

**CELLS OF CONNECTIVE TISSUE**

- Fibroblasts
- Adipocytes
- Macrophages & the Mononuclear Phagocyte System
- Mast Cells
- Plasma Cells
- Leukocytes

- 2. Reticular Fibers
- 3. Elastic Fibers

**GROUND SUBSTANCE**  
**TYPES OF CONNECTIVE TISSUE**

- Connective Tissue Proper
- Reticular Tissue
- Muroid Tissue

**FIBERS**

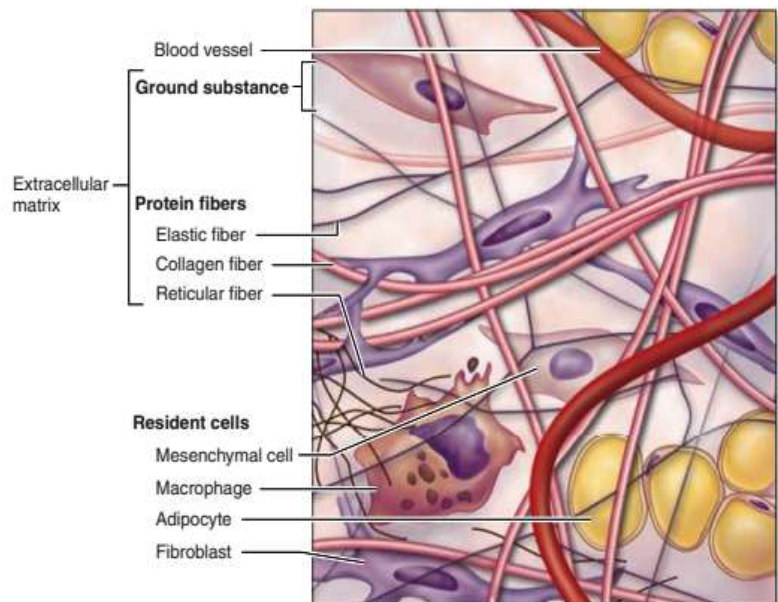
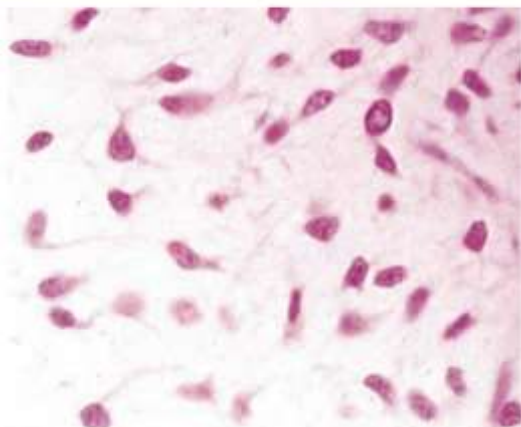
- 1. Collagen

**CELLS OF CONNECTIVE TISSUE**

Fibroblasts are the key cells in connective tissue proper (Figure 5–2 and Table 5–1). Fibroblasts originate locally from **mesenchymal cells** and are permanent residents of connective tissue. Other cells found here, such as **macrophages**, **plasma cells**, and **mast cells**, originate from **hematopoietic stem cells** in bone marrow, circulate in the blood, and then move into connective tissue where they function. These and other white blood cells (leukocytes) are transient cells of most connective tissues, where they perform various functions for a short period as needed and then die by **apoptosis**.

