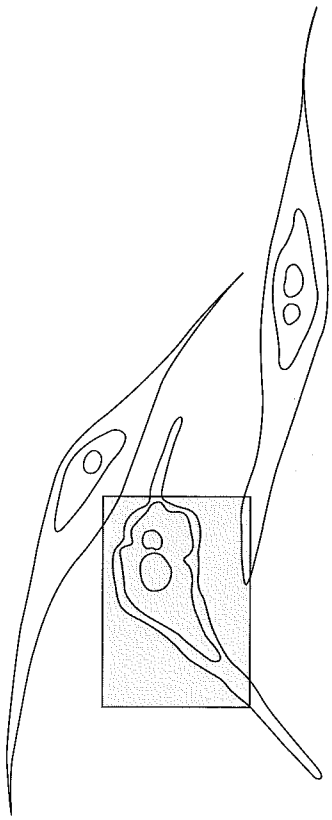
FIBROBLAST



Connective tissue cells, unlike epithelial cells, are not tightly packed, but instead are surrounded by **extracellular fibers** and **ground substance.** The **fibroblast** (F, micrograph), the major cell of connective tissue proper, synthesizes **collagen** and **elastic** fibers and the **proteoglycan** component of the ground substance.

Collagen (c, micrograph) is the most abundant protein in the body and typically the predominant extracellular component of connective tissue. The process of collagen synthesis is complex and involves both intracellular and extracellular steps. Procollagen polypeptide chains are synthesized in the rough ER (r, micrograph), with hydroxylation of specific proline and lysine residues occurring on the growing chains. Sugars are added in both the rough ER and the Golgi complex (not shown).

Assembly of the **triple helical procollagen** molecule occurs in the Golgi and is facilitated by nonhelical peptides at both amino and carboxyl termini. These terminal peptides ensure solubility of the molecule within the cell and prevent the intracellular polymerization into fibrils. From the Golgi procollagen is packaged into secretory vesicles (arrows, micrograph). During or soon after exocytosis, the terminal nonhelical peptides are cleaved by procollagen peptidases and procollagen is converted to tropocollagen. Striated collagen fibrils (arrowheads) consist of precisely arranged tropocollagen molecules (see Connective Tissue, page 72, for details) assembled after cleavage.

Several different types of collagen have been identified. These types differ somewhat in amino acid content but retain such common characteristics as a high glycine content and the presence of two unusual amino acids, hydroxyproline and hydroxylysine. Hydroxyproline forms cross-links that provide intra- and intermolecular stability to collagen fibrils. A deficiency in vitamin C, an essential cofactor in the formation of hydroxyproline, leads to scurvy, a degenerative connective tissue disorder.

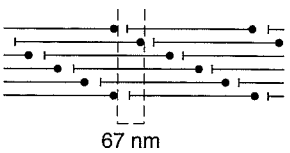
The most common types of collagen are Types I, II, III, and IV. Type I, shown in the micrograph, predominates in bone, tendon, and skin; Type II in cartilage; Type III (reticular fibers) in lymphoid organs, vessels, and muscle; and Type IV in basal lamina. Type IV does not organize as fibrils, but instead forms a network of individual tropocollagen molecules associated with other matrix proteins.

Proteoglycans are not observable in connective tissue proper on routine electron micrographs. They occupy the clear areas (*, micrograph) surrounding the cell and between collagen fibers and also maintain the spacing between individual collagen fibrils. Proteoglycans consist of a core protein covalently attached to a series of highly sulfated repeating disaccharide units called **glycosaminoglycans** (GAGs) (see Connective Tissue, page 84, for details). Proteoglycans associate with collagen and form a hydrated network that is important in the movement of cells and molecules through loose connective tissue.

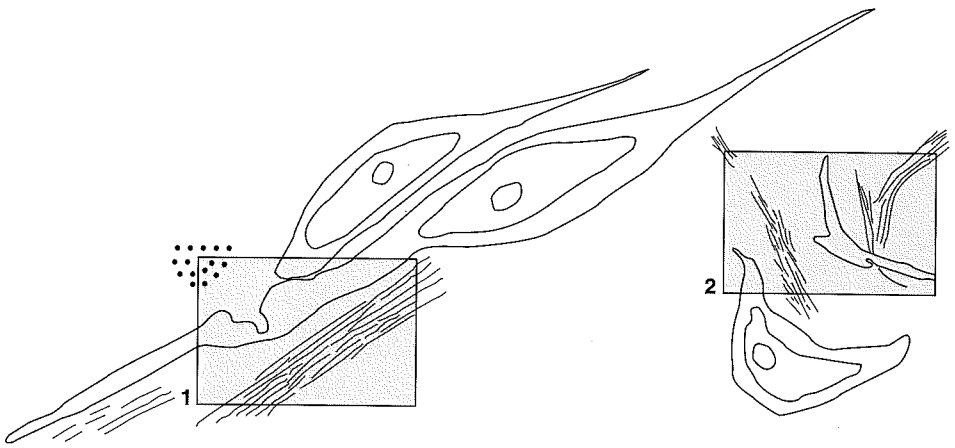

F
c
r
c

Collagen and Elastic Fibers

Collagen, visible in the light microscope as fibers with a diameter of 0.5 to 20 μm, is made up of smaller units, **fibrils.** In electron micrographs of loose and dense connective tissue, collagen fibrils (c, micrograph 1), with a diameter of 20 to 100 nm, are visible in cross section. In longitudinal section striations with a periodicity of 67 nm are apparent. These are created by the staggered arrangement of the individual tropocollagen molecules separated end to end by a gap of 35 nm. Additional narrow bands within this pattern represent stain binding to polar residues of tropocollagen lined up in register.



