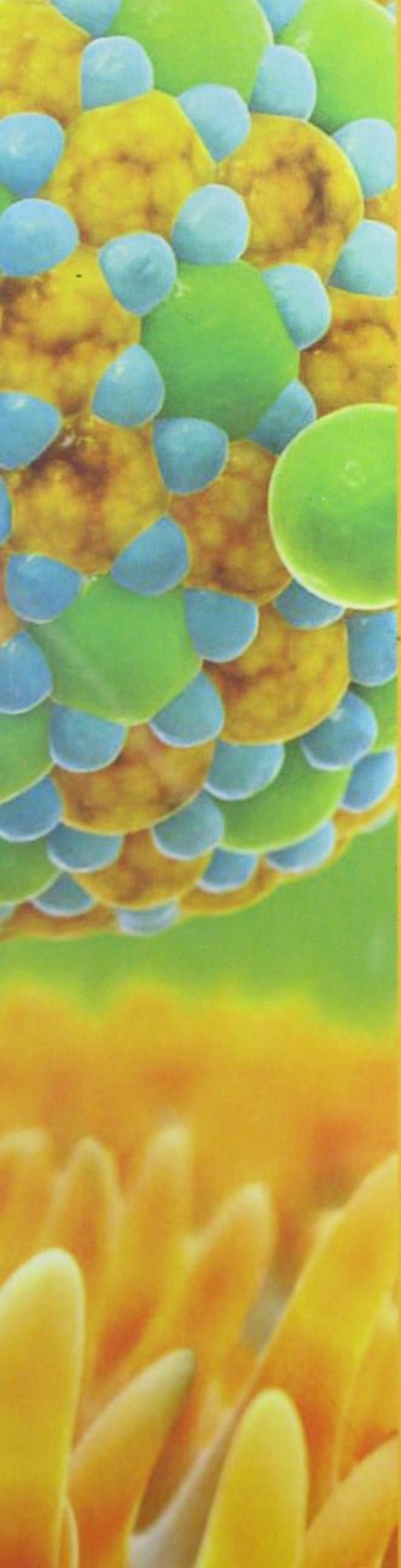

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Clinical Immunology & Allergology

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In the textbook the basic themes on clinical immunology and allergology are presented in accordance with the standard Program approved by Ministry of Health of Ukraine in 2013. Considering a clinical orientation of the Program, problems of diagnostics, features of pathogenesis and disease courses, etc. are stated generally in each chapter. To make it easier to master material the textbook contains many illustrations, tests and control questions.

The textbook is intended for English-speaking students and lecturers of higher educational medical institutions of III–IV accreditation levels.

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Contents

Preface

Clinical immunology is a section of immunology, which studies the functioning of the immune system of man under the conditions of the norm and pathology. In the recent decades, due to appearance of new high-precision methods of immunodiagnostics, effective immunotropic preparations and expansion of ideas about immunoprophylaxis, clinical immunology received wide acceptance. Today it is the actively developing field of scientific investigations in the sphere of medicine and biology. **Allergology** is a section of clinical medicine (medical specialty), which studies problems of etiology, pathogenesis, diagnostics and treatment of allergic diseases. Allergology is tightly connected with clinical immunology, since the allergic reactions are hyperimmune reactions, therefore the study of clinical immunology and allergology is united in one academic discipline.

The academic discipline "Clinical immunology and allergology" is tightly interconnected with other academic disciplines. The basis for its successful study is knowledge, obtained during the study of biology, microbiology, histology, pathologic physiology and anatomy as well as other fundamental disciplines. Clinical immunology and allergology reveals in detail the immunological aspects of pathogenesis, diagnostics and treatment of the diseases, which are studied by students in profile-oriented and related clinical disciplines (internal medicine, obstetrics – gynecology, pediatrics, oncology, infectious diseases, ENT diseases and others).

Basic tasks of the discipline are: to teach a student to use methods of immunodiagnostics, immunotropic therapy and immunoprophylaxis; to acquaint him with modern aspects of etiopathogenesis, diagnosis and treatment of immune disturbances (immunodeficient states; autoimmune, allergic and oncologic diseases; the immunopathologic states of reproduction).

