

Inflammation and Repair

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OVERVIEW OF INFLAMMATION: DEFINITIONS AND GENERAL FEATURES

Inflammation is a response of vascularized tissues that delivers leukocytes and molecules of host defense from the circulation to sites of infection and cell damage. Although in common medical and lay parlance inflammation suggests a harmful reaction, it is actually a protective response that is essential for survival. It serves to rid the host of both the initial cause of cell injury (e.g., microbes, toxins) and the consequences of such injury (e.g., necrotic cells and tissues). The mediators of defense include phagocytic leukocytes, antibodies, and complement proteins. Most of these normally circulate in a resting state in the blood, where they are sequestered from tissues and unable to cause damage and from where they can be rapidly recruited to any site in the body when they are needed. Some of the cells involved in inflammatory responses also reside in tissues, where they function as sentinels on the lookout for threats. The process of inflammation delivers circulating cells and proteins to tissues and activates the recruited and resident cells

as well as some soluble molecules, which then function to get rid of the harmful or unwanted substances. Without inflammation, infections would go unchecked, wounds would never heal, and injured tissues might remain permanent festering sores. The suffix *-itis* after an organ denotes inflammation in that site, such as appendicitis, conjunctivitis, or meningitis.

The typical inflammatory reaction develops through a series of sequential steps (Fig. 3.1):

- **Recognition** of the noxious agent that is the initiating stimulus for inflammation. The cells involved in inflammation (tissue-resident sentinel cells such as dendritic cells, phagocytes, and others) are equipped with receptors that recognize microbial products and substances released from damaged cells. These receptors are described in more detail later. Engagement of the receptors leads to the production of mediators of inflammation, which then trigger the subsequent steps in the inflammatory response.
- **Recruitment** of leukocytes and plasma proteins into the tissues. Since blood perfuses every tissue, leukocytes and proteins (e.g.,

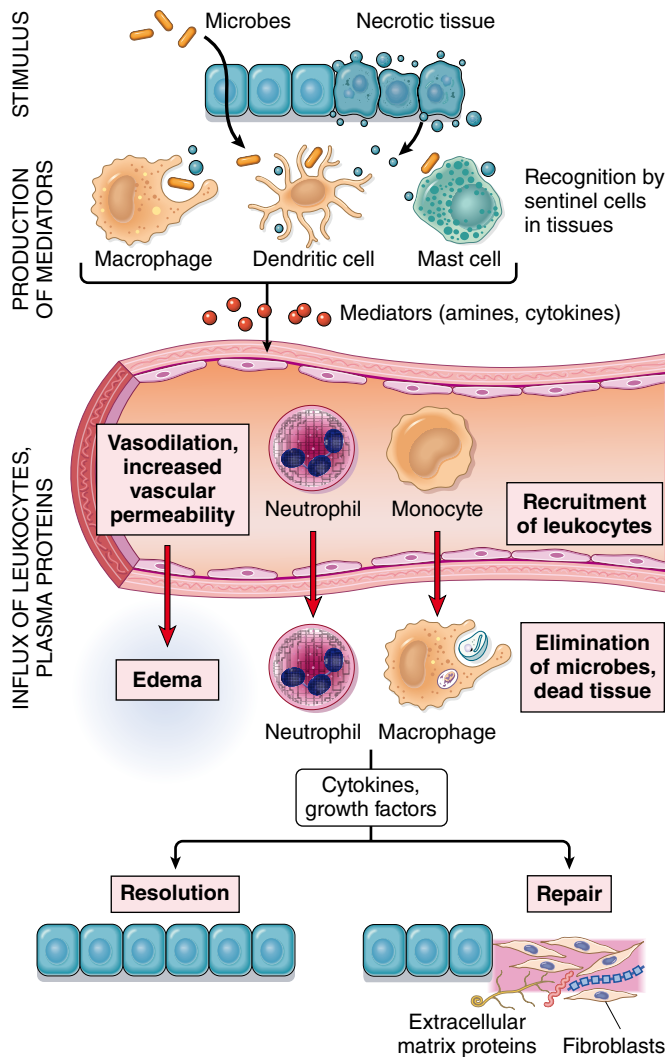


Fig. 3.1 Sequence of events in an inflammatory reaction. Sentinel cells in tissues (macrophages, dendritic cells, and other cell types) recognize microbes and damaged cells and liberate mediators, which trigger the vascular and cellular reactions of inflammation. Plasma proteins involved in inflammation are not shown.

complement proteins) can be delivered to any site of microbial invasion or tissue injury. When pathogenic microbes invade the tissues, or tissue cells die, leukocytes (first mainly neutrophils, later monocytes and lymphocytes) and plasma proteins are rapidly recruited from the circulation to the extravascular site where the offending agent is located. The exodus of cells and plasma proteins from blood requires coordinated changes in blood vessels and secretion of mediators, described in detail later.

- **Removal** of the stimulus for inflammation is accomplished mainly by phagocytic cells, which ingest and destroy microbes and dead cells.
- **Regulation** of the response is important for terminating the reaction when it has accomplished its purpose.
- **Repair** consists of a series of events that heal damaged tissue. In this process the injured tissue is replaced through regeneration of surviving cells and filling of residual defects with connective tissue (scarring).

Before discussing the mechanisms, functions, and pathology of the inflammatory response, it is useful to review some of its fundamental properties.

- **Components of the inflammatory response.** The major participants in the inflammatory reaction in tissues are blood vessels, leukocytes, and various plasma proteins (see Fig. 3.1). As will be discussed in more detail later, blood vessels respond to inflammatory stimuli by dilating and by increasing their permeability, enabling circulating proteins to enter the site of infection or tissue damage. In addition, the endothelium lining blood vessels changes, such that circulating leukocytes adhere and then migrate into the tissues. Leukocytes, once recruited, are activated and acquire the ability to ingest and destroy microbes and dead cells, as well as foreign bodies and other unwanted materials in the tissues.
- **Acute and chronic inflammation.** The distinction between acute and chronic inflammation was originally based on the duration of the reaction, but we now know that they differ in several ways (Table 3.1). Acute inflammation is a rapid, often self-limited, response to offending agents that are readily eliminated, such as many bacteria and fungi, and dead cells. It typically develops within minutes or hours and is of short duration (several hours to a few days). It is characterized by the exudation of fluid and plasma proteins (edema) and the emigration of leukocytes, predominantly neutrophils. If the offending stimulus is eliminated, the reaction subsides, and residual injury is repaired.

Chronic inflammation may follow acute inflammation or arise de novo. It is a response to agents that are difficult to eradicate, such as some bacteria (e.g., tubercle bacilli) and other pathogens (such as viruses and fungi), as well as self antigens and environmental antigens. Chronic inflammation is of longer duration and is associated with more tissue destruction and scarring (fibrosis). Sometimes, chronic inflammation may coexist with unresolved acute inflammation, for example, in peptic ulcers.

- **Harmful consequences of inflammation.** Protective inflammatory reactions to infections are often accompanied by local tissue damage and its associated signs and symptoms (e.g., pain and functional impairment). Typically, however, these harmful consequences are self-limited and resolve as the inflammation abates, leaving little or no permanent damage. In contrast, there are many diseases in which the inflammatory reaction is misdirected (e.g., against self tissues in autoimmune diseases), occurs against normally harmless environmental substances (e.g., in allergies), or is inadequately controlled. In these cases, the normally protective inflammatory reaction becomes the cause of the disease, and the damage it causes is the dominant feature. In clinical medicine, great attention is given to the injurious consequences of inflammation (Table 3.2). Inflammatory reactions underlie common chronic diseases such as rheumatoid arthritis, atherosclerosis, and lung fibrosis, as well as life-threatening hypersensitivity reactions to insect bites, foods, drugs, and toxins. For this reason, pharmacies abound

TABLE 3.1 Features of Acute and Chronic Inflammation

Feature	Acute	Chronic
Onset	Fast: minutes or hours	Slow: days
Cellular infiltrate	Mainly neutrophils	Monocytes/macrophages and lymphocytes
Tissue injury	Usually mild and self-limited	May be severe and progressive
Fibrosis	None	May be severe and progressive
Local and systemic signs	Prominent	Less

TABLE 3.2 Examples of Diseases Caused by Inflammatory Reactions

Disorders	Cells and Molecules Involved in Injury
Acute	
Acute respiratory distress syndrome	Neutrophils
Glomerulonephritis, vasculitis	Antibodies and complements; neutrophils, monocytes
Septic shock	Cytokines
Chronic	
Rheumatoid arthritis	Lymphocytes, macrophages; antibodies?
Asthma	Eosinophils; IgE antibodies

Listed are selected examples of diseases in which the inflammatory response plays a significant role in tissue injury. Some, such as asthma, can present with acute inflammation or a chronic illness with repeated bouts of acute exacerbation. These diseases and their pathogenesis are discussed in relevant chapters.

IgE, Immunoglobulin E.

with antiinflammatory drugs, which ideally would control the harmful sequelae of inflammation yet not interfere with its beneficial effects. Inflammation also may contribute to a variety of diseases that are thought to be primarily metabolic, degenerative, or genetic, such as type 2 diabetes, Alzheimer disease, and cancer.

- **Local and systemic inflammation.** Most of this chapter focuses on the inflammatory response to a localized infection or tissue damage. Although even local reactions may have systemic manifestations (e.g., fever in the setting of bacterial or viral pharyngitis), the inflammation is largely confined to the site of infection or damage. In rare situations, such as some disseminated bacterial infections, the inflammatory reaction is systemic and causes widespread pathologic abnormalities. This reaction is called *sepsis*. *Systemic inflammatory response syndrome* (SIRS) is a sepsis-like syndrome not caused by microbial infections. It can be caused by burns, trauma, and pancreatitis (Chapter 4).
- **Mediators of inflammation.** The vascular and cellular reactions of inflammation are triggered by soluble factors that are produced by various cells or derived from plasma proteins and are generated or activated in response to the inflammatory stimulus. Microbes, necrotic cells (whatever the cause of cell death), and even hypoxia can trigger the elaboration of inflammatory mediators and thus elicit inflammation. Such mediators initiate and amplify the inflammatory response and determine its pattern, severity, and clinical and pathologic manifestations.

Historical Highlights

Although clinical features of inflammation were described in an Egyptian papyrus dated around 3000 BCE, Celsus, a Roman writer of the first century CE, first listed the four cardinal signs of inflammation in his treatise *De Medicina*: *rubor* (redness), *tumor* (swelling), *calor* (heat), and *dolor* (pain). These signs are hallmarks of acute inflammation. A fifth clinical sign, loss of function (*functio laesa*), was added by Rudolf Virchow, known as the father of modern pathology, in the nineteenth century. In 1793, the Scottish surgeon John Hunter noted what is now considered an obvious fact: inflammation is not a disease but a stereotypic response that has a salutary effect on its host. In the 1880s, the Russian biologist Elie Metchnikoff discovered the process of *phagocytosis* by observing the ingestion of rose thorns by amebocytes of starfish larvae and of bacteria by mammalian leukocytes. He concluded that the purpose of inflammation was to bring phagocytic cells to the injured area to engulf invading

bacteria. Sir Thomas Lewis, studying the inflammatory response in skin, established the concept that chemical substances, such as histamine (produced locally in response to injury), mediate the vascular changes of inflammation. This fundamental concept underlies the important discoveries of chemical mediators of inflammation and the clinical use of antiinflammatory drugs to inhibit these mediators.

Causes of Inflammation

Inflammatory reactions may be triggered by a variety of stimuli:

- **Infections** (bacterial, viral, fungal, parasitic) and microbial toxins are among the most common and medically important causes of inflammation. Different infectious pathogens elicit varied inflammatory responses, from mild acute inflammation that causes little or no lasting damage and successfully eradicates the infection, to severe systemic reactions that can be fatal, to prolonged chronic reactions that cause extensive tissue injury. The outcomes are determined largely by the type of pathogen and the host response and, to some extent, by other, poorly defined characteristics of the host.
- **Tissue necrosis** elicits inflammation regardless of the cause of cell death. Cells may die because of ischemia (reduced blood flow, the cause of myocardial infarction), trauma, or physical and chemical injury (e.g., thermal injury, as in burns or frostbite; irradiation; exposure to some environmental chemicals). Several molecules released from necrotic cells are known to trigger this type of sterile inflammation; some of these are described later.
- **Foreign bodies** (splinters, dirt, sutures) may elicit inflammation by themselves or because they cause traumatic tissue injury or carry microbes. Even endogenous substances can be harmful if they deposit in tissues; such substances include urate crystals (in gout) and cholesterol crystals (in atherosclerosis).
- **Immune reactions** (also called *hypersensitivity*) are reactions in which the normally protective immune system damages the individual's own tissues. The injurious immune responses may be inappropriately directed against self antigens, causing autoimmune diseases, or may be reactions against environmental substances, as in allergies, or against microbes. Inflammation is a major cause of tissue injury in these diseases (Chapter 6). Because the stimuli for autoimmune and allergic responses (self antigens and environmental antigens, respectively) cannot be eliminated, these reactions tend to be persistent and difficult to cure, are often associated with chronic inflammation, and are important causes of morbidity and mortality. Inflammation in these disorders is induced largely by cytokines produced by T lymphocytes and other cells of the immune system (Chapter 6).

Recognition of Microbes and Damaged Cells

Recognition of microbial components or substances released from damaged cells is the initiating step in inflammatory reactions. The cells and receptors that perform this function evolved to protect multicellular organisms from microbes in the environment, and the responses they trigger are critical for the survival of the organisms. Several cellular receptors and circulating proteins are capable of recognizing microbes and products of cell damage and triggering inflammation.

- **Cellular receptors for microbes.** Cells express receptors in the plasma membrane (for extracellular microbes), the endosomes (for ingested microbes), and the cytosol (for intracellular microbes) that enable the cells to sense the presence of foreign invaders in any cellular compartment. The best defined of these receptors belong to the family of *Toll-like receptors* (TLRs); these and other cellular receptors of innate immunity are described in Chapter 6. The receptors are expressed on many cell types, including epithelial cells (which are often the first line of defense against microbes entering from the external environment), dendritic cells, macrophages, and other leukocytes

(which may encounter microbes in various tissues). Engagement of these receptors triggers production of molecules involved in inflammation, including adhesion molecules on endothelial cells, cytokines, and other mediators.

- **Sensors of cell damage.** All cells have cytosolic receptors, such as *NOD-like receptors* (NLRs), that recognize diverse molecules that are liberated or altered because of cell damage. These molecules include uric acid (a product of DNA breakdown), adenosine triphosphate (ATP) (released from damaged mitochondria), reduced intracellular K^+ concentrations (reflecting loss of ions because of plasma membrane injury), DNA when it is released into the cytoplasm and not sequestered in nuclei (as it should be normally), and many others. These receptors activate a multiprotein cytosolic complex called the *inflammasome* (Chapter 6), which induces the production of the cytokine interleukin-1 (IL-1). IL-1 recruits leukocytes that get rid of the damaged cells, which serve no purpose and could be harmful by providing a nidus for infection. Gain-of-function mutations in the genes encoding some of the receptors are the cause of rare diseases grouped under *autoinflammatory syndromes* that are characterized by spontaneous IL-1 production and inflammation; IL-1 antagonists are effective treatments for these disorders. The inflammasome has also been implicated in inflammatory reactions to urate crystals (the cause of gout), lipids (in metabolic syndrome and obesity-associated type 2 diabetes), cholesterol crystals (in atherosclerosis), and even amyloid deposits in the brain (in Alzheimer disease). These disorders are discussed later in this and other chapters.
- **Other cellular receptors involved in inflammation.** In addition to directly recognizing microbes, many leukocytes express receptors for the Fc tails of antibodies and for complement proteins. These receptors recognize microbes coated with antibodies and complement (the coating process is called *opsonization*) and promote ingestion and destruction of the microbes as well as inflammation.
- **Circulating proteins.** The *complement system* reacts against microbes and produces mediators of inflammation (discussed later). A circulating protein called *mannose-binding lectin* recognizes microbial sugars and promotes ingestion of the microbes and the activation of the complement system. Other proteins called *collectins* also bind to and combat microbes.

KEY CONCEPTS

General Features and Causes of Inflammation

- Inflammation is a beneficial host response to foreign invaders and necrotic tissue, but it may also cause tissue damage.
- The main components of inflammation are a vascular reaction and a cellular response, both activated by mediators derived from plasma proteins and various cells.
- The steps of the inflammatory response can be remembered as the five R's: (1) recognition of the injurious agent, (2) recruitment of leukocytes, (3) removal of the agent, (4) regulation (control) of the response, and (5) repair (resolution).
- Acute and chronic inflammation differ in the kinetics of the reaction, the principal cells involved, and the degree of injury. The outcome of acute inflammation is either elimination of the noxious stimulus followed by decline of the reaction and repair of the damaged tissue or persistent injury resulting in chronic inflammation.
- Causes of inflammation include infections, tissue necrosis, foreign bodies, trauma, and immune responses.
- Epithelial cells, tissue macrophages and dendritic cells, leukocytes, and other cell types express receptors that sense the presence of microbes and substances released from damaged cells. Circulating proteins recognize microbes that have entered the blood.

ACUTE INFLAMMATION

Acute inflammation has three major components: (1) dilation of small vessels leading to an increase in blood flow; (2) increased permeability of the microvasculature enabling plasma proteins and leukocytes to leave the circulation; and (3) emigration of leukocytes from the microcirculation, their accumulation in the focus of injury, and their activation to eliminate the offending agent (see Fig. 3.1).

When an individual encounters an injurious agent, such as a microbe, or dead cells, phagocytes that reside in tissues try to eliminate these. At the same time, phagocytes and other sentinel cells in the tissues recognize the presence of the foreign or abnormal substance and react by liberating cytokines, lipid messengers, and other mediators of inflammation. Some of these mediators act on small blood vessels in the vicinity and promote the efflux of plasma and the recruitment of circulating leukocytes to the site where the offending agent is located.

Reactions of Blood Vessels in Acute Inflammation

The vascular reactions of acute inflammation consist of changes in the flow of blood and the permeability of vessels, both designed to maximize the movement of plasma proteins and leukocytes out of the circulation and into the site of infection or injury. The escape of fluid, proteins, and blood cells from the vascular system into the interstitial tissue or body cavities is known as *exudation* (Fig. 3.2). An *exudate* is an extravascular fluid that has a high protein concentration and may contain cellular debris or, occasionally, some blood cells. Its presence implies the existence of an inflammatory process that has increased the permeability of small blood vessels. In contrast, a *transudate* is a fluid with low protein content (most of which is albumin) and little or no cellular material. It is essentially an ultrafiltrate of blood plasma that is produced as a result of osmotic or hydrostatic imbalance across the vessel wall without an increase in vascular permeability (Chapter 4). Edema denotes an excess of fluid in the interstitial tissue or serous cavities; it can be either an exudate or a transudate. Pus, a purulent exudate, is an inflammatory exudate rich in leukocytes (mostly neutrophils), the debris of dead cells, and, in many cases, microbes.

Changes in Vascular Flow and Caliber

Changes in vascular flow and caliber begin early after injury and consist of:

- **Vasodilation** is induced by the action of several mediators, notably histamine, on vascular smooth muscle. It is one of the earliest manifestations of acute inflammation. Vasodilation involves mainly postcapillary venules and results in increased blood flow, which is the cause of heat and redness (erythema) at the site of inflammation.
- Vasodilation is quickly followed by *increased permeability of the microvasculature*, with the outpouring of protein-rich fluid into the extravascular tissues; this process is described in detail later.
- The loss of fluid and increased vessel diameter lead to slower blood flow (*stasis*) and increased concentration of red cells in small vessels. These changes result in engorgement of small vessels with slowly moving red cells, which causes redness of the inflamed area and is seen histologically as vascular congestion.
- As stasis develops, blood leukocytes, principally neutrophils, accumulate along the endothelium lining the vessels. At the same time, endothelial cells are activated by mediators produced at sites of infection and tissue damage and express increased levels of adhesion molecules. Leukocytes then adhere to the endothelium, and soon afterward they migrate through the vascular wall into the interstitial tissue in a sequence that is described later.

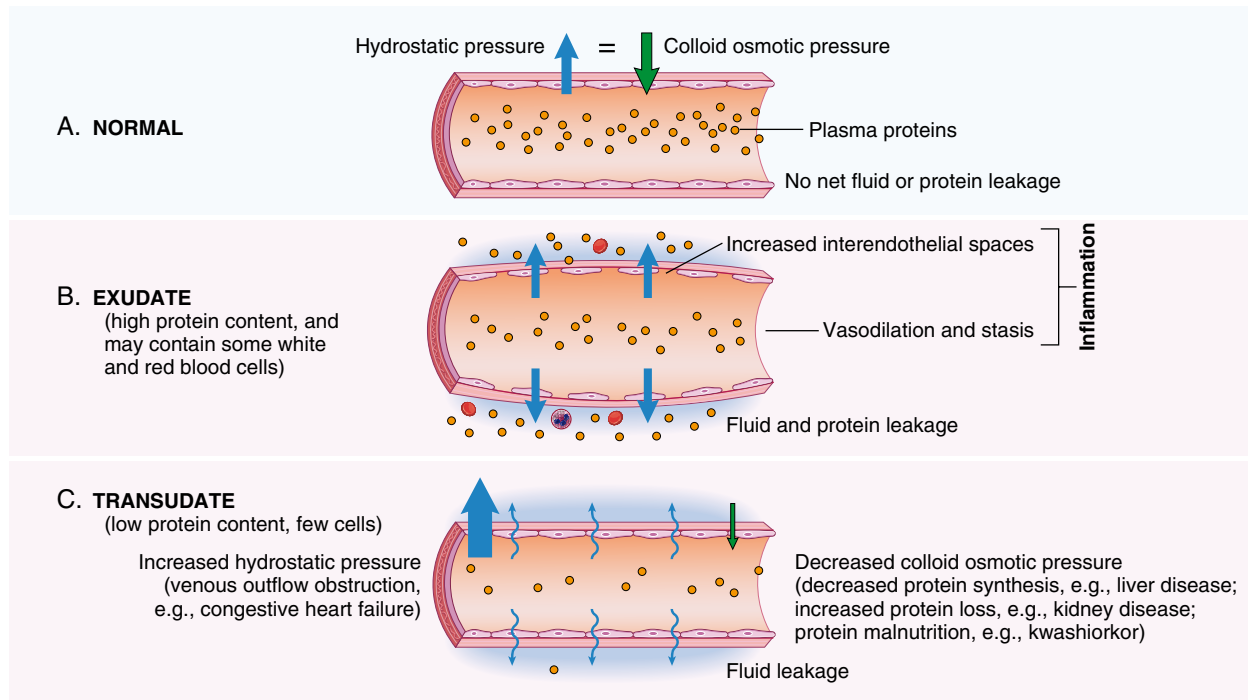


Fig. 3.2 Formation of exudates and transudates. (A) Normal hydrostatic pressure (blue arrow) is about 32 mm Hg at the arterial end of a capillary bed and 12 mm Hg at the venous end; the mean colloid osmotic pressure of tissues is approximately 25 mm Hg (green arrow), which is equal to the mean capillary pressure. Therefore, the net flow of fluid across the vascular bed is almost nil. (B) An exudate is formed in inflammation because vascular permeability increases due to increased interendothelial spaces. (C) A transudate is formed when fluid leaks out because of increased hydrostatic pressure or decreased osmotic pressure.

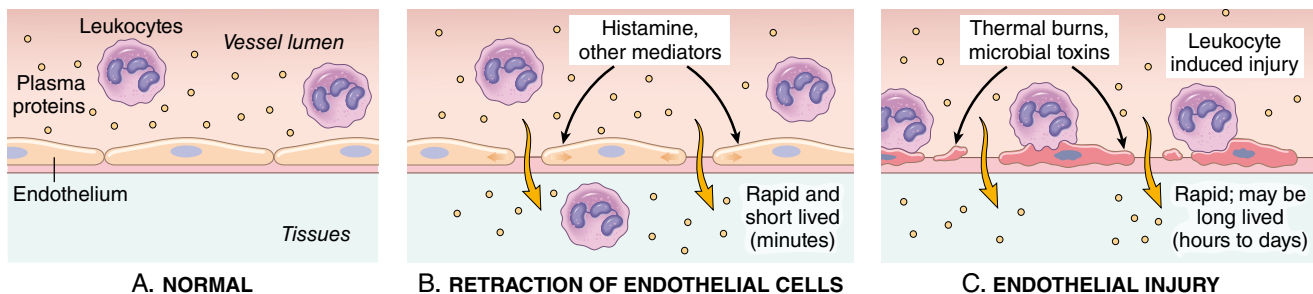


Fig. 3.3 Principal mechanisms of increased vascular permeability in inflammation. Vascular permeability is increased by chemically induced retraction of endothelial cells (B) or endothelial injury (C) compared with normal endothelium (A).

Increased Vascular Permeability (Vascular Leakage)

Several mechanisms are responsible for the increased permeability of postcapillary venules, a hallmark of acute inflammation (Fig. 3.3).

- *Contraction of endothelial cells* resulting in opening of interendothelial gaps is the most common mechanism of vascular leakage. It is elicited by histamine, bradykinin, leukotrienes, and other chemical mediators. It is called the *immediate transient response* because it occurs rapidly after exposure to the mediators and is usually short lived (15 to 30 minutes). In some forms of mild injury (e.g., after burns, irradiation or ultraviolet radiation, and exposure to certain bacterial toxins), vascular leakage begins after a delay of 2 to 12 hours and lasts for several hours or even days; this *delayed prolonged leakage* may be caused by contraction of endothelial cells or mild endothelial damage. Sunburn is a classic example of damage that results in late-appearing vascular leakage. Often the immediate and delayed responses occur along a continuum.

- *Endothelial injury* resulting in endothelial cell necrosis and detachment. Direct damage to the endothelium is encountered in severe physical injuries, for example, in thermal burns, or is induced by the actions of microbes and microbial toxins that damage endothelial cells. Neutrophils that adhere to the endothelium during inflammation may also injure endothelial cells and thus amplify the reaction. In most instances, leakage starts immediately after injury and is sustained for several hours until the damaged vessels are thrombosed or repaired.

Although these mechanisms of increased vascular permeability are described separately, all probably contribute in varying degrees to responses to most stimuli. For example, at different stages of a thermal burn, leakage results from chemically mediated endothelial contraction and direct and leukocyte-dependent endothelial injury. The vascular leakage induced by these mechanisms can cause life-threatening loss of fluid in severely burned patients.

Responses of Lymphatic Vessels and Lymph Nodes

In addition to blood vessels, lymphatic vessels also participate in acute inflammation. The system of lymphatics and lymph nodes filters the extravascular fluids and clears microbes and other foreign substances. Lymphatics drain the small amount of extravascular fluid that seeps out of capillaries in the healthy state. In inflammation, lymph flow is increased and helps drain edema fluid that accumulates because of increased vascular permeability. In addition to fluid, leukocytes and cell debris, as well as microbes, may find their way into lymph. Lymphatic vessels, like blood vessels, proliferate during inflammatory reactions to handle the increased load. The lymphatics may become secondarily inflamed (*lymphangitis*), as may the draining lymph nodes (*lymphadenitis*). Inflamed lymph nodes are often enlarged because of hyperplasia of the lymphoid follicles and increased numbers of lymphocytes and macrophages. This constellation of pathologic changes is termed *reactive*, or *inflammatory*, *lymphadenitis* (Chapter 13). The presence of red streaks near a skin wound is a telltale sign of bacterial infection. The streaks represent inflamed lymphatic channels and are diagnostic of lymphangitis; it may be accompanied by painful enlargement of the draining lymph nodes, indicating lymphadenitis.

