears that is known as **granulation tissue**. This specialized tissue is composed of proliferating fibroblasts and newly formed blood vessels. The process resulting in the development of these newly formed blood vessels is called **angiogenesis** or **neovascularization**. This process is important in healing and is also involved in the progressive growth of parenchymatous tumors. It occurs in four basic steps:

- enzymatic degradation of the basement membrane of the parent vessel,
- migration of endothelial cells toward the angiogenic stimulus,
- proliferation of endothelial cells,
- maturation of endothelial cells and organization into capillary tubes.

These newly formed vessels have leaky inter-endothelial junctions, thus granulation tissue tends to be edematous.

Granulation tissue will generally have considerable numbers of macrophages. Initially, their main purpose is to eliminate injuring agents, macrophages also remove extracellular debris and ultimately they participate in "blanching" of the wound, a process by which the excess granulation tissue is removed. In addition, granulation tissue may have varying numbers of neutrophils, lymphocytes, and eosinophils.

First intention healing (Primary Union)

This type of healing occurs when there is no contamination of the wound and the edges of the wound are approximated, thus closing the wound. The best example of this situation is the surgical incision where contamination of the wound is minimized and the wound is closed by suturing.

Once the wound is sutured, the incision space fills with blood, which contains fibrin and blood cells and which subsequently clots. The surface of this clot becomes dehydrated and forms a **scab**. *Within 24 hours*, **neutrophils** appear at the edges of the incision and the epithelium at the edges of the incision begins to proliferate. It migrates under the scab and forms a thin continuous epithelial layer. *By 72 hours*, **macrophages** are usually the most numerous inflammatory cells and granulation tissue starts to develop. Collagen fibers are present but do not bridge the incision site. The epithelial cells continue to proliferate under the scab and the epidermal covering over the incision becomes thicker. *By day 5*, the incision space is filled with granulation tissue and collagen fibers begin to bridge the incision. The epidermis returns to its normal thickness and keratinized architecture. *During the second week*, there is continued accumulation of collagen fibers and proliferation of fibroblasts. Inflammatory cells and edema disappear and the process of blanching begins. *By the end of one month*, there is a connective tissue scar that is devoid of inflammatory cells and is covered by an intact epidermis.

Second intention healing (Secondary Union)

This type of healing occurs when injuries result in more extensive loss of tissues such as with infarction, inflammatory ulceration, and large surface wounds. In these situations, due to the

large tissue defect, repair by regeneration is minimal and the defect is filled by granulation tissue.

Second intention healing differs from first intention healing in several ways. ***First***, the greater injury invokes a more intense inflammatory response. ***Secondly***, much more granulation tissue is formed. And ***thirdly***, wounds that are repaired by second intention healing undergo a phenomenon known as "**wound contraction**" whereby specialized granulation tissue fibroblasts called myofibroblasts contract and dramatically reduce the size of the wound.

Common aberrations (complications) of the healing process

- **Exuberant Granulation**

  Exuberant granulation is characterized by excessive formation of granulation tissue such that a mass of granulation tissue protrudes from the wound and prevents re-epithelialization. Such excesses are commonly referred to as **"proud flesh".** This is a commonly encountered problem in the management of wounds in **horses**.

- **Keloid Formation**

  Keloid formation also refers to an aberration of wound healing resulting in the formation of large bulging scars but it differs from granulation tissue in that it is caused by **excessive collagenization** of the wound and not excessive formation of granulation tissue. This phenomenon is a common problem in darker people.

Mechanisms involved in repair:

The mechanisms regulating repairs are becoming better understood and the more important features involved in this control include:

- the role of growth factors,
- cell to cell and cell to matrix interactions,
- extracellular matrix synthesis and collagenization.

❖ Growth Factors

The cell proliferation is controlled by a delicate counterbalance between **growth stimulators** and **growth inhibitors**. Numerous growth factors have been reported. Some

of these growth factors stimulate DNA synthesis directly in competent cells and are referred to as **progression factors**, while others merely make cells competent to be stimulated for DNA synthesis and are referred to as **competence factors**.

