Subject IV HEMOPOIESIS. LYMPHOID SYSTEM

Program of lecture

- **Embryonic and postembryonic hematopoiesis: characteristics.**
- **Organs of hematopoiesis.**
- **Immunity, immune defense reactions.**
- **Immunocytes: classification, morpho-functional features.**
- **Immunoglobulins and plasma cells.**
- **Macrophageal system.**
- **Functional relations between hemotopoetic and immune organs, T- and B-zones. Age changes. Regeneration and involution features.**
- **Conception about thymico-lymphatic status**.

The lymphoid system is responsible for the immunological defense of the body. Some of its component organs-**lymph nodes, thymus,** and **spleen**-are surrounded by connective tissue capsules, whereas its other components, members of the **diffuse lymphoid system,** are not encapsulated. The cells of the lymphoid system protect the body against foreign macromolecules, viruses, bacteria, and other invasive microorganisms, and they kill virally transformed cells.

The lymphoid organs are classified into two categories:

- **1 Primary (central) lymphoid organs** are responsible for the development and maturation of lymphocytes into mature, immunocompetent cells.
- **2 Secondary (peripheral) lymphoid organs** are responsible for the proper environment in which immunocompetent cells can react with each other, as well as with antigens and other cells, to mount an immunological challenge against invading antigens or pathogens.

The immune system provides the second and the third lines of defense against invading pathogens. The first line of defense is the epithelial barrier, namely skin and mucosa, which forms a complete lining and covering of the body surfaces. Once this physical barrier is breached by a cut, tear, or abrasion, or even if foreign substances are able to penetrate, but have not yet penetrated, the intact barrier, the second and the third lines of defense may become activated; these are the innate and the adaptive immune systems.

The **innate immune system** (natural immune system) is nonspecific and is composed of (1) a system of blood-borne macromolecules known as **complement;** (2) groups of cells known as **macrophages** and **neutrophils,** which phagocytose invaders; and (3) another group of cells, **natural killer (NK) cells,** which kill tumor cells, virally infected cells, bacteria, and parasites.

The **adaptive immune system** (acquired immune system) is responsible for eliminating threats from specific invaders. Whereas a macrophage can phagocytose most bacteria, the adaptive immune system not only reacts against one specific antigenic component of a pathogen, but also its ability to react against that particular component improves with subsequent confrontations with it.

Embryonic hemopoiesis

Blood cell formation begins 2 weeks after conception **(mesoblastic phase)** in the mesoderm of the yolk sac, where mesenchymal cells aggregate into clusters known as **blood**

islands. The peripheral cells of these islands form the vessel wall, and the remaining cells become **erythroblasts,** which differentiate into nucleated **erythrocytes.**

The mesoblastic phase begins to be replaced by the **hepatic phase** by the 6th week of gestation. The erythrocytes still have nuclei, and leukocytes appear by the 8th week of gestation. The **splenic phase** begins during the second trimester, and both hepatic and splenic phases continue until the end of gestation (Fig. 1).

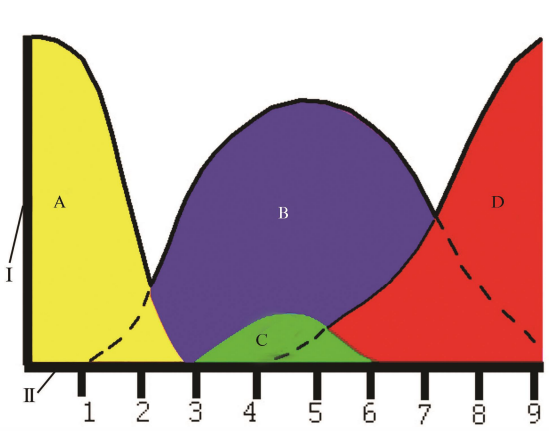

Fig. 1

Hemopoiesis begins in the bone marrow **(myeloid phase)** by the end of the second trimester. As the skeletal system continues to develop, the bone marrow assumes an increasing role in blood cell formation. Although postnatally the liver and the spleen are not active in hemopoiesis, they can revert to forming new blood cells if the need arises.

Postnatal (postembryonic) hemopoiesis

Because all blood cells have a finite life span, they must be replaced continuously. This replacement is accomplished by hemopoiesis, starting from a common population of stem cells within the bone marrow. On a daily basis, more than 10^{11} blood cells are produced in the marrow to replace cells that leave the bloodstream, die, or are destroyed. During hemopoiesis, stem cells undergo multiple cell divisions and differentiate through several intermediate stages, eventually giving rise to the mature blood cells discussed earlier. The entire process is regulated by various growth factors and cytokines that act at different steps to control the type of cells formed and their rate of formation (Fig. 2).

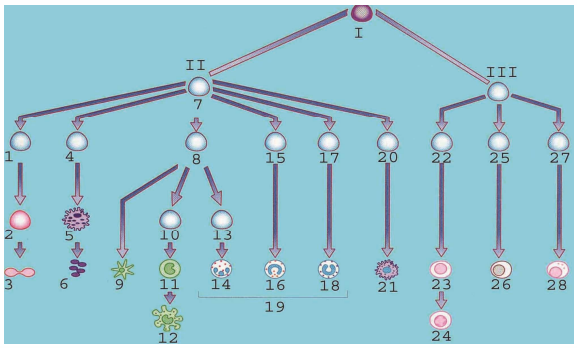

Fig. 2

Stem Cells, Progenitor Cells, and Precursor Cells (Fig. 3).

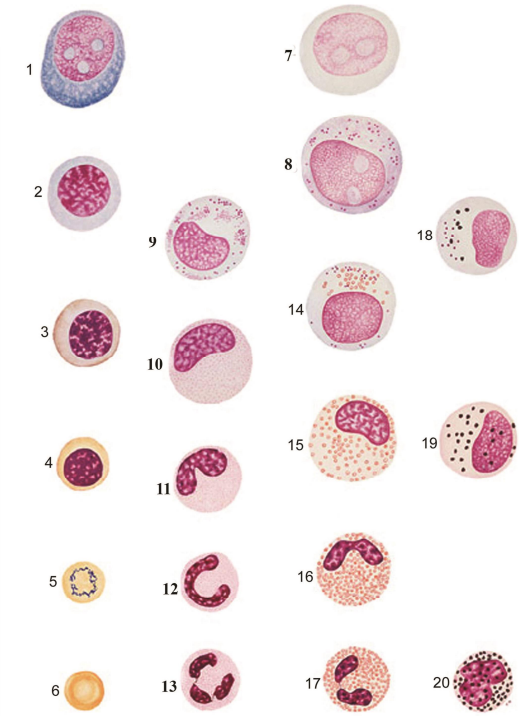

Fig. 3

All blood cells arise from **pluripotential hemopoietic stem cells (PHSCs),** which account for about 0.1% of the nucleated cell population of bone marrow. They are usually amitotic but may undergo bursts of cell division, giving rise to more PHSCs as well as to two types of **multipotential hemopoietic stem cells (MHSCs): colony-forming unit-lymphocyte (CFU-Ly) cells** and **colony-forming unit-granulocyte, erythrocyte, monocyte, megakaryocyte (CFU-GEMM) cells**, previously known as colony-forming unit-spleen [CFU-S] cells. These two populations of MHSCs are responsible for the formation of various progenitor cells. CFU-GEMM cells are predecessors of the **myeloid cell lines** (erythrocytes, granulocytes, monocytes, and platelets); CFU-Ly cells are predecessors of the **lymphoid cell lines** (T cells and B cells). Both PHSCs and MHSCs resemble lymphocytes and constitute a small fraction of the null-cell population of circulating blood.

Stem cells are commonly in the G_0 stage of the cell cycle but can be driven into the G_1 stage by various growth factors and cytokines. Early stem cells may be recognized because they express the specific marker molecules CD34, p170 pump, and c-*kit* on their plasma membranes. **Homeobox genes** may be active in the differentiation of the early stages of hemopoietic cells, specifically *Hox1* in the myeloid (but not erythroid) cell lines and certain members of the *Hox2* group in the erythroid (but not myeloid) cell lines.