This textbook is the first edition on the discipline "Clinical immunology and allergology" in Ukraine intended for English-speaking foreign students of higher medical institutions of III–IV levels of accreditation. It gives the basic themes of the program material on the discipline

(Standard program approved by the Central Methodical Commission for Higher Medical Education of MH of Ukraine in 2013). In contrast to fundamental textbooks on the immunology and microbiology the textbook is not intended for the instruction of students in modern postulates and the latest achievements of immunology and immunogenetics. The authors consciously limited the volume of the theoretical information because of the limited course of the discipline study. Therefore the textbook is orientated to the acquaintance with the applied aspects of immunology and allergology. In this case much attention is paid to questions of pathogenesis, diagnostics, clinical course of diseases, principles of treatment of the immunopathologic states. The book is illustrated with Fig. and tables, which considerably facilitate mastering of the new training material by the students. Tables and figures are borrowed from different training and electronic publications and resources on immunology and are used in the book exclusively for the educational purposes. In the accordance with the requirements of the credit-modular system of the educational process organization, both class and independent work of students is of importance. Therefore each theme of the textbook includes such sections as test tasks and control questions for the self-assessment of the mastering level of the theoretical material and ability to use it in practical activity. The list of the theoretical questions and skills is an important reference point for the students while preparing to practical classes and final modular control. Taking into account the significance of immunology in the clinical practice, the book gives a glossary with a brief interpretation of the basic immunological terms. The most authoritative publications on immunology are given in the list of the recommended literature for more advanced study of clinical immunology to English-speaking students, which are used by the leading universities while training. Since this textbook is published for the first time – there may be some drawbacks. The authors will be grateful for friendly advice and wishes on its correction.

CHAPTER I

STRUCTURE AND PRINCIPLES OF FUNCTIONING OF THE IMMUNE SYSTEM

1.1. Structure of the immune system

The immune system is a specialized system of cells, tissues and organs, basic components of which are central (thymus and bone marrow) and peripheral lymphoid organs (spleen, lymph nodes, group of lymphatic follicles of the bowels – Peyer's patches, tonsils), blood and lymphatic vessels (Fig. 1.1). In the morphological aspect the immune system is considered as the totality of lymphocytes, macrophages, and a number of cells similar to the macrophages, including dendritic cells and epithelial cells of Langerhans (white dendritic epidermocytes).

The chief characteristics of the immune system: prevalence in the organism, constant re-circulation of the corresponding immune cells and their ability to produce specific molecules – antibodies regarding each antigen.

Basic tasks:

1. Protection of the organism from the penetration of genetically foreign external agents;
2. Control of constancy of the internal medium of the organism: destruction of the proper organism cells, which were subjected to genetic mutations; utilization of the proper organism cells destructed as a result of apoptosis; utilization of the cells (fragments, proteins), damaged as a result of inflammatory-destructive processes in the organism (injury, inflammation).