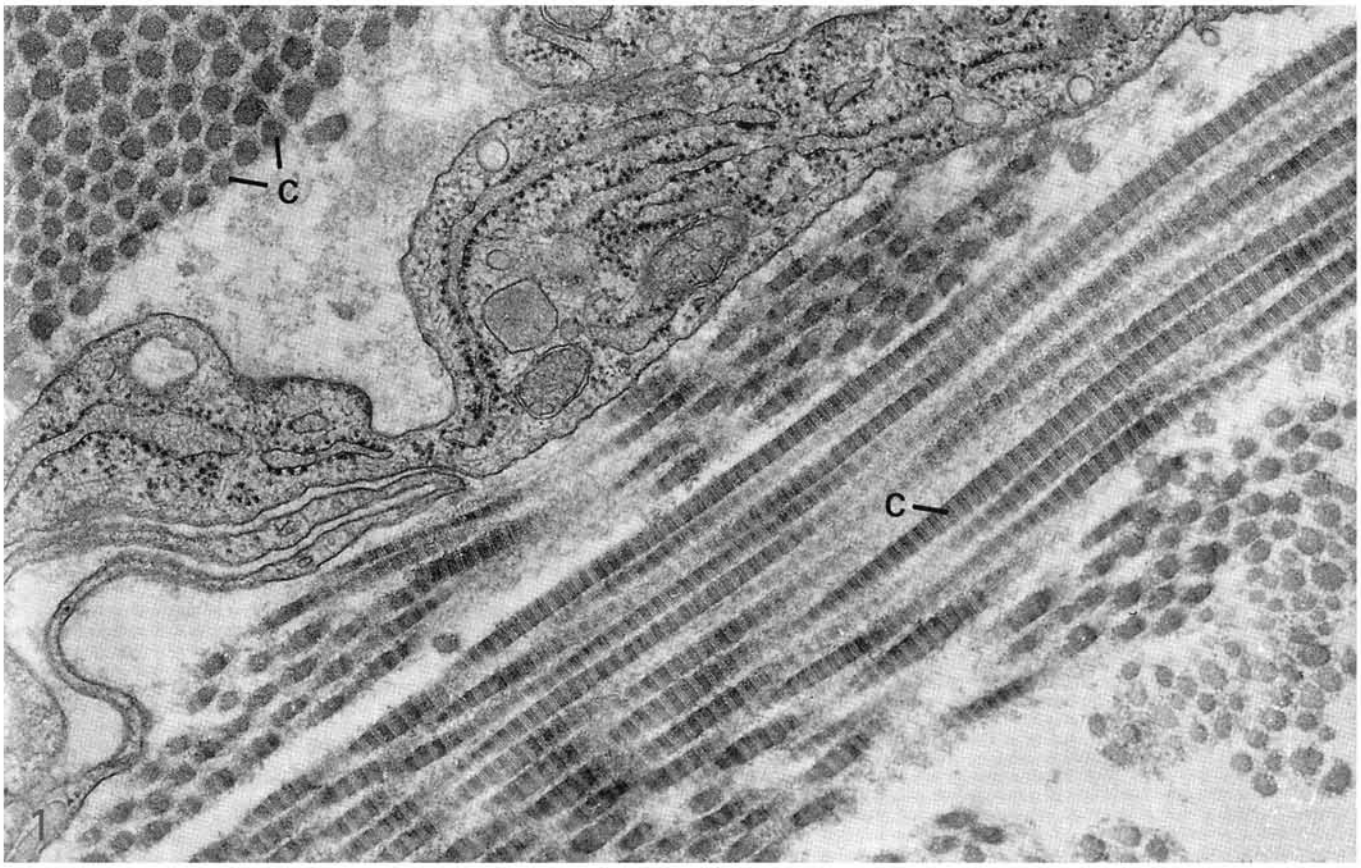
Modified from L. C. Junquiera et al. *Basic Histology,* 7th ed., Appleton and Lange Medical Publications, Norwalk, Conn., 1992.



Like collagen, **elastic fibers** are synthesized by fibroblasts and are ubiquitous. In contrast to most collagen, these relatively thin (0.2–1 μm) branching fibers are not composed of smaller striated fibrils. One elastic fiber viewed under the light microscope is seen as a single structure in routine electron micrographs. The arrows in micrograph 2 identify an elastic fiber that appears to be branching. Each fiber consists of (1) **elastin** (e), a unique protein with primarily hydrophobic, nonpolar amino acids, which does not stain and appears as an amorphous strip, and (2) stained 10-nm **microfibrillar proteins** (arrowheads) containing hydrophilic residues that form a sheet around the elastin.

The resilient character of elastin, which is particularly important in the lung, aorta, and skin, is due in part to the intermolecular cross-links that form extracellularly between tropoelastin molecules. A defect in lysyloxidase, an enzyme necessary for both the cross-linking of tropocollagen and tropoelastin, results in hyperextensible skin and joints, one type of Ehlers–Danlos syndrome.

Extracellular fibers attach to cells either directly via membrane receptors (as with elastin) or indirectly via other molecules such as laminin (in the case of collagen Type IV) and fibronectin (with collagen Types I–III). This association can influence gene expression and functions in many diverse ways. The elastic fiber receptor holds recently synthesized tropoelastin in proper orientation for cross-linking and final fiber formation.