Progenitor cells also resemble small lymphocytes but are **unipotential** (i.e., committed to forming a single cell line, such as eosinophils). Their mitotic activity and differentiation are controlled by specific hemopoietic factors. These cells have only limited capacity for self-renewal.

Precursor cells arise from progenitor cells and are incapable of self-renewal. They have specific morphological characteristics that permit them to be recognized as the first cell of a particular cell line. Precursor cells undergo cell division and differentiation, eventually giving rise to a clone of mature cells. As cell maturation and differentiation proceed, succeeding cells become smaller, their nucleoli disappear, their chromatin network becomes denser, and the morphological characteristics of their cytoplasm approximate those of the mature cells.

Researchers studying hemopoiesis have isolated individual lymphocyte-like cells that, under proper conditions, occasionally give rise to groups (*colonies*) of cells composed of granulocytes, erythrocytes, monocytes, lymphocytes, and platelets. Thus, it has been shown that all blood cells are derived from a single **pluripotential stem cell.** More frequently, however, isolated individual cells give rise to only erythrocytes or eosinophils or another type of blood cell. Because these experiments used the spleen as the site of hemopoiesis, the individual lymphocyte-like cells were originally called colony-forming units-spleen (CFU-S), but have been renamed to describe their function to CFU-GEMM. Careful observations have shown that, as stated previously, there are two types of multipotential cells (CFU-GEMM and CFU-Ly) that give rise to the myeloid series of cells and lymphocytes, respectively. Newer research has demonstrated that each precursor cell has a unipotential CFU as its predecessor. Precursor cells undergo a series of cell divisions and differentiations to yield the mature cell.

Hemopoietic Growth Factors (Colony-Stimulating Factors)

Hemopoiesis is regulated by numerous growth factors produced by various cell types. Each factor acts on specific stem cells, progenitor cells, and precursor cells, generally inducing rapid mitosis, differentiation, or both (Table 10-6). Some of these growth factors also promote the functioning of mature blood cells. Most hemopoietic growth factors are glycoproteins.

Three routes are used to deliver growth factors to their target cells: (1) transport via the bloodstream (as endocrine hormones), (2) secretion by stromal cells of the bone marrow near the hemopoietic cells (as paracrine hormones), and (3) direct cell-to-cell contact (as surface signaling molecules).

Erythropoiesis

The process of erythropoiesis, red blood cell formation, generates 2.5×10^{11} erythrocytes every day. In order to produce such a tremendous number of cells, two types of unipotential progenitor cells arise from the CFU-GEMM: the **burst-forming units-erythrocyte (BFU-E)** and **colony-forming units-erythrocyte (CFU-E).**

If the circulating red blood cell level is low, the kidney produces a high concentration of **erythropoietin,** which, in the presence of IL-3, IL-9, steel factor, and GM-CSF, induces CFU-GEMM to differentiate into BFU-E. These cells undergo a "burst" of mitotic activity, forming a large number of CFU-E. Interestingly, this transformation requires the loss of IL-3 receptors

CFU-E require a low concentration of erythropoietin not only to survive but also to form the first recognizable erythrocyte precursor, the **proerythroblast**. The proerythroblasts and their progeny form spherical clusters around macrophages **(nurse cells)** which phagocytose extruded nuclei and excess or deformed erythrocytes. Nurse cells may also provide growth factors to assist erythropoiesis.

Granulocytopoiesis

Although the granulocytic series usually is discussed under a single heading, as it is here, the three types of granulocytes are actually derived from their own unipotential (or bipotential, as with neutrophils) stem cells. Each of these stem cells is a descendant of the pluripotential stem cell CFU-GEMM. Thus, CFU-Eo, of the eosinophil lineage, and CFU-Ba, of the basophil lineage, each undergo cell division, giving rise to the precursor cell, or **myeloblast.** Neutrophils originate from the bipotential stem cell, **CFU-GM,** whose mitosis produces two unipotential

stem cells, **CFU-G** (of the neutrophil line) and **CFU-M,** responsible for the monocyte lineage. Similar to CFU-Ba and CFU-Eo, CFU-G divides to give rise to myeloblasts.

The proliferation and differentiation of these stem cells are under the influence of G-CSF, GM-CSF, and IL-5. Therefore, these three factors facilitate the development of neutrophils, basophils, and eosinophils. In turn, IL-1, IL-6, and TNF-Ŭ are cofactors necessary for the synthesis and release of G-CSF and GM-CSF. In addition, IL-5 may also play a role in the activation of eosinophils.

Myeloblasts are precursors of all three types of granulocytes, and they cannot be differentiated from one another. It is not known whether a single myeloblast can produce all three types of granulocytes or whether there is a specific myeloblast for each type of granulocyte. Myeloblasts undergo mitosis, giving rise to promyelocytes, which in turn divide to form myelocytes. It is at the myelocyte step that specific granules are present and the three granulocyte lines may be recognized. Each day, the average adult produces approximately 800,000 neutrophils, 170,000 eosinophils, and 60,000 basophils.

Newly formed neutrophils leave the hemopoietic cords by *piercing* the endothelial cells lining the sinusoids rather than by *migrating* between them. Once neutrophils enter the circulatory system, they **marginate;** that is, they adhere to the endothelial cells of the blood vessels and remain there until they are needed. The process of margination requires the sequential expression of various transmembrane adhesion molecules and integrins by the neutrophils as well as of specific surface receptor molecules by the endothelial cells, the description of which is beyond the scope of this textbook. Because of the process of margination, there are always many more neutrophils in the circulatory system than in the circulating blood.

Monocytopoiesis

Monocytes share their bipotential cells with neutrophils. CFU-GM undergoes mitosis and gives rise to CFU-G and **CFU-M (monoblasts).** The progeny of CFU-M are **promonocytes,** large cells (16 to 18 ε m in diameter) that have a kidney-shaped, acentrically located nucleus. The cytoplasm of promonocytes is bluish and houses numerous azurophilic granules.

Electron micrographs of promonocytes disclose a well-developed Golgi apparatus, abundant RER, and numerous mitochondria. The azurophilic granules are lysosomes, about 0.5 ε m in diameter. Every day, the average adult forms more than 10^{10} monocytes, most of which enter the circulation. Within a day or two, the newly formed monocytes enter the connective tissue spaces of the body and differentiate into **macrophages.**

Platelet Formation

The unipotential platelet progenitor, **CFU-Meg,** gives rise to a very large cell, the **megakaryoblast** (25 to 40 ε m in diameter), whose single nucleus has several lobes. These cells undergo **endomitosis,** whereby the cell does not divide; instead, it becomes larger and the nucleus becomes polyploid, as much as 64 N. The bluish cytoplasm accumulates azurophilic granules. These cells are stimulated to differentiate and proliferate by thrombopoietin.

Megakaryoblasts differentiate into **megakaryocytes**, which are large cells (40 to 100 ε m in diameter), each with a single lobulated nucleus. Electron micrographs of megakaryocytes display a well-developed Golgi apparatus, numerous mitochondria, abundant RER, and many lysosomes.

Megakaryocytes are located next to sinusoids, into which they protrude their cytoplasmic processes. These cytoplasmic processes fragment along complex, narrow invaginations of the

plasmalemma, known as **demarcation channels,** into clusters of **proplatelets.** Shortly after the proplatelets are released, they disperse into individual platelets. Each megakaryocyte can form several thousand platelets. The remaining cytoplasm and nucleus of the megakaryocyte degenerate and are phagocytosed by macrophages.

The Innate Immune System

Although the innate immune system is much older than the adaptive immune system, it responds rapidly, usually within a few hours, to an antigenic invasion; it responds in a nonspecific manner; and has no immunological memory. The critical components of the innate immune system are complement, antimicrobial peptides, cytokines, macrophages, neutrophils, NK cells, and Toll-like receptors (TLRs).