- **Epidermal growth factor: (EGF)** is a polypeptide that is mitogenic for a variety of epithelial cells and fibroblasts ***in vitro*.**
- **Platelet-derived Growth Factor: (PDGF**) is primarily found in the alpha granules of platelets from which it is released subsequent to platelet activation. It is also produced by activated macrophages, endothelium, smooth muscle cells, and a variety of tumor cells. **PDGF** causes proliferation and migration of fibroblasts and smooth muscle cells.
- **Fibroblast Growth Factor(s): (FGFs**) are a family of polypeptide growth factors that have numerous activities including stimulation of fibroblast proliferation and angiogenesis.
- **Transforming Growth Factor Alpha and Transforming Growth Factor Beta:** Transforming growth factor alpha is similar to EGF structurally. It binds to EGF receptors and produces similar biologic effects as EGF. Transforming growth factor beta is produced by different cell types including platelets, endothelium, T cells, and macrophages. **It inhibits growth** in most cell types; however, it stimulates fibroblast chemotaxis and the production of collagen and fibronectin by cells.
- **Interleukin-1 (IL-1) and Tumor Necrosis Factor: (TNF)** stimulate fibroblastic proliferation and the synthesis of collagen and collagenase. They are believed to play a role in fibroplasia and remodeling of inflammatory connective tissue. In addition**,** **TNF** has been shown to have angiogenic properties in vivo.

❖ Growth Inhibitors

A number of growth inhibitors are known to be produced in inflammation. Transforming growth factor beta has already been described and others include alpha interferon, prostaglandin E2, and heparin.

❖ Cell to Cell and Cell to Matrix Interactions

Normal cells in tissue cultures tend to proliferate until a confluent monolayer is formed at which point proliferation ceases. This density-dependent regulation is controlled by either

- (1) limitation of necessary materials in the environment,
- (2) alterations in the number of receptor sites for growth factors,
- (3) accumulation of growth inhibitors.

This same phenomenon occurs in vivo and is at least partly responsible for the regulation of cell proliferation in healing. It has been shown that transforming growth factor beta is responsible for limiting proliferation of hepatocytes following partial hepatectomy.

The nature of the matrix appears to influence cell proliferation and differentiation. Such factors include:

- the type of collagen,
- the presence of fibronectin or laminin, and
- the nature of the proteoglycans.

Endothelial cells grown in culture and exposed to growth factors, proliferate faster when grown on type I collagen or laminin than when grown on type IV collagen. On the other hand, when grown on type IV collagen, they tend to form tube-like structures. Fibronectin of fibronectin fragments promote migration of fibroblasts and endothelial cells into an area of injury. This cell to cell interaction seems to be mediated through cell surface receptors which interact with the cytoskeleton to signal locomotion or differentiation. This group of receptors includes **integrins** which are primarily adhesion receptors such as fibronectin receptors, platelet glycoprotein receptors, and leukocyte adhesion molecules.

Collagenization of a wound:

Collagen ultimately provides the tensile strength of healing wounds. It is produced by the proliferating fibroblasts that are a part of the healing process. The fundamental unit of collagen is the collagen molecule which is called tropocollagen. Based on biochemical composition of the molecules, 11 types of collagen are recognized; however, types I, II, and III are the interstitial or fibrillar collagens.

Common Defects Involving Collagen

A critical modification of collagen occurs in the rough endoplasmic reticulum depending on ascorbic acid **(vitamin C**) that is necessary to hold the collagen molecules in the RER. A dietary deficiency of ascorbic acid leads to inadequate collagen production and causes a disease known as scurvy.

There are inherited diseases characterized by defective collagen production as well. One group of such diseases is referred to as Ehlers-Danlos Syndrome **(EDS)**. Based on the underlying biochemical defect, at least 20 variants of EDS are recognized and most are characterized by hyperextensible skin and hypermobile joints.

Another inherited disease involves oxidation of lysine of collagen molecules, a critical modification that occurs extracellularly. In humans, this disease is referred to as Marfan's Syndrome. Marfan's Syndrome is characterized by malformations in the skeleton, skin, and blood vessels.

Factors that might modify the repair response:

Many factors, including **nutrition** and the presence of other disease states, tend to influence the repair response.

Prolonged protein starvation tends to retard the development of tensile strength in healing wounds and conversely, a high protein diet accelerates the rate of tensile strength gain.

In addition, a dietary deficiency of ascorbic acid (***Vitamin C***) will also reduce collagenization of a healing and result in retardation of the acquisition of wound strength.

Any phenomenon that prolongs infections in a healing wound will result in a decreased rate of wound healing. This includes defects in either numbers or function of neutrophils or macrophages. It also includes defects resulting in increased bleeding because certain blood elements often serve as nutrients for bacteria.

Other influences include the presence of systemic diseases such as diabetes, corticosteroid therapy, adequacy of blood supply, the presence of foreign bodies, and the nature of the tissue involved.