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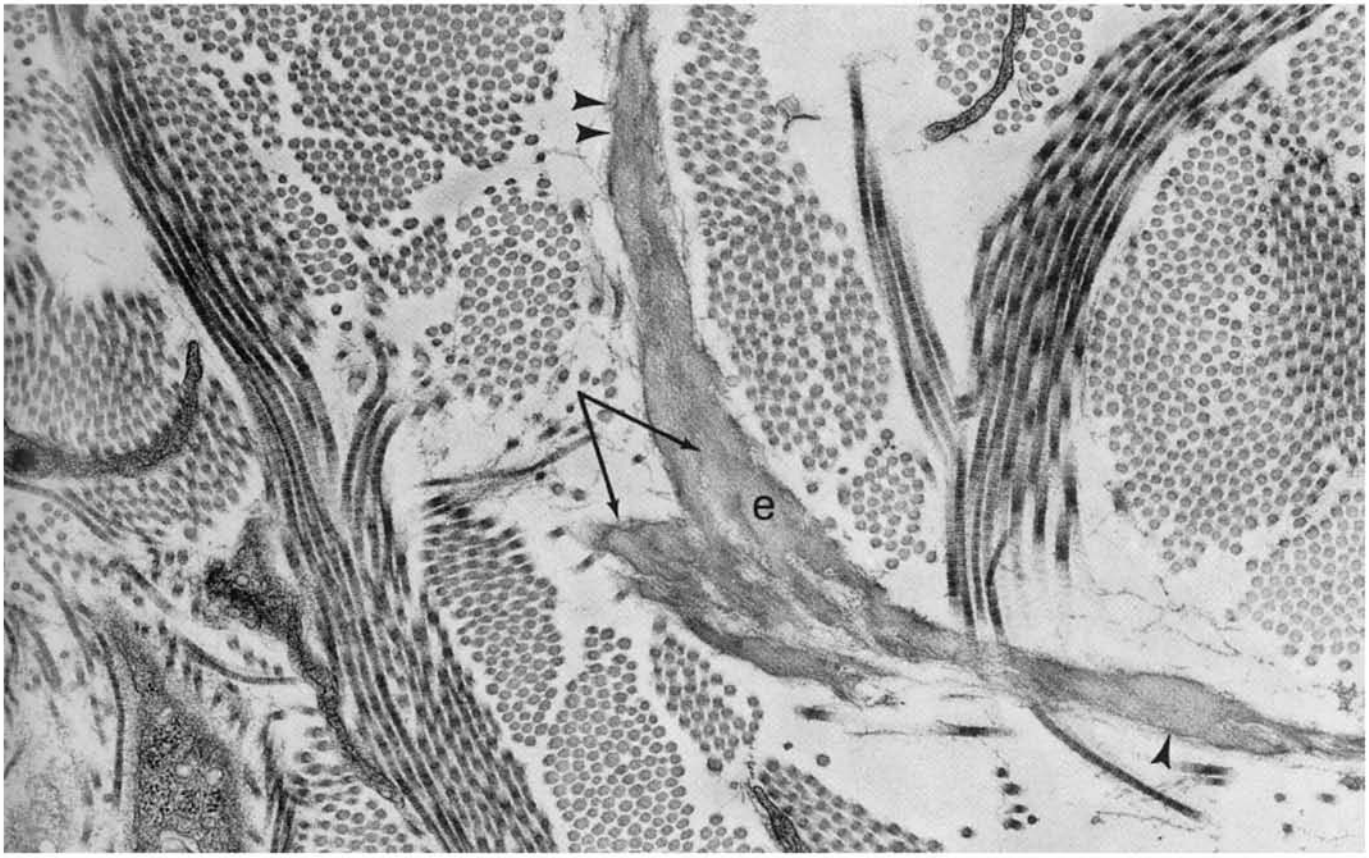
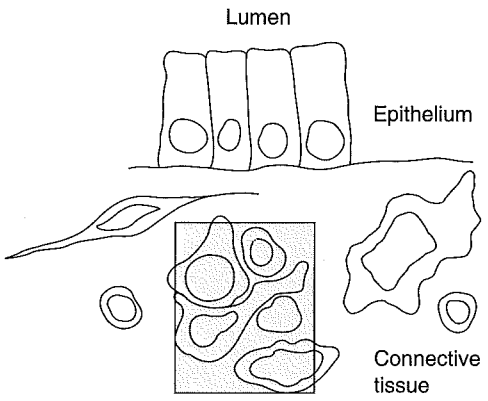


MACROPHAGE

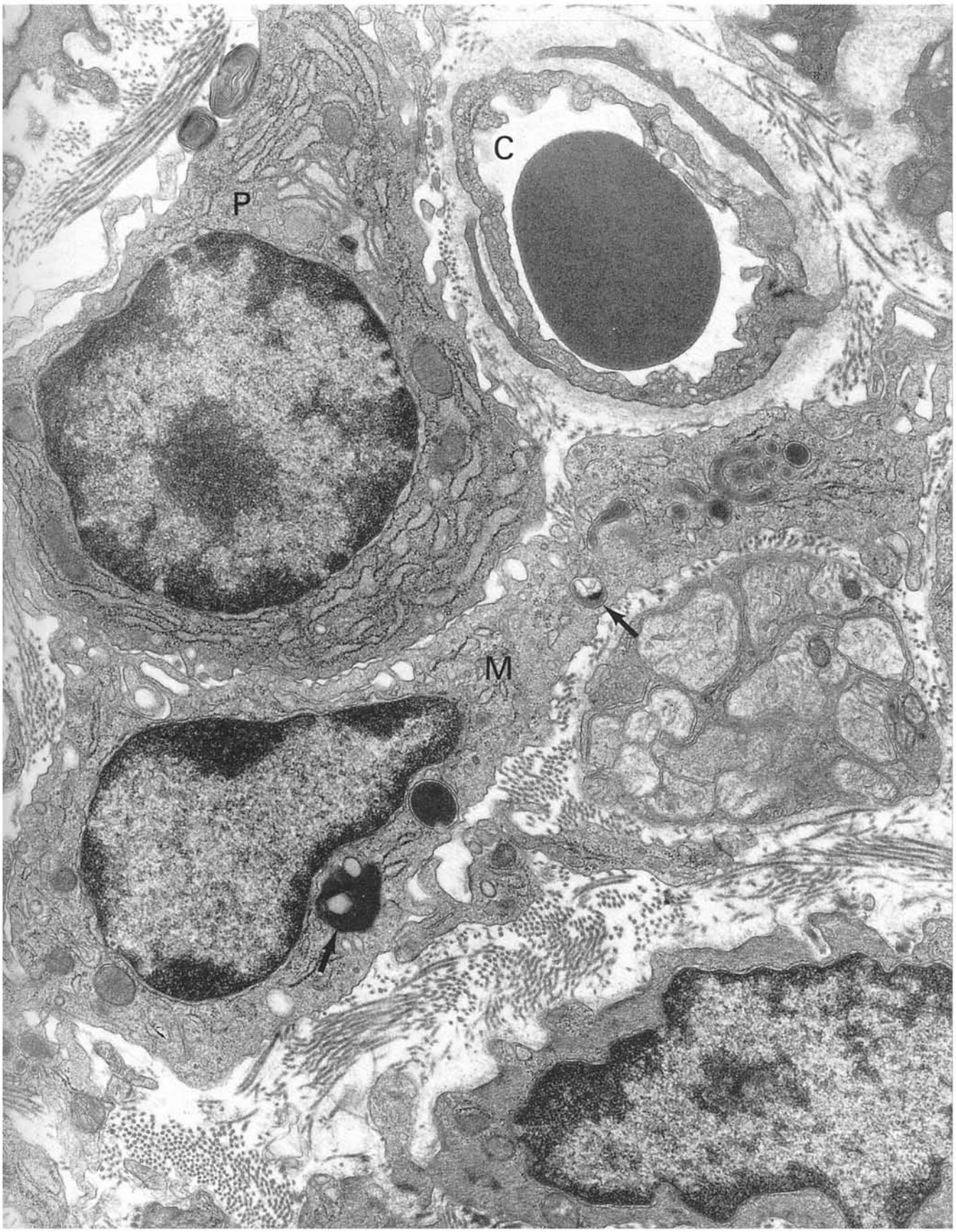


Macrophages (M, micrograph) are a diverse group of specialized cells that carry out a wide range of functions. The most common role of these cells is everyday scavenging as avid phagocytes. Other, more specialized functions include antigen presentation and the secretion of mediators of the immune response.