KEY CONCEPTS

Vascular Reactions in Acute Inflammation

- Vasodilation is induced by chemical mediators such as histamine (described later) resulting in erythema and increased blood flow.
- Increased vascular permeability is induced by histamine, kinins, and other mediators that produce gaps between endothelial cells and by direct or leukocyte-induced endothelial injury. It allows plasma proteins and leukocytes, the mediators of host defense, to enter sites of infection or tissue damage. Fluid leak from blood vessels results in edema.
- Lymphatic vessels and lymph nodes are also involved in inflammation and often show redness and swelling.

Leukocyte Recruitment to Sites of Inflammation

The changes in blood flow and vascular permeability are quickly followed by an influx of leukocytes into the tissue. These leukocytes perform the key function of eliminating the offending agents. The most important leukocytes in typical inflammatory reactions are the ones capable of phagocytosis, namely neutrophils and macrophages. They ingest and destroy bacteria and other microbes, as well as necrotic tissue and foreign substances. Macrophages also produce growth factors that aid in repair. A price that is paid for the defensive potency of leukocytes is that, when activated, they may cause tissue damage because the leukocyte products that destroy microbes and help “clean up” necrotic tissues can also injure normal host tissues.

The journey of leukocytes from the vessel lumen to the tissue is a multistep process that is mediated and controlled by adhesion molecules and cytokines. This process can be divided into sequential phases (Fig. 3.4):

1. In the vessel lumen: *margination, rolling, and adhesion to endothelium*. Vascular endothelium in its normal state does not bind circulating cells or allow their passage through the vessel wall. In inflammation, the endothelium is activated and can bind leukocytes as a prelude to their exit from blood vessels.
2. *Migration across the endothelium* and vessel wall.
3. *Migration in the tissues* toward a chemotactic stimulus.

Leukocyte Adhesion to Endothelium

In normally flowing blood in venules, red cells are confined to a central axial column, displacing the leukocytes toward the wall of the vessel. Because of

dilation of inflamed postcapillary venules, blood flow slows (stasis) and more white cells assume a peripheral position along the endothelial surface. This process of leukocyte redistribution is called *margination*. The slowed leukocytes sense signals from the endothelium, resulting first in the cells rolling on the vessel wall and then recognizing adhesion molecules expressed on the endothelium that lead to the cells adhering firmly (resembling pebbles over which a stream runs without disturbing them).

The attachment of leukocytes to endothelial cells is mediated by adhesion molecules whose expression is enhanced by cytokines. The cytokines are secreted by sentinel cells in tissues in response to microbes and other injurious agents, thus ensuring that leukocytes are recruited to the tissues where these stimuli are present. The major families of proteins involved in leukocyte adhesion and migration are the selectins and integrins and their ligands (Table 3.3). They are expressed on leukocytes and endothelial cells.

- **Selectins.** The initial rolling interactions are mediated by selectins, of which there are three types: one expressed on leukocytes (L-selectin), one on endothelium (E-selectin), and one in platelets and on endothelium (P-selectin) (see Table 3.3). The ligands for selectins are sialylated oligosaccharides on mucin-like glycoproteins. The expression of selectins and their ligands is regulated by cytokines produced in response to infection and injury. Tissue macrophages, mast cells, and endothelial cells that encounter microbes and dead tissues respond by secreting several cytokines including tumor necrosis factor (TNF), interleukin-1 (IL-1), and chemokines (*chemoattractant cytokines*). (Cytokines are described in more detail later and in Chapter 6.) TNF and IL-1 act on the endothelial cells of postcapillary venules adjacent to the infection and induce the coordinate expression of numerous adhesion molecules. Within 1 to 2 hours the endothelial cells begin to express E-selectin and the ligands for L-selectin. Other mediators such as histamine and thrombin, described later, stimulate the redistribution of P-selectin from its normal intracellular stores in endothelial cell granules (called Weibel-Palade bodies) to the cell surface. Leukocytes express L-selectin at the tips of their microvilli, and ligands for E-selectin and P-selectin, all of which bind to the complementary molecules on the endothelial cells. These are low-affinity interactions with a fast off-rate, so they are easily disrupted by the flowing blood. As a result, the leukocytes bind, detach, and bind again and thus begin to roll along the endothelial surface.
- **Integrins.** The weak rolling interactions slow down the leukocytes and give them the opportunity to bind more firmly to the endothelium. Firm adhesion is mediated by a family of heterodimeric leukocyte surface proteins called *integrins* (see Table 3.3). TNF and IL-1 induce endothelial expression of ligands for integrins, mainly vascular cell adhesion molecule 1 (VCAM-1), the ligand for the $\beta 1$ integrin VLA-4, and intercellular adhesion molecule 1 (ICAM-1), the ligand for the $\beta 2$ integrins LFA-1 and MAC-1. Leukocytes normally express integrins in a low-affinity state. Chemokines that were produced at the site of injury bind to endothelial cell proteoglycans and are displayed at high concentrations on the endothelial surface. These chemokines bind to and activate the rolling leukocytes. One of the consequences of activation is the conversion of VLA-4 and LFA-1 integrins on the leukocytes to a high-affinity state. The combination of cytokine-induced expression of integrin ligands on the endothelium and increased integrin affinity on the leukocytes results in firm integrin-mediated adhesion of the leukocytes to the endothelium at the site of inflammation. The leukocytes stop rolling, their cytoskeleton is reorganized, and they spread out on the endothelial surface.

Leukocyte Migration Through Endothelium

The next step in the process of leukocyte recruitment is migration of the leukocytes through intact endothelium, called *transmigration*

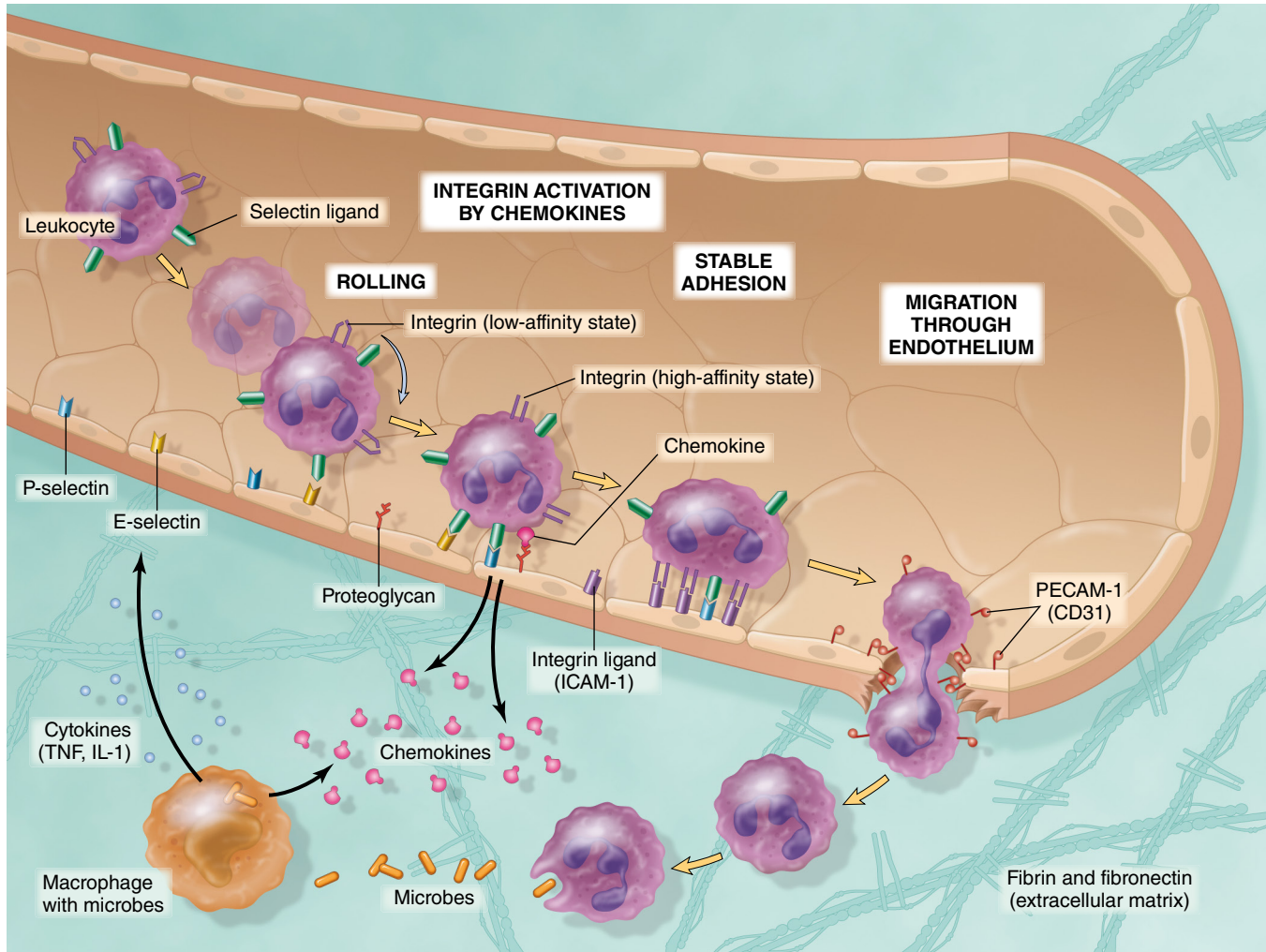


Fig. 3.4 The multistep process of leukocyte migration through blood vessels, shown here for neutrophils. The leukocytes first roll, then become activated and adhere to endothelium, then transmigrate across the endothelium, pierce the basement membrane, and migrate toward chemoattractants emanating from the source of injury. Different molecules play predominant roles in different steps of this process: selectins, in rolling; chemokines (usually displayed bound to proteoglycans), in activating the neutrophils to increase the avidity of integrins; integrins, in firm adhesion; and CD31 (PECAM-1), in transmigration. *ICAM-1*, Intercellular adhesion molecule 1; *IL-1*, interleukin-1; *PECAM-1*, platelet endothelial cell adhesion molecule (also known as CD31); *TNF*, tumor necrosis factor.

or diapedesis. Transmigration of leukocytes occurs mainly in postcapillary venules (the site of increased vascular permeability). Chemokines act on the adherent leukocytes and stimulate the cells to migrate through interendothelial gaps toward the chemical concentration gradient, that is, toward the site of injury or infection where the chemokines are being produced. Several adhesion molecules present in the intercellular junctions between endothelial cells are involved in the migration of leukocytes. These molecules include a member of the immunoglobulin superfamily called *CD31* or *PECAM-1* (platelet endothelial cell adhesion molecule). After traversing the endothelium, leukocytes pierce the basement membrane, probably by secreting collagenases, and enter the extravascular space. After leukocytes pass through, the basement membranes are repaired and become continuous again. The cells that have exited the vessel then migrate toward the chemotactic gradient created by chemokines and other chemoattractants and accumulate in the extravascular site.

The most telling proof of the importance of leukocyte adhesion molecules in the host inflammatory response are genetic deficiencies in these

molecules, which result in increased susceptibility to bacterial infections. These leukocyte adhesion deficiencies are described in Chapter 6.

Chemotaxis of Leukocytes

After exiting the circulation, leukocytes move in the tissues toward the site of injury by a process called *chemotaxis*, which is defined as locomotion along a chemical gradient. Both exogenous and endogenous substances act as chemoattractants. The most common exogenous factors are bacterial products, including peptides with *N*-formylmethionine terminal amino acids and some lipids. Endogenous chemoattractants include several chemical mediators (described later): (1) cytokines, particularly those of the chemokine family (e.g., *IL-8*); (2) components of the complement system, particularly *C5a*; and (3) arachidonic acid (*AA*) metabolites, mainly leukotriene *B₄* (*LTB₄*). All these chemotactic agents bind to specific seven-transmembrane G protein–coupled receptors on the surface of leukocytes. Signals initiated from these receptors result in activation of second messengers that induce polymerization of actin at the leading edge of the cell, and

TABLE 3.3 Endothelial and Leukocyte Adhesion Molecules

Family	Molecule	Distribution	Ligand
Selectin	L-selectin (CD62L)	Neutrophils, monocytes T cells (naïve and central memory) B cells (naïve)	Sialyl-Lewis X/PNAd on various glycoproteins (GlyCAM-1, CD34, MAdCAM-1, others); expressed on endothelium
	E-selectin (CD62E)	Endothelium activated by cytokines (TNF, IL-1)	Sialyl-Lewis X (e.g., CLA) on glycoproteins; expressed on neutrophils, monocytes, T cells (effector, memory)
	P-selectin (CD62P)	Endothelium activated by cytokines (TNF, IL-1), histamine, or thrombin	Sialyl-Lewis X on PSGL-1 and other glycoproteins; expressed on neutrophils, monocytes, T cells (effector, memory)
Integrin	LFA-1 (CD11aCD18)	Neutrophils, monocytes, T cells (naïve, effector, memory)	ICAM-1 (CD54), ICAM-2 (CD102); expressed on endothelium (upregulated on activated endothelium)
	MAC-1 (CD11bCD18)	Monocytes, DCs	ICAM-1 (CD54), ICAM-2 (CD102); expressed on endothelium (upregulated on activated endothelium)
	VLA-4 (CD49dCD29)	Monocytes T cells (naïve, effector, memory)	VCAM-1 (CD106); expressed on endothelium (upregulated on activated endothelium)
	$\alpha 4\beta 7$	Monocytes T cells (gut homing naïve, effector, memory)	VCAM-1 (CD106), MAdCAM-1; expressed on endothelium in gut and gut-associated lymphoid tissues
Ig	CD31	Endothelial cells, leukocytes	CD31 (homotypic interaction)

CLA, Cutaneous lymphocyte antigen-1; GlyCAM-1, glycan-bearing cell adhesion molecule-1; HEV, high endothelial venule; Ig, immunoglobulin; IL-1, interleukin-1; ICAM, intercellular adhesion molecule; LFA-1, leukocyte function associated antigen-1; MAdCAM-1, mucosal addressin cell adhesion molecule-1; PNAd, peripheral node addressin; PSGL-1, P-selectin glycoprotein ligand-1; TNF, tumor necrosis factor; VCAM, vascular cell adhesion molecule; VLA-4, very late antigen-4.

localization of myosin filaments at the back. This reorganization of the cytoskeleton allows the leading edge of the leukocyte to extend filopodia that pull the back of the cell in the direction of extension, much as an automobile with front-wheel drive is pulled by the wheels in front. The net result is that leukocytes migrate in the direction of the chemoattractants that are produced at the site of infection or tissue injury.

The nature of the leukocyte infiltrate varies with the age of the inflammatory response and the type of stimulus. In most forms of acute inflammation, neutrophils predominate in the inflammatory infiltrate during the first 6 to 24 hours and are replaced by monocytes in 24 to 48 hours (Fig. 3.5). There are several reasons for the early preponderance of neutrophils: they are more numerous than are other leukocytes, respond more rapidly to chemokines, and may attach more firmly to the adhesion molecules that are rapidly induced on endothelial cells such as P-selectin and E-selectin. After entering tissues, neutrophils are short lived; most neutrophils in extravascular tissues undergo apoptosis

within a few days. Macrophages, which develop from monocytes, not only survive longer but may also proliferate in the tissues, and thus they become the dominant population in prolonged inflammatory reactions (e.g., after 48 hours). There are, however, exceptions to this stereotypic pattern of cellular infiltration. In certain infections—for example, by *Pseudomonas* bacteria—the cellular infiltrate is dominated by continuously recruited neutrophils for several days; in viral infections, lymphocytes may be the first cells to arrive; some hypersensitivity reactions are dominated by activated lymphocytes, macrophages, and plasma cells (reflecting the immune response); and in helminthic infections and allergic reactions, eosinophils may be the main cell type.

The molecular understanding of leukocyte recruitment and migration has led to development of drugs for controlling harmful inflammation, including agents that block TNF (discussed later), and antagonists of leukocyte integrins. Predictably, these antagonists not only have the desired effect of controlling the inflammation but can also compromise

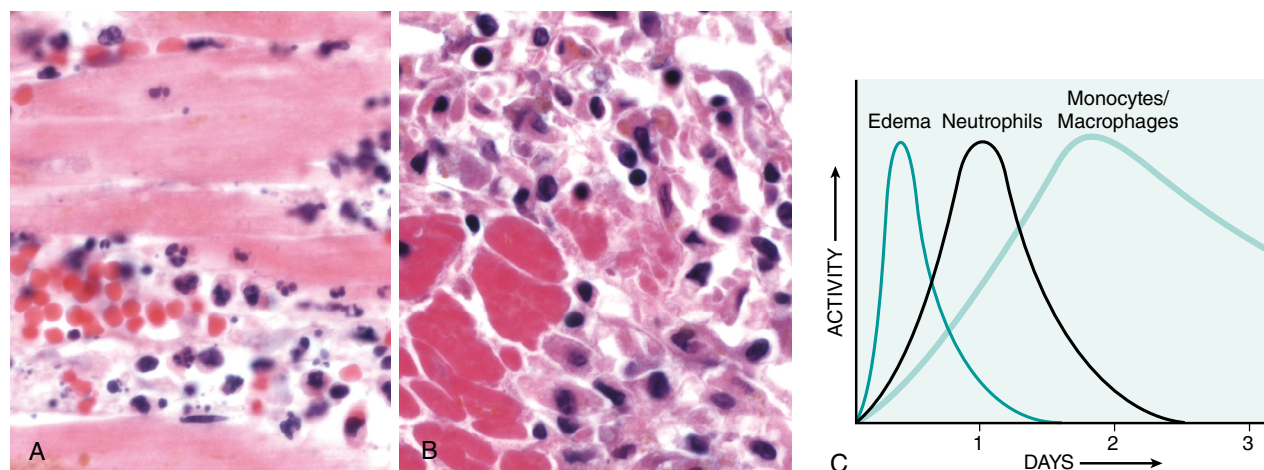


Fig. 3.5 Nature of leukocyte infiltrates in inflammatory reactions. The photomicrographs show an inflammatory reaction in the myocardium after ischemic necrosis (infarction). (A) Early (neutrophilic) infiltrates and congested blood vessels. (B) Later (mononuclear) cellular infiltrates. (C) The approximate kinetics of edema and cellular infiltration. For simplicity, edema is shown as an acute transient response, although secondary waves of delayed edema and neutrophil infiltration can also occur.

the ability of treated patients to defend themselves against microbes, which, of course, is the physiologic function of the inflammatory response.

KEY CONCEPTS

Leukocyte Recruitment to Sites of Inflammation

- Leukocytes are recruited from the blood into the extravascular tissue, where infectious pathogens or damaged tissues may be located, migrate to the site of infection or tissue injury, and are activated to perform their functions.
- Leukocyte recruitment is a multistep process consisting of loose attachment to and rolling on endothelium (mediated by selectins), firm attachment to endothelium (mediated by integrins), and migration through interendothelial spaces.
- Various cytokines promote expression of selectins and integrin ligands on endothelium (e.g., TNF, IL-1), increase the avidity of integrins for their ligands (e.g., chemokines), and promote directional migration of leukocytes (also chemokines). Many cytokines are produced by tissue macrophages and other cells responding to the pathogens or damaged tissues.
- Neutrophils predominate in the early inflammatory infiltrate and are later replaced by monocytes, which differentiate into macrophages in the tissues.

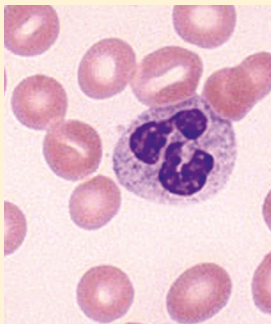
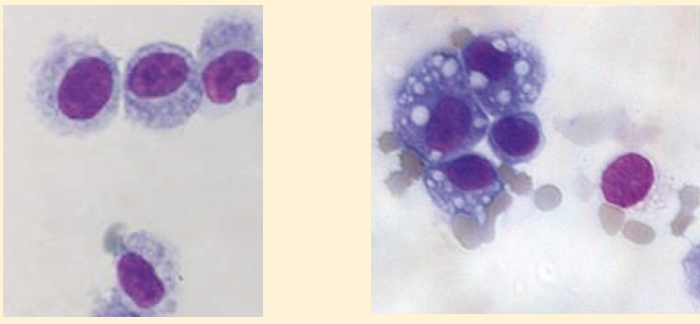
Once leukocytes (particularly neutrophils and monocytes) are recruited to a site of infection or cell death, they must be activated to perform their functions. The responses of these leukocytes consist of recognition of the offending agents by TLRs and other receptors, described earlier, which deliver signals that activate the leukocytes to phagocytose and destroy the offending agents.

Phagocytosis and Clearance of the Offending Agent

The two major phagocytes are neutrophils and macrophages. Although these cell types share many functional properties, they also differ in significant ways (Table 3.4).

Recognition of microbes or dead cells induces several responses in leukocytes that are collectively called *leukocyte activation* (Fig. 3.6). Activation results from signaling pathways that are triggered in leukocytes by various cellular receptors and cytokines. The functional responses that are most important for destruction of microbes and other offenders are phagocytosis and intracellular killing. Several other responses aid in the defensive functions of inflammation and may contribute to its injurious consequences.

TABLE 3.4 Properties of Neutrophils and Macrophages

	Neutrophils	Macrophages
		
	Blood neutrophil	Blood monocytes Macrophages
Origin	HSCs in bone marrow	HSCs in bone marrow (in inflammatory reactions) Many tissue-resident macrophages: stem cells in yolk sac or fetal liver (early in development)
Lifespan in tissues	Several days	Macrophages recruited from blood: days or weeks Tissue-resident macrophages: years
Responses to activating stimuli	Rapid, short lived, mostly degranulation and enzymatic activity	More prolonged, slower, often dependent on new gene transcription
Reactive oxygen species	Rapidly induced by assembly of phagocyte oxidase (respiratory burst)	Less prominent
Nitric oxide	Low levels or none	Produced by transcriptionally activated iNOS
Degranulation	Major response; induced by cytoskeletal rearrangement	Not prominent
Cytokine production	Low levels or none	Major functional activity; requires transcriptional activation of cytokine genes
NET formation	Rapidly induced, by extrusion of nuclear contents	No
Secretion of lysosomal enzymes	Prominent	Less

HSC, Hematopoietic stem cells; iNOS, inducible nitric oxide synthase; NET, neutrophil extracellular trap.

This table lists the major differences between neutrophils and macrophages. The reactions summarized above are described in the text. Note that the two cell types share many features such as phagocytosis, ability to migrate through blood vessels into tissues, and chemotaxis.

Images from Forbes B, O'Lone R, Pribul A, et al. Challenges for inhaled drug discovery and development: induced alveolar macrophage responses. *Adv Drug Deliv Rev.* 2014;71:15-33.

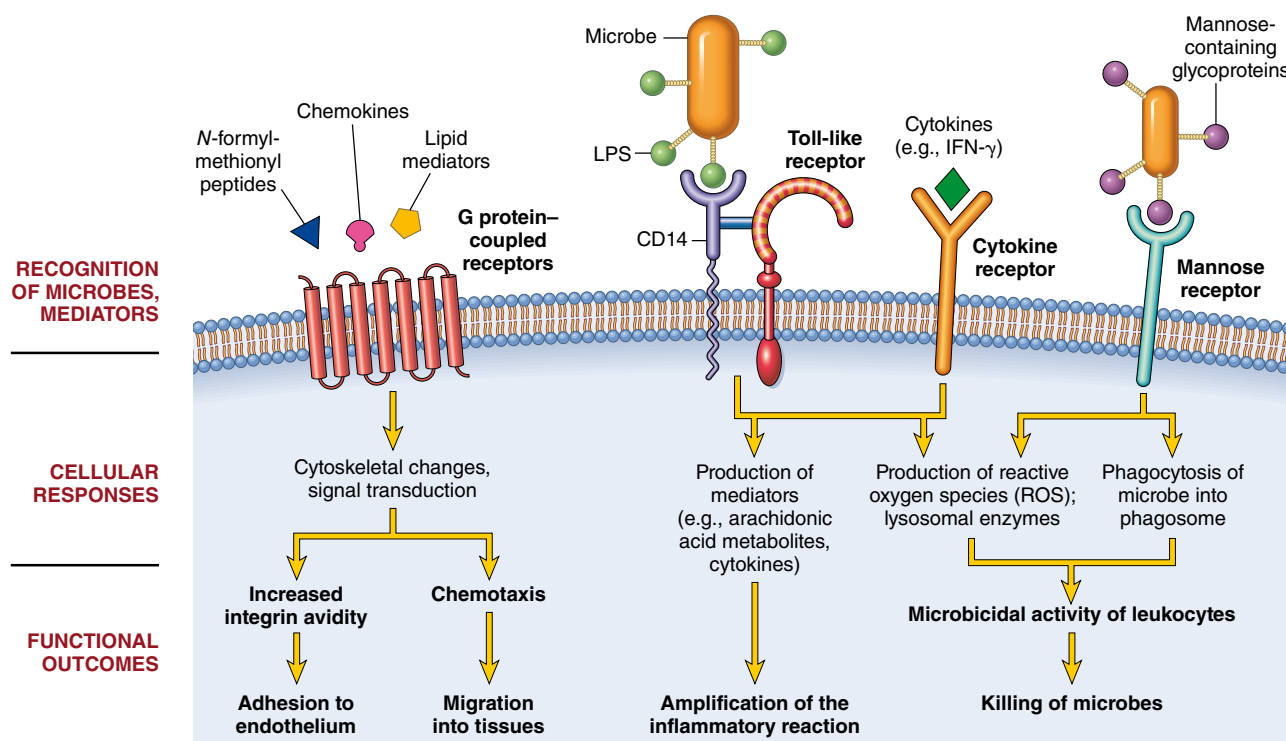


Fig. 3.6 Leukocyte activation. Different classes of cell surface receptors of leukocytes recognize different stimuli. The receptors initiate responses that mediate the functions of the leukocytes. Only some receptors are depicted (see text for details). Lipopolysaccharide (LPS) first binds to a circulating LPS-binding protein (not shown). IFN- γ , Interferon- γ .