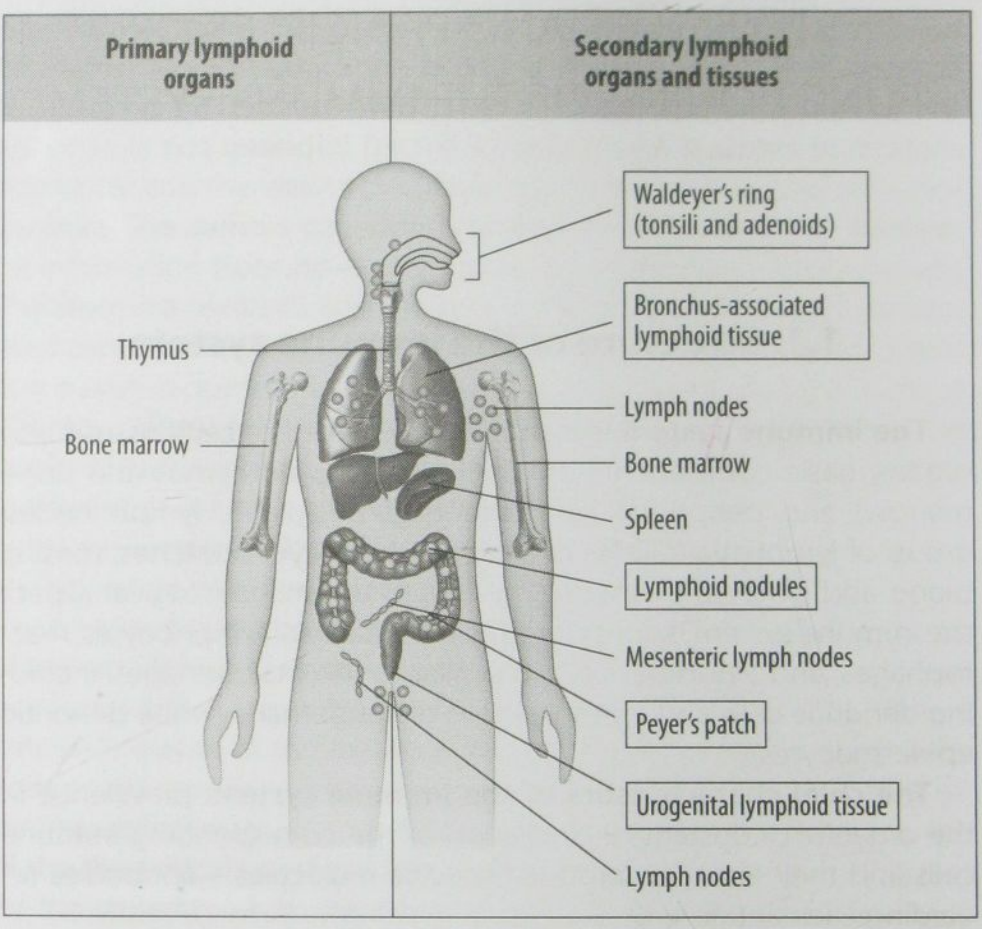

Fig. 1.1. Thymus and bone marrow are primary lymphoid organs. *They are sites of maturation for T and B-cells respectively. Cellular and humeral immune responses occur in the secondary (peripheral) lymphoid organs and tissues. Secondary lymphoid organs can be classified according to the body areas which they defend. The spleen responds predominantly to the blood-borne antigens. Lymph nodes mount immune responses to antigens circulating in the lymph, entering through the skin (subcutaneous lymph nodes) or through mucosal surfaces (visceral lymph nodes). Tonsils, Peyer's patches and other mucosa-associated lymphoid tissues (blue boxes) react to antigens which have entered via the surface mucosal barriers. (I. Roitt et al., Immunology, 2001).*

1.2. Acquired and congenital immunity

The immune response consists in recognition of the foreign genetic material and development of the responses directed at its elimination.

The immune reaction can be **congenital** (innate, nonspecific immunity) and **acquired** (adaptive, specific immunity) (Fig. 1.2).

Nonspecific resistance is achieved by the cellular and humoral factors, which closely interact in the achievement of the final effect – catabolism of the foreign substance: by macrophages, neutrophils, complement, lysozyme, properdin, IgA as well as by other cells and dissoluble factors. The factors of nonspecific resistance are also the skin and mucous membranes of the organism – physiological barriers and the secretion exerting the bactericidal effect. Saliva, gastric juice and digestive enzymes suppress growth and multiplication of the microbes.

Nonspecific immunity provides the first line of protection from the foreign particles and organisms, and it is accomplished by several types of cells. Phagocytes – the blood monocytes and tissue macrophages – absorb and destroy a lot of foreign particles. Polymorphonuclear leukocytes together with the mast cells participate in the protection from the microorganisms, being the most important components of the reaction of acute inflammation. Natural killers provide the first line of protection of the organism affecting its own cells infected with viruses or neoplastic process. The so-called proteins of the acute phase of inflammation and the complement system play an important part in the formation of responses of the nonspecific immunity.

The main characteristics of the acquired immunity **(specific or adaptive immunity)** are the formation of specific antibodies, presence of immunological memory, possible participation of T-killers and learning ability (Fig. 1.3). Instruction and memory are accomplished by the laws of clonal selection, i.e. genesis of the cell clones ready to the subsequent encounter with the identical (specific) antigen. ***Clonal selection*** is a proliferation of cells, caused by the specific antigen.