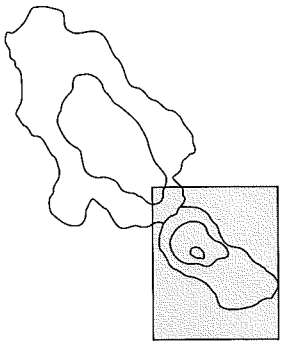
In section, macrophages frequently exhibit an irregular shape as they are caught in some stage of active movement. Characteristically they contain accumulations of heterogeneous bodies in **secondary lysosomes** (arrows, micrograph), which represent phagocytosed material in the process of digestion.

Phagocytosis by the macrophages in loose connective tissue provides defense against foreign antigens entering across epithelial barriers. More specifically, phagocytosis by macrophages (1) kills invading organisms (by using the respiratory burst; see Cell, page 20); (2) removes cellular debris, particularly following inflammation; and (3) is essential prior to presentation of antigens to lymphocytes in the immune responses.

Phagocytosis depends upon receptor-coupled activation, and in many cases the receptor is for the Fc portion of antibodies (see Immune system, page 198). Each antibody class (e.g., IgG, IgA, IgE) produced by plasma cells (P, micrograph) has a unique Fc region and is involved in a different type of immune response. Following secretion, many antibodies enter the vascular system (capillary, C, micrograph) and have effects at distant locations, while others act locally. Certain macrophages have receptors for the Fc portion of both IgG and IgA and can therefore act as scavengers for all antigen bound to both of these antibody classes.


C
P
M

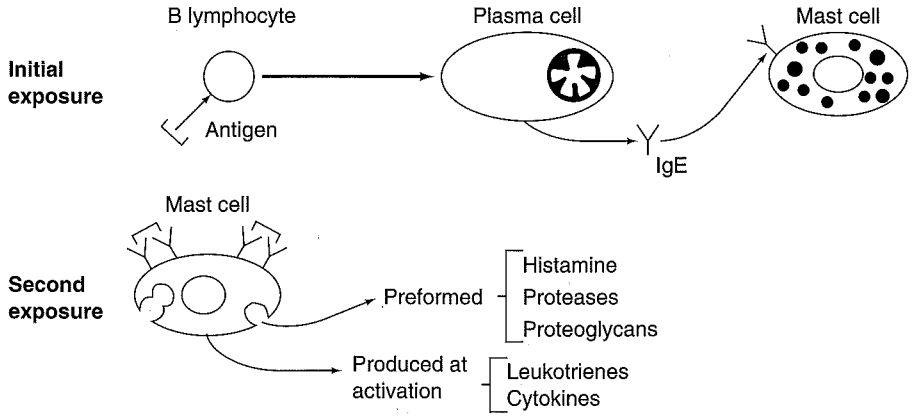
Mast Cell



Mast cells (micrograph) are large cells, 20–30 μm, with a round nucleus (N), conspicuous granules (g), and folds of membrane (arrows) that project from the surface. They are found throughout the body in loose connective tissue, particularly under the epithelium of the respiratory and gastrointestinal tracts, and are frequently concentrated next to blood vessels. Mast cells are a heterogeneous population that can adapt to changes in the immediate environment by altering both type and quantity of synthetic activities.

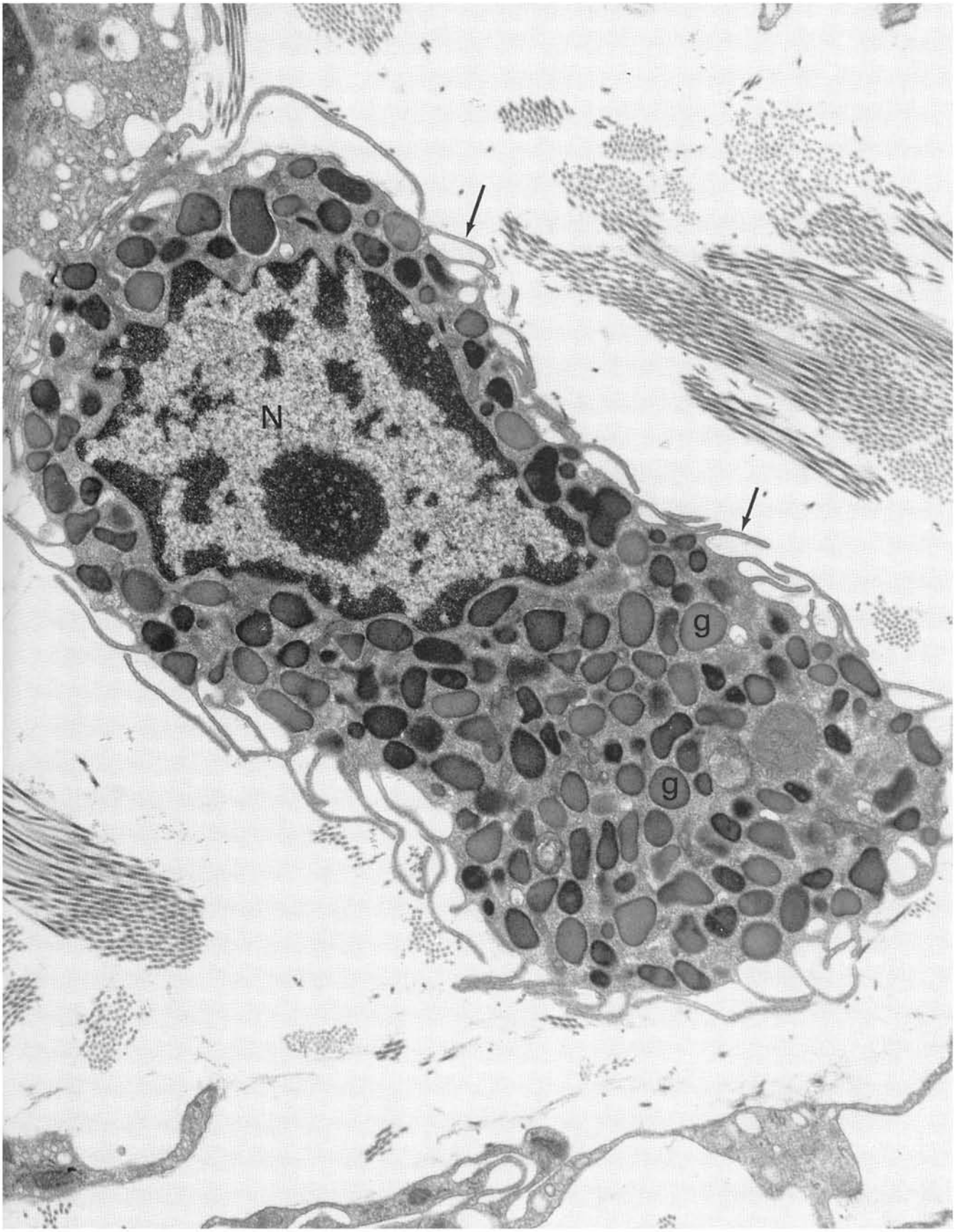
In their role in maintaining health, mast cells facilitate the movement of defense molecules and cells into sites of foreign invasion. They increase vascular permeability allowing plasma exudates containing antibodies and other defense molecules to enter the "threatened" tissue. They also recruit leukocytes (particularly neutrophils and eosinophils) to the area and facilitate their movement across vessel walls. In certain cases the action of mast cells results in an early and late physiological response. The late phase reaction is associated with the presence of large numbers of leukocytes.