Phagocytosis

Phagocytosis involves sequential steps (Fig. 3.7):

1. Recognition and attachment of the particle to be ingested by the leukocyte
2. Engulfment, with subsequent formation of a phagocytic vacuole
3. Killing of the microbe and degradation of the ingested material

Phagocytic Receptors. Mannose receptors, scavenger receptors, and receptors for various opsonins enable phagocytes to bind and ingest microbes. The macrophage *mannose receptor* is a lectin that binds terminal mannose and fucose residues of glycoproteins and glycolipids. These sugars are typically part of molecules found on microbial cell walls, whereas in mammals, glycoproteins and glycolipids are modified to contain terminal sialic acid or N-acetylgalactosamine. Therefore, the mannose receptor recognizes microbes and not host cells. *Scavenger receptors* were originally defined as molecules that bind and mediate endocytosis of oxidized or acetylated low-density lipoprotein (LDL) particles that do not interact with the conventional LDL receptor. Macrophage scavenger receptors bind a variety of microbes in addition to modified LDL particles. Macrophage integrins, notably MAC-1 (CD11b/CD18), may also bind microbes for phagocytosis. The efficiency of phagocytosis is greatly enhanced when microbes are coated with molecules called *opsonins* for which the phagocytes express high-affinity receptors. The major opsonins are immunoglobulin G (IgG) antibodies, the C3b breakdown product of complement, and certain plasma lectins, notably mannose-binding lectin and collectins, all of which are recognized by specific receptors on leukocytes.

Engulfment. After a particle is bound to phagocyte receptors, extensions of the cytoplasm flow around it, and the plasma membrane pinches off to form an intracellular vesicle (phagosome) that encloses

the particle. The phagosome then fuses with a lysosomal granule, which discharges its contents into the phagolysosome (see Fig. 3.7). During this process, the phagocyte may also release lysosomal contents into the extracellular space.

The process of phagocytosis is complex and involves the integration of many receptor-initiated signals that lead to membrane remodeling and cytoskeletal changes, particularly the polymerization of actin filaments.

Intracellular Destruction of Microbes and Debris

Killing of microbes is accomplished by reactive oxygen species (ROS), also called reactive oxygen intermediates, reactive nitrogen species, derived from nitric oxide (NO), and lysosomal enzymes, all of which degrade phagocytosed materials (see Fig. 3.7). This is the final step in the elimination of infectious agents and necrotic cells. The killing and digestion of microbes and dead cell debris within neutrophils and macrophages occur most efficiently after activation of the phagocytes. All these killing mechanisms are normally sequestered in lysosomes, to which phagocytosed materials are brought. Thus, potentially lethal molecules are segregated from the cell's cytoplasm and nucleus to avoid damage to the phagocyte while it is performing its normal function.

Reactive Oxygen Species. Reactive oxygen species (ROS) are produced by the rapid assembly and activation of a multicomponent enzyme called *phagocyte oxidase* (NADPH oxidase), which oxidizes reduced nicotinamide-adenine dinucleotide phosphate (NADPH) and, in the process, converts oxygen to superoxide anion ($O_2^{\cdot -}$), a major ROS. In neutrophils, this oxidative reaction is accompanied by oxygen consumption and is called the *respiratory burst*. Phagocyte oxidase is an enzyme complex consisting of at least seven proteins. In resting neutrophils, different components of the enzyme are located in the plasma membrane and the

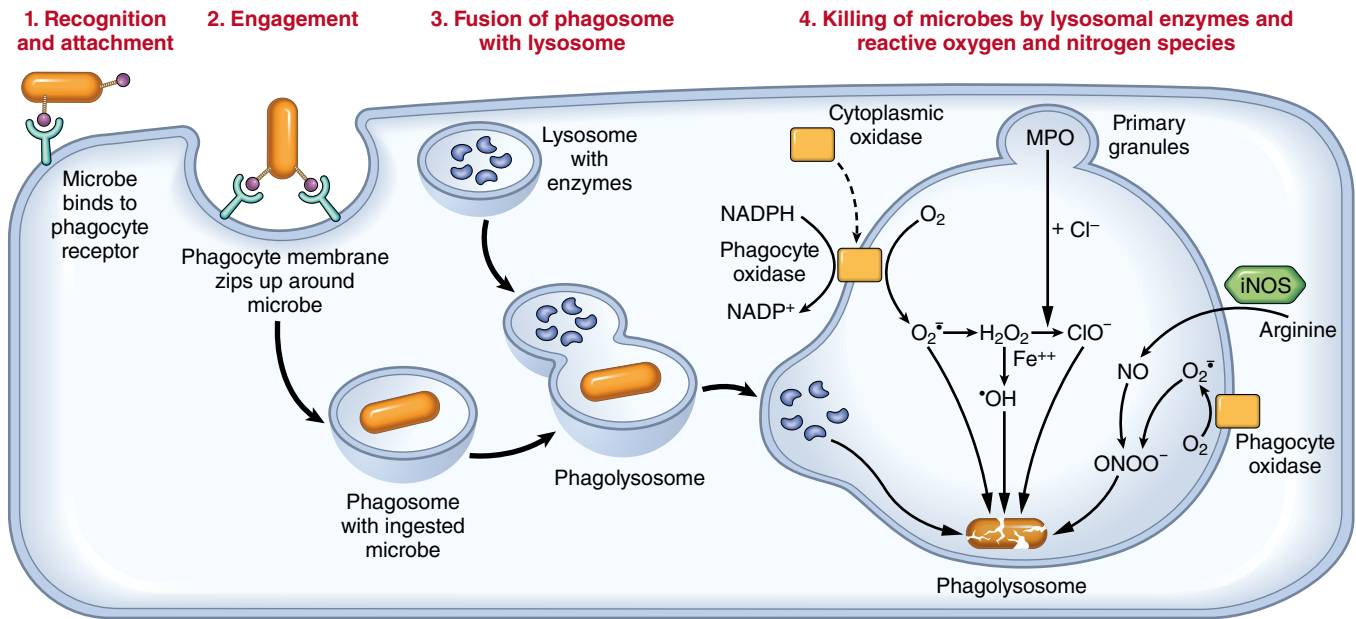


Fig. 3.7 Phagocytosis and intracellular destruction of microbes. Phagocytosis of a particle (e.g., a bacterium) involves its recognition by receptors on the leukocyte membrane and engulfment. This is followed by fusion of the phagocytic vacuoles with lysosomes and destruction of ingested particles within the phagolysosomes by lysosomal enzymes and by microbicidal reactive oxygen and nitrogen species. In activated phagocytes, cytoplasmic components of the phagocyte oxidase enzyme assemble in the membrane of the phagosome to form the active enzyme, which catalyzes the conversion of oxygen into superoxide (O_2^-) and hydrogen peroxide (H_2O_2). MPO, present in the granules of neutrophils, converts H_2O_2 to hypochlorite (ClO^-). Peroxynitrite ($OONO^-$) is generated from nitric oxide (NO) by the enzyme iNOS. During phagocytosis, granule contents may be released into extracellular tissues (not shown). *iNOS*, Inducible nitric oxide synthase; *MPO*, myeloperoxidase; *NADP*, nicotinamide-adenine dinucleotide phosphate.

cytoplasm. In response to activating stimuli, the cytosolic protein components translocate to the phagosomal membrane, where they assemble and form the functional enzyme complex. Thus, the ROS are produced within the phagolysosome, where they can act on ingested particles without damaging the host cell. O_2^- is converted into hydrogen peroxide (H_2O_2), mostly by spontaneous dismutation. H_2O_2 is not able to efficiently kill microbes by itself. However, the azurophilic granules of neutrophils contain the enzyme myeloperoxidase (MPO), which, in the presence of a halide such as Cl^- , converts H_2O_2 to hypochlorite (ClO^-), the active ingredient in household bleach. The latter is a potent antimicrobial agent that destroys microbes by halogenation (in which the halide binds covalently to cellular constituents) or by oxidation of proteins and lipids (lipid peroxidation). The H_2O_2 -MPO-halide system is the most potent bactericidal system of neutrophils. Nevertheless, inherited deficiency of MPO by itself leads to minimal increase in susceptibility to infection, emphasizing the redundancy of microbicidal mechanisms in leukocytes. H_2O_2 is also converted to hydroxyl radical ($\cdot OH$), another powerful destructive agent. As discussed in Chapter 2, these oxygen-derived free radicals bind to and modify cellular lipids, proteins, and nucleic acids and thus destroy cells such as microbes.

Oxygen-derived radicals may be released extracellularly from leukocytes after exposure to microbes, chemokines, and antigen-antibody complexes or following a phagocytic challenge. These ROS are implicated in tissue damage accompanying inflammation.

Plasma, tissue fluids, and host cells possess antioxidant mechanisms that protect healthy cells from these potentially injurious oxygen-derived radicals. These antioxidants are discussed in Chapter 2 and include (1) the enzyme superoxide dismutase, which is found in, or can be activated in, a variety of cell types; (2) the enzyme catalase, which detoxifies H_2O_2 ; (3) glutathione peroxidase, another powerful

H_2O_2 detoxifier; (4) the copper-containing plasma protein ceruloplasmin; and (5) the iron-free fraction of plasma transferrin.

Inherited deficiencies of components of phagocyte oxidase cause an immunodeficiency disease called *chronic granulomatous disease* (CGD), which is discussed in Chapter 6.

Nitric Oxide. NO, a soluble gas produced from arginine by the action of nitric oxide synthase (NOS), also participates in microbial killing. There are three different types of NOS: endothelial (eNOS), neuronal (nNOS), and inducible (iNOS). eNOS and nNOS are constitutively expressed at low levels, and the NO they generate functions to maintain vascular tone and as a neurotransmitter, respectively. iNOS, the type that is involved in microbial killing, is induced when macrophages (and, to a lesser extent, neutrophils) are activated by cytokines (e.g., interferon- γ [IFN- γ]) or microbial products. In macrophages, NO reacts with superoxide (O_2^-) to generate the highly reactive free radical peroxynitrite ($ONOO^-$). These nitrogen-derived free radicals, similar to ROS, attack and damage the lipids, proteins, and nucleic acids of microbes (Chapter 2). Reactive oxygen and nitrogen species have overlapping actions, as shown by the observation that knockout mice lacking either phagocyte oxidase or iNOS are only mildly susceptible to infections, but mice lacking both succumb rapidly to disseminated infections by normally harmless commensal bacteria.

In addition to its role as a microbicidal substance, NO relaxes vascular smooth muscle and promotes vasodilation. It is not clear if this action of NO plays an important role in the vascular reactions of acute inflammation.

Lysosomal Enzymes and Other Lysosomal Proteins. Neutrophils and macrophages contain lysosomal granules filled with enzymes that contribute to microbial killing and, when released, may cause tissue damage.

Neutrophils have two main types of granules. The smaller specific (or secondary) granules contain lysozyme, collagenase, gelatinase, lactoferrin, plasminogen activator, histaminase, and alkaline phosphatase. The larger azurophil (or primary) granules contain MPO, bactericidal proteins (lysozyme, defensins), acid hydrolases, and a variety of neutral proteases (elastase, cathepsin G, nonspecific collagenases, proteinase 3). Both types of granules can fuse with phagocytic vacuoles containing engulfed material, or the granule contents can be released into the extracellular space during “frustrated phagocytosis” (discussed later).

Different granule enzymes serve different functions. Acid proteases degrade bacteria and debris within the phagolysosomes, which are acidified by membrane-bound proton pumps. Neutral proteases are capable of degrading various extracellular components such as collagen, basement membrane, fibrin, elastin, and cartilage, resulting in the tissue destruction that accompanies inflammatory processes. Neutral proteases can also cleave C3 and C5 complement proteins and release a kinin-like peptide from kininogen. The released components of complement and kinins act as mediators of acute inflammation (discussed later). Neutrophil elastase has been shown to degrade virulence factors of bacteria and thus combat bacterial infections. Macrophages also contain acid hydrolases, collagenase, elastase, phospholipase, and plasminogen activator.

Because of the destructive effects of lysosomal enzymes, the initial leukocytic infiltration, if unchecked, can damage host tissues. These harmful proteases, however, are normally controlled by a system of antiproteases in the serum and tissue fluids. Foremost among these is α_1 -antitrypsin, which is the major inhibitor of neutrophil elastase. A deficiency of these inhibitors may lead to sustained action of leukocyte proteases, as is the case in patients with α_1 -antitrypsin deficiency, who are at risk for emphysema due to destruction of elastic support fibers in the lung because of uncontrolled elastase activity (Chapter 15). α_2 -Macroglobulin is another antiprotease found in serum and various secretions.

Other microbicidal granule contents include defensins, cationic arginine-rich granule peptides that are toxic to microbes; cathelicidins,

antimicrobial proteins found in neutrophils and other cells; lysozyme, which hydrolyzes the muramic acid-*N*-acetylglucosamine bond found in the glycopeptide coat of all bacteria; lactoferrin, an iron-binding protein present in specific granules; and major basic protein, a cationic protein of eosinophils, which has limited bactericidal activity but is cytotoxic to many helminthic parasites.

Neutrophil Extracellular Traps

Neutrophil extracellular traps (NETs) are extracellular fibrillar networks that concentrate antimicrobial substances at sites of infection and trap microbes, helping to prevent their spread. They are produced by neutrophils in response to infectious pathogens (mainly bacteria and fungi) and inflammatory mediators (e.g., chemokines, cytokines [mainly interferons], complement proteins, and ROS). The extracellular traps consist of a viscous meshwork of nuclear chromatin that binds and concentrates granule proteins such as antimicrobial peptides and enzymes (Fig. 3.8). NET formation starts with ROS-dependent activation of an arginine deaminase that converts arginines to citrulline, leading to chromatin decondensation. Other enzymes that are produced in activated neutrophils, such as MPO and elastase, enter the nucleus and cause further chromatin decondensation, culminating in rupture of the nuclear envelope and release of chromatin. In this process, the nuclei of the neutrophils are lost, leading to death of the cells. NETs have also been detected in the blood during sepsis. NETs may play roles in host defense by providing high local concentrations of antimicrobial substances; by trapping large microbes such as fungi, which may be too large to be phagocytosed; and by preventing systemic dissemination of microbes. NETs may also be involved in some diseases. The nuclear chromatin in NETs, which includes histones and associated DNA, has been postulated to be a source of nuclear antigens in systemic autoimmune diseases, particularly lupus, in which individuals react against their own DNA and nucleoproteins (Chapter 6).

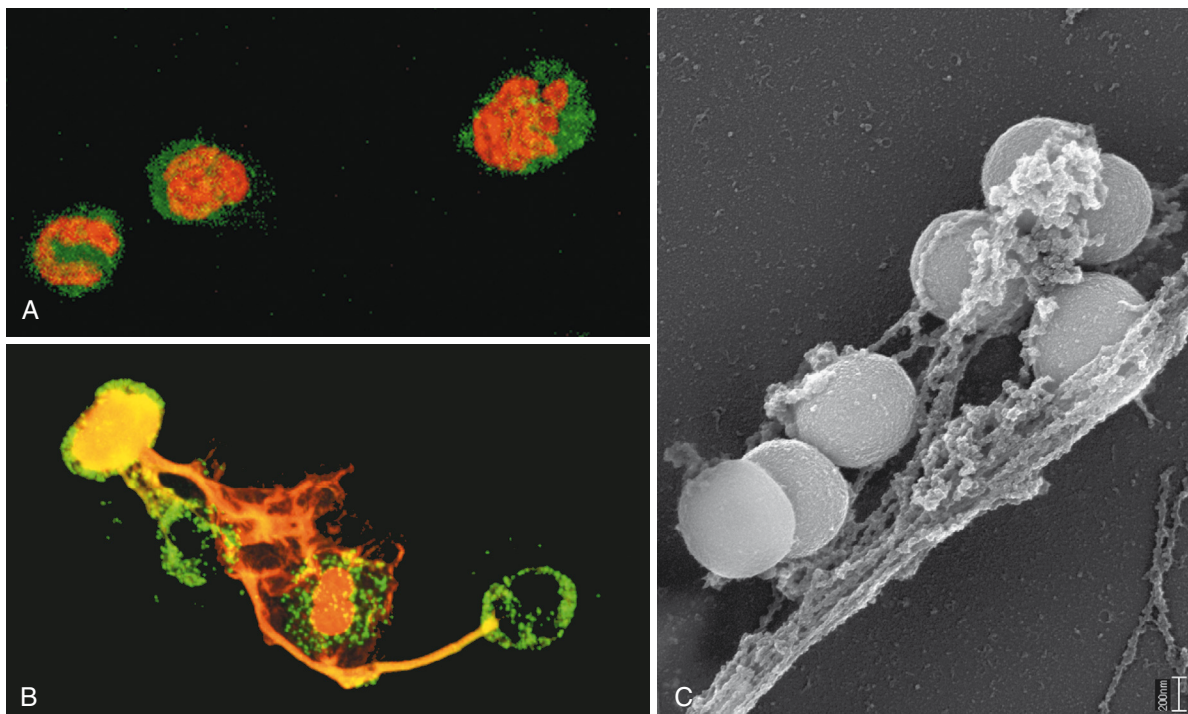


Fig. 3.8 Neutrophil extracellular traps (NETs). (A) Healthy neutrophils with nuclei stained red and cytoplasm stained green. (B) Release of nuclear material from neutrophils (note that two have lost their nuclei), forming extracellular traps. (C) Electron micrograph of bacteria (staphylococci) trapped in NETs. (From Brinkmann V, Zychlinsky A. Beneficial suicide: why neutrophils die to make NETs. *Nat Rev Microbiol.* 2007;5:577-582.)

Leukocyte-Mediated Tissue Injury

Leukocytes are important causes of injury to normal cells and tissues under several circumstances.

- As part of a normal defense reaction against pathogenic microbes, when adjacent tissues suffer collateral damage. In some infections that are difficult to eradicate, such as tuberculosis and certain viral diseases, the prolonged host response contributes more to the pathology than does the microbe itself.
- When the inflammatory response is inappropriately directed against host tissues, as in certain autoimmune diseases.
- When the host reacts excessively against usually harmless environmental substances, as in allergic diseases, including asthma.

In all these situations, **the mechanisms by which leukocytes damage normal tissues are the same as the mechanisms involved in antimicrobial defense** because once the leukocytes are activated, their effector mechanisms do not distinguish between offender and host. During activation and phagocytosis, neutrophils and macrophages produce microbicidal substances (ROS, NO, and lysosomal enzymes) within the phagolysosome; under some circumstances, these substances are also released into the extracellular space. These released substances are capable of damaging host cells and may thus amplify the effects of the initial injurious agent. If unchecked or inappropriately directed against host tissues, the leukocyte infiltrate itself becomes the offender, and indeed leukocyte-dependent inflammation and tissue injury underlie many acute and chronic human diseases (see Table 3.1). This fact becomes evident in the discussion of specific disorders throughout this book.

The contents of lysosomal granules are secreted by leukocytes into the extracellular milieu by several mechanisms. Controlled secretion of granule contents is a normal response of activated leukocytes. If phagocytes encounter materials that cannot be easily ingested, such as immune complexes deposited on large surfaces (e.g., glomerular basement membrane), the inability of the leukocytes to surround and ingest these substances (frustrated phagocytosis) triggers strong activation and the release of lysosomal enzymes into the extracellular environment. Some phagocytosed substances, such as urate crystals, may damage the membrane of the phagolysosome, also leading to the release of lysosomal granule contents.

Other Functional Responses of Activated Leukocytes

In addition to eliminating microbes and dead cells, activated leukocytes play several other roles in host defense. Importantly, these cells, especially macrophages, produce cytokines that can either amplify or limit inflammatory reactions, growth factors that stimulate the proliferation of endothelial cells and fibroblasts and the synthesis of collagen, and enzymes that remodel connective tissues. Because of these activities, macrophages are also critical cells of chronic inflammation and tissue repair after inflammation has subsided. These functions of macrophages are discussed later in the chapter.

In this discussion of acute inflammation, we emphasize the importance of neutrophils and macrophages. However, it has recently become clear that some T lymphocytes, which are cells of adaptive immunity, also contribute to acute inflammation. The most important of these cells are those that produce the cytokine IL-17 (so-called Th17 cells), discussed in more detail in Chapter 6. IL-17 induces the secretion of chemokines that recruit other leukocytes. In the absence of effective Th17 responses, individuals are susceptible to fungal and bacterial infections and tend to develop “cold abscesses,” particularly in the skin, that lack the classic features of acute inflammation, such as warmth and redness.

Termination of the Acute Inflammatory Response

Such a powerful system of host defense, with its inherent capacity to cause tissue injury, needs tight controls to minimize damage. In part, inflammation declines after the offending agents are removed simply because the

mediators of inflammation are produced for only as long as the stimulus persists, have short half-lives, and are degraded after their release. Neutrophils also have a short half-life in tissues and die by apoptosis within several hours or a few days after leaving the blood. In addition, as inflammation develops, the process itself triggers a variety of stop signals that actively terminate the reaction. These active termination mechanisms include a switch in the type of arachidonic acid metabolite produced, from proinflammatory leukotrienes to antiinflammatory lipoxins (described later), and the liberation of antiinflammatory cytokines, including transforming growth factor- β (TGF- β) and IL-10, from macrophages and other cells. Other control mechanisms that have been demonstrated experimentally include neural impulses (cholinergic discharge) that inhibit the production of TNF in macrophages.

KEY CONCEPTS

Leukocyte Activation and Removal of Offending Agents

- Leukocytes eliminate microbes and dead cells by phagocytosis, followed by their destruction in phagolysosomes.
- Destruction is caused by free radicals (ROS, NO) generated in activated leukocytes and lysosomal enzymes.
- Neutrophils can extrude their nuclear contents to form extracellular nets that trap and destroy microbes.
- Enzymes and ROS may be released into the extracellular environment.
- The mechanisms that function to eliminate microbes and dead cells (the physiologic role of inflammation) are also capable of damaging normal tissues (the pathologic consequences of inflammation).
- The acute inflammatory reaction subsides as the initiating stimulus is removed mainly because the cells and mediators of inflammation have short half-lives. Antiinflammatory substances may also contribute to the termination of the inflammatory response when no longer needed.

Mediators of Inflammation

Inflammatory mediators are the substances that initiate and regulate inflammatory reactions. Many mediators have been identified and targeted therapeutically to limit inflammation. We begin our discussion of mediators by reviewing their shared properties and the general principles that govern their production and actions.

- The most important mediators of acute inflammation are vasoactive amines, lipid products (prostaglandins and leukotrienes), cytokines (including chemokines), and products of complement activation (Table 3.5). These mediators induce various components of the inflammatory response, typically by distinct mechanisms, which is why inhibiting each has been therapeutically beneficial. However, there is also some overlap (redundancy) in the actions of the mediators.
- Mediators are either secreted by cells or generated from plasma proteins. *Cell-derived mediators* are normally sequestered in intracellular granules and can be rapidly secreted by granule exocytosis (e.g., histamine in mast cell granules) or are synthesized de novo (e.g., prostaglandins and leukotrienes, cytokines) in response to a stimulus. The major cell types that produce mediators of acute inflammation are the sentinels that detect invaders and tissue damage, that is, macrophages, dendritic cells, and mast cells, but platelets, neutrophils, endothelial cells, and most epithelia can also be induced to elaborate some of the mediators. *Plasma-derived mediators* (e.g., complement proteins) are produced mainly in the liver and are present in the circulation as inactive precursors that must be activated, usually by a series of proteolytic cleavages, to acquire their functional properties.
- Active mediators are produced only in response to offending stimuli. These stimuli include microbial products and substances released from injured or necrotic cells. Some of the stimuli trigger well-defined receptors and signaling pathways, described earlier,

TABLE 3.5 Principal Mediators of Inflammation

Mediator	Principal Sources	Actions
Histamine	Mast cells, basophils, platelets	Vasodilation, increased vascular permeability, endothelial activation
Prostaglandins	Mast cells, leukocytes	Vasodilation, pain, fever
Leukotrienes	Mast cells, leukocytes	Increased vascular permeability, chemotaxis, leukocyte adhesion and activation
Cytokines (TNF, IL-1, IL-6)	Macrophages, endothelial cells, mast cells	Local: endothelial activation (expression of adhesion molecules) Systemic: fever, metabolic abnormalities, hypotension (shock)
Chemokines	Leukocytes, activated macrophages	Chemotaxis, leukocyte activation
Platelet-activating factor	Leukocytes, mast cells	Vasodilation, increased vascular permeability, leukocyte adhesion, chemotaxis, degranulation, oxidative burst
Complement	Plasma (produced in liver)	Leukocyte chemotaxis and activation, direct target killing (membrane attack complex), vasodilation (mast cell stimulation)
Kinins	Plasma (produced in liver)	Increased vascular permeability, smooth muscle contraction, vasodilation, pain

IL, Interleukin; TNF, tumor necrosis factor.

but we still do not know how other stimuli induce the secretion of mediators (e.g., from mast cells in response to cell injury or mechanical irritation). The usual requirement for microbes or dead tissues as the initiating stimulus ensures that inflammation is normally triggered only when and where it is needed.

- Most mediators are short lived. They quickly decay, or are inactivated by enzymes, or they are otherwise scavenged or inhibited. Thus, there is a system of checks and balances that regulates mediator actions. These built-in control mechanisms are discussed with each class of mediator.
- One mediator can stimulate the release of other mediators. For instance, products of complement activation stimulate the release of histamine, and the cytokine TNF acts on endothelial cells to stimulate the production of another cytokine, IL-1, and many chemokines. The secondary mediators may have the same actions as the initial mediators but may also have different and even opposing activities. Such cascades provide mechanisms for amplifying—or, in certain instances, counteracting—the initial action of a mediator.

We next discuss the more important mediators of acute inflammation, focusing on their mechanisms of action and roles in acute inflammation.

Vasoactive Amines: Histamine and Serotonin

The two major vasoactive amines, so named because they have important actions on blood vessels, are *histamine* and *serotonin*. They are stored as preformed molecules in cells and are therefore among the first mediators to be released during inflammation.

The richest source of histamine are mast cells that are normally present in the connective tissue adjacent to blood vessels. It is also found in basophils and platelets. Histamine is stored in mast cell granules and is released by mast cell degranulation in response to a variety of stimuli, including (1) physical injury (such as trauma), cold, and heat, all by unknown mechanisms; (2) binding of antigen to IgE antibodies displayed on the surfaces of mast cells, which underlies immediate hypersensitivity (allergic) reactions (Chapter 6); and (3) products of complement called *anaphylatoxins* (C3a and C5a), described later. Antibodies and complement products bind to specific receptors on mast cells and trigger signaling pathways that induce rapid degranulation. Neuropeptides (e.g., substance P) and cytokines (IL-1, IL-8) may also trigger release of histamine. Serotonin (5-hydroxytryptamine) is a preformed vasoactive mediator present in platelets and certain neuroendocrine cells, such as in the gastrointestinal tract. Its primary function is as a neurotransmitter in the gastrointestinal tract and the central nervous system. It is also a vasoconstrictor, but the importance of this action in inflammation is unclear.

Histamine causes dilation of arterioles and increases the permeability of venules. Histamine is considered to be the principal mediator of the immediate transient phase of increased vascular permeability, producing interendothelial gaps in postcapillary venules, as discussed earlier. Histamine also causes contraction of some smooth muscles. There are four types of histamine receptors: H1 receptors are linked to allergic responses, H2 to gastric acid regulation, H3 to neurotransmitter release modulation, and H4 to immune system function. All belong to the G protein–coupled receptor family. Its vasoactive effects are mediated mainly via binding to H1 receptors on microvascular endothelial cells. Drugs that block H1 receptors are used to treat allergic reactions, whereas H2 blockers reduce acid secretion in the stomach.