Complement is a series of blood-borne proteins that attack microbes that found their way into the bloodstream. As they precipitate on the surface of these invading pathogens, they form a membrane attack complex (MAC) that damage the microbe's cell membrane. Phagocytic cells, such as neutrophils and macrophages, of the host have receptors for a specific moeity of complement (i.e., C3b) and the presence of C3b on the microbial surface facilitates phagocytosis of microbes by these host defense cells.

The Adaptive Immune System

The adaptive immune response exhibits four distinctive properties: **specificity, diversity, memory,** and **self/nonself recognition**-that is, the ability to distinguish between structures that belong to the organism, self, and those that are foreign, nonself. **T lymphocytes, B lymphocytes,** and specialized macrophages known as **antigen-presenting cells (APCs)** participate in the (adaptive) immune response. These cells communicate with members of the innate immune system as well as with each other by signaling molecules **(cytokines),** which are released in response to encounters with foreign substances called **antigens.**

Recognition of a substance as foreign by the immune system stimulates a complex sequence of reactions that result either in the production of **immunoglobulins** (also known as **antibodies**), which bind to the antigen, or in the induction of a group of cells that specialize in cytotoxicity, namely the killing of the foreign cell or altered self-cell (e.g., tumor cell). The immune response that depends on the formation of antibodies is called the **humoral immune response,** whereas the cytotoxic response is known as the **cell-mediated immune response.**

The cells that constitute the functional components of the innate and adaptive immune system (T cells, B cells, macrophages, and their subcategory, APCs) are all formed in the bone marrow. B cells become immunocompetent in the bone marrow, whereas T cells migrate to the thymus to become immunocompetent; therefore, bone marrow and the thymus are called the **primary (central) lymphoid organs**. After lymphocytes become immunocompetent in the bone marrow or in the thymus, they migrate to the **secondary (peripheral) lymphoid organs**-diffuse lymphoid tissue, lymph nodes, and spleen-where they come into contact with antigens.

A foreign structure that can elicit an immune response in a particular host is known as an immunogen; an antigen is a molecule that can react with an antibody irrespective of its ability to elicit an immune response. Although not all antigens are immunogens, in this textbook the two terms are considered synonymous, and only the term *antigen* is used.

The region of the antigen that reacts with an antibody, or T-cell receptor, is known as its **epitope,** or antigenic determinant. Each epitope is a small portion of the antigen molecule and consists of only 8 to 12 or 15 to 22 hydrophilic amino acid or sugar residues that are accessible to the immune apparatus. Large foreign invaders such as bacteria have several epitopes, each capable of binding to a different antibody.

Immunoglobulins **(antibodies)** are glycoproteins that inactivate antigens (including viruses) and elicit an extracellular response against invading microorganisms. The response may

involve phagocytosis in the connective tissue spaces by macrophages (or neutrophils) or the activation of the blood-borne **complement system (Fig. 4).**

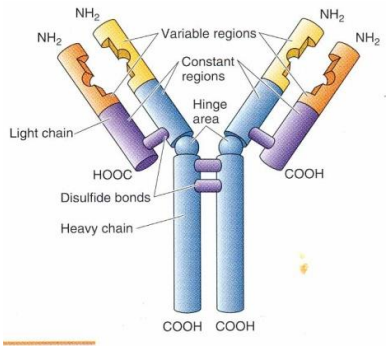

Fig. 4

Immunoglobulins are manufactured in large numbers by plasma cells, which release them into the lymph or blood vascular system. The typical antibody is immunoglobulin G (IgG). Each IgG is a Y-shaped molecule, composed of two long, identical 55- to 70-kDa polypeptides, known as **heavy chains,** and two shorter, identical 25-kDa polypeptides, the **light chains.** The four chains are bound to each other by several disulfide bonds and noncovalent bonds in such a way that the stem of the Y is composed only of heavy chains and the diverging arms consist of both light and heavy chains (Fig.4, 5).

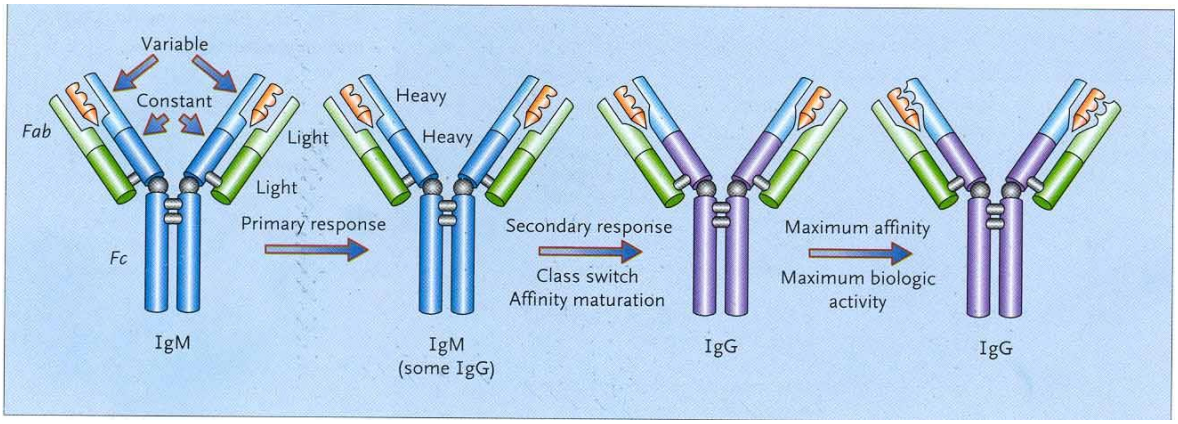

Fig. 5

The region in the vicinity of the sulfide bonds between the two heavy chains-the **hinge region**-is flexible and permits the arms to move away from or toward each other. The distal regions on the tips of the arms (the four amino-terminal segments) are responsible for binding to the epitope; hence, each antibody molecule can bind two *identical* epitopes. The enzyme papain cleaves the antibody molecule at its hinge regions (see Fig. 12-1), forming three fragments: one **Fc fragment** composed of the stem of the Y and containing equal parts of the two heavy chains, and two **Fab fragments,** each composed of the remaining part of one heavy chain and one entire light chain. Fc fragments are easily crystallized (hence the "c" designation), whereas the Fab fragment is the *a*ntigen-*b*inding region of the antibody (hence the "ab" designation) (Fig. 5).

The amino acid sequence of the Fc fragment is mostly constant in its class; thus the stem of an antibody binds to Fc receptors of many different cells. The amino acid sequence of the Fab region is variable, and it is the alterations of that sequence that determine the **specificity** of the antibody molecule for its specific antigen. The amino acid sequence of the Fc fragment is mostly constant in its class; thus the stem of an antibody binds to Fc receptors of many different cells. The amino acid sequence of the Fab region is variable, and it is the alterations of that sequence that determine the **specificity** of the antibody molecule for its specific antigen.