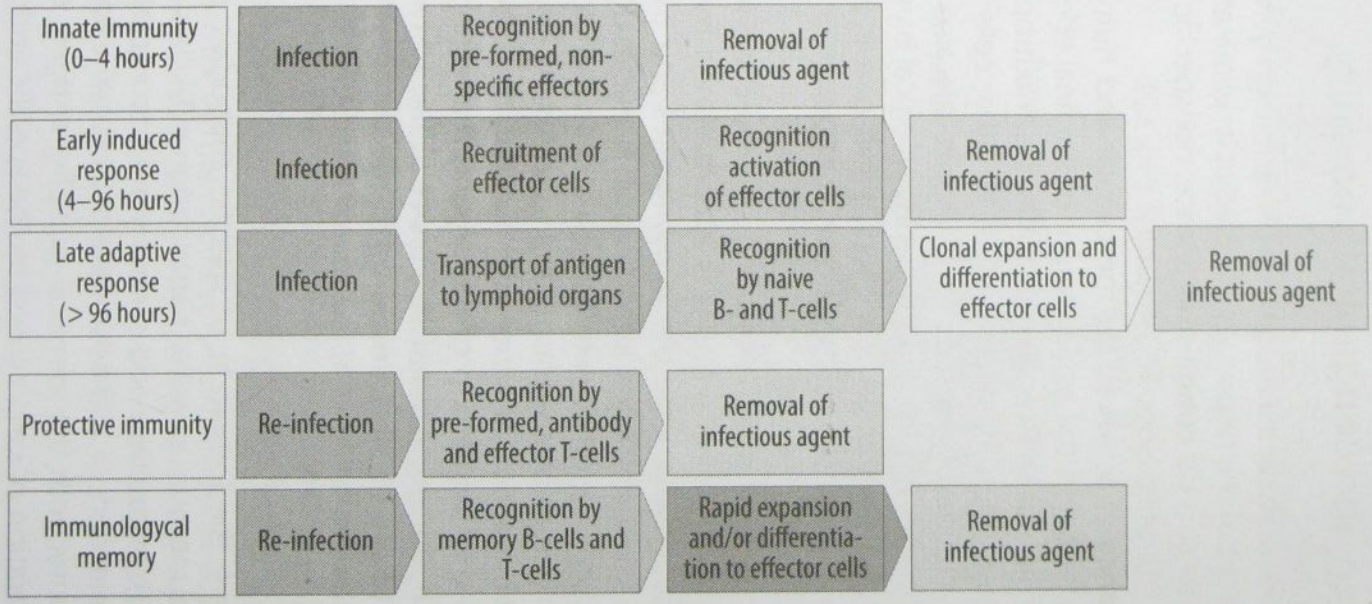

Fig. 1.2. The response to an initial infection occurs in three phases. *The effector mechanisms that remove the infectious agent (e.g. phagocytes, NK-cells, complement) are similar or identical in each phase but the recognition mechanisms differ. Adaptive immunity occurs late, because rare antigen-specific cells must undergo clonal expansion before they can differentiate into effector cells. After an adaptive immune response to a pathogen, the response to re-infection is much more rapid; pre-formed antibodies and effector cells act immediately on the pathogen, and immunological memory speeds a renewed adaptive response. (Charles A. Janeway et al., Immunobiology, 1999).*

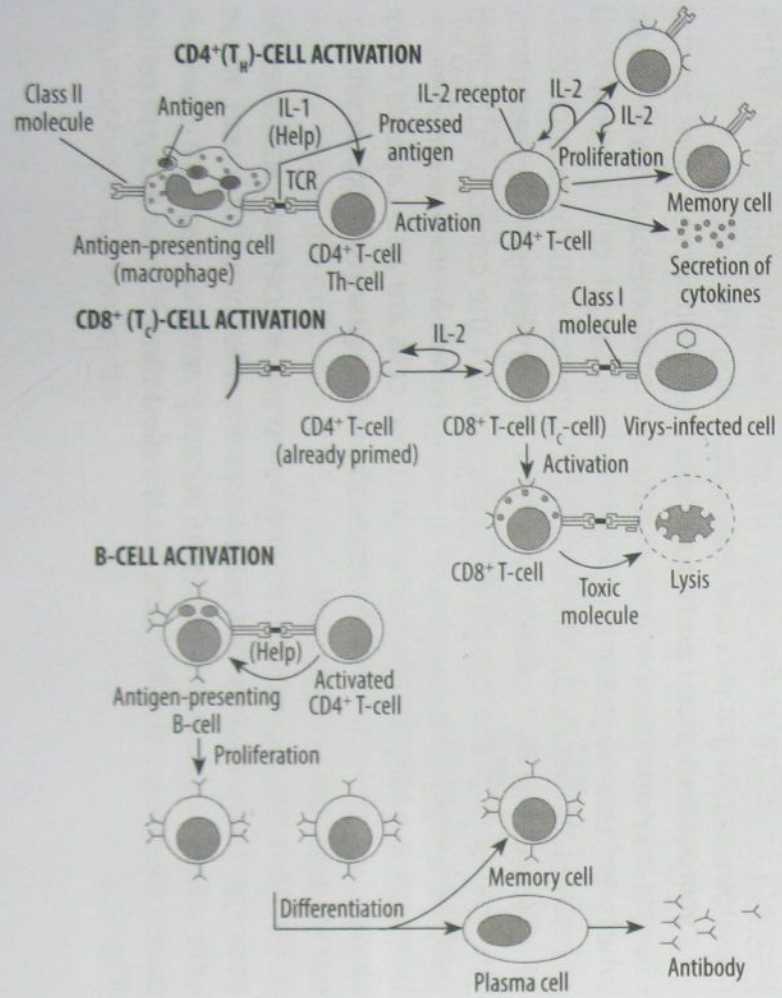

Fig. 1.3. Development of the immune response. *The details of this response will be discussed in subsequent chapters. When an antigen is introduced into a host, antigen-presenting cells (macrophages) process and present the antigen to CD4+ T-cells (usually considered Th-cells). This process and the release of interleukin-1 (IL-1), a helper monokine, activate the CD4+ T-cells. Activated CD4+ T-cells help themselves and T-cells (usually considered Th-cells) by releasing IL-2. Activated T-cells respond in cell-mediated immunity reactions by their direct participation as effector cells. Antigen-activated B-cells, after processing and presenting antigen to activated B-cells, convert to plasma cells with help through direct interaction with T-cells and by cytokines like IL-4 and IL-5. The plasma cells secrete specific antibodies against the inducing agent and thereby provide humoral immunity. Some cells become memory cells and react more rapidly to the next exposure. The T-cell receptors (TCR) of CD4+ T-cells recognize antigen only when associated with class II or I molecules HLA, respectively. (Klaus D. Elgert, Immunology, 1996).*

The second immune response in comparison with the primary one is always more rapid and stronger (phenomenon of instruction at the level of the cellular populations).