Even though cytokines and neuropeptides can directly activate mast cells, the most well-documented mechanism is via the antibody **IgE.** IgE, formed by plasma cells during initial exposure to certain antigens, binds (by the Fc portion) to receptors on the mast cell. Mast cells, packed with granules and coated with IgE specific to the antigen, migrate to regions under epithelia where they present concentrated antibody in an area where the same antigen may reappear. At the second exposure to the particular antigen, receptor aggregation resulting from the binding of a single antigen molecule to two or more IgE molecules activates the mast cell.



On activation mast cells release three classes of bioactive molecules that mediate their action: those preformed and stored in the granules, newly formed membrane-derived molecules (e.g., leukotrienes), and newly formed cytokines (e.g., tumor necrosis factor-α, or TNF-α). The most well-defined associations between mediator and defense actions are the increase in vascular permeability due to histamine release and the recruitment of leukocytes by leukotrienes and TNF-α. Proteoglycans function primarily to bind and concentrate the mediators within the granules. Secretory granules of other cell types possess proteoglycans that serve a similar packaging function.

> The combined effects of mast cells and invited inflammatory cells on bronchiole constriction and airway mucus secretion can be life threatening, as in severe allergy and bronchial asthma.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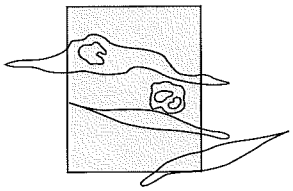
g