- **IgM,** which resembles five IgG molecules bound to each other (pentameric form of immunoglobulin)
- **IgA,** which resembles two IgG molecules bound to each other (dimeric form of immunoglobulin)
- **IgG,** the monomeric form of immunoglobulin described earlier
- **IgD,** which is present in very low concentration in the blood, but is found on the B-cell surface as a monomeric form of immunoglobulin known as surface IgD (sIgD)
- **IgE,** a monomeric form of immunoglobulin present on the surface of basophils and mast cells **B lymphocytes** B lymphocytes, also known as **B cells,** are small lymphocytes that both originate and become **immunocompetent** in the bone marrow. However, in birds, in which B cells were first identified, they become immunocompetent in a diverticulum of the cloaca, known as the **bursa of Fabricius** (hence "B" cells). During the process of becoming immunocompetent, each cell manufactures 50,000 to 100,000 IgM and IgD immunoglobulins and inserts these in its plasma membrane so that the epitope-binding sites of the antibodies face the extracellular space. The Fc region of the antibody is embedded in the phospholipid bilayer with the assistance of two pairs of transmembrane proteins, Igβ and IgŬ, whose carboxyl termini are in contact with intracellular protein complexes. Every member of a particular clone of B cells has antibodies that bind to the same epitope (Fig. 6).When the surface immunoglobulin reacts with its epitope, the Igβ and IgŬ transduce (relay) the information to the intracellular protein complex with which they are in contact, initiating a chain of events that results in **activation** of the B cell. The activated B cell undergoes mitosis, forming antibody-producing **plasma cells** and **B memory cells,** as discussed earlier. Because the antibodies produced by plasma cells are released either into the blood or into the lymph circulation, B cells are responsible for the **humorally mediated immune response.**

T lymphocytes

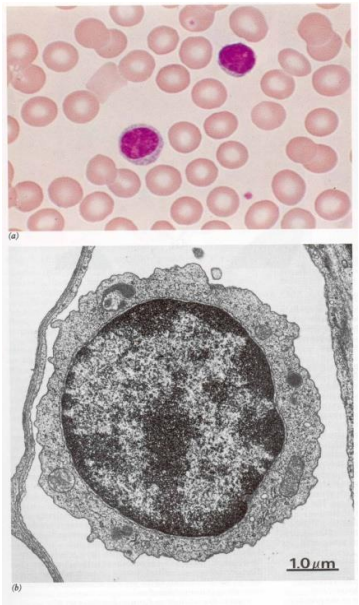

Fig. 6

T lymphoctes **(T cells)** also are formed in the bone marrow, but they migrate to the thymic cortex, where they become immunocompetent by expressing specific molecules on their cell membranes that permit them to perform their functions (Fig. 7).

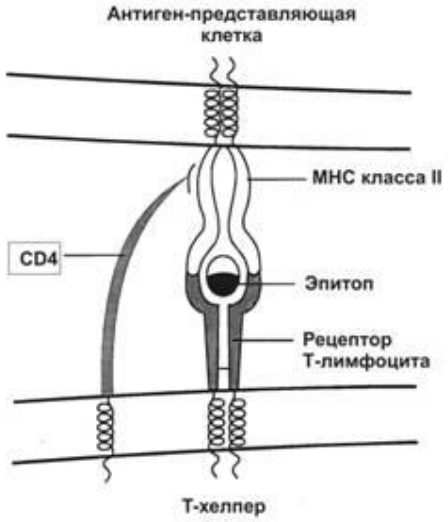

Fig. 7

Although histologically T cells appear to be identical to B cells, there are important differences between them:

- T cells have TCRs rather than sIgs on their cell surface.
- T cells recognize only epitopes presented to them by other cells (APCs).
- T cells respond only to protein antigens.
- T cells perform their functions only at short distances

Similar to sIgs on B cells, **TCRs** on the plasmalemma of T cells function as antigen receptors. The **constant regions** of the TCR are membrane-bound, whereas the variable **amino-terminal regions** containing the antigen-binding sites extend from the cell surface. In addition to TCR molecules, T cells express **clusters of differentiation proteins (CD molecules or CD markers)** on their plasmalemma. These accessory proteins bind to specific ligands on target cells. Although almost 200 CD molecules are known, The membrane-bound portion of the TCR associates with the membrane proteins, CD3, and either CD4 or CD8, forming the **TCR complex.** Several other membrane proteins play roles in signal transduction and in strengthening the interaction between the TCR and an epitope, thus facilitating antigen-stimulated T-cell activation (Fig. 8).

A TCR can recognize an epitope only if the epitope is a polypeptide (composed of amino acids) and if the epitope is bound to a **major histocompatibility complex (MHC) molecule,** such as those in the plasmalemma of an APC. There are two classes of these glycoproteins: MHC class I and MHC class II. Most nucleated cells express MHC I molecules on their surface, whereas APCs (discussed later) can express both MHC I and MHC II molecules on their plasmalemma. The MHC molecules are unique in each individual (except for identical twins), and to be activated, T cells must recognize not only the foreign epitope but also the MHC molecule as self. If a T cell recognizes the epitope but not the MHC molecule, it does not become stimulated; hence, the T cell's capacity to act against an epitope is **MHC-restricted.**

There are three types of T cells, some with two or more subtypes:

- Naïve T cells

- Memory T cells
- Effector T cells

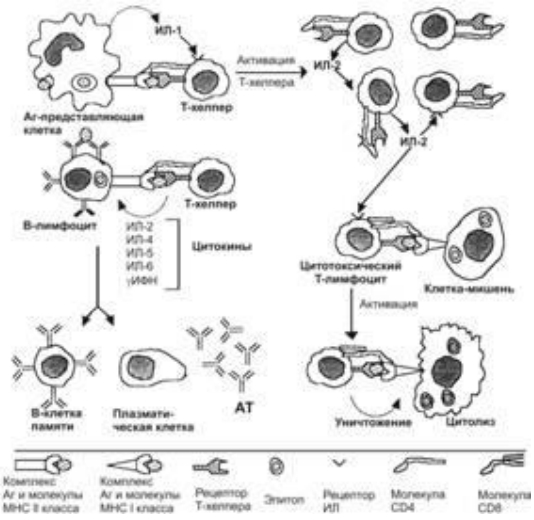

Fig. 8

Major Histocompatibility Complex Molecules

The prime importance of MHC molecules is to permit APCs and cells under viral attack (or cells already virally transformed) to present the epitopes of the invading pathogen to the T cells. These epitopes are short polypeptides that fit into a groove on the surface of the MHC molecule.

There are two classes of MHC molecules:

- **MHC I molecules** function in presenting short polypeptide fragments (8 to 12 amino acids in length) derived from endogenous proteins (i.e., proteins manufactured by the cell).
- **MHC II molecules** function in presenting longer polypeptide fragments (13 to 25 amino acids in length) derived from exogenous proteins (i.e., proteins that were phagocytosed and cleaved by these cells from the extracellular space).

Antigen-Presenting Cells (APCs)

APCs phagocytose, catabolize, and process antigens, attach their epitopes to MHC II molecules, and present this complex to T cells. Most APCs are derived from monocytes and therefore belong to the mononuclear phagocyte system. APCs include macrophages, dendritic cells (such as Langerhans cells of the epidermis and oral mucosa), and two types of non-monocyte-derived cells (B cells and epithelial reticular cells of the thymus).

Similar to T_H cells, APCs manufacture and release **cytokines.** These signaling molecules are needed to activate target cells to perform their specific functions, not only in the immune response but also in other processes.

Interaction among the Lymphoid Cells

Cells of the lymphoid system interact with each other to effect an immune response. The process of interaction is regulated by recognition of surface molecules; if the molecules are not

recognized, the cell is eliminated to prevent an incorrect response. If the surface molecules are recognized, the lymphocytes proliferate and differentiate. The initiation of these two responses is called **activation.** At least two signals are required for activation:

- Recognition of the antigen (or epitope)
- Recognition of a second, costimulatory signal, which may be mediated by a cytokine or by a membrane-bound signaling molecule

T-Helper Cell-Mediated (T_H2 cells) Humoral Immune Response

Except for thymus-independent antigens, B cells can respond to an antigen only if instructed to do so by the T_H2 cell subtype. When the B cell binds antigens on its sIgs, it internalizes the antigen-antibody complex, removes the epitope and attaches it to MHC II molecules, and places the epitope-MHC II complex on its surface and presents it to a T_H2 cell.

- **Signal 1.** The T_H2 cell not only must recognize the epitope with its TCR but also must recognize the MHC II molecule with its CD4 molecule.
- **Signal 2.** The T_H2 cell's CD40 receptor must bind to the B cell's CD40 molecule, and the T_H2 cell's CD28 has to contact the B cell's CD80 molecule

T-Helper Cell-Mediated (T_H1 cells) Killing of Virally Transformed Cells

In most cases, CTLs need to receive a signal from a T_H1 cell to be capable of killing virally transformed cells. Before that signal can be given, however, the T_H1 cell must be activated by an APC that offers the proper epitope.