Any **immune response** has two basic phases:

- recognition of the antigen;
- reactions, directed at its elimination.

The immune system has a number of mechanisms for destroying the pathogenic microbes, and each of them corresponds to this type of infection and concrete stage of the life cycle of the agent.

Neutralization. The antibodies are bound with the specific agent to oppose its vital activity. Antibodies to the external proteins of the capsid of some rhino-viruses prevent binding of the viral particles with the cells of organism and their infection.

Phagocytosis. The antibodies realize their effect by activating complement or acting as the opsonins, which intensify absorption of the microbes by phagocytes and their processing (Fig. 1.4).

Cytotoxic reactions and apoptosis. The cytotoxic reactions are the effector immune mechanisms directed at intact cells, usually at those, which are too large for phagocytosis. This target cell is recognized either by the specific antibodies, which interact with the components of its surface or by T-cells by means of the antigenspecific receptors. The granules of the cytotoxic T-cells contain the compounds called perforins, which are capable of creating canals in the external membrane of the target cells. Some cytotoxic cells are also capable of including the program of self-destruction of the target cell by their signal – the process of apoptosis.

In the normal condition the immune system cells are scattered all over the body tissues, but when there is a focus of infection, these cells, their byproducts and the complement system are concentrated precisely in it. This process is called the inflammatory reaction (Fig. 1.5).

Phases of the immune response

(Charles A. Janeway et al., Immunobiology, 1999, as amended)

Immune response	Immediate (0–4 hours)	Early (4–96 hours)	Late (after 96 hours)
	Non-specific Innate No memory No specific T-cells	Non-specific + specific Inducible No memory No specific T-cells	Specific Inducible Memory Specific T-cells
Barrier functions	Skin, epithelia	Local inflammation (C5a) Local TNF-α	IgA antibody in luminal spaces IgE antibody on mast cells
Response to extracellular pathogens	Phagocytes Alternative complement pathway	Mannan-binding lectin C-reactive protein T-cell independent B-cell antibody + complement	IgG antibody and Fc receptor-bearing cells IgG, IgM antibody + classical complement pathway
Response to intracellular bacteria	Macrophages	Activated NK-dependent macrophage activation IL-1, IL-6, TNF-α, IL-12	T-cell activation of macrophages by IFN-γ
Response to virus-infected cells	Natural killer (NK) cells	Interferon-α and -β IL-12-activated NK-cells	Cytotoxic T-cells IFN-γ

Chemotaxis and migration of the cells (Fig. 1.6). Having penetrated into the tissue, the cells migrate towards the infection focus under the effect of the chemical attraction, called chemotaxis. Active migration of the specific chemotaxic compounds by the concentration gradient is inherent to phagocytes. Especially intense chemotaxis is caused by a fragment of one of the complement components – C5a, attracting neutrophils and monocytes.