**FIGURE 5–2 Cellular and extracellular components of connective tissue.**

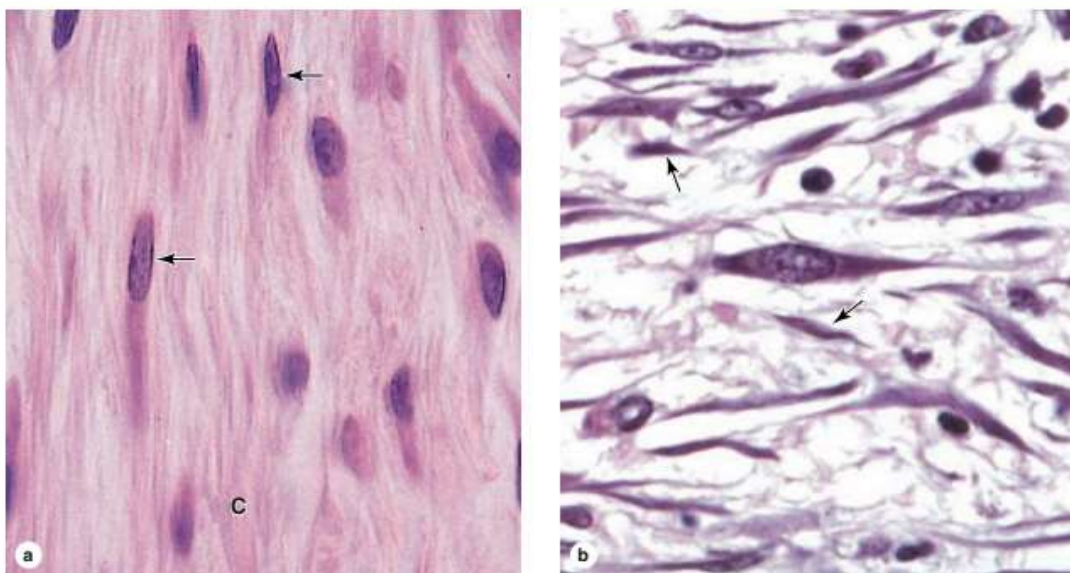
**FIGURE 5–1 Embryonic mesenchyme.**



## 1. Fibroblasts

Fibroblasts (Figure 5–3), the most common cells in connective tissue proper, produce and maintain most of the tissue’s extracellular components. Fibroblasts synthesize and secrete **collagen** (the most abundant protein of the body) and **elastin**, which both form large fibers, as well as the GAGs, proteoglycans, and multiadhesive glycoproteins that comprise the ground substance. Most of the secreted ECM components undergo further modification outside the cell before assembling as a **matrix**. Distinct levels of fibroblast activity can be observed histologically (Figure 5–3b). Cells with intense synthetic activity are morphologically different from the quiescent fibroblasts that are scattered within the matrix they have already synthesized. Some histologists reserve the term “**fibroblast**” to denote the active cell and “**fibrocyte**” to denote the quiescent cell. The active fibroblast has more abundant and irregularly branched cytoplasm, containing much rough endoplasmic reticulum (RER) and a well-developed Golgi apparatus, with a large, ovoid, euchromatic nucleus and a prominent nucleolus. The quiescent cell is smaller than the active fibroblast, is usually spindle-shaped with fewer processes, much less RER, and a darker, more heterochromatic nucleus. **Fibroblasts** involved in wound healing, sometimes called **myofibroblasts**, have a well-developed contractile function and are enriched with a form of actin also found in smooth muscle cells.

FIGURE 5–3 Fibroblasts.



(a) Fibroblasts typically have large active nuclei and eosinophilic cytoplasm that tapers off in both directions along the axis of the nucleus, a morphology often referred to as “spindle-shaped.” Nuclei (arrows) are clearly seen, but the eosinophilic cytoplasmic processes resemble the collagen bundles (C) that fill the ECM and are difficult to distinguish in H&E-stained sections.

(b) Both active and quiescent fibroblasts may sometimes be distinguished, as in this section of dermis. Active fibroblasts have large, euchromatic nuclei and basophilic cytoplasm, while inactive fibroblasts (or fibrocytes) are smaller with more heterochromatic nuclei (arrows). The round, very basophilic round cells are in leukocytes. (Both X400; H&E)