Arachidonic Acid Metabolites

The lipid mediators *prostaglandins* and *leukotrienes* are produced from arachidonic acid (AA) present in membrane phospholipids and stimulate vascular and cellular reactions in acute inflammation. AA is a 20-carbon polyunsaturated fatty acid (5,8,11,14-eicosatetraenoic acid) that is derived from dietary sources or by synthesis from a precursor molecule, the essential fatty acid linoleic acid. Mechanical, chemical, and physical stimuli or other mediators (e.g., C5a) release AA from membrane phospholipids through the action of cellular phospholipases, mainly phospholipase A₂. AA-derived mediators, also called *eicosanoids* (because they are derived from 20-carbon fatty acids; Greek *eicosa* = 20), are synthesized by two major classes of enzymes: *cyclooxygenases* (which generate prostaglandins) and *lipoxygenases* (which produce leukotrienes and lipoxins) (Fig. 3.9). Eicosanoids bind to G protein–coupled receptors on many cell types and can mediate virtually every step of inflammation (Table 3.6).

Prostaglandins. Prostaglandins (PGs) are produced by mast cells, macrophages, endothelial cells, and many other cell types, and are involved in the vascular and systemic reactions of inflammation. They are generated by the actions of two cyclooxygenases, called COX-1 and COX-2. COX-1 is constitutively expressed in most tissues, where it may serve various homeostatic functions (e.g., fluid and electrolyte balance in the kidneys, cytoprotection in the gastrointestinal tract), and is also induced by inflammatory stimuli. By contrast, COX-2 expression is mainly confined to cells that are participating in inflammatory reactions.

Prostaglandins are named based on common structural features coded by a letter (PGD, PGE, PGF, PGG, and PGH) and a subscript numeral (e.g., 1, 2) that indicates the number of double bonds in the compound. The most important ones in inflammation are PGE₂, PGD₂, PGF_{2α}, PGI₂ (prostacyclin), and thromboxane A₂ (TxA₂), each of which is synthesized by a specific enzyme acting on an intermediate

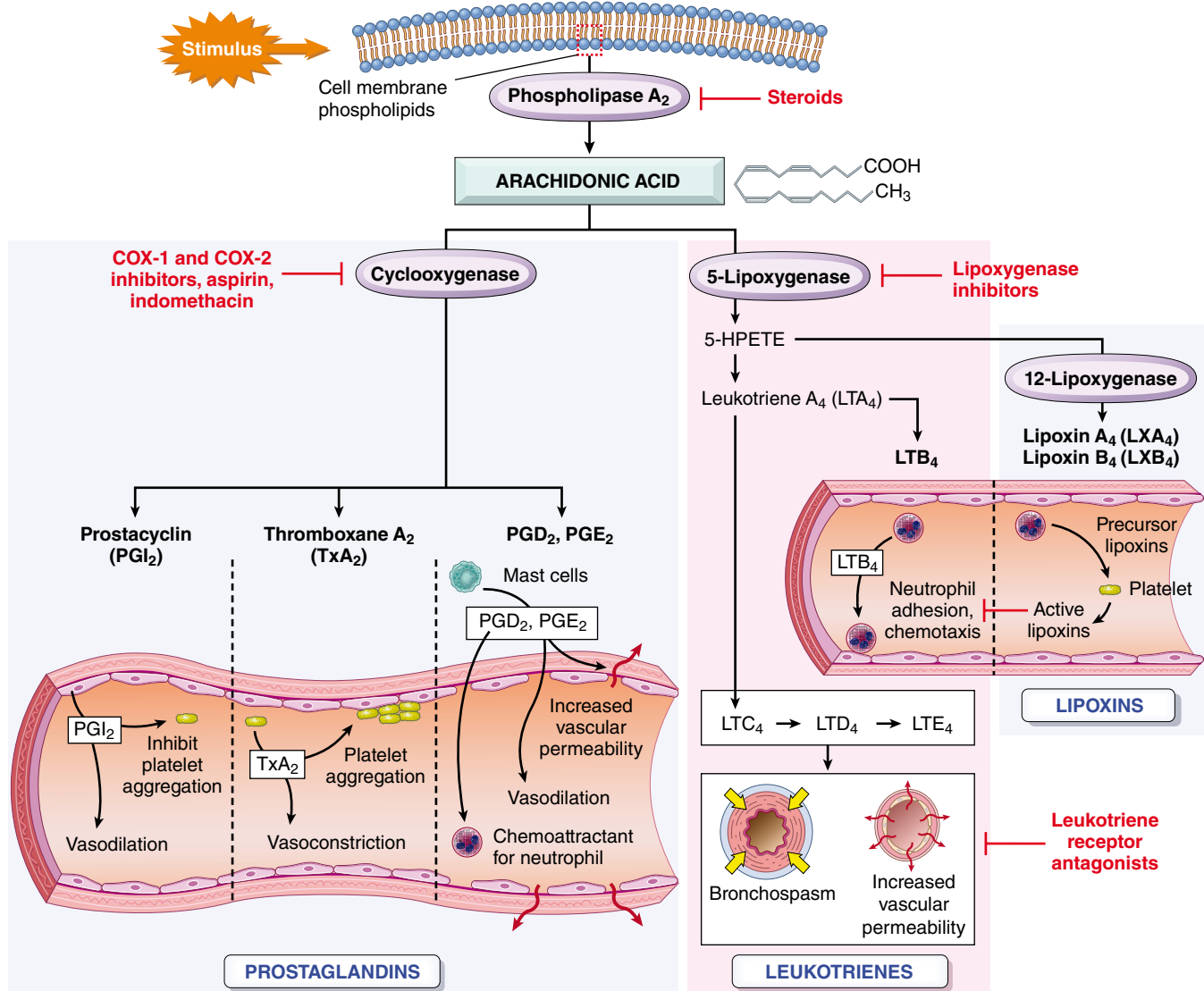


Fig. 3.9 Production of arachidonic acid metabolites and their roles in inflammation. Note the enzymatic activities whose inhibition through pharmacologic intervention blocks major pathways (in red). COX-1, COX-2, Cyclooxygenase 1 and 2; HETE, hydroxyeicosatetraenoic acid; HPETE, hydroperoxyeicosatetraenoic acid.

TABLE 3.6 Principal Actions of Arachidonic Acid Metabolites in Inflammation

Action	Eicosanoid
Vasodilation	Prostaglandins PGI ₂ (prostacyclin), PGE ₁ , PGE ₂ , PGD ₂
Vasoconstriction	Thromboxane A ₂ , leukotrienes C ₄ , D ₄ , E ₄
Increased vascular permeability	Leukotrienes C ₄ , D ₄ , E ₄
Chemotaxis, leukocyte adhesion	Leukotrienes B ₄ , HETE

HETE, Hydroxyeicosatetraenoic acid.

in the pathway. Some of these enzymes have restricted tissue distributions. The prostaglandins have numerous functions.

- PGD₂ is the major prostaglandin made by mast cells; along with PGE₂ (which is more widely distributed), it causes vasodilation and increases the permeability of postcapillary venules, thus

potentiating edema formation. PGD₂ is also a chemoattractant for neutrophils.

- Platelets contain the enzyme thromboxane synthase, and hence thromboxane A₂ is the major eicosanoid produced in these cells. Thromboxane A₂ is a potent platelet-aggregating agent and vasoconstrictor. It is unstable and rapidly converted to its inactive form. Vascular endothelium lacks thromboxane synthase but contains prostacyclin synthase, which is responsible for the formation of prostacyclin (PGI₂) and its stable end product PGF_{1α}.
- Prostacyclin is a vasodilator and a potent inhibitor of platelet aggregation, and markedly potentiates the permeability-increasing and chemotactic effects of other mediators. A thromboxane-prostacyclin imbalance has been implicated as an early event in thrombus formation in coronary and cerebral blood vessels.
- In addition to their local effects, prostaglandins are involved in the pathogenesis of pain and fever in inflammation. PGE₂ is hyperalgesic, making the skin hypersensitive to painful stimuli such as intradermal injection of suboptimal concentrations of histamine and

bradykinin. It is involved in cytokine-induced fever during infections (described later).

Leukotrienes. Leukotrienes are produced in leukocytes and mast cells by the action of lipoxygenase and are involved in vascular and smooth muscle reactions and leukocyte recruitment. There are three different lipoxygenases, 5-lipoxygenase being the predominant one in neutrophils. This enzyme converts AA to 5-hydroxyeicosatetraenoic acid, which is chemotactic for neutrophils and is the precursor of leukotrienes. Leukotriene B₄ (LTB₄) is a potent chemotactic agent and activator of neutrophils, causing aggregation and adhesion of the cells to venular endothelium, generation of ROS, and release of lysosomal enzymes. The cysteinyl-containing leukotrienes LTC₄, LTD₄, and LTE₄ cause bronchospasm (important in asthma) and increased permeability of venules.

Lipoxins. Lipoxins are also generated from AA by the lipoxygenase pathway, but unlike prostaglandins and leukotrienes, the lipoxins suppress inflammation by inhibiting neutrophil chemotaxis and adhesion to endothelium. They are also unusual in that two cell populations are required for the transcellular biosynthesis of these mediators. Neutrophils synthesize precursors of active lipoxins and pass these to platelets, where they are converted to mature lipoxins.

Various other antiinflammatory AA-derived mediators have been described and given names (e.g., *resolvins*, because they are postulated to resolve the active phase of acute inflammation). The role of these compounds in the inflammatory response is a topic of active study.

Pharmacologic Inhibitors of Prostaglandins and Leukotrienes. The importance of eicosanoids in inflammation has driven attempts to develop drugs that inhibit their production or actions and thus suppress inflammation. These antiinflammatory drugs include:

- **Cyclooxygenase inhibitors** such as aspirin and other nonsteroidal anti-inflammatory drugs (NSAIDs). They inactivate both COX-1 and COX-2 and thus inhibit prostaglandin synthesis (hence their efficacy in treating pain and fever); aspirin does this by irreversibly acetylating and inactivating cyclooxygenases. Selective COX-2 inhibitors are 200- to 300-fold more potent in blocking COX-2 than COX-1. There has been great interest in COX-2 as a therapeutic target because of the possibility that COX-1 is responsible for the production of prostaglandins that are involved in both inflammation and physiologic protective functions, for example, protecting the gastric epithelium,

whereas COX-2 generates prostaglandins that are involved only in inflammatory reactions. If this idea is correct, selective COX-2 inhibitors should be antiinflammatory without having the toxicities of the nonselective inhibitors, such as gastric ulceration. However, these distinctions are not absolute, as COX-2 also seems to play a role in cytoprotection. Furthermore, selective COX-2 inhibitors may increase the risk of cardiovascular and cerebrovascular events, possibly because they impair endothelial cell production of PGI₂ (prostacyclin), a vasodilator and inhibitor of platelet aggregation, but leave intact the COX-1-mediated production by platelets of thromboxane A₂, which activates platelets and constricts blood vessels. Thus, selective COX-2 inhibition may tilt the balance toward thromboxane and promote vascular thrombosis, especially in individuals with other factors that increase the risk of thrombosis. Nevertheless, these drugs are still used in individuals who do not have risk factors for cardiovascular disease when their benefits outweigh their risks.

- **Lipoxygenase inhibitors.** 5-Lipoxygenase is not affected by NSAIDs, and many new inhibitors of this enzyme pathway have been developed. Pharmacologic agents that inhibit leukotriene production are useful in the treatment of asthma.
- **Corticosteroids** are broad-spectrum antiinflammatory agents that reduce the transcription of genes encoding many proteins involved in inflammation, including COX-2, phospholipase A₂, proinflammatory cytokines (e.g., IL-1 and TNF), and iNOS.
- **Leukotriene receptor antagonists** block leukotriene receptors and inhibit the actions of leukotrienes. These drugs are useful in the treatment of asthma.
- Another approach to manipulating inflammatory responses has been to modify the intake and content of dietary lipids by increasing the consumption of fish oil. The proposed explanation for the effectiveness of this approach is that the polyunsaturated fatty acids in fish oil are poor substrates for conversion to active metabolites by the cyclooxygenase and lipoxygenase pathways but are better substrates for the production of antiinflammatory lipid products, including resolvins.

Cytokines and Chemokines

Cytokines are proteins produced by many cell types (principally activated lymphocytes, macrophages, and dendritic cells, but also endothelial, epithelial, and connective tissue cells) that mediate and regulate immune and inflammatory reactions. By convention, growth factors that act on epithelial and mesenchymal cells are not

TABLE 3.7 Cytokines in Inflammation

Cytokine	Principal Sources	Principal Actions in Inflammation
In Acute Inflammation		
TNF	Macrophages, dendritic cells, mast cells, T lymphocytes	Stimulates expression of endothelial adhesion molecules and secretion of other cytokines; systemic effects
IL-1	Macrophages, dendritic cells, some epithelial cells	Similar to TNF; greater role in fever
IL-6	Macrophages, other cells	Systemic effects (acute-phase response)
Chemokines	Macrophages, endothelial cells, T lymphocytes, mast cells, other cell types	Recruitment of leukocytes to sites of inflammation; migration of cells in normal tissues
IL-17	T lymphocytes	Recruitment of neutrophils and monocytes
In Chronic Inflammation		
IL-12	Dendritic cells, macrophages	Increased production of IFN- γ
IFN- γ	T lymphocytes, NK cells	Activation of macrophages (increased ability to kill microbes and tumor cells)
IL-17	T lymphocytes	Recruitment of neutrophils and monocytes

The most important cytokines involved in inflammatory reactions are listed. Many other cytokines may play lesser roles in inflammation. There is also considerable overlap between the cytokines involved in acute and chronic inflammation. Specifically, all the cytokines listed under acute inflammation may also contribute to chronic inflammatory reactions.

IFN- γ , Interferon- γ ; IL, interleukin; NK, natural killer; TNF, tumor necrosis factor.

grouped under cytokines. The general properties and functions of cytokines are discussed in Chapter 6. The cytokines involved in acute inflammation are discussed here (Table 3.7).

Tumor Necrosis Factor and Interleukin-1. TNF and IL-1 serve critical roles in leukocyte recruitment by promoting adhesion of leukocytes to endothelium and their migration through blood vessels. These cytokines are produced mainly by activated macrophages and dendritic cells; TNF is also produced by T lymphocytes and mast cells, and IL-1 is produced by some epithelial cells as well. The secretion of TNF and IL-1 can be stimulated by microbial products, dead cells, immune complexes, foreign bodies, physical injury, and a variety of other inflammatory stimuli. The production of TNF is induced by signals through TLRs and other microbial sensors. The synthesis of IL-1 is stimulated by the same signals, but the generation of the biologically active form of this cytokine is dependent on the inflammasome (described earlier).

The actions of TNF and IL-1 contribute to the local and systemic reactions of inflammation (Fig. 3.10). The most important roles of these cytokines in inflammation are:

- **Endothelial activation.** Both TNF and IL-1 act on endothelial cells to induce a spectrum of changes referred to as endothelial activation. These changes include increased expression of adhesion molecules, mostly E-selectin and P-selectin and ligands for leukocyte integrins; increased production of various mediators, including other cytokines and chemokines, growth factors, and eicosanoids; and increased procoagulant activity of the endothelium.
- **Activation of leukocytes and other cells.** TNF augments responses of neutrophils to other stimuli such as bacterial endotoxin and stimulates the microbicidal activity of macrophages, in part by inducing production of NO. IL-1 activates fibroblasts to synthesize collagen and stimulates proliferation of synovial and other mesenchymal

cells. IL-1 also stimulates Th17 responses, which in turn induce acute inflammation.

- **Other actions.** IL-1 and TNF (as well as IL-6) induce the systemic reactions associated with infection or injury, discussed later. TNF regulates energy balance by promoting lipid and protein mobilization and by suppressing appetite. Therefore, sustained production of TNF contributes to cachexia, a pathologic state characterized by weight loss and anorexia that is seen in some chronic infections and neoplastic diseases.

TNF antagonists have been remarkably effective in the treatment of chronic inflammatory diseases, particularly rheumatoid arthritis, psoriasis, and some types of inflammatory bowel disease. One of the complications of this therapy is that patients become susceptible to mycobacterial infection, reflecting the reduced ability of macrophages to kill intracellular microbes. Although many of the actions of TNF and IL-1 seem to overlap, IL-1 antagonists are not as effective for reasons that remain unclear. Also, blocking either cytokine has no effect on the outcome of sepsis, perhaps because multiple cytokines contribute to this systemic inflammatory reaction.

Chemokines. Chemokines are a family of small (8 to 10 kDa) proteins that act primarily as chemoattractants for specific types of leukocytes. About 40 different chemokines and 20 different receptors for chemokines have been identified. They are classified into four major groups, according to the arrangement of cysteine (C) residues in the proteins.

- **C-X-C chemokines** have one amino acid residue separating the first two of four conserved cysteine residues. A subset of these chemokines acts primarily on neutrophils. IL-8 (now called CXCL8) is typical of this group. It is secreted by activated macrophages, endothelial cells, and other cell types and causes activation and

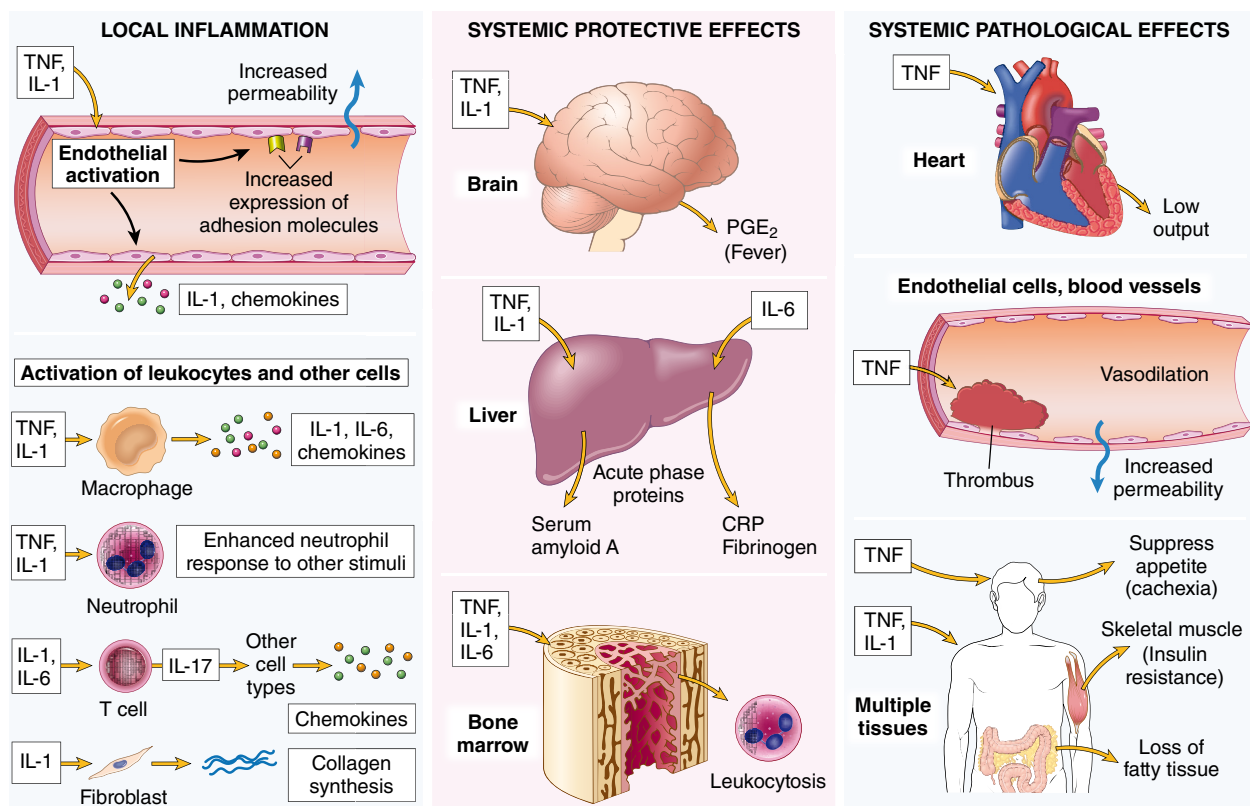


Fig. 3.10 Major roles of cytokines in acute inflammation. CRP, C-reactive protein; IL, interleukin; PG, prostaglandin; TNF, tumor necrosis factor.

chemotaxis of neutrophils, with limited activity on monocytes and eosinophils. Its most important inducers are microbial products and other cytokines, mainly IL-1 and TNF.

- *C-C chemokines* have the first two conserved cysteine residues adjacent. The C-C chemokines, which include monocyte chemoattractant protein (MCP-1 or CCL2), eotaxin (CCL11), macrophage inflammatory protein-1 α (MIP-1 α or CCL3), and others, generally attract monocytes, eosinophils, basophils, and lymphocytes, but are less potent chemoattractants for neutrophils. Although most of the chemokines in this class have overlapping actions, eotaxin selectively recruits eosinophils.
- *C chemokines* lack the first and third of the four conserved cysteines. The C chemokines (e.g., lymphotactin or XCL1) are relatively specific for lymphocytes.
- *CX₃C chemokines* contain three amino acids between the first two cysteines. The only known member of this class is called *fractalkine* (CX3CL1). This chemokine exists in two forms: a cell surface-bound protein induced on endothelial cells by inflammatory cytokines that promotes strong adhesion of monocytes and T cells, and a soluble form derived by proteolysis of the membrane-bound protein that has potent chemoattractant activity for the same cells.

Chemokines mediate their activities by binding to seven-transmembrane G protein-coupled receptors. Most leukocytes express more than one receptor type. As discussed in Chapter 6, certain chemokine receptors (CXCR4, CCR5) act as coreceptors for a viral envelope glycoprotein of human immunodeficiency virus (HIV), the cause of AIDS, and are thus involved in binding and entry of the virus into cells.

Chemokines may be displayed at high concentrations attached to proteoglycans on the surface of endothelial cells and in the extracellular matrix (ECM). They have two main functions.

- *In acute inflammation.* Inflammatory chemokines are the ones whose production is induced by microbes and other stimuli. These chemokines stimulate leukocyte attachment to endothelium by acting on leukocytes to increase the affinity of integrins, and they stimulate migration (chemotaxis) of leukocytes in tissues to the site of infection or tissue damage.
- *Maintenance of tissue architecture.* Some chemokines are produced constitutively in tissues and are sometimes called *homeostatic chemokines*. These organize various cell types in different anatomic regions of the tissues, such as T and B lymphocytes in discrete areas of the spleen and lymph nodes (Chapter 6).

Although the role of chemokines in inflammation is well established, it has proved difficult to develop antagonists that are effective therapeutic agents.

Other Cytokines in Acute Inflammation. The list of cytokines implicated in inflammation is huge and constantly growing. In addition to the ones described earlier, two that have received considerable recent interest are IL-6, made by macrophages and other cells, which is involved in local and systemic reactions, and IL-17, produced mainly by T lymphocytes, which promotes neutrophil recruitment. Antagonists against both are effective for the treatment of inflammatory diseases, such as juvenile arthritis (anti-IL-6 receptor) and psoriasis (anti-IL-17). Cytokines also play key roles in chronic inflammation; these are described later.

Complement System

The complement system is a collection of plasma proteins that function mainly in host defense against microbes and in pathologic inflammatory reactions. The system consists of more than 20 proteins,

some of which are numbered C1 through C9. The complement proteins participate in both innate and adaptive immunity for defense against microbial pathogens. In the process of complement activation, several cleavage products of complement proteins are elaborated that cause increased vascular permeability, chemotaxis, and opsonization. The activation and functions of complement are outlined in Fig. 3.11.

Complement proteins are present in inactive forms in the plasma, and many of them are activated to become proteolytic enzymes that degrade other complement proteins, thus forming an enzymatic cascade capable of tremendous amplification. The critical step in complement activation is the proteolysis of the third (and most abundant) component, C3. **Cleavage of C3 can occur by one of three pathways:**

- The *classical pathway*, which is triggered by binding of C1 to antibody (IgM or IgG) that has combined with antigen, leading to sequential binding of C2 and C4 and culminating in the formation of a multiprotein enzyme that cleaves C3.
- The *alternative pathway*, which can be triggered by microbial surface molecules (e.g., endotoxin, or lipopolysaccharide [LPS]), complex polysaccharides, cobra venom, and other substances, in the absence of antibody. In this pathway, spontaneous cleavage of C3 is amplified and stabilized on microbes and other activating substrates.
- The *lectin pathway*, in which plasma mannose-binding lectin binds to carbohydrates on microbes and activates a protease that is homologous to C1. The subsequent steps are as in the classical pathway.

All three pathways of complement activation lead to the formation of an active enzyme called C3 convertase, which splits C3 into two functionally distinct fragments, C3a and C3b. C3a is released, and C3b becomes covalently attached to the cell or molecule where complement is being activated. More C3b then binds to the previously generated fragments to form C5 convertase, which cleaves C5 to release C5a and leave C5b attached to the cell surface. C5b binds the late components (C6–C9), culminating in the formation of the membrane attack complex (composed of multiple C9 molecules).

The complement system has three main functions (see Fig. 3.11):

- *Inflammation.* C5a, C3a, and, to a lesser extent, C4a are cleavage products of the corresponding complement components that stimulate histamine release from mast cells and thereby increase vascular permeability and cause vasodilation. They are called *anaphylatoxins* because they have effects similar to those of mast cell mediators that are involved in the allergic reaction called *anaphylaxis* (Chapter 6). C5a is also a chemotactic agent for neutrophils, monocytes, eosinophils, and basophils. In addition, C5a activates the lipoxygenase pathway of AA metabolism in neutrophils and monocytes, causing further release of inflammatory mediators.
- *Opsonization and phagocytosis.* C3b and its cleavage product inactive C3b (iC3b), when fixed to a microbial cell wall, act as opsonins and promote phagocytosis by neutrophils and macrophages, which bear cell surface receptors for the complement fragments.
- *Cell lysis.* The deposition of the membrane attack complex on cells makes these cells permeable to water and ions and results in osmotic lysis of the cells. This role of complement is particularly important for killing microbes with thin cell walls, such as *Neisseria* bacteria, and deficiency of the terminal components of complement predisposes to *Neisseria* infections.