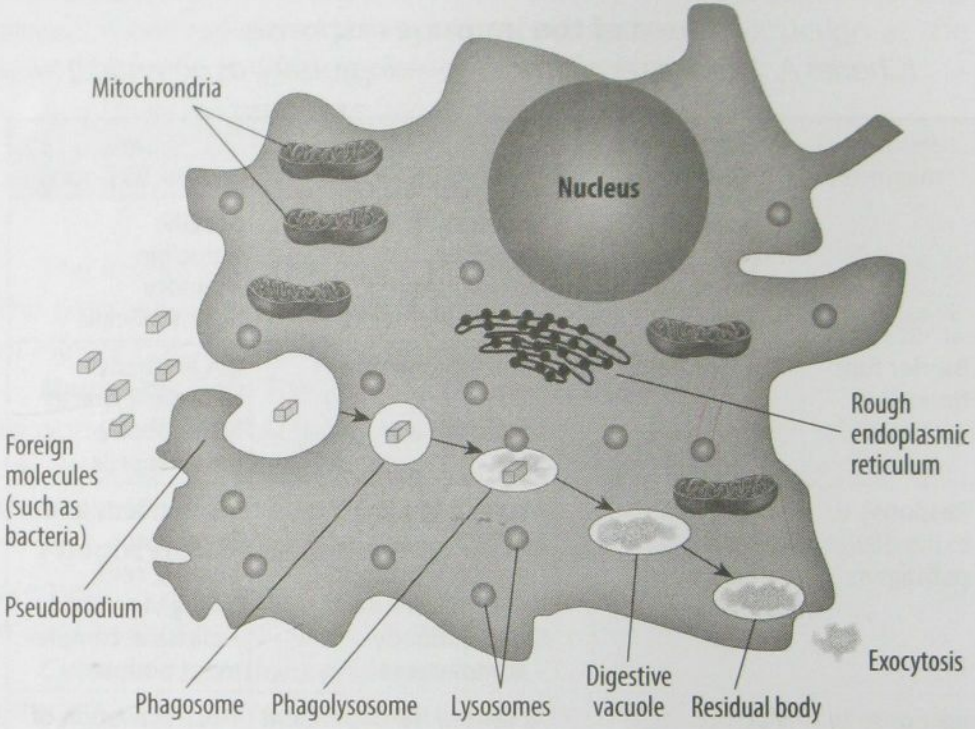

Fig. 1.4. Phagocytosis. *Cells of the mononuclear phagocyte system are attracted to the site of infection by factors released by pathogens, damaged host cells, and other blood components. The phagocytes engulf the pathogens, using pseudopodia, and internalize them as membrane-bound organelles (phagosomes) within the phagocyte. The phagosomes fuse with other organelles (lysosomes) containing hydrolytic enzymes. These organelles are then called phagolysosomes. Inside the phagolysosome, azurophile and specific granules discharge two groups of toxic substances into the organelle: (1) oxygen-dependent products formed by reactive oxygen metabolites and (2) oxygen-independent reactants such as proteases, lactoferrin, and phospholipase A_2. Organisms are killed by the action of superoxide ions, hypochlorite, and hydrogen peroxide. The phagocyte's activity is enhanced by other parts of the immune system (Klaus D. Elgert, Immunology, 1996).*

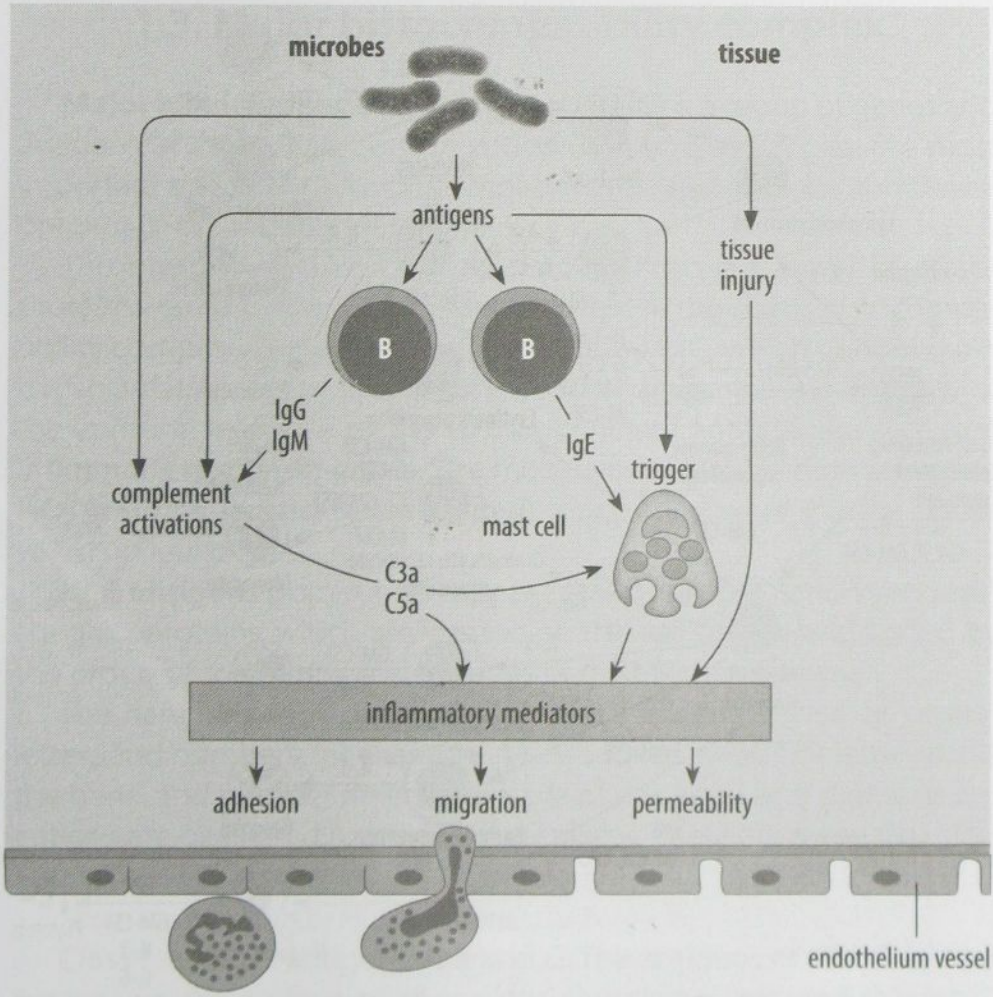

Fig. 1.5. The adaptive immune system modulates inflammatory processes via the complement system. *Antigens (e.g. from microorganisms) stimulate B-cells to produce antibodies including IgE binding to mast cells, while IgG and IgM activate a complement The complement can also be activated directly via the alternative pathway. When triggered by antigen, the sensitized mast cells release their granule-associated mediators and eicosanoids (products of arachidonic acid metabolism, including prostaglandins and leukotrienes). In association with complement (which can also trigger mast cells via C3a and C5a) the mediators induce local inflammation, facilitating the arrival of leucocytes and more plasma enzyme system molecules. (I. Roitt et al., Immunology, 2001).*