TABLE 5-1

## Functions of cells in connective tissue proper.

Cell Type	Major Product or Activity
Fibroblasts (fibrocytes)	Extracellular fibers and ground substance
Plasma cells	Antibodies
Lymphocytes (several types)	Various immune/defense functions
Eosinophilic leukocytes	Modulate allergic/vasoactive reactions and defense against parasites
Neutrophilic leukocytes	Phagocytosis of bacteria
Macrophages	Phagocytosis of ECM components and debris; antigen processing and presentation to immune cells; secretion of growth factors, cytokines, and other agents
Mast cells and basophilic leukocytes	Pharmacologically active molecules (eg, histamine)
Adipocytes	Storage of neutral fats

## 2. Adipocytes

Adipocytes, or fat cells, are found in the connective tissue of many organs. These large, **mesenchymally** derived cells are specialized for cytoplasmic storage of lipid as neutral fats, or less commonly for the production of heat. Tissue with a large population of adipocytes, called **adipose connective tissue**, serves to cushion and insulate the skin and other organs. Adipocytes have major metabolic significance with considerable medical importance.

## 3. Macrophages & the Mononuclear Phagocyte

Macrophages have highly developed phagocytic ability and specialize in turnover of protein fibers and removal of dead cells, tissue debris, or other particulate material, being especially abundant at sites of inflammation. Size and shape vary considerably, corresponding to their state of functional activity. A typical macrophage measures between 10 and 30  $\mu\text{m}$  in diameter and has an eccentrically located, oval or kidney-shaped nucleus. Macrophages are present in the connective tissue of most organs and are sometimes referred to by pathologists as "**histiocytes.**"

**Macrophages** derive from bone marrow precursor cells called **monocytes** that circulate in the blood. These cells cross the epithelial wall of small venules to enter connective tissue, where they differentiate, mature, and acquire the morphologic features of phagocytic cells. Therefore, monocytes and macrophages are the same cell at different stages of maturation. Macrophages play a very important role in the early stages of repair and inflammation after tissue damage. Under such conditions these cells accumulate in connective tissue by local proliferation of macrophages and recruitment of more monocytes from the blood. Macrophages are distributed throughout the body and are normally present in the stroma of most organs. Along with other monocyte derived cells, they comprise a family of cells called the mononuclear phagocyte system (Table 5–2). All of these macrophage-like cells are derived from monocytes, but have different names in various organs, for example, **Kupffer cells in the liver**, **microglial cells in the central nervous system**, **Langerhans cells in the skin**, and **osteoclasts in bone**. All are long-living cells and may survive in the tissues for months. In addition to debris removal, these cells are highly important for the **uptake, processing, and presentation of antigens for lymphocyte activation**. The transformation from monocytes to macrophages in connective tissue involves increases in **cell size, increased protein synthesis, and increases in the number of Golgi complexes and lysosomes**.

Cell Type	Major Location	Main Function
Monocyte	Blood	Precursor of macrophages
Macrophage	Connective tissue, lymphoid organs, lungs, bone marrow, pleural and peritoneal cavities	Production of cytokines, chemotactic factors, and several other molecules that participate in inflammation (defense), antigen processing, and presentation
Kupffer cell	Liver (perisinusoidal)	Same as macrophages
Microglial cell	Central nervous system	Same as macrophages
Langerhans cell	Epidermis of skin	Antigen processing and presentation
Dendritic cell	Lymph nodes, spleen	Antigen processing and presentation
Osteoclast (from fusion of several macrophages)	Bone	Localized digestion of bone matrix
Multinuclear giant cell (several fused macrophages)	In connective tissue under various pathological conditions	Segregation and digestion of foreign bodies

#### 4. Mast Cells

Mast cells are oval or irregularly shaped cells of connective tissue, between 7 and 20 µm in diameter, filled with basophilic secretory granules which often

obscure the central nucleus (Figure 5–5). These granules are electron-dense and of variable size, ranging from 0.3 to 2.0  $\mu\text{m}$  in diameter.