The activation of complement is tightly controlled by cell-associated and circulating regulatory proteins. Different regulatory proteins inhibit the production of active complement fragments or remove fragments that deposit on cells. These regulators are expressed on normal host cells and are thus designed to prevent healthy tissues from being injured at sites of complement activation. Regulatory proteins can be overwhelmed when large

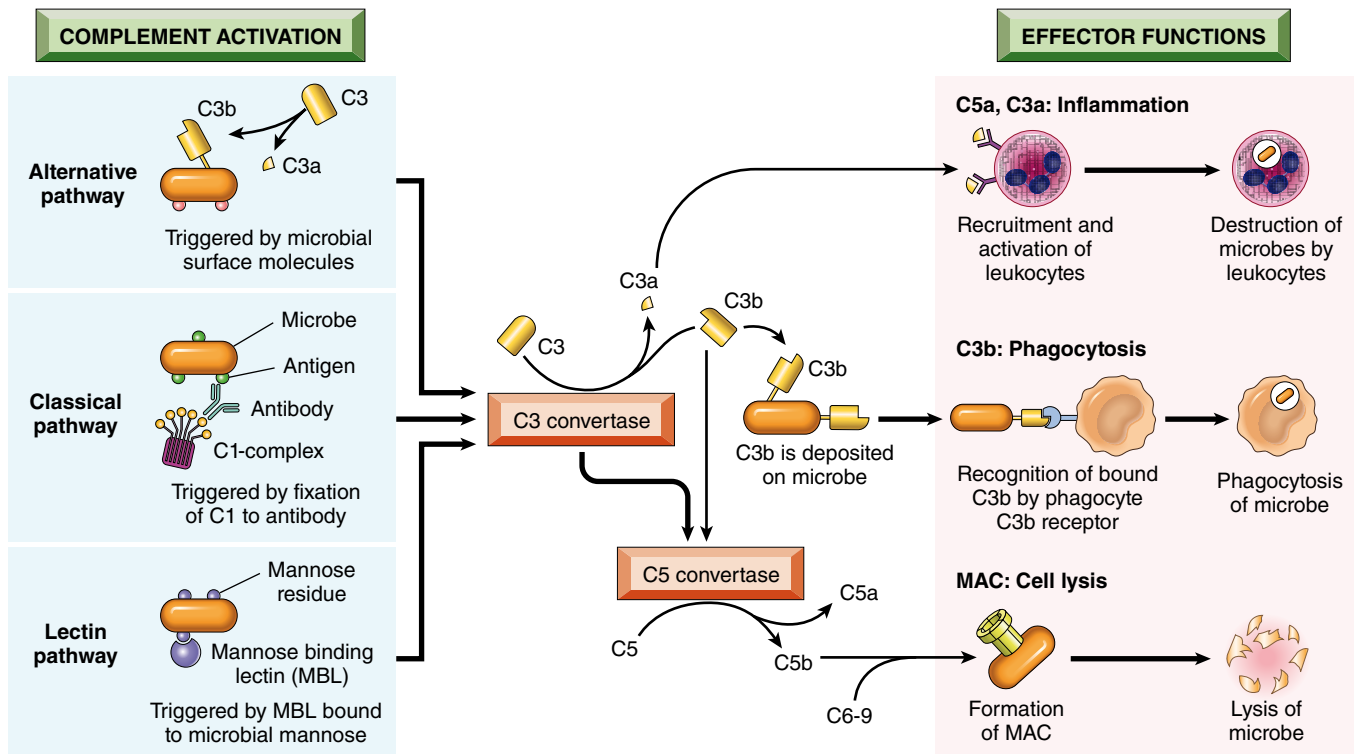


Fig. 3.11 Activation and functions of the complement system. Activation of complement by different pathways leads to cleavage of C3. The functions of the complement system are mediated by breakdown products of C3 and other complement proteins and by the membrane attack complex (MAC).

amounts of complement are deposited on host cells and in tissues, as happens in autoimmune diseases, in which individuals produce complement-fixing antibodies against their own cell and tissue antigens (Chapter 6). The most important of these regulatory proteins are:

- *C1 inhibitor* (C1 INH) blocks the activation of C1, the first protein of the classical complement pathway. Inherited deficiency of this inhibitor is the cause of hereditary angioedema.
- *Decay accelerating factor* (DAF or CD55) and *CD59* are two proteins that are linked to plasma membranes by a glycosylphosphatidylinositol (GPI) anchor. DAF prevents formation of C3 convertases, and CD59 inhibits formation of the membrane attack complex. An acquired deficiency of the enzyme that creates GPI anchors leads to deficiency of these regulators and excessive complement activation and lysis of red cells (which are sensitive to complement-mediated cell lysis) in the disease paroxysmal nocturnal hemoglobinuria (PNH) (Chapter 14).
- *Complement Factor H* is a circulating glycoprotein that inhibits the alternative pathway of complement activation by promoting the cleavage and destruction of C3b and the turnover of the C3 convertases. Inherited defects in Factor H and several other regulatory proteins that interact with Factor H cause an atypical form of hemolytic uremic syndrome (Chapter 20), in which complement deposits in glomerular vessels, leading to endothelial damage and formation of platelet-rich thrombi. Polymorphisms in the *Factor H* gene have also been linked to age-related macular degeneration (Chapter 29), an important cause of vision loss in older adults.

The complement system contributes to disease in several ways. The activation of complement by antibodies or antigen-antibody complexes deposited on host cells and tissues is an important mechanism of cell and tissue injury (Chapter 6). Inherited deficiencies of complement proteins cause increased susceptibility to infections (Chapter 6),

and, as mentioned earlier, deficiencies of regulatory proteins cause a variety of disorders resulting from excessive complement activation. Inhibitors of complement, specifically C5 and C3, activation are now approved for several of these disorders.

Other Mediators of Inflammation

Platelet-Activating Factor. Platelet-activating factor (PAF) is a phospholipid-derived mediator that was discovered as a factor that caused platelet aggregation, but it is now known to have multiple inflammatory effects. A variety of cell types, including platelets, basophils, mast cells, neutrophils, macrophages, and endothelial cells, can elaborate PAF in both secreted and cell-bound forms. In addition to platelet aggregation, PAF causes vasoconstriction and bronchoconstriction, and at low concentrations it induces vasodilation and increased venular permeability. Despite these actions, the use of PAF antagonists in various inflammatory diseases has not been found to be useful.

Products of Coagulation. A link between the coagulation pathway and inflammation is supported by the presence of protease-activated receptors (PARs) on leukocytes that are activated by thrombin (the protease that cleaves fibrinogen to produce fibrin, which forms the clot). It is, however, likely that the major role of the PARs is in platelet activation during clotting (Chapter 4). There is also some evidence that cleavage products of fibrin (fibrinopeptides) can stimulate inflammation. However, many forms of tissue injury are associated with both clotting and inflammation, and it is not established if products of the coagulation cascade themselves stimulate inflammation in some situations.

Kinins. Kinins are vasoactive peptides derived from plasma proteins, called *kininogens*, by the action of specific proteases called *kallikreins*.

The enzyme kallikrein cleaves a plasma glycoprotein precursor, high-molecular-weight kininogen, to produce bradykinin. **Bradykinin increases vascular permeability and causes contraction of smooth muscle, dilation of blood vessels, and pain when injected into the skin.** These effects are similar to those of histamine. The action of bradykinin is short lived because it is quickly inactivated by an enzyme called *kininase*. Bradykinin has been implicated as a mediator in some forms of allergic reaction, such as anaphylaxis (Chapter 6).

Neuropeptides. Neuropeptides are secreted by sensory nerves and various leukocytes and may play a role in the initiation and regulation of inflammatory responses. These small peptides, such as substance P and neurokinin A, are produced in the central and peripheral nervous systems. Nerve fibers containing substance P are prominent in the lung and gastrointestinal tract. Substance P has many activities that may be important in inflammation, including the transmission of pain signals and increasing vascular permeability. Leukocytes express receptors for many neuropeptides, so these neural products could provide a mechanism for cross-talk between the nervous system and immune and inflammatory reactions. For instance, activation of the efferent vagus nerve inhibits the production of proinflammatory cytokines such as TNF, providing a mechanism for suppressing inflammation. This observation has led to clinical trials of vagus nerve stimulation in patients with rheumatoid arthritis.

When Lewis discovered the role of histamine in inflammation, one mediator was thought to be enough. Now, we are wallowing in them! Yet, from this large compendium, it is likely that a few mediators are most important for the reactions of acute inflammation *in vivo*, and these are summarized in Table 3.8. The redundancy of the mediators and their actions ensures that this protective response is robust and not readily subverted.

KEY CONCEPTS

Actions of the Principal Mediators of Inflammation

- Vasoactive amines, mainly histamine: Vasodilation and increased vascular permeability.
- Arachidonic acid metabolites (prostaglandins and leukotrienes): Several forms exist and are involved in vascular reactions, leukocyte chemotaxis, and other reactions of inflammation; antagonized by lipoxins.
- Cytokines: Proteins produced by many cell types; usually act at short range; mediate multiple effects, mainly leukocyte recruitment and migration; principal ones in acute inflammation are TNF, IL-1, and chemokines.
- Complement proteins: Activation of the complement system by microbes or antibodies leads to the generation of multiple breakdown products, which are responsible for leukocyte chemotaxis, opsonization, phagocytosis of microbes and other particles, and cell killing.
- Kinins: Produced by proteolytic cleavage of precursors; mediate vascular reaction, pain.

Morphologic Patterns of Acute Inflammation

The morphologic hallmarks of acute inflammatory reactions are **dilation of small blood vessels and accumulation of leukocytes and fluid in the extravascular tissue.** However, special morphologic patterns are often superimposed on these general features, depending on the severity of the reaction, its etiology, and the particular tissue and site involved. The importance of recognizing gross and microscopic patterns is that they often provide valuable clues about the underlying cause.

TABLE 3.8 Role of Mediators in Different Reactions of Inflammation

Reaction of Inflammation	Principal Mediators
Vasodilation	Histamine
Increased vascular permeability	Histamine and serotonin C3a and C5a (by liberating vasoactive amines from mast cells, other cells) Leukotrienes LTC ₄ , D ₄ , E ₄
Chemotaxis, leukocyte recruitment and activation	TNF, IL-1 Chemokines C3a, C5a Leukotriene LTB ₄
Fever	IL-1, TNF Prostaglandins
Pain	Prostaglandins Bradykinin Substance P
Tissue damage	Lysosomal enzymes of leukocytes Reactive oxygen species

IL-1, Interleukin-1; TNF, tumor necrosis factor.

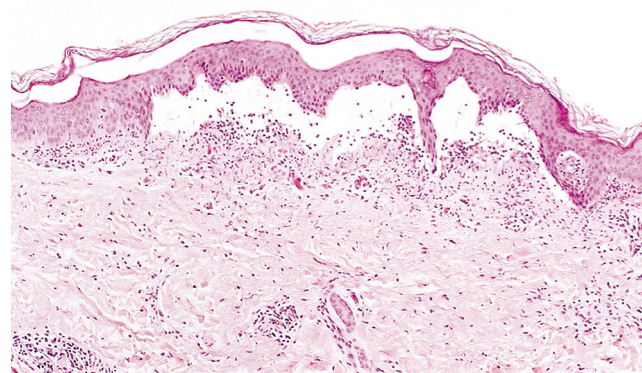


Fig. 3.12 Serous inflammation. Low-power view of a cross section of a skin blister showing the epidermis separated from the dermis by a focal collection of serous effusion.

Serous Inflammation

Serous inflammation is marked by the exudation of serum-like fluid into spaces created by cell injury or into body cavities lined by the peritoneum, pleura, or pericardium. Typically, the fluid in serous inflammation does not contain microbes or large numbers of leukocytes (which tend to produce purulent inflammation, described later). The skin blister resulting from a burn or viral infection represents accumulation of serous fluid within or immediately beneath the damaged epidermis of the skin (Fig. 3.12). In body cavities the fluid may be derived from plasma (as a result of increased vascular permeability) or from the secretions of mesothelial cells (as a result of local irritation); accumulation of fluid in these cavities is called an *effusion*. (Effusions also occur in noninflammatory conditions, such as reduced blood outflow in heart failure or reduced plasma protein levels in some kidney and liver diseases.)

Fibrinous Inflammation

With greater increase in vascular permeability, large molecules such as fibrinogen pass out of the blood, and fibrin is formed and deposited in the extracellular space. **A fibrinous exudate develops when large molecules leak from vessels and there is a local procoagulant stimulus.** A fibrinous exudate is characteristic of inflammation in the lining of body

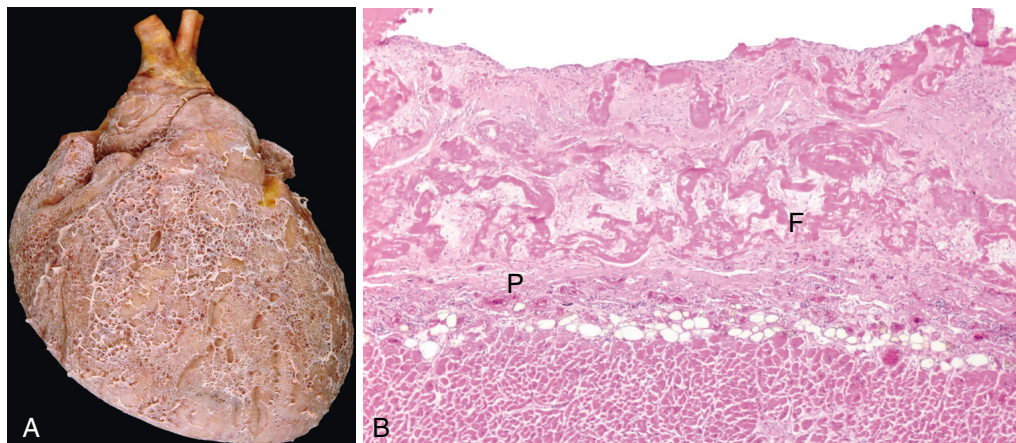


Fig. 3.13 Fibrinous pericarditis. (A) Deposits of fibrin on the pericardium. (B) A pink meshwork of fibrin exudate (F) overlies the pericardial surface (P). (Courtesy Dr. Joseph J. Maleszewski, Mayo Clinic, Rochester, MN.)

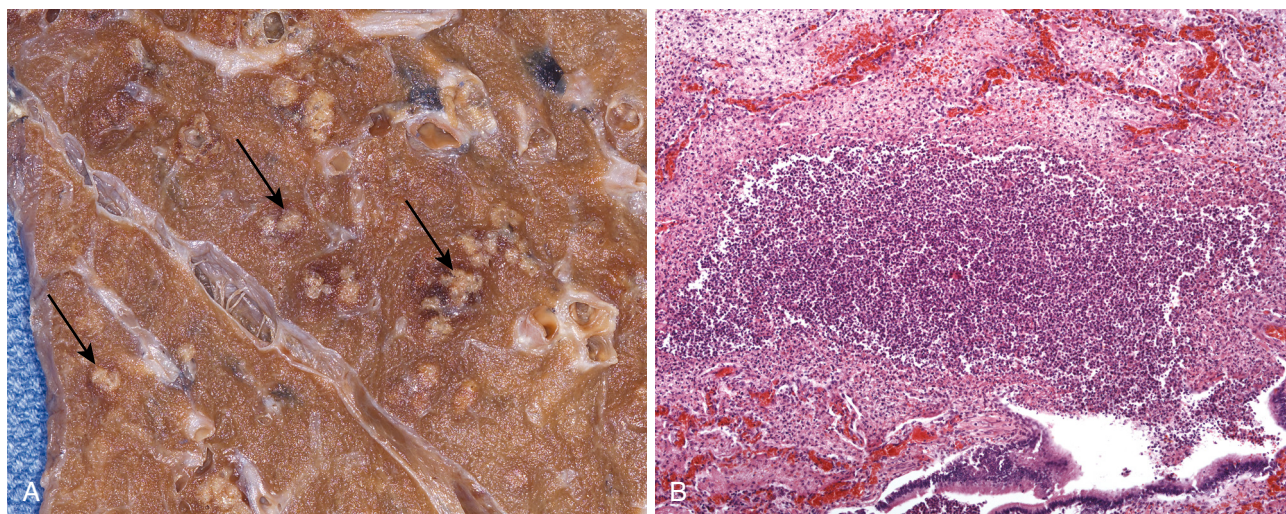


Fig. 3.14 Purulent inflammation. (A) Multiple bacterial abscesses (arrows) in the lung in a case of broncho-pneumonia. (B) The abscess contains neutrophils and cellular debris and is surrounded by congested blood vessels.

cavities, such as the meninges, pericardium (Fig. 3.13A), and pleura. Histologically, fibrin appears as an eosinophilic meshwork of threads or sometimes as an amorphous coagulum (Fig. 3.13B). Fibrinous exudates may be dissolved by fibrinolysis and cleared by macrophages. If the fibrin is not removed, over time it may stimulate the ingrowth of fibroblasts and blood vessels and thus lead to scarring. Conversion of the fibrinous exudate to scar tissue (*organization*) within the pericardial sac leads to opaque fibrous thickening of the pericardium and epicardium in the area of exudation and, if the fibrosis is extensive, obliteration of the pericardial space.

Purulent (Suppurative) Inflammation and Abscess

Purulent inflammation is characterized by the production of pus, an exudate consisting of neutrophils, the liquefied debris of necrotic cells, and edema fluid. The most frequent cause of purulent (also called *suppurative*) inflammation is infection with bacteria that cause liquefactive tissue necrosis, such as staphylococci; these pathogens are referred to as pyogenic (pus-producing) bacteria. A common example of acute suppurative inflammation is acute appendicitis.

Abscesses are localized collections of pus caused by suppuration buried in a tissue, an organ, or a confined space. They are produced by

seeding of pyogenic bacteria into a tissue (Fig. 3.14). Abscesses have a central liquefied region composed of necrotic leukocytes and tissue cells. There is usually a zone of preserved neutrophils around this necrotic focus, and outside this region there may be vascular dilation and parenchymal and fibroblastic proliferation, indicating chronic inflammation and repair. In time the abscess may become walled off and ultimately replaced by connective tissue.

Ulcers

An ulcer is a local defect, or excavation, of the surface of an organ or tissue that is produced by the sloughing (shedding) of inflamed necrotic tissue (Fig. 3.15). Ulceration can occur only when tissue necrosis and resultant inflammation exist on or near a surface. It is most common in (1) the mucosa of the mouth, stomach, intestines, or genitourinary tract, and (2) the skin and subcutaneous tissue of the lower extremities in individuals with disorders that predispose to vascular insufficiency, such as diabetes, sickle cell anemia, and peripheral vascular disease.

Ulcerations are best exemplified by peptic ulcer of the stomach or duodenum, in which acute and chronic inflammation coexist. During the acute stage there is intense polymorphonuclear infiltration and

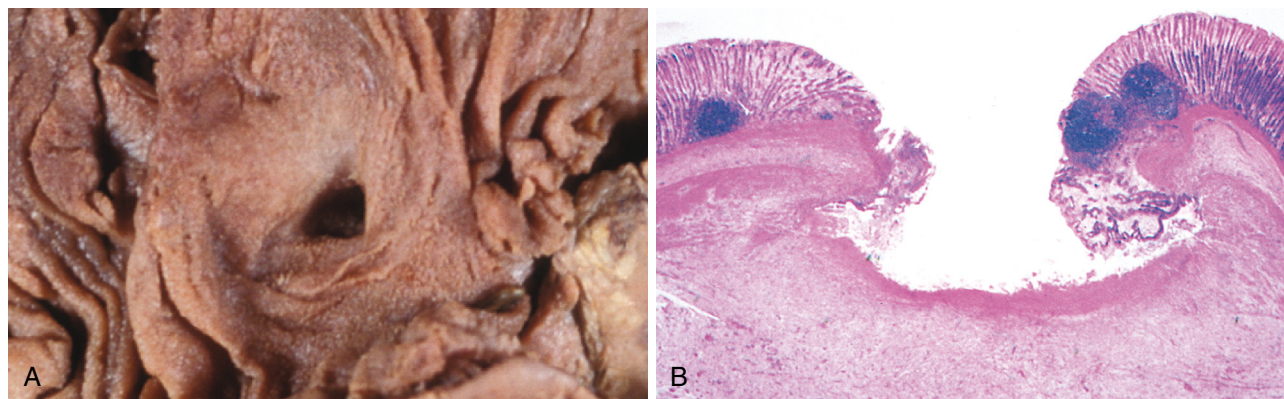


Fig. 3.15 Ulcer. (A) A chronic duodenal ulcer. (B) Low-power cross-section view of a duodenal ulcer crater with an acute inflammatory exudate in the base.

vascular dilation in the margins of the defect. With chronicity, the margins and base of the ulcer develop fibroblastic proliferation, scarring, as well as the accumulation of lymphocytes, macrophages, and plasma cells.

Outcomes of Acute Inflammation

Although, as might be expected, many variables modify the basic process of inflammation, including the nature and intensity of the injury, the site and tissue affected, and the responsiveness of the host, **acute inflammatory reactions typically have one of three outcomes** (Fig. 3.16).

- **Complete resolution.** In a perfect world, all inflammatory reactions, once they have succeeded in eliminating the offending agent, would end with restoration of the site of acute inflammation to normal. This is called resolution and is the usual outcome when the injury is limited or short lived or when there has been little tissue destruction, and the damaged parenchymal cells can regenerate. Resolution involves removal of cellular debris and microbes by macrophages and resorption of edema fluid by lymphatics, followed by regeneration of the damaged tissue.
- **Healing by connective tissue replacement (scarring, or fibrosis).** This occurs after substantial tissue destruction, when the inflammatory injury involves tissues that are incapable of regeneration, or when there is abundant fibrin exudation in tissue or in serous cavities (pleura, peritoneum) that cannot be adequately cleared. In all these situations, connective tissue grows into the area of damage or exudate, converting it into a mass of fibrous tissue, a process also called organization.
- **Progression of the response to chronic inflammation** (discussed later). Acute to chronic transition occurs when the acute inflammatory response cannot be resolved, because of either the persistence of the injurious agent or some interference with the normal process of healing.

Summary of Acute Inflammation

Now that we have described the components, mediators, and pathologic manifestations of acute inflammatory responses, it is useful to summarize the main features of a typical response of this type. When a host encounters an injurious agent, such as a microbe or dead cells, resident phagocytes and phagocytes recruited from the blood try to eliminate these agents. At the same time, sentinel cells, phagocytes, and other host cells react to the presence of the foreign or abnormal substance by liberating cytokines, lipid messengers, and other mediators of inflammation.

Some of these mediators act on small blood vessels in the vicinity and promote the efflux of plasma proteins and the recruitment of circulating leukocytes to the site where the offending agent is located. The recruited leukocytes are activated by molecules derived from microbes and injured cells and by locally produced mediators, and the activated leukocytes try to remove the offending agent by phagocytosis. As the injurious agent is eliminated and antiinflammatory mechanisms become active, the process subsides, and the host returns to a normal state of health. If the injurious agent cannot be quickly eliminated, the result may be chronic inflammation.

The vascular and cellular reactions account for the cardinal signs of inflammation: *rubor*, *calor*, *tumor*, *dolor*, and *functio laesa*. The increased blood flow to the injured area and increased vascular permeability lead to the accumulation of extravascular fluid (exudate) rich in plasma proteins, known as *edema*. The redness (*rubor*), warmth (*calor*), and swelling (*tumor*) are caused by the increased blood flow and edema. Circulating leukocytes, initially predominantly neutrophils, adhere to the endothelium via adhesion molecules, traverse the endothelium, and migrate to the site of injury under the influence of chemotactic agents. Leukocytes that are activated by the offending agent and by endogenous mediators may release toxic metabolites and proteases extracellularly, causing tissue damage. Due to the damage and the liberation of prostaglandins, neuropeptides, and cytokines, one of the local symptoms is pain (*dolor*). Loss of function (*functio laesa*) results from pain and injury to the tissues.

KEY CONCEPTS

Morphology and Outcomes of Acute Inflammation

- Serous inflammation is accumulation of protein-rich exudate in body cavities and spaces created by tissue necrosis.
- Fibrinous inflammation is characterized by formation of fibrin, usually on the surfaces of organs such as the heart and lungs.
- Purulent inflammation is characterized by formation of pus, which consists of dead cells, neutrophils, and microbes, and is most often caused by bacterial infection. An abscess is localized purulent inflammation.
- An ulcer is a discontinuity in an epithelium, with underlying acute and chronic inflammation.
- The acute inflammatory reaction may resolve or may progress to chronic inflammation if the initiating stimulus cannot be removed. Replacement by a connective tissue scar is most often seen in foci of chronic inflammation.

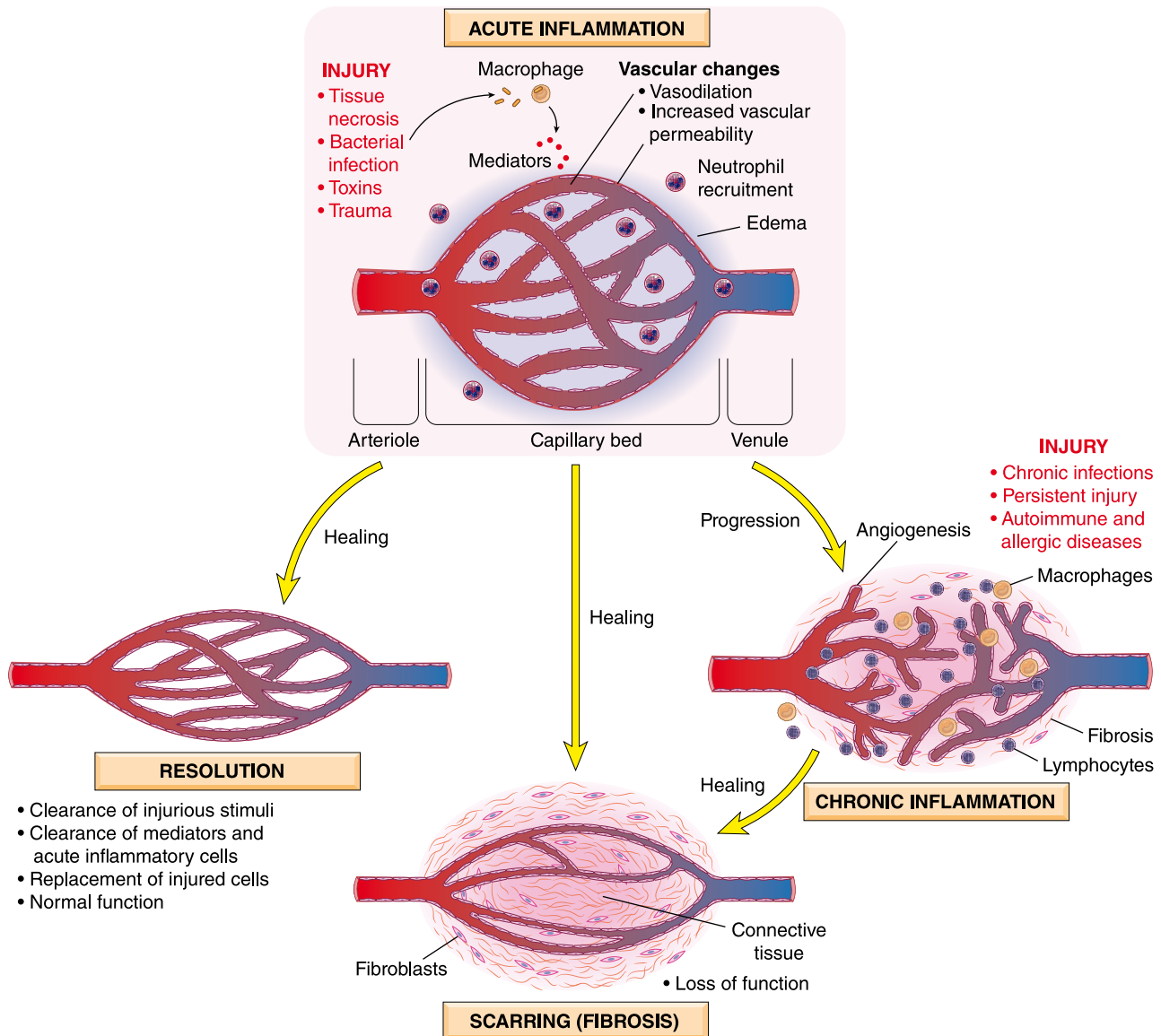


Fig. 3.16 Outcomes of acute inflammation: resolution, healing by fibrosis resulting in scarring, or chronic inflammation. The components of the various reactions and their functional outcomes are listed.