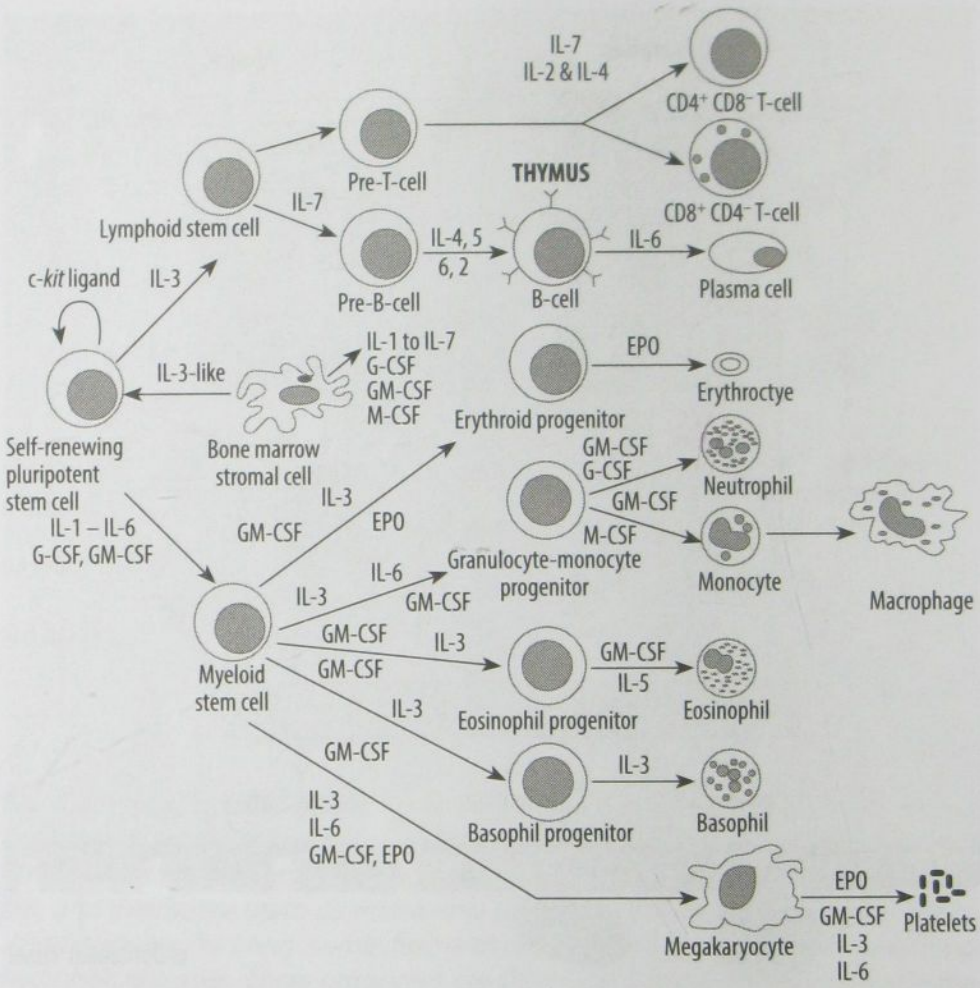

Fig. 1.6. Maturation of immune system cells. *Myeloid and lymphoid cells develop in adults from pluripotent (many different potentials) stem cells in the bone marrow.This development is driven by colony-stimulating factors. Nonlymphoid stem cells give rise to elements of the peripheral blood, such as erythrocytes, platelets, granulocytes (basophils, eosinophils, or neutrophils), and monocytes (precursor cells for macrophages). Lymphoid stem cells can develop along two pathways. If these stem cells migrate through the thymus, they become T-lymphocytes or T-cells, represented by CD4+ T- and CD8+ T-cells. If the lymphoid stem cells mature in the bone marrow, the cells become a population of lymphocytes, called B-lymphocytes or B-cells (Klaus D. Elgert, Immunology, 1996).*

1.3. Major histocompatibility complex

Major histocompatibility complex (MHC) is a group of genes and antigens of the cellular surface coded by them, which play the most important role in the recognition of the foreign substance and development of the immune response.