Mast Cell–Eosinophil Interaction

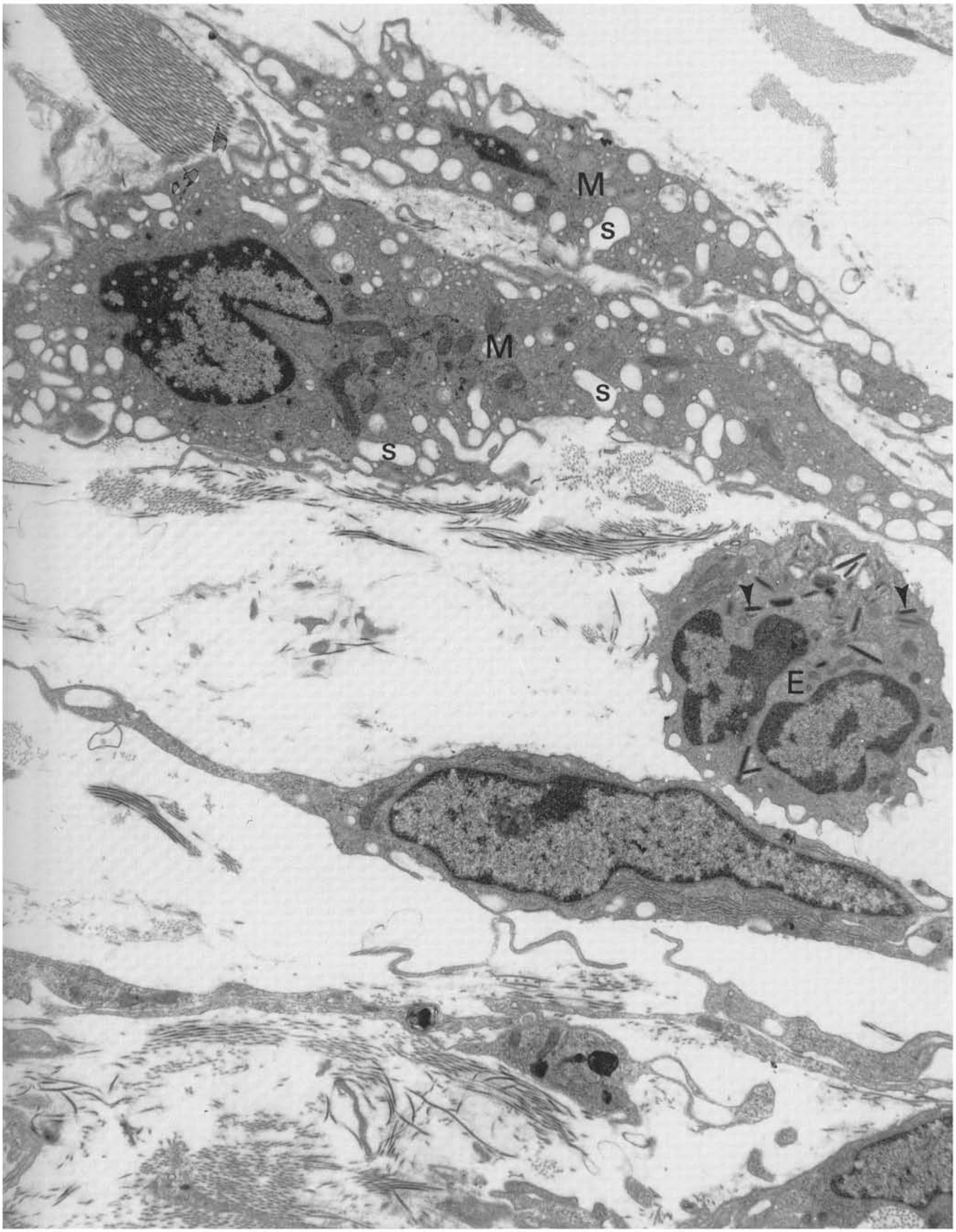


When **mast cells** are **activated,** the contents of their granules are released at once in a process known as compound exocytosis. During this event granules fuse with one another, forming channels to the cell surface for quick release. The two activated, degranulated mast cells (M) in the micrograph are easily recognized by the spaces (s) that represent sections through these channels after activation. Excess cell membrane, the result of fusion of the granule membrane with the cell membrane at exocytosis, is the source of the characteristic mast cell surface folds.

Eosinophils (E, micrograph) are found at the sites of mast cell activity, attracted from nearby blood vessels to the region by factors released during mast cell activation. Recruitment is an important means of bringing eosinophils to areas where they are most effective in defense, such as destruction of parasitic larval schistosomes (see Blood, page 176). Once at the site, eosinophils appear to modulate and neutralize the potentially deleterious effects of mast cell action.

Eosinophils inhibit the synthesis of certain mast cell mediators and also degrade mediators following their release. An eosinophil histaminase deaminates mast cell histamine, and a peroxidase converts certain mast cell leukotrienes to isomers that lack vasoactive bronchoconstrictive activity.