CHRONIC INFLAMMATION

Chronic inflammation is a response of prolonged duration (weeks or months) in which inflammation, tissue injury, and attempts at repair coexist. It may follow acute inflammation, as described earlier, or chronic inflammation may begin insidiously, as a prolonged response without a preceding acute reaction.

Causes of Chronic Inflammation

Chronic inflammation arises in the following settings.

- Persistent infections** by microorganisms that are difficult to eradicate, such as mycobacteria and certain viruses, fungi, and parasites. These organisms often evoke a T lymphocyte reaction called delayed-type hypersensitivity (Chapter 6). Some chronic inflammatory responses develop a specific pattern called a granulomatous reaction (discussed later). In other cases, an unresolved acute inflammation may evolve into chronic inflammation, as may occur in acute bacterial infection of the lung that progresses to a chronic lung abscess.

- Hypersensitivity diseases.** Chronic inflammation plays an important role in a group of diseases that are caused by excessive and inappropriate activation of the immune system. Under certain conditions, immune reactions develop against the individual's own tissues, leading to autoimmune diseases (Chapter 6). In these diseases, autoantigens evoke a self-perpetuating reaction of T and B lymphocytes that results in chronic tissue damage and inflammation; examples of such diseases include rheumatoid arthritis and multiple sclerosis. In other cases, chronic inflammation is the result of unregulated immune responses against microbes, as in inflammatory bowel disease. Immune responses against common environmental substances are the cause of allergic diseases, such as bronchial asthma (Chapter 6). Because these autoimmune and allergic reactions are triggered against antigens that are normally harmless, the reactions serve no useful purpose and only cause disease. Such diseases may show morphologic patterns of mixed acute and chronic inflammation because they are characterized by repeated bouts of inflammation. Fibrosis may dominate the late stages.

- **Toxic substances.** Prolonged exposure to potentially toxic agents, either exogenous or endogenous. An example of an exogenous agent is particulate silica, a nondegradable inanimate material that, when inhaled in sufficient quantities, results in an inflammatory lung disease called silicosis (Chapter 15). Atherosclerosis (Chapter 11) is a chronic inflammatory process of the arterial wall induced, at least in part, by excessive production and tissue deposition of endogenous cholesterol and other lipids.

Morphologic Features

In contrast to acute inflammation, which is manifested by vascular changes, edema, and predominantly neutrophilic infiltration, **chronic inflammation is characterized by:**

- **Infiltration with mononuclear cells**, which include macrophages, lymphocytes, and plasma cells (Fig. 3.17).
- **Tissue destruction**, induced by the persistent offending agent or by the inflammatory cells.
- **Attempts at healing** by connective tissue replacement of damaged tissue, accomplished by *angiogenesis* (proliferation of small blood vessels) and, in particular, *fibrosis*.

Because angiogenesis and fibrosis are components of wound healing and repair, they are discussed later, in the context of tissue repair.

Cells and Mediators of Chronic Inflammation

The combination of leukocyte infiltration, tissue damage, and fibrosis that characterize chronic inflammation is the result of the local activation of several cell types and the production of mediators.

Role of Macrophages

The dominant cells in most chronic inflammatory reactions are macrophages, which contribute to the reaction by secreting cytokines and growth factors that act on various cells, destroying foreign invaders and tissues, and activating other cells, notably T lymphocytes. Macrophages are professional phagocytes that eliminate microbes and damaged tissues. They also serve important roles in the repair of injured tissues. Here we review the development and functions of macrophages.

Macrophages are tissue cells derived from hematopoietic stem cells in the bone marrow in postnatal life and from progenitors in

the embryonic yolk sac and fetal liver during early development (Fig. 3.18). Circulating cells of this lineage are known as monocytes. Macrophages are normally diffusely scattered in most connective tissues. In addition, they are present in specific locations in organs such as the liver (where they are called Kupffer cells), spleen and lymph nodes (called sinus histiocytes), central nervous system (microglia), and lungs (alveolar macrophages). Together, these cells comprise the mononuclear phagocyte system, also known by the older (and inaccurate) name reticuloendothelial system.

Bone marrow–derived macrophages arise from committed progenitors in the marrow that give rise to monocytes, which enter the blood, migrate into various tissues, and differentiate into macrophages. This is typical of macrophages at sites of inflammation and in some tissues such as the skin and intestinal tract. The half-life of blood monocytes is about 1 day, whereas the lifespan of tissue macrophages may be several months or years. As discussed earlier, in inflammatory reactions, monocytes begin to emigrate into extravascular tissues quite early, and within 48 hours they may be the predominant cell type. Extravasation of monocytes is governed by the same factors that are involved in neutrophil emigration, that is, adhesion molecules and chemotactic factors. **Tissue-resident macrophages**, such as microglia, Kupffer cells, and alveolar macrophages, arise from progenitors in the yolk sac or fetal liver very early in embryogenesis and migrate to the developing brain, liver, and lung, where they persist throughout life as a stable population of cells.

The products of activated macrophages eliminate injurious agents such as microbes and initiate the process of repair but are also responsible for much of the tissue injury in chronic inflammation. Several functions of macrophages are central to the development and persistence of chronic inflammation and the accompanying tissue injury.

- Macrophages, like the other type of phagocytes, the neutrophils, ingest and destroy microbes and dead tissues.
- Macrophages initiate the process of tissue repair and are involved in scar formation and fibrosis. These processes are discussed later in the chapter.
- Macrophages secrete mediators of inflammation, such as cytokines (TNF, IL-1, chemokines, and others) and eicosanoids. Thus, macrophages contribute to the initiation and propagation of inflammatory reactions.

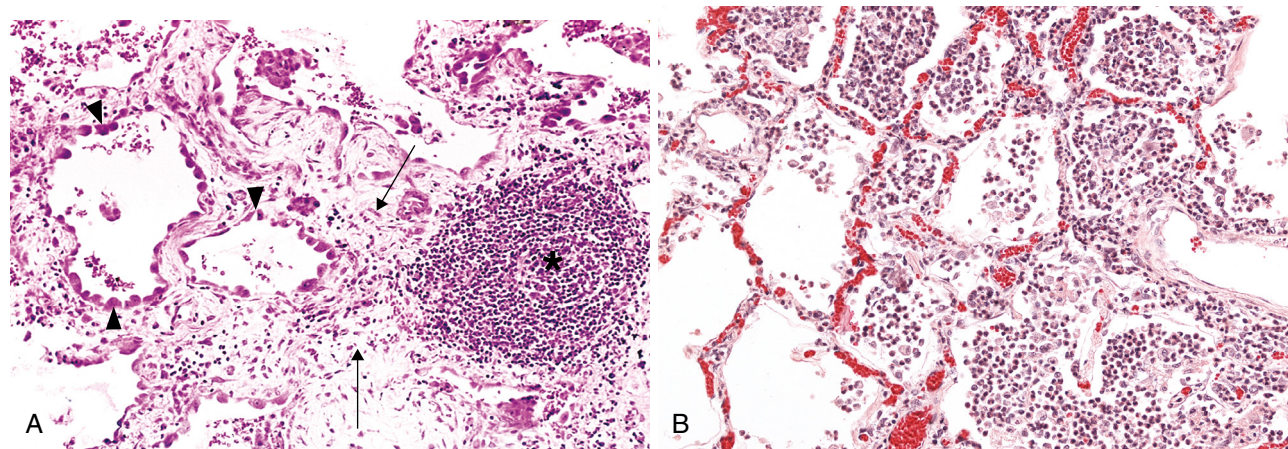


Fig. 3.17 (A) Chronic inflammation in the lung, showing all three characteristic histologic features: (1) collection of chronic inflammatory cells (*asterisk*), (2) destruction of parenchyma (normal alveoli are replaced by spaces lined by cuboidal epithelium) (*arrowheads*), and (3) replacement by connective tissue (fibrosis) (*arrows*). (B) In contrast, in acute inflammation of the lung (acute bronchopneumonia), neutrophils fill the alveolar spaces, and blood vessels are congested.

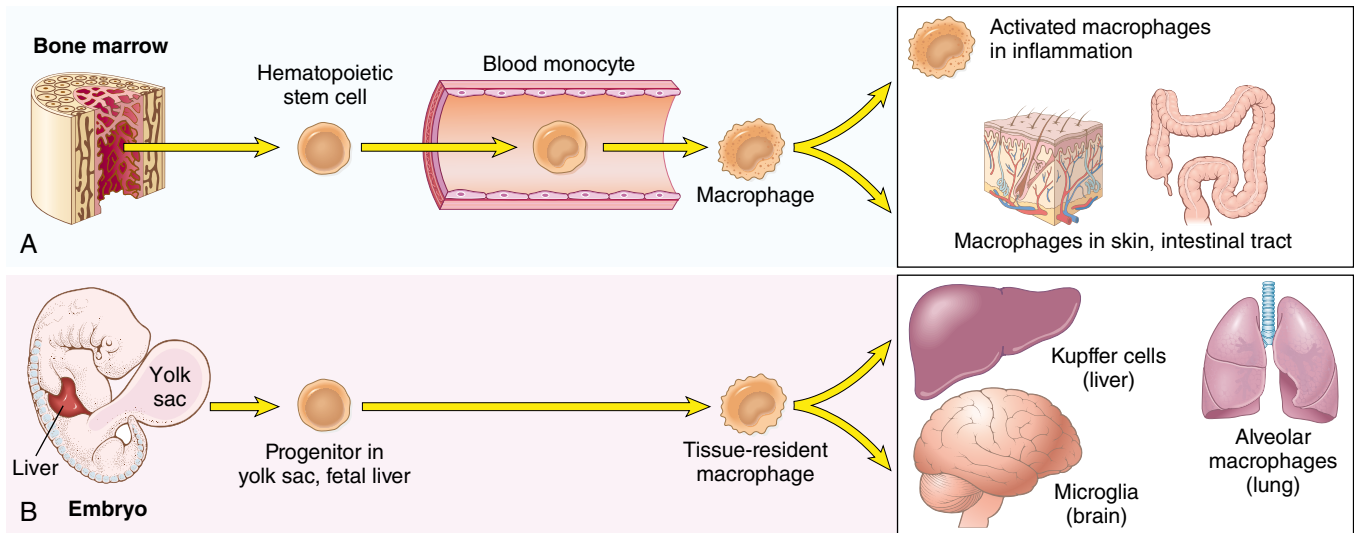


Fig. 3.18 Maturation of mononuclear phagocytes. (A) During inflammatory reactions, most tissue macrophages are derived from hematopoietic precursors. (B) Some long-lived tissue-resident macrophages are derived from embryonic precursors that populate the tissues early in development.

- Macrophages display antigens to T lymphocytes and respond to signals from T cells, thus setting up a feedback loop that is essential for defense against many microbes by cell-mediated immune responses. These interactions are described further in the subsequent discussion of the role of lymphocytes in chronic inflammation and in more detail in Chapter 6 where cell-mediated immunity is considered.

There are two major pathways of macrophage activation, called *classical* and *alternative*, that endow macrophages with different functional activities (Fig. 3.19).

- Classical macrophage activation** may be induced by microbial products such as endotoxin, which engage TLRs and other sensors; by T cell–derived signals, importantly the cytokine $\text{IFN-}\gamma$, in immune responses; or by foreign substances, including crystals and particulate matter. Classically activated (also called M1) macrophages produce NO, ROS, and lysosomal enzymes, which enhance their ability to kill ingested organisms, and secrete cytokines that stimulate inflammation. The main role of these macrophages in host defense is to destroy microbes and promote the inflammatory response.
- Alternative macrophage activation** is induced by cytokines other than $\text{IFN-}\gamma$, such as IL-4 and IL-13, produced by T lymphocytes and other cells. Alternatively activated macrophages (also called M2) are not actively microbicidal; instead, their principal functions are to terminate inflammation and promote tissue repair.

It seems plausible that in response to most injurious stimuli the first activation pathway is the classical one, designed to destroy the offending agents, and this is followed by alternative activation, which initiates tissue repair. However, such a precise sequence is not documented in most inflammatory reactions, and most reactions contain varying numbers of both types of macrophages. Also, the M1 and M2 phenotypes are extreme forms that provide a conceptual framework for understanding the types of activated macrophages, but there may be many intermediate phenotypes that are difficult to characterize as typical M1 or M2. Because the definition of these subsets is imprecise due to the lack of defining markers, some authorities refer to them as M1-like and M2-like.

Their impressive arsenal of mediators makes macrophages powerful allies in the body's defense against unwanted invaders, but the same

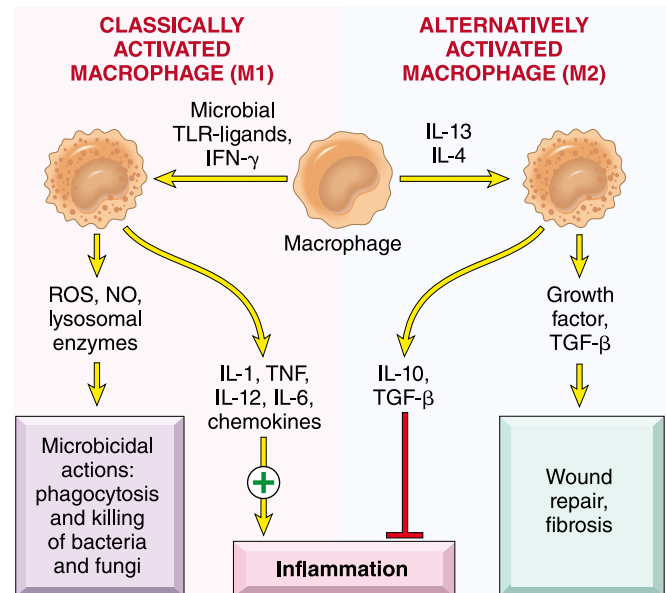


Fig. 3.19 Classical and alternative macrophage activation. Different stimuli activate monocytes/macrophages to develop into functionally distinct populations. Classically activated macrophages are induced by microbial products and cytokines, particularly interferon- γ ($\text{IFN-}\gamma$). They phagocytose and destroy microbes and dead tissues and can potentiate inflammatory reactions. Alternatively activated macrophages are induced by other cytokines and are important in tissue repair and resolution of inflammation. IL, Interleukin; NO, nitric oxide; ROS, reactive oxygen species; $\text{TGF-}\beta$, transforming growth factor- β ; TLR, toll-like receptor.

weaponry can also induce considerable tissue destruction when macrophages are inappropriately or excessively activated. It is largely because of these activities of macrophages that tissue destruction is one of the hallmarks of chronic inflammation.

In some instances, if the irritant is eliminated, macrophages eventually disappear (either dying off or making their way into the lymphatics and lymph nodes). In others, macrophage accumulation persists

because of continuous recruitment from the circulation and local proliferation at the site of inflammation.

Role of Lymphocytes

Microbes and other environmental antigens activate T and B lymphocytes, which amplify and propagate chronic inflammation. Activation of these cells is often prominent in chronic inflammatory reactions, and when they are involved, the inflammation tends to be persistent and severe, in part because lymphocyte activation leads to the generation of long-lived memory cells. Some of the persistent chronic inflammatory reactions, such as granulomatous inflammation, described later, are dependent on generation of memory lymphocyte responses. Lymphocytes may be the dominant population in the chronic inflammation seen in various autoimmune diseases.

By virtue of their ability to secrete cytokines, CD4⁺ T lymphocytes promote inflammation and influence the nature of the inflammatory reaction. There are three subsets of CD4⁺ T cells that secrete different types of cytokines and elicit different types of inflammation.

- Th1 cells produce the cytokine IFN- γ , which activates macrophages by the classical pathway.
- Th2 cells secrete IL-5, which recruits and activates eosinophils, and IL-4 and IL-13, which stimulate IgE antibody production and are responsible for the alternative pathway of macrophage activation. The role of these cells in allergic disease is discussed in Chapter 6.
- Th17 cells secrete IL-17 and other cytokines, which induce the secretion of chemokines responsible for recruiting neutrophils (and monocytes) into the reaction.

Both Th1 and Th17 cells are involved in defense against many types of bacteria and viruses and in autoimmune diseases in which tissue injury is caused by chronic inflammation. Th2 cells are important in defense against helminthic parasites and in allergic inflammation. These T-cell subsets and their functions are described in more detail in Chapter 6.

Lymphocytes and macrophages interact in a bidirectional way, and these interactions play an important role in propagating chronic inflammation (Fig. 3.20). Macrophages display antigens to T cells, express membrane molecules called costimulators, and produce cytokines (IL-12 and others) that stimulate T-cell responses (Chapter 6). Activated T lymphocytes, in turn, produce cytokines, described earlier, which recruit and activate macrophages, promoting more antigen presentation and cytokine secretion. The result is a cycle of cellular reactions that fuel and sustain chronic inflammation. A typical consequence of such a chronic T cell–macrophage reaction is the formation of granulomas, described later.

Activated B lymphocytes and antibody-secreting plasma cells are also often present at sites of chronic inflammation. The antibodies produced may be specific for persistent foreign antigens or self-antigens in the inflammatory site or against altered tissue components. However, the contribution of antibodies to most chronic inflammatory disorders is unclear.

In some chronic inflammatory reactions, the accumulated lymphocytes, antigen-presenting cells, and plasma cells cluster together to form organized lymphoid follicles resembling those seen in lymph nodes. These are called *tertiary lymphoid structures*; they may be seen in the synovium of patients with long-standing rheumatoid arthritis, in the thyroid in Hashimoto thyroiditis, and in the infiltrates around some tumors. It has been postulated that the local formation of lymphoid follicles may perpetuate the immune reaction, but the significance of these structures is not established.

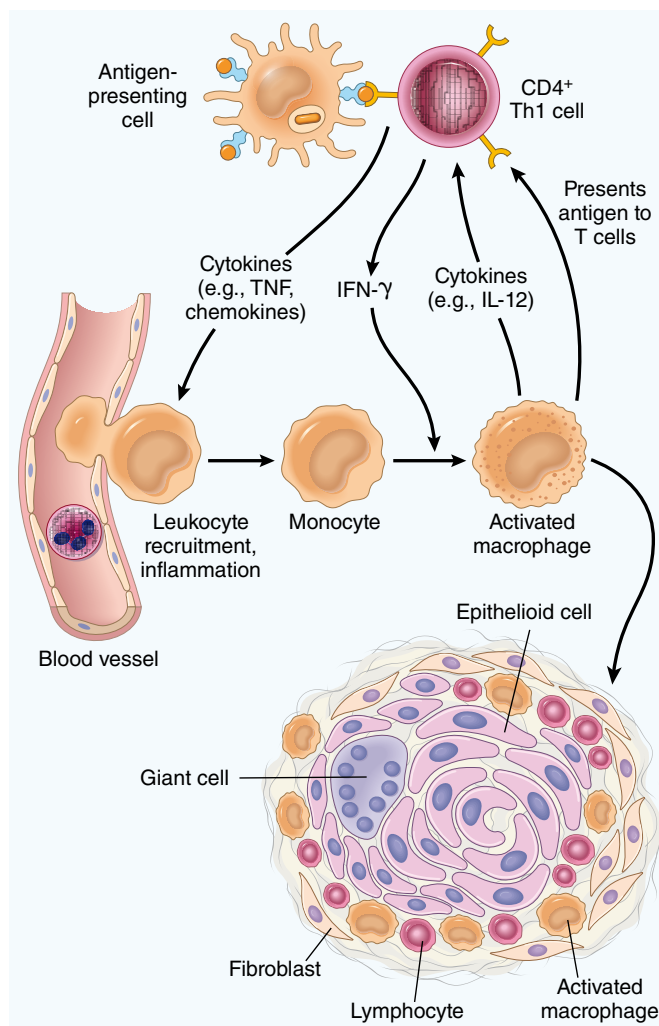


Fig. 3.20 Macrophage-lymphocyte interactions in chronic inflammation. Activated T cells produce cytokines that recruit macrophages (tumor necrosis factor [*TNF*], interleukin-17 [*IL-17*], chemokines) and others that activate macrophages (interferon- γ [*IFN-γ*]). Activated macrophages in turn stimulate T cells by presenting antigens and via cytokines such as IL-12. Prolonged reactions involving T cells and macrophages may result in granuloma formation.

Other Cells in Chronic Inflammation

Other cell types may be prominent in chronic inflammation induced by particular stimuli.

- **Eosinophils** are abundant in immune reactions mediated by Th2 cells, such as parasitic infections and some allergies (Fig. 3.21). Their recruitment is driven by adhesion molecules similar to those used by neutrophils and by specific chemokines (e.g., eotaxin) derived from leukocytes and epithelial cells. Eosinophils have granules that contain major basic protein, a highly cationic protein that is toxic to helminths but also may injure host epithelial cells. This is why eosinophils are of benefit in controlling helminth infections, yet they also contribute to tissue damage in immune reactions such as allergies (Chapter 6).
- **Mast cells** are widely distributed in connective tissues and participate in both acute and chronic inflammatory reactions. A receptor called Fc ϵ RI that binds the Fc portion of IgE antibody is expressed on the surface of mast cells. In immediate hypersensitivity reactions, IgE antibodies bound to the mast cells' Fc receptors specifically

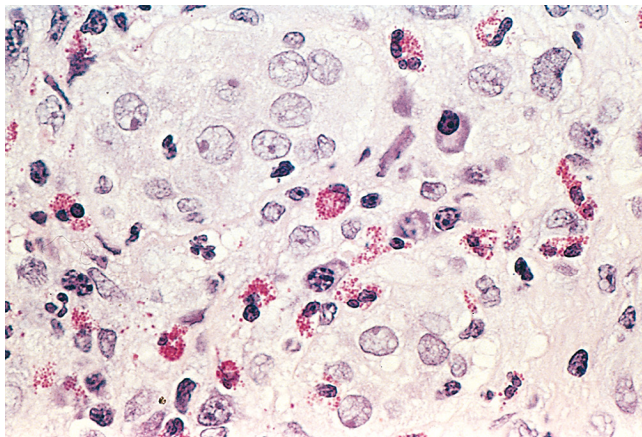


Fig. 3.21 Focus of inflammation containing numerous eosinophils.

recognize antigen, and the cells respond to receptor engagement by degranulating and releasing mediators such as histamine and prostaglandins (Chapter 6). This type of response occurs during allergic reactions to foods, insect venom, or drugs, sometimes with catastrophic results (e.g., anaphylactic shock). Mast cells are also present in chronic inflammatory reactions, and because they secrete a plethora of cytokines, they may promote inflammatory reactions in different situations.

- Although *neutrophils* are characteristic of acute inflammation, many forms of chronic inflammation continue to show neutrophils, induced either by persistent microbes or by mediators produced by activated macrophages and T lymphocytes. In chronic bacterial infection of bone (osteomyelitis), a neutrophilic exudate can persist for many months. Neutrophils are also important in the chronic damage induced in lungs by smoking and other irritant stimuli (Chapter 15).

Granulomatous Inflammation

Granulomatous inflammation is a form of chronic inflammation characterized by collections of activated macrophages, often with T lymphocytes, and sometimes associated with necrosis. The name granuloma derives from the granular macroscopic appearance of these inflammatory foci. Granuloma formation is a cellular attempt to contain an offending agent that is difficult to eradicate; it often involves strong activation of macrophages and T lymphocytes, which can cause injury to normal tissues. The activated macrophages may develop abundant cytoplasm and begin to resemble epithelial cells, and hence are called **epithelioid cells**. Some activated macrophages may fuse, forming multinucleate giant cells.

There are two types of granulomas, which differ in their pathogenesis:

- *Foreign body granulomas* are reactions to inert foreign bodies, which induce inflammation in the absence of T cell–mediated immune responses. Typically, foreign body granulomas form around materials such as talc (associated with intravenous drug use) (Chapter 9), suture material, or other fibers that are large enough to preclude phagocytosis by macrophages and are not immunogenic, so they do not incite any specific immune response. Epithelioid cells and giant cells are apposed to the surface of the foreign body. The foreign material can usually be identified in the center of the granuloma, sometimes within the giant cells, particularly if viewed with polarized light, in which it may appear refractile.
- *Immune granulomas* are caused by a variety of agents that can induce a persistent T cell–mediated immune response. This type of

immune response usually produces granulomas when the inciting agent is a persistent microbe or other chronic stimulus that activates T cells. In such responses, Th1 cells produce cytokines such as IFN- γ , which activates macrophages (see Fig. 3.20). In some parasitic infections, such as schistosomiasis, the granulomas are associated with strong Th2 responses and eosinophils.

MORPHOLOGY

In the usual hematoxylin and eosin preparations, the activated macrophages in granulomas have pink granular cytoplasm with indistinct cell boundaries and are called **epithelioid cells** (Fig. 3.22). The aggregates of epithelioid macrophages are surrounded by a collar of lymphocytes. Older granulomas may have a rim of fibroblasts and connective tissue. Frequently, but not invariably, multinucleated **giant cells** 40 to 50 μm in diameter are found in granulomas; these are called **Langhans giant cells**. They consist of a large mass of cytoplasm and many nuclei, and they derive from the fusion of multiple activated macrophages. In granulomas associated with certain infectious organisms (most classically *Mycobacterium tuberculosis*), a combination of hypoxia and free radical–mediated injury leads to a central zone of necrosis. Grossly, this has a granular, cheesy appearance and is therefore called **caseous necrosis**. Microscopically, this necrotic material appears as amorphous, structureless, eosinophilic, granular debris, with complete loss of cellular details (as opposed to coagulative necrosis, in which cell outlines are preserved). The granulomas in Crohn's disease, sarcoidosis, and foreign body reactions tend to not have necrotic centers and are said to be noncaseating. Healing of granulomas is accompanied by fibrosis that may be extensive in involved organs.

Recognition of granulomatous inflammation is important because of the limited number of conditions (some life-threatening) that cause it (Table 3.9). **Tuberculosis is the prototype of a granulomatous disease caused by infection and should always be excluded as the cause when granulomas are identified.** In this disease the granulomas are referred to as tubercles. Granulomas may also develop in some immune-mediated inflammatory diseases. Notable among these are Crohn's disease, one type of inflammatory bowel disease and an important cause of granulomatous inflammation in the United States, and sarcoidosis, a disorder of unknown etiology. The morphologic patterns in the various granulomatous diseases may be sufficiently different to allow reasonably accurate diagnosis by an experienced pathologist

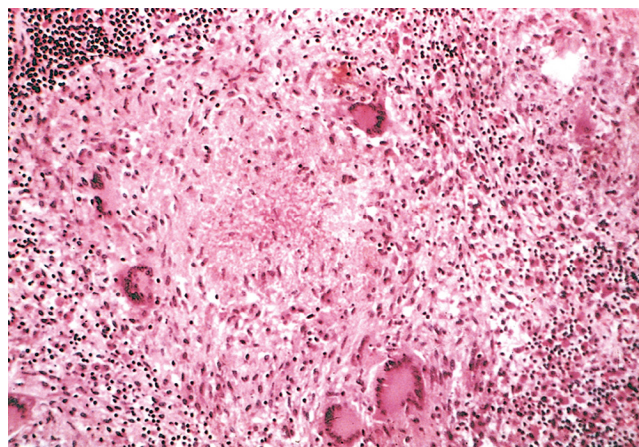


Fig. 3.22 Granulomatous inflammation. Typical tuberculous granuloma showing an area of central necrosis surrounded by multiple giant cells, epithelioid cells, and lymphocytes.