- **Signal 1.** The TCR and the CD4 molecule of the T_H1 cell must recognize the epitope-MHC II complex on the surface of an APC. If these events occur, the APC expresses a molecule called **B7** on its surface.
- **Signal 2.** The CD28 molecule of the T_H1 cell binds to the B7 molecule of the APC

The T_H1 cell is now activated and releases IL-2, IFN-ɔ, and TNF. **IFN-ɔ** causes activation and proliferation of the CTL if that CTL is bound to the same APC and if the following conditions are met:

- **Signal 1.** The TCR and the **CD8 molecule** of the CTL must recognize the **epitope-MHC I complex** of the APC; also, the CD28 molecule of the CTL must bind with the B7 molecule of the APC.
- **Signal 2.** IL-2 released by the T_H1 cell binds to the IL-2 receptors of the CTL

T_H1 Cells Assist Macrophages in Killing Bacteria

Bacteria that are phagocytosed by macrophages can readily proliferate within the phagosome (becoming infected) because macrophages cannot destroy these microorganisms unless they are activated by T_H1 cells

- **Signal 1.** The TCR and CD4 molecules of the T_H1 cell must recognize the epitope-MHC II complex of the macrophage that phagocytosed the bacteria.
- **Signal 2.** The T_H1 cell expresses IL-2 receptors on its surface and releases IL-2, which binds to the receptors, thus activating itself

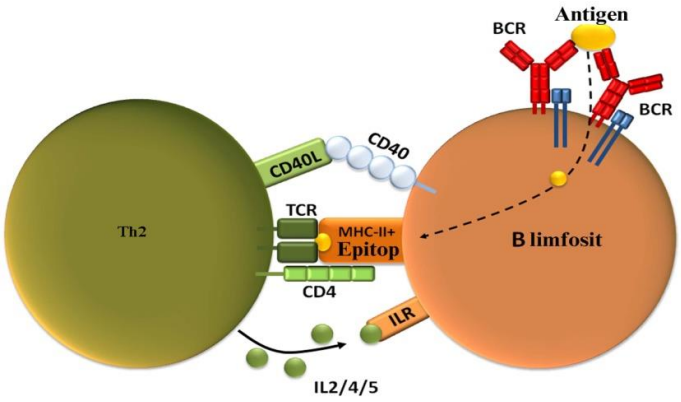

Fig. 9

BONE MARROW

The medullary cavity of long bones and the interstices between trabeculae of spongy bones house the soft, gelatinous, highly vascular, and cellular tissue known as marrow. Bone marrow is isolated from bone by the endosteum (composed of osteoprogenitor cells, osteoblasts, and occasional osteoclasts). Bone marrow constitutes almost 5% of the total body weight. It is responsible for the formation of blood cells **(hemopoiesis)** and their delivery into the circulatory system, and it performs this function from the fifth month of prenatal life until the person dies. Bone marrow also provides a microenvironment for much of the maturation process of B lymphocytes and for the initial maturation of T lymphocytes (Fig. 10).

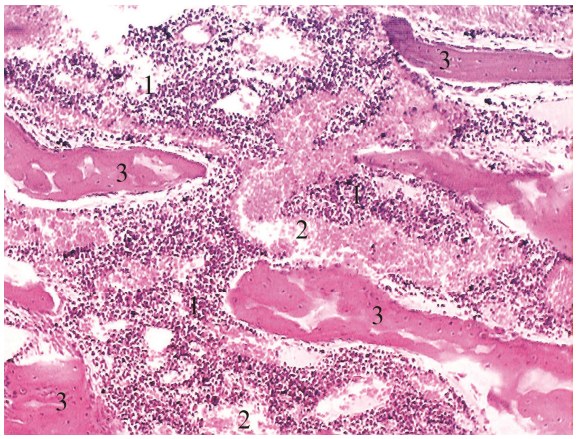

Fig. 10

The marrow of the newborn is called **red marrow** because of the great number of erythrocytes being produced there. By age 20 years, however, the diaphyses of long bones house only **yellow marrow** because of the accumulation of large quantities of fat and the absence of hemopoiesis in the shafts of these bones.

The vascular supply of bone marrow is derived from the nutrient arteries that pierce the diaphysis via the nutrient foramina, tunnels leading from the outside surface of bone into the medullary cavity. These arteries enter the marrow cavity and give rise to a number of small, peripherally located vessels that provide numerous branches both centrally, to the marrow, and peripherally, to the cortical bone. Vessels entering the cortical bone are distributed through the haversian and Volkmann canals to serve the compact bone (Fig. 11).

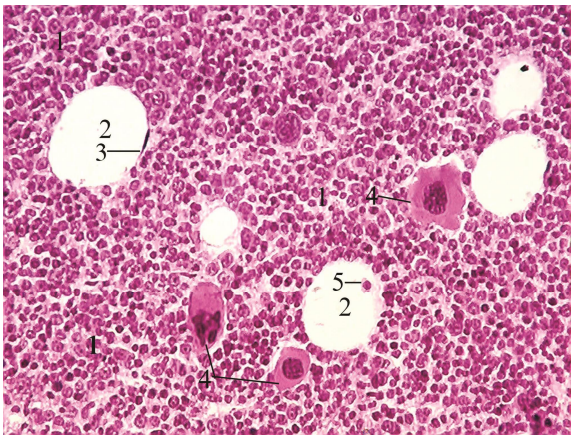

Fig. 11

The centrally directed branches deliver their blood to the extensive network of large **sinusoids** (45 to 80 εm in diameter). The sinusoids drain into a **central longitudinal vein,** which is drained by veins leaving the bone via the nutrient canal (Fig. 11).

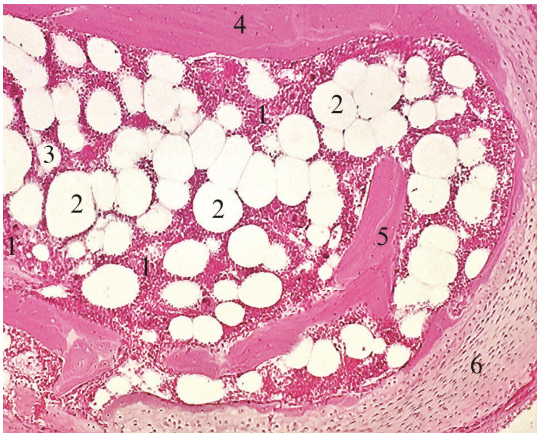

Fig. 12

It is interesting that the veins are *smaller* than the arteries, thus establishing high hydrostatic pressure within the sinusoids, thus preventing their collapse. The veins, arteries, and sinusoids form the **vascular compartment,** and the intervening spaces are filled with pleomorphic **islands of hemopoietic cells** that merge with each other, forming the **hemopoietic compartment** (Fig. 12).

Thymus

The thymus, situated in the superior mediastinum and extending over the great vessels of the heart, is a small encapsulated organ composed of two **lobes.** Each lobe arises separately in the third (and possibly fourth) pharyngeal pouches of the embryo. The T lymphocytes that enter the thymus to become instructed to achieve immunological competence arise from mesoderm (Fig. 13).

The thymus originates early in the embryo and continues to grow until puberty, when it may weigh as much as 35 to 40 g. After the first few years of life, the thymus begins to **involute** (atrophy) and becomes infiltrated by adipose cells. However, it may continue to function even in older adults.

The capsule of the thymus, composed of dense, irregular collagenous connective tissue, sends septa into the lobes, subdividing them into incomplete **lobules** (Fig. 14). Each lobule is composed of a cortex and a medulla, although the medullae of adjacent lobules are confluent with each other.