Aside from dampening mast cell activity, eosinophils seem to complicate defense reactions in certain instances. The release of basic proteins that make up the crystalline core (arrowheads, micrograph) of the eosinophil granule is the major factor responsible for certain allergic complications. The respiratory epithelial damage associated with asthma is characterized by the infiltration of both mast cells and eosinophils.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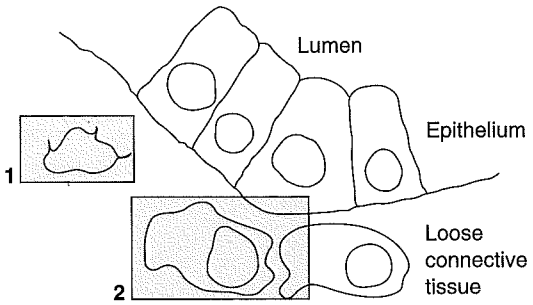
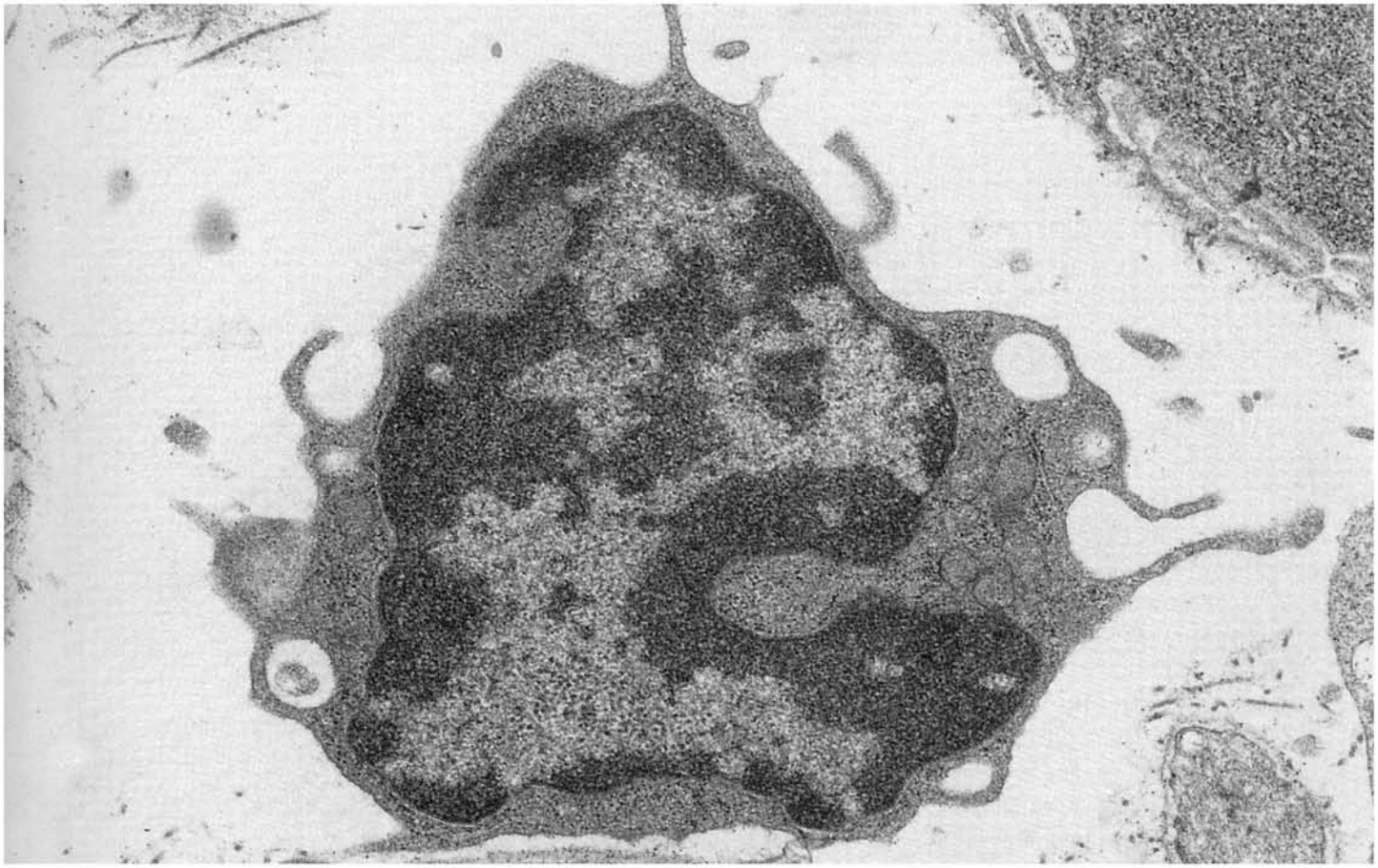
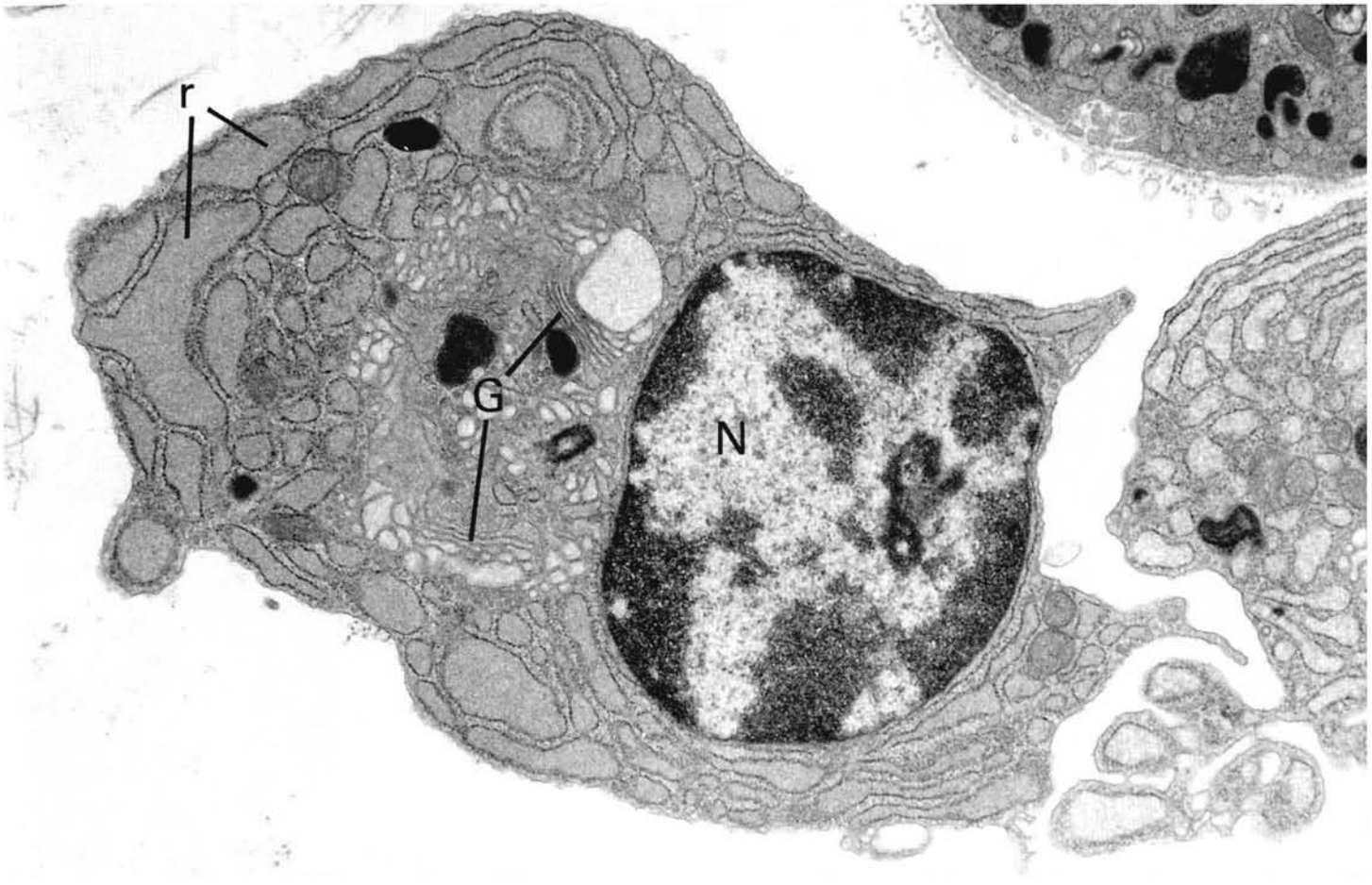
Lymphocytes and Plasma Cells



Lymphocytes are the principal cells of the immune system. In connective tissue, lymphocytes are organized into follicles or scattered between other connective tissue elements. Most lymphocytes (micrograph 1) have a similar ultrastructure; they are small cells (7 – 9 μm) with a thin rim of cytoplasm. In routine electron micrographs it is not possible to distinguish B from T types, or immature (virgin) from most products of activation (e.g., memory, T cytotoxic cells). One exception, however, is the **plasma cell** (micrograph 2), a product of B cell activation that synthesizes and secretes antibodies. This cell has an eccentric nucleus (N), a well-developed Golgi (G), and a cytoplasm packed with dilated rough endoplasmic reticulum (r) containing many copies of a single type of antibody.