TABLE 3.9 Examples of Diseases With Granulomatous Inflammation

Disease	Etiology	Tissue Reaction
Tuberculosis	<i>Mycobacterium tuberculosis</i> infection	Caseating granulomas (tubercles): foci of activated macrophages (epithelioid cells), rimmed by fibroblasts, lymphocytes, histiocytes, occasional Langhans giant cells; central necrosis with amorphous granular debris; acid-fast bacilli
Leprosy	<i>Mycobacterium leprae</i> infection	Acid-fast bacilli in macrophages; noncaseating granulomas
Syphilis	<i>Treponema pallidum</i> infection	Gumma: microscopic to grossly visible lesion, enclosing wall of histiocytes; plasma cell infiltrate; central cells are necrotic without loss of cellular outline
Cat-scratch disease	<i>Bartonella henselae</i> (gram-negative bacillus) infection	Rounded or stellate granuloma containing central granular debris and recognizable neutrophils; giant cells uncommon
Sarcoidosis	Unknown etiology	Noncaseating granulomas with abundant activated macrophages
Crohn's disease (inflammatory bowel disease)	Immune reaction against intestinal bacteria, possibly self-antigens	Occasional noncaseating granulomas in the wall of the intestine, with dense chronic inflammatory infiltrate

(see Table 3.9); however, there are so many atypical presentations that it is always necessary to identify the specific etiologic agent by special stains for organisms (e.g., acid-fast stains for tubercle bacilli), by culture methods (e.g., in tuberculosis and fungal diseases), by molecular techniques (e.g., the polymerase chain reaction in tuberculosis), and by serologic studies (e.g., in syphilis).

KEY CONCEPTS

Chronic Inflammation

- Chronic inflammation is a prolonged host response to persistent stimuli.
- It is caused by microbes that resist elimination, immune responses against self-antigens and environmental antigens, and some toxic substances (e.g., silica), and it underlies many medically important diseases.
- It is characterized by coexisting inflammation, tissue injury, and attempted repair by scarring.
- The cellular infiltrate consists of macrophages, lymphocytes, plasma cells, and other leukocytes.
- It is mediated by cytokines produced by macrophages and lymphocytes (notably T lymphocytes); bidirectional interactions between these cells tend to amplify and prolong the inflammatory reaction.
- Granulomatous inflammation is a pattern of chronic inflammation induced by T-cell and macrophage activation in response to an agent that is resistant to eradication.

SYSTEMIC EFFECTS OF INFLAMMATION

Inflammation, even if localized, is associated with cytokine-induced systemic reactions. Anyone who has suffered through a severe bout of a viral illness (e.g., influenza) has experienced the systemic manifestations of acute inflammation. These changes are reactions to cytokines whose production is stimulated by bacterial products and by other inflammatory stimuli. The cytokines TNF, IL-1, and IL-6 are important mediators of the systemic reaction; other cytokines, notably interferons, also contribute.

The systemic response in inflammation consists of several clinical and pathologic changes.

- Fever**, characterized by an elevation of body temperature, usually by 1°C to 4°C, is one of the most prominent manifestations of the systemic response, especially when inflammation is associated with infection. Substances that induce fever are called *pyrogens* and include bacterial products (exogenous pyrogens, e.g., LPS) and cytokines, principally IL-1 and TNF (called endogenous pyrogens). Exogenous pyrogens act by stimulating immune cells to release

IL-1 and TNF, which upregulate cyclooxygenases, the enzymes that synthesize prostaglandins. Among the cells that respond to IL-1 and TNF are vascular and perivascular cells of the hypothalamus. Prostaglandins released by these cells, especially PGE₂, stimulate the production of neurotransmitters by the hypothalamus that reset the body's steady-state temperature to a higher level by reducing heat loss (via vasoconstriction) and increasing heat generation (through effects on brown fat and skeletal muscle). NSAIDs, including aspirin, reduce fever by inhibiting prostaglandin synthesis. An elevated body temperature has been shown to help amphibians ward off microbial infections, and it is possible that fever is a protective host response in mammals as well.

- Leukocytosis** is a common feature of inflammatory reactions, especially those induced by bacterial infections. The leukocyte count usually climbs to 15,000 or 20,000 cells/mL, but sometimes it may reach extraordinarily high levels of 40,000 to 100,000 cells/mL. These extreme elevations are sometimes called *leukemoid reactions* because they are similar to the white blood cell counts observed in leukemia, from which they must be distinguished. The leukocytosis occurs initially because of accelerated release of granulocytes from the bone marrow (caused by cytokines, including TNF and IL-1) and is therefore associated with a rise in the number of both mature and immature neutrophils in the blood; the presence of immature cells is referred to as a shift to the left (for arcane reasons having to do with the way these cells were counted manually by technicians in days past). Prolonged infection also induces proliferation of precursors in the bone marrow, caused by increased production of colony-stimulating factors (CSFs) mainly from activated macrophages and marrow stromal cells. The increase in the bone marrow output of leukocytes compensates for the loss of these cells in the inflammatory reaction. (See also the discussion of leukocytosis in Chapter 13.) Most bacterial infections induce an increase in the blood neutrophil count, called neutrophilia. Viral infections, such as infectious mononucleosis, mumps, and German measles, cause an absolute increase in the number of lymphocytes (lymphocytosis). In some allergies and helminth infestations, there is an increase in the absolute number of eosinophils, creating eosinophilia. Certain infections (typhoid fever and infections caused by some viruses, rickettsiae, and certain protozoa) are associated with fewer circulating white cells (leukopenia), in part because of sequestration of activated leukocytes in vascular spaces and tissues.
- The **acute-phase response** consists of the production of proteins called acute-phase proteins. They are mostly synthesized in the liver, and their plasma concentrations may increase several hundred-fold as part of the response to inflammatory stimuli. Despite their name, plasma levels of these proteins also increase in chronic inflammation.

Three of the best known of the proteins are C-reactive protein (CRP), fibrinogen, and serum amyloid A (SAA) protein. Synthesis of these molecules in hepatocytes is stimulated by cytokines, especially IL-6 (for CRP and fibrinogen) and IL-1 or TNF (for SAA). Many acute-phase proteins, such as CRP and SAA, bind to microbial cell walls, and they may act as opsonins and fix complement. Some of these proteins bind phospholipids and nuclear components, possibly aiding in clearing apoptotic cells and fragments. Fibrinogen binds to red cells and causes them to form stacks (rouleaux) that sediment more rapidly at unit gravity than do individual red cells. This is the basis for measuring the *erythrocyte sedimentation rate*, a simple test for an inflammatory response caused by any stimulus. Some of these proteins may also promote tissue repair. Prolonged production of acute-phase proteins (especially SAA) in states of chronic inflammation may cause secondary amyloidosis (Chapter 6). Elevated serum levels of CRP have been proposed as a marker for increased risk of myocardial infarction in patients with coronary artery disease. It is postulated that inflammation involving atherosclerotic plaques in the coronary arteries may predispose to thrombosis and subsequent infarction. Two other liver proteins that are released in increased amounts as part of the acute-phase response often lead to altered blood counts. *Hepcidin* is a small protein that regulates the uptake of iron from the gut and the release of iron from macrophages, which recycle iron from effect red cells. Chronic inflammation and the attendant elevation of hepcidin sequesters iron from red cell progenitors, leading to anemia (Chapter 14). *Thrombopoietin*, the major growth factor for megakaryocytes (platelet precursors) in the bone marrow, is also upregulated and as a result systemic inflammation may be associated with an elevated platelet count (thrombocytosis).

- Other manifestations of the systemic response include increased pulse and blood pressure; decreased sweating, mainly because of redirection of blood flow from cutaneous to deep vascular beds, to minimize heat loss through the skin; rigors (shivering), chills (search for warmth), anorexia, somnolence, and malaise, probably because of the actions of cytokines on brain cells.
- In severe bacterial infections (sepsis), the large amounts of bacteria and their products in the blood stimulate the production of enormous quantities of several cytokines, notably TNF and IL-1. High blood levels of cytokines cause various clinical manifestations such as disseminated intravascular coagulation, hypotensive shock, and metabolic disturbances, including insulin resistance and hyperglycemia. This clinical triad is known as *septic shock* and is discussed in more detail in Chapter 4.

KEY CONCEPTS

Systemic Effects of Inflammation

- Fever: cytokines (TNF, IL-1) stimulate production of prostaglandins in the hypothalamus.
- Leukocytosis: caused by release of cells from the bone marrow; cytokines (colony-stimulating factors) stimulate production of leukocytes from precursors in the bone marrow.
- Production of acute-phase proteins: C-reactive protein, others; synthesis stimulated by cytokines (IL-6, others) acting on liver cells.
- In some severe infections, septic shock: fall in blood pressure, disseminated intravascular coagulation, metabolic abnormalities; induced by high levels of TNF and other cytokines.

Whereas excessive inflammation is the underlying cause of many human diseases, defective inflammation results mainly in increased susceptibility to infections. The most common cause of

defective inflammation is leukocyte deficiency due to replacement of the bone marrow by leukemias and metastatic tumors and suppression of the marrow by therapies for cancer and graft rejection. Inherited genetic abnormalities of leukocyte adhesion and microbicidal function are rare but informative; these are described in Chapter 6, in the context of immunodeficiency diseases. Deficiencies of the complement system are mentioned earlier and are described further in Chapter 6.

TISSUE REPAIR

Repair, also called healing, refers to the restoration of tissue architecture and function after an injury. By convention, the term *repair* is used for parenchymal and connective tissues and *healing* for surface epithelia, but these distinctions are not based on biology, and we use the terms interchangeably. Critical to the survival of an organism is the ability to repair the damage caused by toxic insults and inflammation. Hence the inflammatory response to microbes and injured tissues not only serves to eliminate these dangers but also sets into motion the process of repair.

Repair of damaged tissues occurs by two processes: regeneration, which restores normal cells, and scarring, the deposition of connective tissue (Fig. 3.23).

- **Regeneration.** Some tissues can replace the damaged components and essentially return to a normal state; this process is called *regeneration*. Regeneration may occur by proliferation of differentiated cells that survive the injury and retain the capacity to proliferate, such as hepatocytes in the liver. In other tissues, particularly the epithelia of the skin and intestines, tissue stem

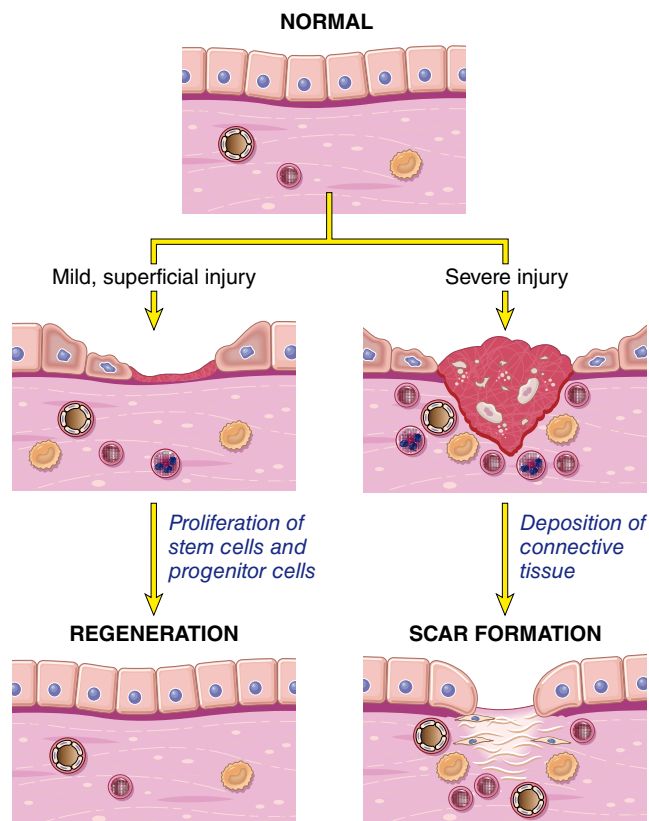


Fig. 3.23 Mechanisms of tissue repair: regeneration and scar formation. Following mild injury, which damages the epithelium but not the underlying tissue, resolution occurs by regeneration. More severe injury damages the connective tissue, and repair is by scar formation.

cells and their progenitors contribute to the restoration of damaged tissues. However, mammals have a limited capacity to regenerate most damaged tissues and organs, and only some components of these tissues can fully restore themselves.

- **Scar formation.** When the injured tissues are incapable of complete restitution, or when the supporting structures of the tissue are damaged, repair occurs by the laying down of connective (fibrous) tissue, a process known as scarring. Although the fibrous scar is not normal, it usually provides enough structural stability that the injured tissue is able to function. The term *fibrosis* is often used to describe the deposition of collagen that occurs in the lungs, liver, kidney, and other organs as a consequence of chronic inflammation or in the myocardium after extensive ischemic necrosis (infarction). If fibrosis develops in a tissue space occupied by an inflammatory exudate, it is called *organization* (as in organizing pneumonia affecting the lung).

After many common types of injury, both regeneration and scar formation contribute in varying degrees to the ultimate repair. Both processes involve the proliferation of cells and close interactions between cells and the ECM. We first discuss the general mechanisms of cellular proliferation and regeneration, then the salient features of scar formation.

Cell and Tissue Regeneration

The regeneration of injured cells and tissues involves cell proliferation, which is driven by growth factors and is critically dependent on the integrity of the ECM, and by the development of mature cells from tissue stem cells. Several cell types proliferate during tissue repair. These include the remnants of the injured tissue (which attempt to restore normal structure), vascular endothelial cells (to create new vessels that provide the nutrients needed for the repair process), and fibroblasts (the source of the fibrous tissue that forms the scar to fill defects that cannot be corrected by regeneration). Before describing examples of repair by regeneration, the general principles of cell proliferation are discussed.

The ability of tissues to repair themselves is determined, in part, by their intrinsic proliferative capacity. Based on these criteria, the tissues of the body are divided into three groups.

- **Labile (continuously dividing) tissues.** Cells of these tissues are continuously being lost and replaced by maturation from tissue stem cells and by proliferation of mature cells. Labile cells include hematopoietic cells in the bone marrow and the majority of surface epithelia, such as the stratified squamous epithelia of the skin, oral cavity, esophagus, vagina, and cervix; the cuboidal epithelia of the ducts draining exocrine organs (e.g., salivary glands, pancreas, biliary tract); the columnar epithelium of the gastrointestinal tract, uterus, and fallopian tubes; and the transitional epithelium of the urinary tract. These tissues can readily regenerate after injury as long as the pool of stem cells is preserved.
- **Stable tissues.** Cells of these tissues are quiescent (in the G_0 stage of the cell cycle) and have only minimal proliferative activity in their normal state. However, these cells can divide in response to injury or loss of tissue mass. Stable cells constitute the parenchyma of most solid organs, such as liver, kidney, and pancreas. They also include endothelial cells, fibroblasts, and smooth muscle cells; the proliferation of these cells is particularly important in wound healing. With the exception of the liver, stable tissues have a limited capacity to regenerate after injury.
- **Permanent tissues.** The cells of these tissues are considered to be terminally differentiated and nonproliferative in postnatal life. Most neurons and cardiac muscle cells belong to this category. Thus, injury to the brain or heart is irreversible and results in a scar because neurons and cardiac myocytes cannot regenerate. Limited

stem cell replication and differentiation occur in some areas of the adult brain, and there is evidence that heart muscle cells may proliferate after myocardial necrosis. Nevertheless, whatever proliferative capacity exists in these tissues, it is insufficient to produce tissue regeneration after injury. Skeletal muscle is usually classified as a permanent tissue, but satellite cells attached to the endomysial sheath provide some regenerative capacity for muscle. In permanent tissues, repair is typically dominated by scar formation.

Cell proliferation is driven by signals provided by growth factors and from the ECM. Many different growth factors have been described; some act on multiple cell types, and others are cell-type specific (see Table 1.1 in Chapter 1). Growth factors are typically produced by cells near the site of damage. The most important sources of these growth factors are macrophages that are activated by tissue injury, but epithelial and stromal cells also produce some of these factors. Several growth factors are displayed at high concentrations bound to ECM proteins. All growth factors activate signaling pathways that stimulate DNA replication (Chapter 1) and also foster changes in metabolism that promote the biosynthesis of other cellular components (membranes, organelles, proteins) needed for a “mother” cell to produce two daughter cells. In addition to responding to growth factors, cells use integrins to bind to ECM proteins, and signals from the integrins can also stimulate cell proliferation.

In the process of regeneration, proliferation of residual cells is supplemented by development of mature cells from stem cells. Stem cells were described in Chapter 1. Tissue stem cells live in specialized niches, and injury triggers signals that stimulate their proliferation and differentiation into mature cells that repopulate the injured tissue. Their contribution to regeneration of injured tissues is greatest when the differentiated cells that survive the injury have limited or no intrinsic proliferative capacity.

Liver Regeneration

The liver has a unique capacity to regenerate, as demonstrated by its growth after partial hepatectomy, which may be performed for tumor resection or for living-donor hepatic transplantation. It is, therefore, an informative model for tissue regeneration. The mythologic image of liver regeneration is found in the story of Prometheus, whose liver was eaten every day by an eagle sent by Zeus as punishment for stealing the secret of fire and grew back overnight. The reality, although less dramatic, is still quite impressive.

Regeneration of the liver occurs by the same two mechanisms described earlier: proliferation of remaining hepatocytes and repopulation from stem cells. Which mechanism plays the dominant role depends on the nature of the injury.

- **Proliferation of hepatocytes following partial hepatectomy.** In humans, up to 90% of the liver can be regenerated by the proliferation of residual hepatocytes. Hepatocyte proliferation in the regenerating liver is triggered by the combined actions of cytokines and polypeptide growth factors. Cytokines such as IL-6 produced mainly by Kupffer cells act on hepatocytes to make the parenchymal cells competent to receive and respond to growth factor signals. Growth factors such as hepatocyte growth factor (HGF) and TGF- α , produced by many cell types, act on the primed hepatocytes to stimulate their proliferation.
- **Liver regeneration from stem cells.** In situations in which the proliferative capacity of hepatocytes is impaired, such as after chronic liver injury or inflammation, stem cells in the liver contribute to repopulation. Some of these stem cells reside in specialized niches called *canals of Hering*, where bile canaliculi connect with larger bile ducts.

KEY CONCEPTS

Repair by Regeneration

- Different tissues consist of continuously dividing labile cells (epithelia, hematopoietic), normally quiescent stable cells capable of proliferation (most parenchymal organs), and nondividing permanent cells (neurons, skeletal and cardiac muscle). The regenerative capacity of a tissue depends on the proliferative potential of its constituent cells.
- Continuously dividing labile tissues contain stem cells that differentiate to replenish lost cells and maintain tissue homeostasis.
- Cell proliferation is stimulated by growth factors and interactions of cells with the ECM.
- Regeneration of the liver is a classic example of repair by regeneration. It is triggered by cytokines and growth factors produced in response to loss of liver mass and inflammation. In different situations, regeneration may occur by proliferation of surviving hepatocytes or repopulation from progenitor cells.

Repair by Scarring

If repair cannot be accomplished by regeneration alone, it occurs by replacement of the injured cells with connective tissue, leading to the formation of a scar. Scarring may be prominent in stable or permanent tissues, whose cells have a limited capacity to proliferate, or if there is damage to the connective tissue framework, which normally provides the scaffold to support regenerating cells. In contrast to regeneration, which involves the restitution of tissue components, scar formation is a response that “patches” rather than restores the tissue. The term *scar* is most often used in connection to wound healing in the skin but may also be used to describe the replacement of parenchymal cells in any tissue by collagen, as in the heart after myocardial infarction.

Steps in Scar Formation

Repair by connective tissue deposition consists of sequential processes that follow tissue injury, best illustrated by healing of skin wounds. Within minutes after injury, a hemostatic plug composed of platelets (Chapter 4) is formed, which stops bleeding and provides a scaffold for the formation of a stable clot. The subsequent steps are summarized (Fig. 3.24):

- **Inflammation.** Over the next 6 to 48 hours, neutrophils and then monocytes are recruited to the site to eliminate the offending

agents and clear the debris. As the injurious agents and necrotic cells are cleared, the inflammation resolves.

- **Cell proliferation.** In the next stage, which takes up to 10 days, several cell types, including epithelial cells, endothelial and other vascular cells, and fibroblasts, proliferate and migrate to close the now clean wound. Each cell type serves unique functions.
 - *Epithelial cells* respond to locally produced growth factors and migrate over the wound to cover it up.
 - *Endothelial and other vascular cells* proliferate to form new blood vessels, a process known as *angiogenesis*. Because of the importance of this process in physiologic host responses and in many pathologic conditions, it is described in more detail subsequently.
 - *Fibroblasts* proliferate and migrate into the site of injury and lay down collagen fibers that form the scar.
- **Formation of granulation tissue.** Migration and proliferation of fibroblasts and deposition of loose connective tissue, together with the vessels and interspersed mononuclear leukocytes, form granulation tissue (not to be confused with granuloma). The term *granulation tissue* derives from the pink, soft, granular gross appearance, such as that seen beneath the scab of a skin wound. Its histologic appearance is characterized by proliferation of fibroblasts and new thin-walled, delicate capillaries (angiogenesis) in a loose ECM, often with admixed inflammatory cells, mainly macrophages (Fig. 3.25A). Granulation tissue progressively fills the site of injury; the amount of granulation tissue that is formed depends on the size of the tissue defect created by the wound and the intensity of inflammation.
- **Deposition of connective tissue.** Granulation tissue is progressively replaced by deposition of collagen. The amount of connective tissue increases in the granulation tissue, eventually resulting in the formation of a stable fibrous scar (Fig. 3.25B).

Macrophages play a central role in repair by clearing offending agents and dead tissue, providing growth factors for the proliferation of various cells, and secreting cytokines that stimulate fibroblast proliferation and connective tissue synthesis and deposition. The macrophages that are involved in repair are mostly of the alternatively activated (M2) type. It is not clear how the classically activated (M1) macrophages, which dominate during inflammation and are involved in getting rid of microbes and dead tissues are gradually replaced by

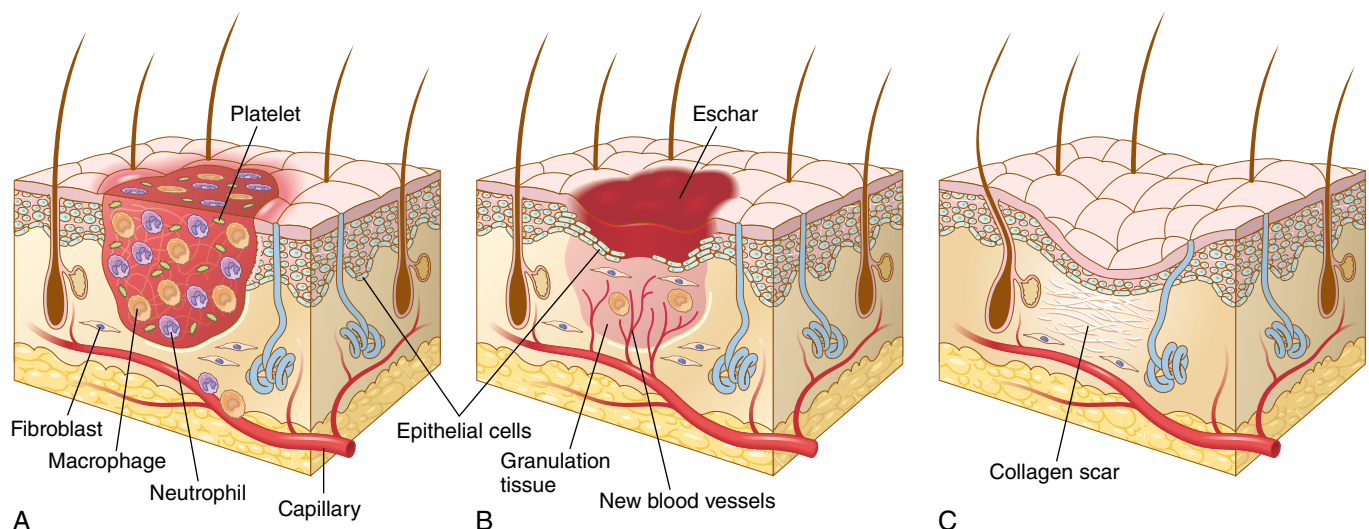


Fig. 3.24 Steps in repair by scar formation: wound healing in the skin. (A) Inflammation. (B) Proliferation of epithelial cells; formation of granulation tissue by vessel growth and proliferating fibroblasts. (C) Remodeling to produce the fibrous scar.

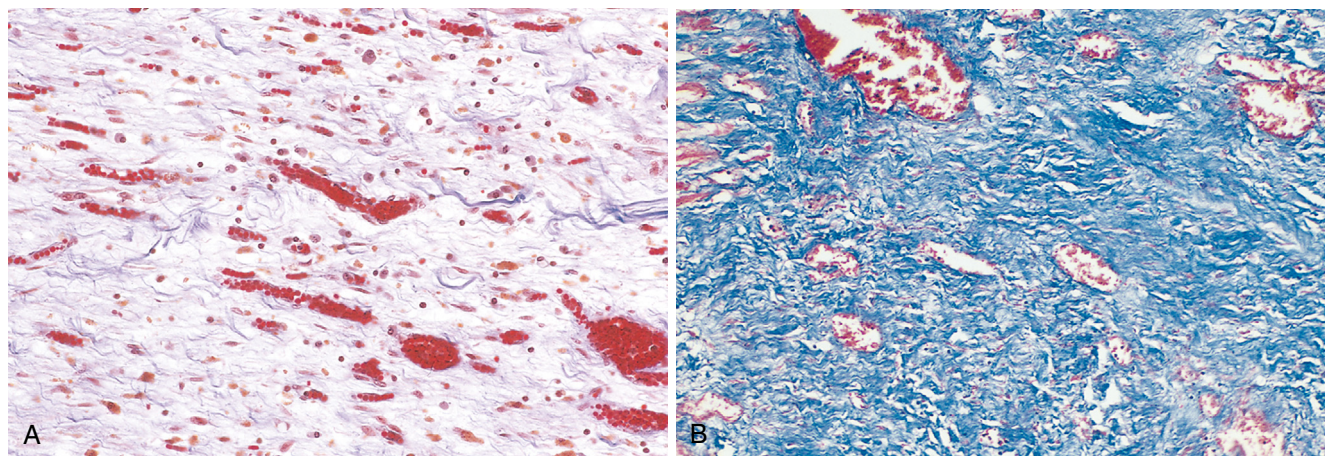


Fig. 3.25 (A) Granulation tissue showing numerous blood vessels, edema, and a loose extracellular matrix containing occasional inflammatory cells. Collagen is stained blue by the trichrome stain; minimal mature collagen can be seen at this point. (B) Trichrome stain of mature scar, showing dense collagen, with only scattered vascular channels.

alternatively activated macrophages that serve to terminate inflammation and induce repair.