The cortex of the thymus appears much darker histologically than does the medulla because of the presence of a large number of **T lymphocytes (thymocytes)**. Immunologically incompetent T cells leave the bone marrow and migrate to the periphery of the thymic cortex, where they undergo extensive proliferation and instruction to become immunocompetent T cells. In addition to the lymphocytes, the cortex houses macrophages and **epithelial reticular cells.** It is believed that in humans epithelial reticular cells are derived from the endoderm of the third (and possibly fourth) pharyngeal pouch.

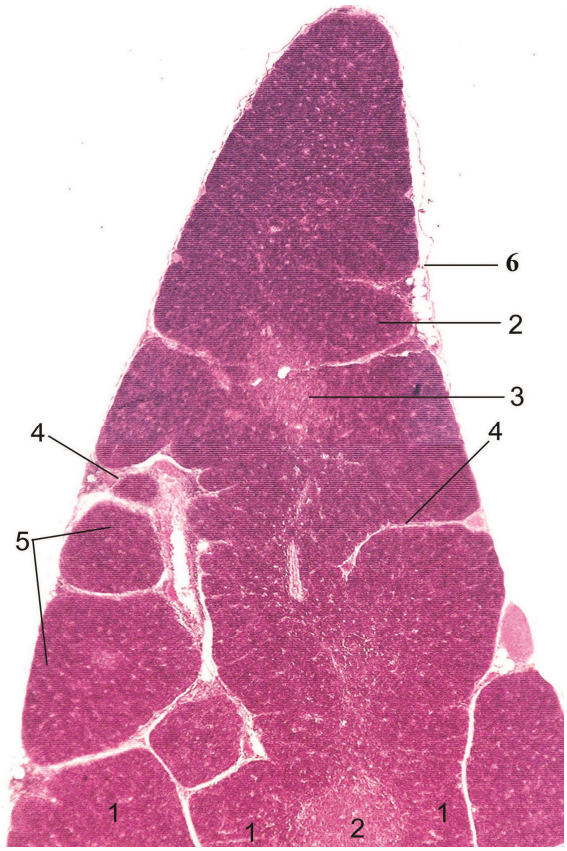

Fig. 13

The capillaries of the cortex are of the **continuous** type, possess a thick basal lamina, and are invested by a sheath of type I epithelial reticular cells that form a **blood-thymus barrier.** Thus, the developing T cells of the cortex are protected from contacting blood-borne macromolecules. However, self-macromolecules are permitted to cross the blood-thymus barrier (probably controlled by the epithelial reticular cells), possibly to eliminate those T cells that are programmed against self-antigens. The cortical capillary network drains into small venules in the medulla (Fig. 14, 15).

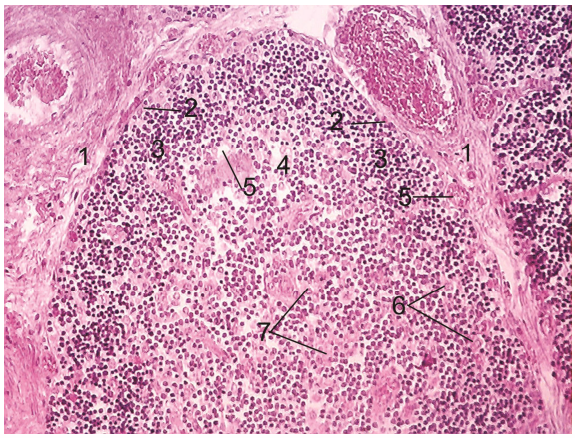

Fig. 14

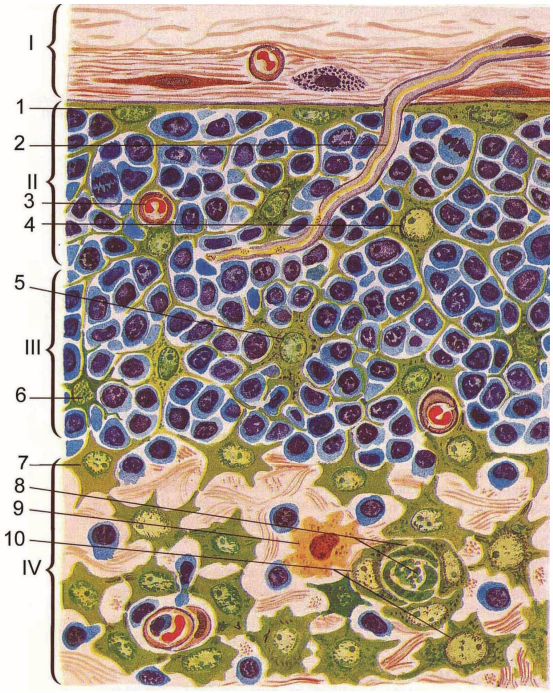

Fig. 15

Newly formed, immunologically incompetent T cells arriving from the bone marrow leave the vascular supply at the corticomedullary junction and migrate to the periphery of the cortex. As these cells mature, they move deeper into the cortex and enter the medulla as naïve but immunocompetent cells. They leave the medulla via veins draining the thymus (Fig. 16, 17).

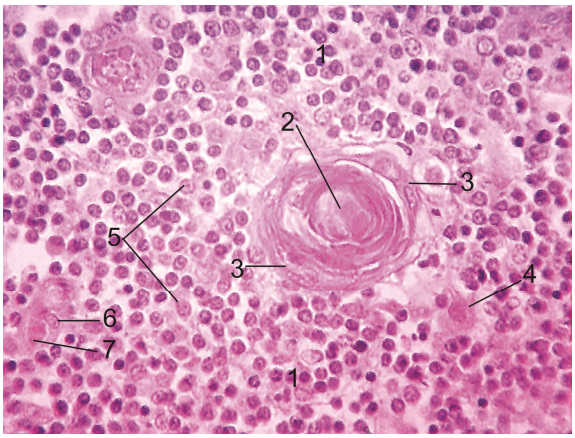

Fig. 16

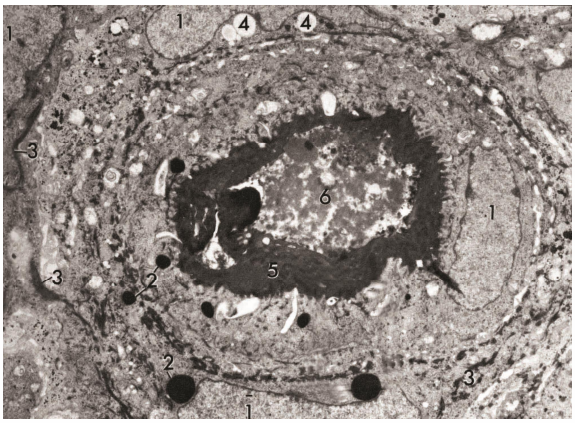

Fig. 17

The developing T cells proliferate extensively in the cortex, begin to express their surface markers, and are tested for their ability to recognize **self-MHC molecules** and **self-epitopes.** T cells that are unable to recognize self-MHC I and self-MHC II molecules are destroyed by being driven into apoptosis. Additionally, those T lymphocytes whose TCRs are programmed against self-macromolecules are also destroyed.

The process of testing for self-MHC molecules and self-epitopes is believed to be a function of type II and type III epithelial reticular cells and of bone marrow-derived dendritic cells, because these three cell types express both classes of the epitope-MHC molecule complex on their surface.

Spleen

The spleen, the largest lymphoid organ in the body, is located in the peritoneum in the upper left quadrant of the abdominal cavity. Its dense, irregular fibroelastic connective tissue capsule, occasionally housing **smooth muscle cells,** is surrounded by visceral peritoneum. The simple squamous epithelium of the peritoneum provides a smooth surface for the spleen. The spleen functions not only in the immunological capacity of antibody formation and T-cell and B-cell proliferation but also as a filter of the blood in destroying old erythrocytes. During fetal development, the spleen is a hemopoietic organ; if necessary, it can resume that function in the adult. Additionally, in some animals (but not in humans), the spleen acts as a reservoir of red blood cells, which may be released into circulation as the need arises (Fig. 18, 19).