Mast cells function in the localized release of many bioactive substances important in the **local inflammatory response**, **innate immunity**, and **tissue repair**. A partial list of molecules released from these cells' secretory granules includes the following:

- **Heparin**, a sulfated GAG that acts locally as an anticoagulant
- **Histamine**, which promotes increased vascular permeability and smooth muscle contraction
- **Serine proteases**, which activate various mediators of inflammation
- **Eosinophil** and **neutrophil** chemotactic factors, which attract those leukocytes
- **Cytokines**, polypeptides directing activities of leukocytes and other cells of the immune system
- **Phospholipid precursors**, which are converted to prostaglandins, leukotrienes, and other important lipid mediators of the inflammatory response.

## **5. Plasma Cells**

Plasma cells are lymphocyte derived, antibody producing cells. These relatively large, ovoid cells have basophilic cytoplasm rich in RER and a large Golgi apparatus near the nucleus that may appear pale in routine histologic preparations (Figure 5–7). The nucleus of the plasma cell is generally spherical but eccentrically placed. Many of these nuclei contain compact, peripheral regions of heterochromatin alternating with lighter areas of euchromatin. At least a few plasma cells are present in most connective tissues. Their average lifespan is only **10-20 days**.

## **6. Leukocytes**

Other white blood cells, or leukocytes, besides macrophages and plasma cells normally comprise a population of wandering cells in connective tissue. Derived from circulating blood cells, they leave blood by migrating between the endothelial cells of venules to enter connective tissue. This process increases greatly during inflammation, which is a vascular and cellular defensive response to injury or foreign substances, including pathogenic bacteria or irritating chemical substances. Inflammation begins with the local release of chemical mediators from various cells, the ECM, and blood plasma proteins. These substances act on local blood vessels, mast cells, macrophages, and other cells to induce events characteristic of inflammation, for example, increased blood flow and vascular permeability, entry and migration of leukocytes, and activation of macrophages for phagocytosis. Most leukocytes function in connective tissue only for a few hours or days and then undergo apoptosis. Some lymphocytes and phagocytic antigen-presenting cells

normally leave the interstitial fluid of connective tissue, enter blood or lymph, and move to selected lymphoid organs.

## **FIBERS**

The fibrous components of connective tissue are elongated structures formed from proteins that polymerize after secretion from fibroblasts (Figure 5–2). The three main types of fibers include **collagen**, **reticular**, and **elastic fibers**. **Collagen** and **reticular** fibers are both formed by proteins of the **collagen** family, and elastic fibers are composed mainly of the protein elastin. These fibers are distributed unequally among the different types of connective tissue, with the predominant fiber type conferring most specific tissue properties.

### **1. Collagen**

The collagens constitute a family of proteins selected during evolution for their ability to form various extracellular fibers, sheets, and networks, all of which extremely strong and resistant to normal shearing and tearing forces. Collagen is a key element of all connective tissues, as well as epithelial basement membranes and the external laminae of muscle and nerve cells. Collagen is the most abundant protein in the human body, representing 30% of its dry weight. A major product of fibroblasts, collagens are also secreted by several other cell types and are distinguishable by their molecular compositions, morphologic characteristics, distribution, functions, and pathologies. Collagen turnover and renewal in normal connective tissue is generally a very slow but ongoing process. In some organs, such as **tendons** and **ligaments**. To be renewed, the collagen must first be degraded. Degradation is initiated by specific enzymes called **collagenases**, which are members of an enzyme class called matrix metalloproteinases (MMPs). Various MMPs are secreted by **macrophages** and **play an important role in remodeling the extracellular matrix ECM during tissue repair**.

### **2. Reticular Fibers**

Found in delicate connective tissue of many organs, notably in the **immune system**, reticular fibers consist mainly of collagen type III, which forms an extensive network (reticulum) of thin fibers for the support of many different cells. Reticular fibers are seldom visible in hematoxylin and eosin (H&E) preparations but are characteristically stained black after impregnation with silver salts and are thus termed **argyrophilic** (Gr. argyros, silver). Reticular fibers are also periodic acid-Schiff (PAS) positive, which, like argyrophilia, is due to the high content of sugar chains bound to type III collagen  $\alpha$  chains.