Angiogenesis

Angiogenesis is the process of new blood vessel development from existing vessels. It is critical in healing at sites of injury, in the development of collateral circulations at sites of ischemia, and in allowing tumors to increase in size beyond the constraints of their original blood supply. Much work has been done to understand the mechanisms underlying angiogenesis, and therapies to either augment the process (e.g., to improve blood flow to a heart ravaged by coronary atherosclerosis) or inhibit it (to frustrate tumor growth or block pathologic vessel growth such as in diabetic retinopathy) have been developed.

Angiogenesis involves sprouting of new vessels from existing ones and consists of (Fig. 3.26):

- *Vasodilation* in response to nitric oxide (NO) and increased permeability induced by vascular endothelial growth factor (VEGF).
- *Separation of pericytes* from the abluminal surface and breakdown of the basement membrane to allow formation of a vessel sprout.
- *Migration of endothelial cells* toward the area of tissue injury.
- *Proliferation of endothelial cells* just behind the leading front (“tip”) of migrating cells.
- *Remodeling into capillary tubes.*
- *Recruitment of periendothelial cells* (pericytes for small capillaries and smooth muscle cells for larger vessels) to form the mature vessel.
- *Suppression of endothelial proliferation* and migration and deposition of the basement membrane.

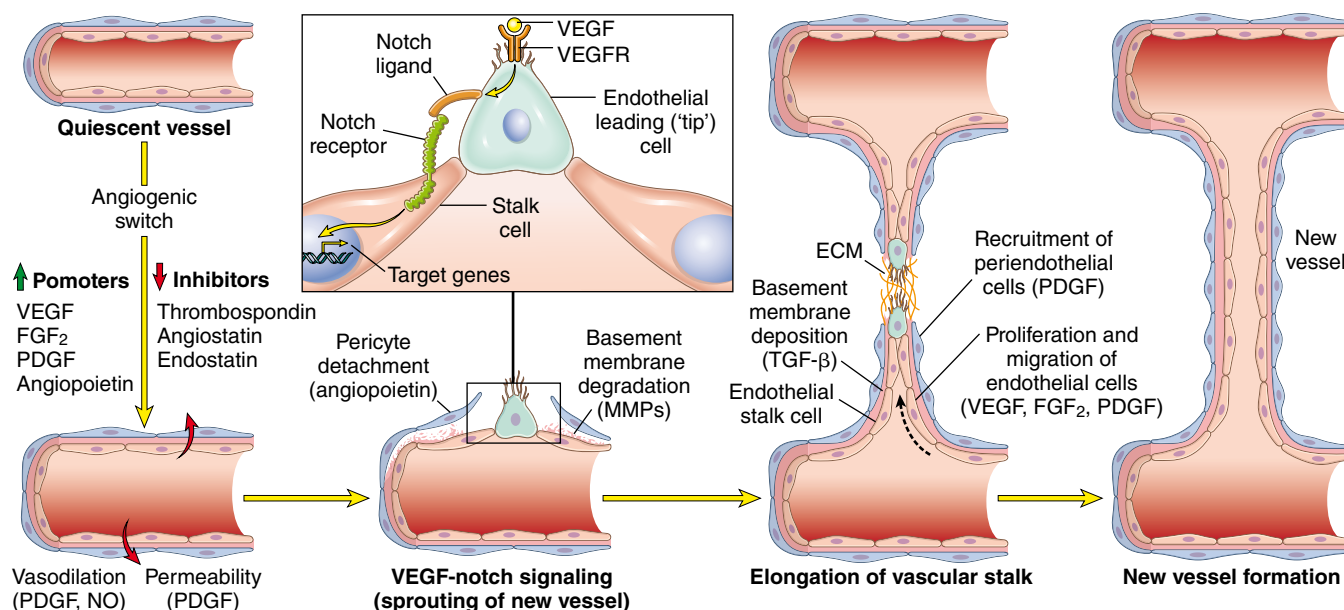


Fig. 3.26 Angiogenesis. In tissue repair, angiogenesis occurs mainly by sprouting of new vessels. The steps in the process and the major signals involved are illustrated. The newly formed vessel joins up with other vessels (not shown) to form the new vascular bed. *ECM*, Extracellular matrix; *FGF*, fibroblast growth factor; *MMPs*, matrix metalloproteinases; *NO*, nitric oxide; *PDGF*, platelet-derived growth factor; *TGF*, transforming growth factor; *VEGF*, vascular endothelial growth factor.

Mechanisms of Angiogenesis. The process of angiogenesis involves several signaling pathways, cell-cell interactions, ECM proteins, and tissue enzymes. *VEGFs*, mainly VEGF-A (Chapter 1), stimulate both migration and proliferation of endothelial cells, thus initiating the process of capillary sprouting in angiogenesis (sometimes called the angiogenic switch). VEGF promotes vasodilation by stimulating the production of NO and contributes to the formation of the vascular lumen. Fibroblast growth factors (FGFs), mainly FGF-2, stimulate the proliferation of endothelial cells. They also promote the migration of macrophages and fibroblasts to the damaged area and stimulate epithelial cell migration to cover epidermal wounds. Angiopoietins 1 and 2 (Ang 1 and Ang 2) are growth factors that play a role in angiogenesis and the structural maturation of new vessels. Newly formed vessels need to be stabilized by pericytes and smooth muscle cells and by the deposition of connective tissue. Platelet-derived growth factor (PDGF) and TGF- β also participate in the stabilization process: PDGF recruits smooth muscle cells and TGF- β suppresses endothelial proliferation and migration and enhances the production of ECM proteins.

The Notch signaling pathway regulates the sprouting and branching of new vessels and thus ensures that the new vessels that are formed have the proper spacing to effectively supply the healing tissue with blood. VEGF stimulates the expression of Notch ligands, which bind to the Notch receptor on endothelial cells and regulate the pattern of vessel branching.

ECM proteins participate in the process of vessel sprouting in angiogenesis, largely through interactions with integrin receptors in endothelial cells and by providing the scaffold for vessel growth. Enzymes in the ECM, notably the matrix metalloproteinases (MMPs), degrade the ECM to permit remodeling and extension of the vascular tube.

Deposition of Connective Tissue

The laying down of connective tissue occurs in two steps: migration and proliferation of fibroblasts into the site of injury, and deposition of ECM proteins produced by these cells (Fig. 3.27). These processes are orchestrated by locally produced cytokines and growth factors including PDGF, FGF-2, and TGF- β . The major sources of these factors are inflammatory cells, particularly alternatively activated (M2) macrophages, which are present at sites of injury and in granulation tissue. Sites of inflammation are also rich in mast cells, and in the appropriate chemotactic milieu, lymphocytes may also be present. Each of these can secrete cytokines and growth factors that contribute to fibroblast proliferation and activation.

TGF- β is the most important cytokine for the synthesis and deposition of connective tissue proteins. It is produced by most of the cells in granulation tissue, including alternatively activated macrophages. The levels of TGF- β in tissues are primarily regulated not by the transcription of the gene but by the posttranscriptional activation of latent TGF- β , the rate of secretion of the active molecule, and molecules that make up or that interact with the ECM, notably integrins and microfibrils, that enhance or diminish TGF- β activity. TGF- β stimulates fibroblast migration and proliferation, increases synthesis of collagen and fibronectin, and decreases degradation of ECM by inhibiting metalloproteinases. TGF- β is involved not only in scar formation after injury but also in the development of fibrosis in the lung, liver, and kidneys in response to chronic inflammation. TGF- β is also an antiinflammatory cytokine that serves to limit and terminate inflammatory responses. It does this by inhibiting lymphocyte proliferation and the activity of other leukocytes.

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 deposition is critical for the development of strength

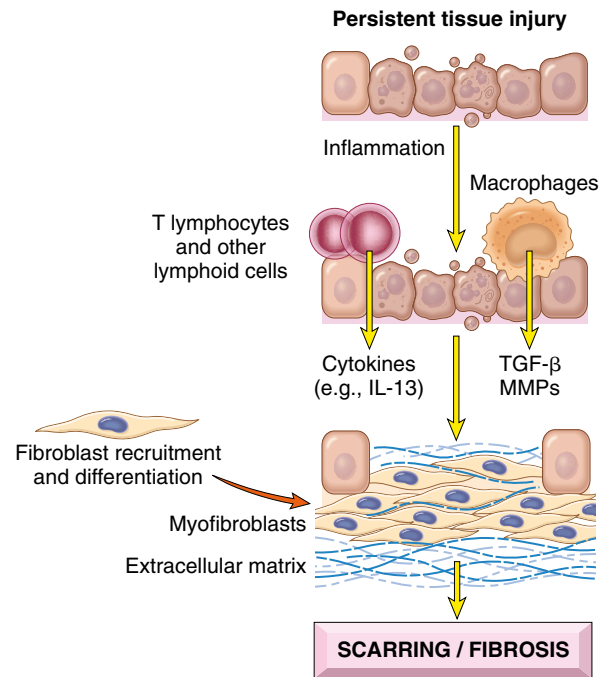


Fig. 3.27 Mechanisms of connective tissue deposition. Persistent tissue injury leads to chronic inflammation and loss of tissue architecture. Cytokines produced by macrophages and other leukocytes stimulate the migration and proliferation of fibroblasts and myofibroblasts and the deposition of collagen and other extracellular matrix proteins. The net result is replacement of normal tissue by connective tissue. *IL*, Interleukin; *MMPs*, matrix metalloproteinases; *TGF*, transforming growth factor.

in a healing wound site. As the scar matures, there is progressive vascular regression, which eventually transforms the highly vascularized granulation tissue into a pale, largely avascular scar. Some of the fibroblasts also acquire features of smooth muscle cells, including the presence of actin filaments, and are called *myofibroblasts*. These cells contribute to the contraction of the scar over time.

Remodeling of Connective Tissue

The outcome of the repair process is influenced by a balance between synthesis and degradation of ECM proteins. After its deposition, the connective tissue in the scar continues to be modified and remodeled. The degradation of collagens and other ECM components is accomplished by a family of matrix metalloproteinases (MMPs), so called because they are dependent on metal ions (e.g., zinc) for their activity. MMPs include interstitial collagenases (MMP-1, MMP-2, and MMP-3), which cleave fibrillar collagen; gelatinases, which degrade amorphous collagen and fibronectin; and stromelysins, which degrade a variety of ECM constituents, including proteoglycans, laminin, fibronectin, and amorphous collagen.

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. The activity of the MMPs is tightly controlled. They are produced as inactive precursors (zymogens) that must be activated; this is accomplished by proteases (e.g., plasmin) likely to be present only at sites of injury. In turn, active MMPs can be rapidly inhibited by specific *tissue inhibitors of metalloproteinases (TIMPs)*, produced by most mesenchymal cells. Thus, during scar formation, MMPs are activated to remodel the deposited ECM, and then their activity is shut down by the TIMPs.

KEY CONCEPTS

Repair by Scar Formation

- Tissues are repaired by replacement with connective tissue and scar formation if the injured tissue is not capable of proliferation or if the structural framework is damaged and cannot support regeneration.
- The main steps in repair by scarring are clot formation, inflammation, angiogenesis with formation of granulation tissue, migration and proliferation of fibroblasts, collagen synthesis, and connective tissue remodeling.
- Macrophages are critical for orchestrating the repair process, by eliminating offending agents and producing cytokines and growth factors that stimulate the proliferation of the cell types involved in repair.
- TGF- β is a potent fibrogenic agent; ECM deposition depends on the balance between fibrogenic agents, MMPs that digest ECM, and TIMPs.

Healing of Skin Wounds

Many of the principles of repair by regeneration or scarring that we have described are well illustrated by the healing of skin wounds because the process can be readily observed and defined with precision. Skin wound healing can be classified into two types depending on the extent of tissue injury and the dominant mechanism of healing (Table 3.10).

- Healing by first intention (primary union).* This occurs when there is minimal scarring and restitution is by epithelial regeneration. It is typically seen in well-apposed surgical incisions.
- Healing by second intention (secondary union).* In larger wounds, especially if they are infected, healing is by a combination of regeneration and scarring. The process follows a characteristic sequence (Table 3.11).

Factors That Influence Tissue Repair

Tissue repair may be altered by several factors, that impact the quality or adequacy of the reparative process (Fig. 3.28). Variables that modify healing may be extrinsic (e.g., infection) or intrinsic to the injured tissue and systemic or local:

- Infection* is clinically one of the most important causes of delayed healing; it prolongs inflammation and potentially increases the local tissue injury.
- Diabetes* is a metabolic disease that compromises tissue repair for many reasons (Chapter 24) and is one of the most important systemic causes of abnormal wound healing.
- Nutritional status* has profound effects on repair; protein deficiency and vitamin C deficiency inhibit collagen synthesis and retard healing.
- Aging* reduces the capacity to heal injured tissues, for several reasons.

TABLE 3.10 Characteristics of Healing of Skin Wounds: First and Second Intention

	Healing by First Intention (Primary Union)	Healing by Second Intention (Secondary Union)
Nature of wound	Small incision space Surgically clean wound margins Generally uninfected	Large tissue defect Irregular wound margins Often infected
Inflammatory response	Limited inflammation and less granulation tissue formation	Marked inflammatory reaction and exuberant granulation tissue formation
Wound contraction	Less	Significant
Outcome	Shorter healing process; healed wound appears as neat linear scar; rare complications (e.g., epidermal inclusion cyst formation)	Longer healing process; healed wound appears contracted and irregular; frequent complications (e.g., suppuration)

TABLE 3.11 Kinetics of Healing of Skin Wounds

Time	Sequence of Events in Wound Healing
Immediate	Formation of blood clot
24 hours	Neutrophils at incision margins
24–48 hours	Epithelial cells from both edges proliferate to form a thin continuous layer
Day 3	Neutrophils replaced by macrophages; early granulation tissue appears; initiation of angiogenesis and ECM deposition
Day 5	Maximum neovascularization and granulation tissue; collagen and ECM deposition
Day 14	Continued collagen deposition and fibroblast proliferation
Day 30	Complete scar formation

ECM, Extracellular membrane.

- Glucocorticoids (steroids)* have well-documented antiinflammatory effects, and their administration may result in weakness of the scar due to inhibition of TGF- β production and diminished fibrosis. In some instances, however, these effects of glucocorticoids are

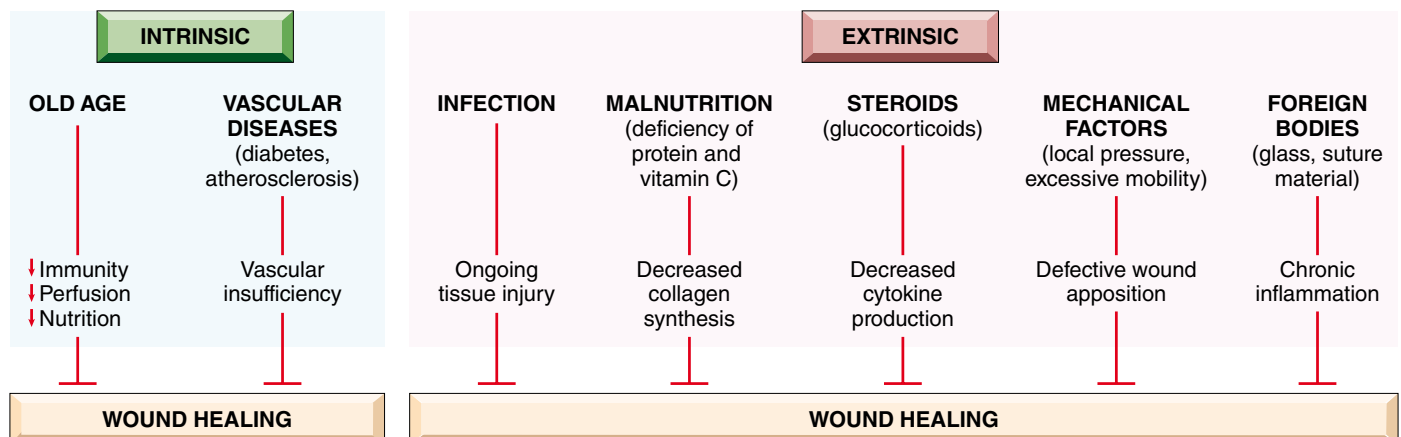


Fig. 3.28 Factors that influence tissue repair.

desirable. For example, in corneal infections, glucocorticoids are sometimes prescribed (along with antibiotics) to reduce the likelihood of opacity that may result from scarring.

- **Mechanical factors** such as excessive mobility may interfere with apposition of wound edges (as in fractures and skin grafts). Increased local pressure or torsion may cause wounds to pull apart.
- **Poor perfusion**, due to peripheral vascular disease, arteriosclerosis, and diabetes or due to obstructed venous drainage (e.g., in varicose veins), also impairs healing.
- **Foreign bodies** such as fragments of steel, glass, or even bone impede healing by perpetuating chronic inflammation.
- **The type and extent of tissue injury** and the character of the tissue in which the injury occurs affect the subsequent repair. Complete restoration can occur only in tissues composed of stable and labile cells. Injury to tissues composed of permanent cells inevitably results in scarring and some loss of function.
- **The location of the injury** is also important. For example, inflammation arising in tissue spaces (e.g., pleural, peritoneal, synovial cavities) develops extensive exudates. Subsequent repair may occur by digestion of the exudate, initiated by the proteolytic enzymes of leukocytes, and resorption of the liquefied exudate. This is called *resolution*, and in the absence of cellular necrosis, normal tissue architecture is generally restored. However, in the setting of larger accumulations, granulation tissue grows into the exudate, and a fibrous scar ultimately forms. This is called *organization*.

Abnormalities in Tissue Repair

Complications in tissue repair can arise from abnormalities in any of the basic components of the process, including deficient scar formation, excessive formation of the repair components, and formation of contractures.

Defects in Healing: Chronic Wounds

These are seen in numerous clinical situations, because of local and systemic factors. Some common examples are:

- **Venous leg ulcers** (Fig. 3.29A) develop most often in elderly people because of chronic venous hypertension, which may be caused by severe varicose veins or congestive heart failure. Deposits of iron pigment (hemosiderin) are common, resulting from red cell breakdown, and there may be accompanying chronic inflammation. These ulcers fail to heal because of poor delivery of oxygen to the site of the ulcer.
- **Arterial ulcers** (Fig. 3.29B) develop in individuals with atherosclerosis of peripheral arteries, especially associated with diabetes. The ischemia results in atrophy and then necrosis of the skin and underlying tissues. These lesions can be quite painful.
- **Diabetic ulcers** (Fig. 3.29C) affect the lower extremities, particularly the feet. There is tissue necrosis and failure to heal as a result of vascular disease, causing ischemia, neuropathy, and secondary infections. Histologically, these lesions are characterized by epithelial ulceration (Fig. 3.29E) and extensive granulation tissue in the underlying dermis (Fig. 3.29F).

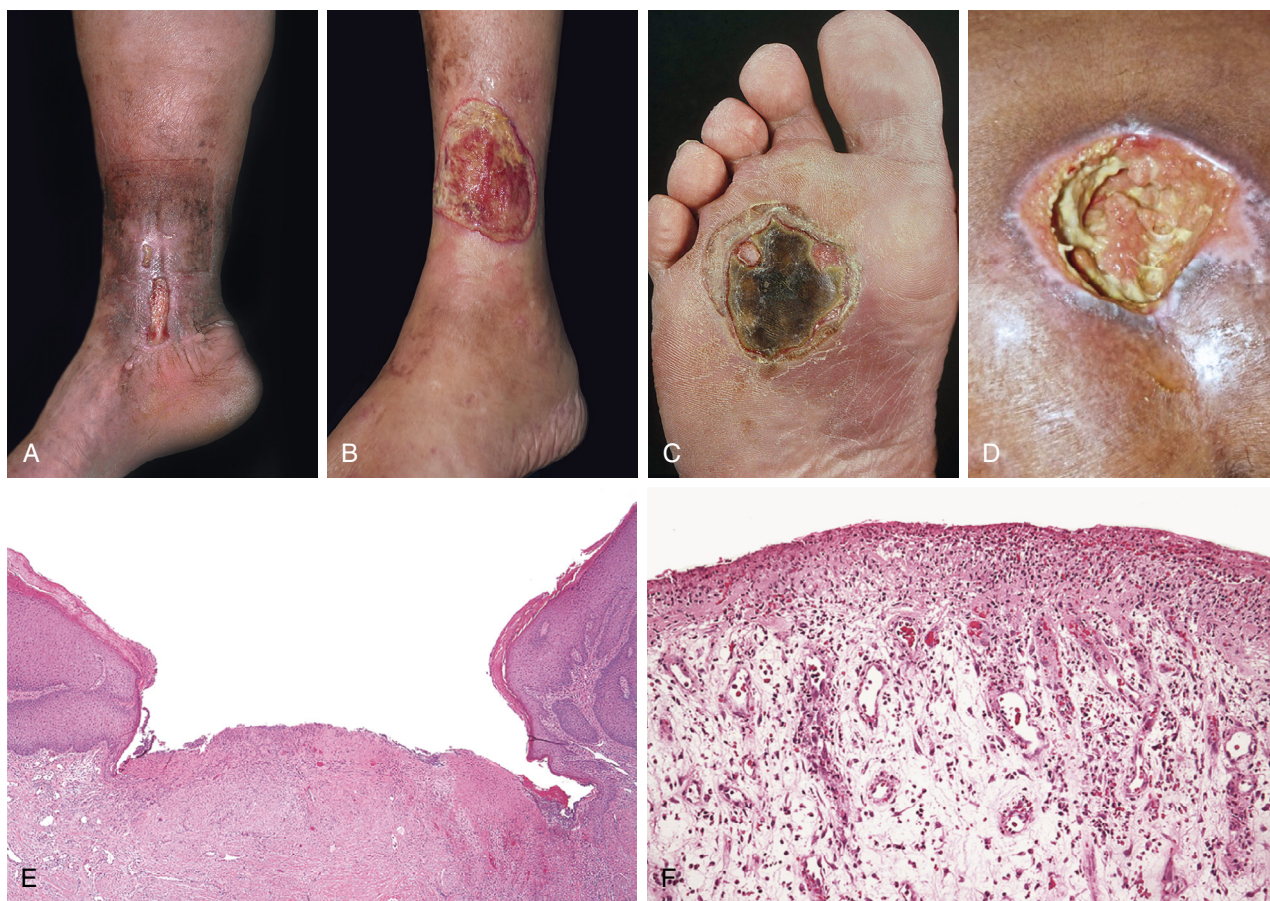


Fig. 3.29 Chronic wounds illustrating defects in wound healing. (A–D) External appearance of skin ulcers. (A) Venous leg ulcer; (B) arterial ulcer, with more extensive tissue necrosis; (C) diabetic ulcer; (D) pressure sore. (E–F) Histologic appearance of a diabetic ulcer. (E) Ulcer crater; (F) chronic inflammation and granulation tissue. (A–D, From Eming SA, Margin P, Tomic-Canic M. Wound repair and regeneration: mechanisms, signaling, and translation. *Sci Transl Med*. 2014;6:265sr6.)

- **Pressure sores** (Fig. 3.29D) are areas of skin ulceration and necrosis of underlying tissues caused by prolonged compression of tissues against a bone (e.g., in elderly patients with numerous morbidities lying in bed without moving). The lesions are caused by mechanical pressure and local ischemia.

When a surgical incision reopens internally or externally it is called wound *dehiscence*. The risk factors for such an occurrence are obesity, malnutrition, infections, vascular insufficiency, and mechanical factors. In abdominal wounds it can be precipitated by vomiting and coughing.

Excessive Scarring

Excessive formation of the components of the repair process can give rise to hypertrophic scars and keloids. The accumulation of excessive amounts of collagen may give rise to a raised scar known as a *hypertrophic scar*. These often grow rapidly and contain abundant myofibroblasts, but they tend to regress over several months. Hypertrophic scars generally develop after thermal or traumatic injury that involves the deep layers of the dermis. If the scar tissue grows beyond the boundaries of the original wound and does not regress, it is called a *keloid* (Fig. 3.30). Keloid formation seems to be an individual predisposition.

Exuberant granulation is another deviation in wound healing consisting of the formation of excessive amounts of granulation tissue, which protrudes above the level of the surrounding skin and blocks re-epithelialization (this process has been called, with a certain literary fervor, proud flesh). Excessive granulation tissue must be removed by cautery or surgical excision to permit restoration of the continuity of the epithelium. Rarely, incisional scars or traumatic injuries may be followed by exuberant proliferation of fibroblasts and other connective tissue elements that may recur after excision. Desmoids, or aggressive fibromatoses, are benign neoplasms composed of connective tissue (Chapter 26).

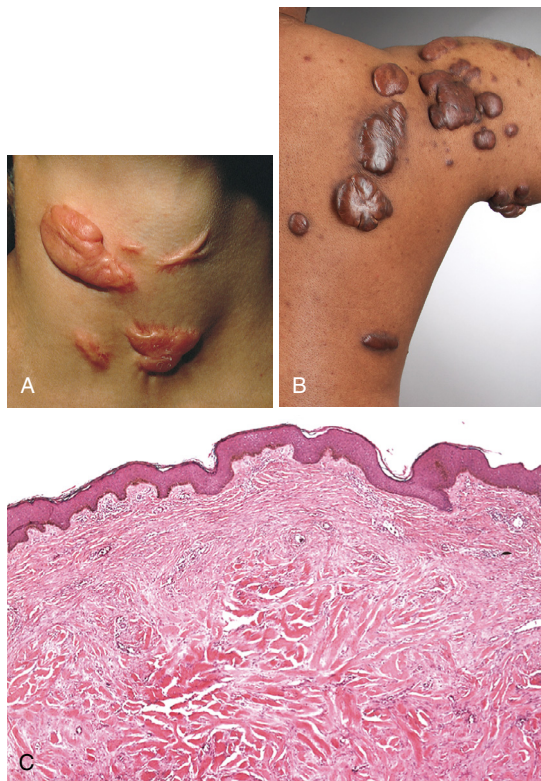


Fig. 3.30 Keloid. Keloids may appear erythematous in light skin tones (A) or hyperpigmented in darker skin (B). (C) Microscopic appearance of a keloid. Note the thick connective tissue deposited in the dermis.

Contraction in the size of a wound is an important part of the normal healing process. An exaggeration of this process gives rise to *contracture* and results in deformities of the wound and the surrounding tissues. Contractures are particularly prone to develop on the palms, the soles, and the anterior aspect of the thorax. Contractures are commonly seen after serious burns and can compromise the movement of joints.

KEY CONCEPTS

Cutaneous Wound Healing and Pathologic Aspects of Repair

- The main phases of cutaneous wound healing are inflammation, formation of granulation tissue, and ECM remodeling.
- Cutaneous wounds can heal by primary union (first intention) or secondary union (secondary intention); secondary healing involves more extensive scarring and wound contraction.
- Wound healing can be altered by many conditions, particularly infection and diabetes; the type, volume, and location of the injury are important factors that influence the healing process.
- Excessive production of ECM can cause keloids in the skin.

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