The spleen has a convex surface as well as a concave aspect known as the **hilum.** The capsule of the spleen is thickened at the hilum, and it is here where arteries and their accompanying nerve fibers enter and veins and lymph vessels leave the spleen. The trabeculae, arising from the capsule, carry blood vessels into and out of the parenchyma of the spleen (Fig. 19). Histologically, the spleen has a three-dimensional network of **reticular fibers** and associated reticular cells. The reticular fiber network is attached to the capsule as well as to the trabeculae and forms the architectural framework of this organ (Fig. 19, 20). The interstices of the reticular tissue network are occupied by **venous sinuses,** trabeculae conveying blood vessels, and the splenic parenchyma. The cut surface of a fresh spleen shows gray areas surrounded by red areas; the former are called **white pulp** and the latter are known as **red pulp.** Central to the appreciation of the organization and function of the spleen is an understanding of its blood supply.

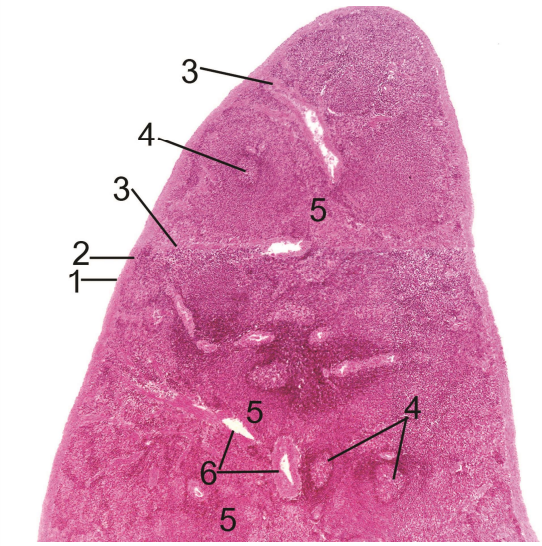

Fig. 18

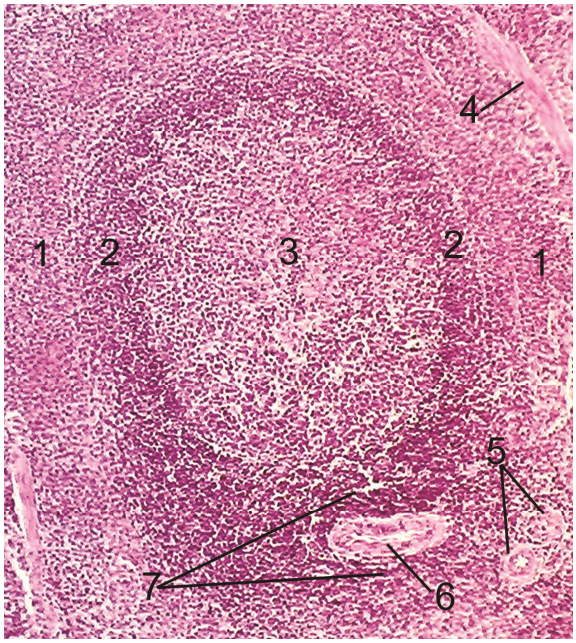

Fig. 19

Vascular Supply of the Spleen

The splenic artery branches repeatedly as it pierces the connective tissue capsule at the hilum of the spleen. Branches of these vessels, **trabecular arteries,** are conveyed into the substance of the spleen by trabeculae of decreasing sizes. When the trabecular arteries are reduced to about 0.2 mm in diameter, they leave the trabeculae. The tunica adventitia of these vessels that left the trabeculae become loosely organized, and they become infiltrated by a sheath of lymphocytes, the **periarterial lymphatic sheath (PALS).** Because this vessel occupies the center of the PALS, it is called the **central artery (Fig. 21).**

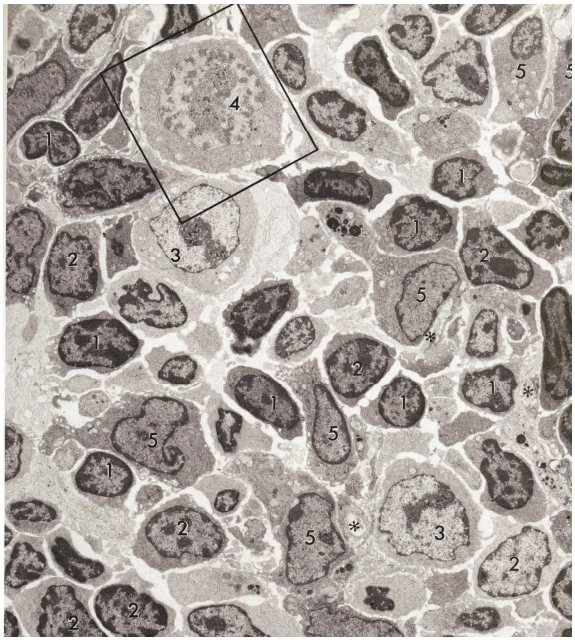

Fig. 20

At its termination, the central splenic artery loses its lymphatic sheath and subdivides into several short, parallel branches, known as **penicillar arteries,** which enter the red pulp. The penicillar arteries have three regions: (1) the **pulp arteriole,** (2) the **sheathed arteriole** (a thickened region of the vessel surrounded by a sheath of macrophages termed the Schweigger-Seidel sheath), and (3) the **terminal arterial capillaries (Fig. 21).**

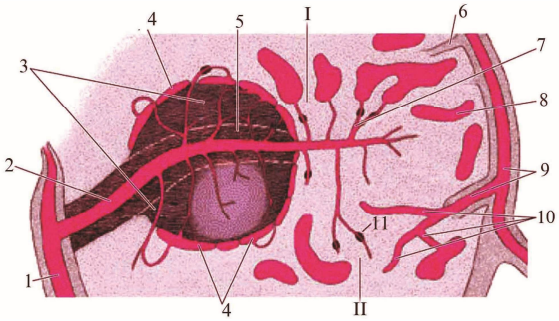

Fig. 21

Although it is known that the terminal arterial capillaries deliver their blood into the splenic sinuses, the method of delivery is not completely understood, which has prompted the formulation of three theories of spleen circulation: (1) closed circulation, (2) open circulation, and (3) a combination of the first two theories.

Proponents of the **closed circulation theory** believe that the endothelial lining of the terminal arterial capillaries is continuous with the sinus endothelium. Investigators who subscribe to the **open circulation theory** believe that the terminal arterial capillaries terminate

prior to reaching the sinusoids, and blood from these vessels percolates through the red pulp into the sinuses. Still other investigators believe that some vessels connect to the sinusoids and that other vessels terminate as open-ended channels in the red pulp, suggesting that the spleen has both **open and closed systems of circulation** (Fig. 22).

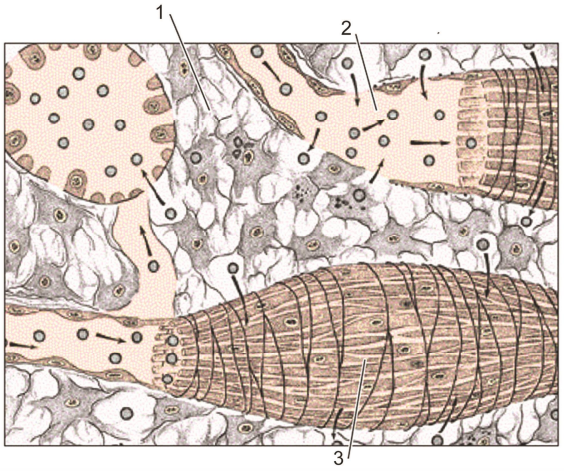

Fig. 22

The structure of the white pulp is closely associated with the central arteriole. The PALS that surrounds the central arteriole is composed of T lymphocytes. Frequently, enclosed within the PALS are **lymphoid nodules,** which are composed of B cells and displace the central arteriole to a peripheral position. Lymphoid nodules may display **germinal centers,** indicative of antigenic challenge. The PALS and lymphoid nodules constitute the white pulp, and as in the lymph node, the T and B cells are stationed in specific locations.