Reticular fibers produced by **fibroblasts** occur in the reticular lamina of basement membranes and typically also surround adipocytes, smooth muscle and nerve fibers, and small blood vessels. Delicate reticular networks serve as the supportive stroma for the parenchymal secretory cells and rich microvasculature of the liver and endocrine glands. Abundant reticular fibers also characterize the stroma of **hemopoietic tissue (bone marrow)**, the **spleen**, and **lymph nodes** where they support rapidly changing populations of proliferating cells and phagocytic cells.

### 3. Elastic Fibers

Elastic fibers are also thinner than the type I collagen fibers and form sparse networks interspersed with collagen bundles in many organs, particularly those subject to regular stretching or bending. As the name implies, elastic fibers have rubberlike properties that allow tissue containing these fibers, such as the stroma of **the lungs**, to be stretched or distended and return to their original shape. In the wall of **large blood vessels**, especially **arteries**, elastin also occurs as fenestrated sheets called elastic lamellae. Elastic fibers and lamellae are not strongly acidophilic and stain poorly with H&E; they are stained more darkly than collagen with other stains such as orcein and aldehyde fuchsin .

Elastic fibers (and lamellae) are a composite of fibrillin, which forms a network of microfibrils, embedded in a larger mass of cross-linked elastin. Both proteins are secreted from fibroblasts (and smooth muscle cells in vascular walls) and give rise to elastic fibers in a stepwise manner. The elastic properties of these fibers and lamellae result from the structure of the elastin subunits and the unique cross-links holding them together. Elastin molecules have many lysine-rich regions interspersed with hydrophobic domains rich in lysine and proline which are thought to form extensible, random-coil conformations. Elastin resists digestion by most proteases, but it is hydrolyzed by **pancreatic elastase**.

### GROUND SUBSTANCE

The ground substance of the ECM is a highly hydrated (with much bound water), transparent, complex mixture of three major kinds of macromolecules: **glycosaminoglycans (GAGs)**, **proteoglycans**, and **multiadhesive glycoproteins**. Filling the space between cells and fibers in connective tissue, ground substance allows diffusion of small molecules and, because it is viscous, acts as both a lubricant and a barrier to the penetration of invaders. Physical properties of ground substance also profoundly influence various cellular activities. Water in the ground substance of connective tissue is referred to as **interstitial fluid** and has an ion composition similar to that of blood plasma. Interstitial fluid

also contains plasma proteins of low molecular weight that pass through the thin walls of the smallest blood vessels, the capillaries. Although only a small proportion of connective tissue proteins are plasma proteins, it is estimated that as much as one-third of the body's plasma proteins are normally found in the interstitial fluid of connective tissue because of its large volume and wide distribution. Capillaries in connective tissue also bring the various nutrients required by cells and carry away their metabolic waste products to the detoxifying and excretory organs, the liver and kidneys. Interstitial fluid is the solvent for these substances.

**Two main forces act on the water in capillaries:**

- **The hydrostatic pressure** of the blood caused by the pumping action of the heart, which forces water out across the capillary wall
- **The colloid osmotic pressure** produced by plasma proteins such as albumin, which draws water back into the capillaries.

## **TYPES OF CONNECTIVE TISSUE**

### **1. Connective Tissue Proper**

Connective tissue proper is broadly classified as “**loose**” or “**dense**,” terms which refer to the amount of collagen present. **Loose connective tissue** is common, forming a layer beneath the epithelial lining of many organs and filling the spaces between fibers of muscle and nerve. Also called **areolar Loose connective tissue**, the loose connective tissue typically contains cells, fibers, and ground substance in roughly equal parts. The most numerous cells are fibroblasts, but the other types of connective tissue cells are also normally found, along with nerves and small blood vessels. Collagen fibers predominate, but elastic and reticular fibers are also present. With at least a moderate amount of ground substance, loose connective tissue has a delicate consistency; it is flexible and not very resistant to stress. **Dense connective tissue** has similar components as loose connective tissue, but with fewer cells, mostly fibroblasts, and a clear predominance of bundled type I collagen fibers over ground substance. The abundance of collagen here protects organs and strengthens them structurally. In **dense irregular connective tissue** bundles of collagen fibers appear randomly interwoven, with no definite orientation. The tough three dimensional collagen networks provide resistance to stress from all directions. Examples of **dense irregular connective tissue** include the deep dermis layer of **skin** and **capsules** surrounding most **organs**. Dense irregular and loose connective tissues are often closely associated; with the two types grading into each other and making distinctions between them somewhat arbitrary. **Dense regular**