Lymphocytes and plasma cells are commonly found in the loose connective tissue under secretory epithelia. An immune response in this region provides defense against foreign material entering the body by crossing an epithelial barrier. Local plasma cells specifically synthesize and release IgA, the antibody of secretions.




r
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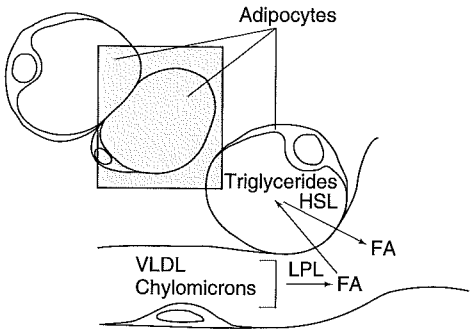
ADIPOCYTE

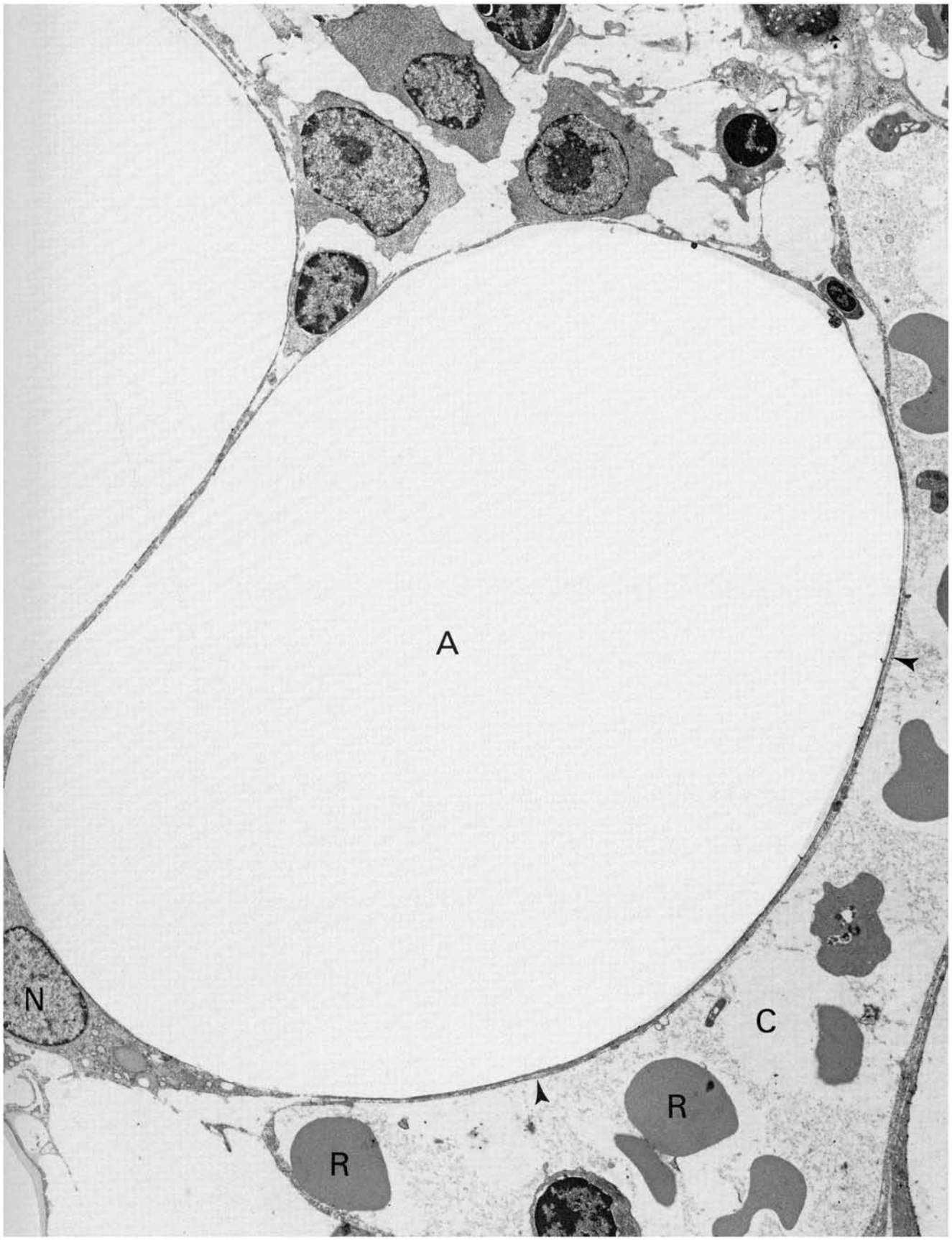


Adipocytes (A, micrograph), cells specialized for fuel storage, are found isolated in loose connective tissue or packed together in groups in certain locations. The fuel, **fatty acids,** is either stored in many small lipid droplets (multilocular, brown fat) or concentrated almost entirely into one large lipid droplet (unilocular, white fat) as in the micrograph. In adults, nearly all adipocytes are of the white type and function as the principal fatty acid energy store in the body. Two main distinguishing morphological characteristics of unilocular adipocytes are illustrated in the electron micrograph: (1) the large size of the cell (compare to the red blood cells, R, in the capillary, C) and (2) the massive lipid droplet that forces the nucleus (N) and cytoplasm to a peripheral location, creating a "signet ring" appearance.

Fatty acids are packaged in the lipid droplet as esters with glycerol, forming **triglycerides** (TGs). TGs are continually being synthesized and broken down as energy demands fluctuate. Some of the fatty acids are synthesized from glucose within the adipose cell itself. Many fatty acids, however, originate in the intestine and liver and are transported to adipocytes and other target organs packaged with proteins. These lipoprotein packages, chylomicrons from the intestines and very low density lipoproteins (VLDLs) from the liver, reach the adipocyte via capillaries. Each package that reaches the adipocyte contains thousands (in VLDLs) to millions (in chylomicrons) of TG molecules.

Fatty acids and monoglycerides are released from chylomicrons and VLDLs by an enzyme, **lipoprotein lipase** (LPL), synthesized by adipocytes and adsorbed (probably via a heparan sulfate) to the surface of the endothelium (arrowheads, micrograph) of adjacent capillaries. The freed fatty acids and monoglycerides are transported to the adipose cell, where TGs are resynthesized within the smooth endoplasmic reticulum and added to the lipid droplet.

> Lipoprotein lipase is found associated with endothelial cells in many different tissues and functions in a similar way in the supply of fatty acids to local cells for storage or oxidation. The activity of this enzyme changes in response to foreign invasion. Cachectin (tumor necrosis factor), a protein synthesized by macrophages in response to pathogens and endotoxins, inhibits lipoprotein lipase, causing the weight loss characteristic of wasting in chronic disease.
>
> The breakdown, or lipolysis, of triglycerides in cells is regulated by a **hormone-sensitive lipase** (HSL) within the adipocyte that is controlled both by hormones (e.g., epinephrine and glucagon) and neurotransmitters (e.g., norepinephrine). Each adipocyte is covered with many different receptors for these mediators.


A
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