The red pulp resembles a sponge, in that the spaces within the sponge represent the sinuses and the sponge material among the spaces denotes the splenic cords.

Lymph nodes

Lymph nodes are located in various regions of the body but are most prevalent in the neck, in the axilla, in the groin, along major vessels, and in the body cavities. Their parenchyma is composed of collections of T and B lymphocytes, APCs, and macrophages. These lymphoid cells react to the presence of antigens by mounting an immunological response in which macrophages phagocytose bacteria and other microorganisms that enter the lymph node by way of the lymph (Fig.23).

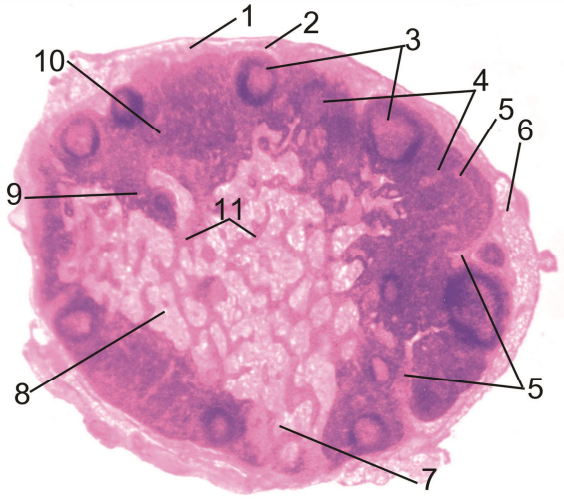

Fig. 23

Each lymph node is a relatively small, soft structure that is less than 3 cm in diameter and that has a fibrous connective tissue capsule, usually surrounded by adipose tissue. It has a convex surface that is perforated by **afferent lymph vessels** that have **valves,** which ensure that lymph from those vessels enters the substance of the node. The concave surface of the node, the **hilum,** is the site of arteries and veins entering and exiting the node. Additionally, lymph leaves the node via the **efferent lymph vessels,** which are also located at the hilum. The efferent lymph vessels have valves that prevent regurgitation of lymph back into the node **(Fig. 24)**.

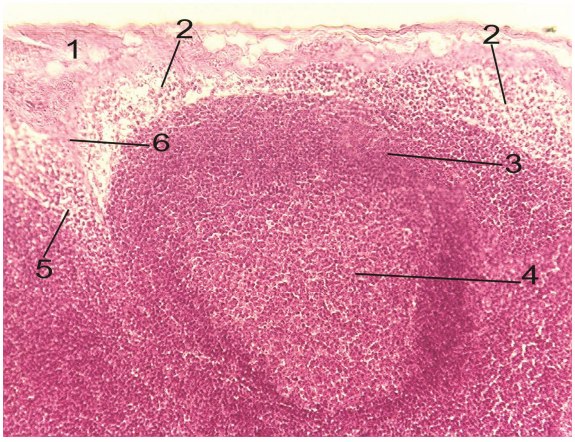

Fig. 24

Lymph Node Cortex (Fig. 25).

The dense, irregular, collagenous connective tissue **capsule** sends **trabeculae** into the substance of the lymph node, subdividing the outer region of the **cortex** into incomplete compartments that extend to the vicinity of the hilum. The capsule is thickened at the hilum, and as vessels enter the substance of the node, they are surrounded by a connective tissue sheath derived from the capsule. Suspended from the capsule and trabeculae is a three-dimensional network of reticular connective tissue that forms the architectural framework of the entire lymph node.

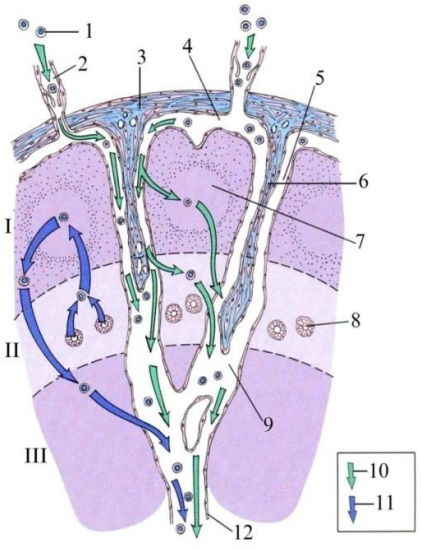

Fig. 25

The afferent lymph vessels pierce the capsule on the convex surface of the node and empty their lymph into the **subcapsular sinus,** which is located just deep to the capsule. This sinus is continuous with the **cortical sinuses (paratrabecular sinuses)** that parallel the trabeculae and deliver the lymph into the **medullary sinuses**, eventually to enter the **efferent lymphatic vessels.** These sinuses have a network of stellate reticular cells whose processes contact those of other cells and the endothelium-like simple squamous epithelium. **Macrophages,** attached to the stellate reticular cells, avidly phagocytose foreign particulate matter. Additionally, lymphoid cells can enter or leave the sinusoids by passing between their squamous cell lining (Fig.25).

The incomplete compartments within the cortex house **primary lymphoid nodules,** which are spherical aggregates of B lymphocytes (both virgin B cells and B memory cells) that are in the process of entering or leaving the lymph node. Frequently, the centers of the lymphoid nodules are stained pale and house **germinal centers,** and these lymph nodules are then known as **secondary lymphoid nodules.** Secondary lymphoid nodules form only in response to an antigenic challenge; it is believed that they are the sites of B memory cell and plasma cell generation.

APCs (e.g., Langerhans cells from skin or dendritic cells from the mucosa) migrate to the paracortex region of the lymph node to present their epitope-MHC II complex to T_H cells. If T_H cells become activated, they proliferate, increasing the width of the paracortex to such an extent that it may intrude deep into the medulla. Newly formed T cells then migrate to the medullary sinuses, leave the lymph node, and proceed to the area of antigenic activity.

High endothelial venules (HEVs) are located in the paracortex. Lymphocytes leave the vascular supply by migrating between the cuboidal cells of this unusual endothelium and enter the substance of the lymph node. B cells migrate to the outer cortex, whereas most T cells remain in the paracortex (Fig.26).

Vascularization of the Lymph Node

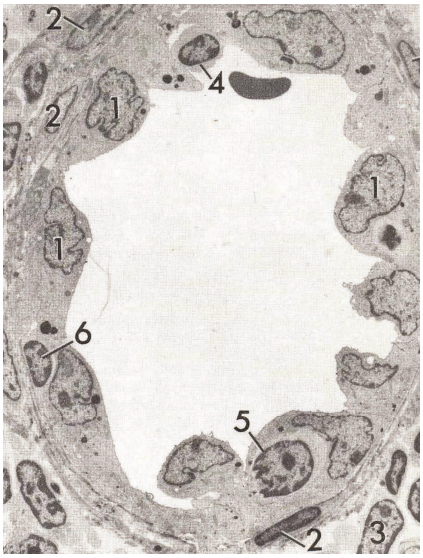

Fig. 26

The arterial supply enters the substance of lymph nodes at the hilum. The vessels course through the medulla within trabeculae and become smaller as they repeatedly branch. Eventually, they lose their connective tissue sheath, travel within the substance of medullary cords, and contribute to the formation of the medullary capillary beds. The small branches of the arteries continue in the medullary cords until they reach the cortex. Here they form a cortical

capillary bed, which is drained by **postcapillary venules.** Blood from postcapillary venules drains into larger veins, which exit the lymph node at the hilum (Fig. 25).

Mucosa-associated lymphoid tissue (MALT)

Mucosa-associated lymphoid tissue (MALT) is composed of a nonencapsulated, localized lymphocyte infiltration and lymphoid nodules in the mucosa of the gastrointestinal, respiratory, and urinary tracts. The best examples of these accumulations are those associated with the mucosa of the gut: **gut-associated lymphoid tissue (GALT), bronchus-associated lymphatic tissue (BALT),** and the **tonsils.**