**connective tissue** consists mostly of type I collagen bundles and fibroblasts aligned in parallel for great resistance to prolonged or repeated stresses from the same direction. The best examples of dense regular connective tissue are the very strong and flexible **tendons, cords** connecting muscles to bones; **aponeuroses**, which are sheet-like **tendons**; and **ligaments**, bands or sheets that hold together components of the skeletal system. Consisting almost entirely of densely packed parallel collagen fibers separated by very little ground substance and having very few **blood vessels**, these inextensible structures are white in the fresh state.

## **2. Reticular Tissue**

Reticular tissue is characterized by abundant fibers of type III collagen forming a delicate network that supports various types of cells. This collagen is also known as reticulin and is produced by modified fibroblasts often called reticular cells that remain associated with and partially cover the fibers. The loose disposition of glycosylated reticular fibers provides a framework with specialized microenvironments for cells in hemopoietic tissue and some lymphoid organs (**bone marrow, lymph nodes, and spleen**). The resulting cell-lined system creates a meshwork for the passage of leukocytes and lymph. Macrophages and dendritic cells (also in the mononuclear phagocyte family) are also dispersed within these reticular tissues to monitor cells formed there or passing through and to remove debris.

## **3. Muroid Tissue**

Muroid (or mucous) connective tissue is the principal component of the fetal umbilical cord, where it is referred to as Wharton's jelly. With abundant ground substance composed chiefly of hyaluronan, muroid tissue is gelatinous, with sparse collagen fibers and scattered fibroblasts. Included among the fibroblastic cells are many mesenchymal stem cells, which are being studied for their potential in regenerative medicine. Muroid connective tissue is similar to the tissue found in the vitreous chambers of eyes and pulp cavities of young teeth.

**TABLE 5-6**

**Classification of connective or supporting tissues.**

	General Organization	Major Functions	Examples
<b>Connective Tissue Proper</b>			
Loose (areolar) connective tissue	Much ground substance; many cells and little collagen, randomly distributed	Supports microvasculature, nerves, and immune defense cells	Lamina propria beneath epithelial lining of digestive tract
Dense irregular connective tissue	Little ground substance; few cells (mostly fibroblasts); much collagen in randomly arranged fibers	Protects and supports organs; resists tearing	Dermis of skin, organ capsules, submucosa layer of digestive tract
Dense regular connective tissue	Almost completely filled with parallel bundles of collagen; few fibroblasts, aligned with collagen	Provide strong connections within musculoskeletal system; strong resistance to force	Ligaments, tendons, aponeuroses, corneal stroma
<b>Embryonic Connective Tissues</b>			
Mesenchyme	Sparse, undifferentiated cells, uniformly distributed in matrix with sparse collagen fibers	Contains stem/progenitor cells for all adult connective tissue cells	Mesodermal layer of early embryo
Mucoid (mucous) connective tissue	Random fibroblasts and collagen fibers in viscous matrix	Supports and cushions large blood vessels	Matrix of the fetal umbilical cord
<b>Specialized Connective Tissues</b>			
Reticular connective tissue (see Chapter 14)	Delicate network of reticulin/collagen III with attached fibroblasts (reticular cells)	Supports blood-forming cells, many secretory cells, and lymphocytes in most lymphoid organs	Bone marrow, liver, pancreas, adrenal glands, all lymphoid organs except the thymus
Adipose Tissue (Chapter 6)			
Cartilage (Chapter 7)			
Bone (Chapter 8)			
Blood (Chapter 12)			