The major molecules of histocompatibility are a family of glycoproteins coded by the genes, which compose the major histocompatibility complex. There are the genes within MHC, which control main transplantation antigens and genes, which determine the intensity of the immune response to this or that concrete antigen, the so-called **Ir (*immune response*) *genes.*** The molecules of MHC in man are called **HLA (*human leucocyte-associated*)**, since they were initially discovered on leukocytes.

HLA antigens (sometimes they are called transplantation antigens) are glycoproteins, which are located on the cell surface and coded by the group of the tightly linked genes of the 6th chromosome.

The names of HLA genes and antigens consist of one or several letters and numbers, for example, A3, B45, DR15, DQ4. The letter marks the gene, and number means the allele of this gene, and digital designations are given in discovery of new alleles. There are more than 100 antigens of HLA today.

There are 3 classes of HLA antigens.

Class I includes antigens A, B and C. The antigens of the class I are present on the surface of all nucleus-containing cells and thrombocytes. They are necessary for the recognition of the transformed cells by the cytotoxic T-lymphocytes. The cytotoxic T-lymphocytes (T-killers) recognize the target cells only in presence of the HLA antigens of the class I of their own genotype on their surface.

Class II comprises antigens DR, DP and DQ. The antigens of the class II are present on the surface of B-lymphocytes, activated T-lymphocytes, monocytes, macrophages and dendritic cells, i.e., on the cells, which participate in the immune responses (lymphocytes, macrophages). The most important function of the HLA antigens of the

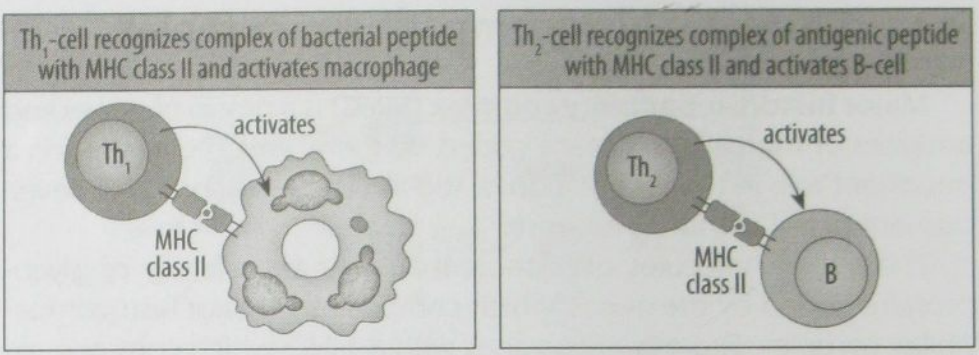

Fig. 1.7. Th_1 and Th_2-cells recognize antigen presented by MHC class II molecules. *Th_1 and Th_2-cells both recognize peptides bound to MHC class II molecules. On recognition of their specific antigen on infected macrophages, Th_1-cells activate the macrophage, leading to the destruction of the intracellular bacteria (left panel). When Th_2-cells recognize antigen on B-cells, helper T-cells activate these cells to proliferate and differentiate into antibody-producing plasma cells (right panel) (Charles A. Janeway et al., Immunobiology, 1999).*

class II is provision of interaction between the T-lymphocytes and macrophages in the process of the immune response. T-helpers recognize foreign antigen only after its processing by macrophages, combining with the HLA antigens of the class II and appearance of this complex on the macrophage surface (Fig. 1.7).

The genes of HLA of the ***class III*** control some components of the complement: C4 and C2, the tumor necrosis factors (TNF-alpha and TNF-beta), i.e., control the synthesis of the proteins, part of which participates in the immune processes. However, in contrast to HLA molecules of the class I and the class II they do not participate in control of the immune response.

The expression of HLA antigens regulates cytokine-interferon-gamma and factor of the tumor necrosis – powerful inductors of the HLA expression by the cells of many types. The majority of the HLA genes are highly polymorphous, i.e., there may be many alleles in the population in the specific locus of HLA. The inheritance of the HLA-genes occurs by the codominant sign, in which in descendents of the

HLA-alleles obtained from each of the parents are expressed in the same degree.

The expression of MHC molecules differs between tissues
(Charles A. Janeway et al., Immunobiology, 1999, as amended)

Tissue	MHC class I	MHC class II
T-cell	+++	+
B-cell	+++	+++
Macrophages	+++	++
Other antigen-presenting cell	+++	+++
Epitelsal sells of thymus	+	–
Neutrophils	+++	+++

1.4. Cellular immunity (Fig. 1.8)

Antigen-processing cell (antigen-presenting cell). For an antigen to be recognised by a T-lymphocyte, it must be first processed and presented in a formthat the antigen can recognise. This is the function of an antigen-processing cell. The process by which this takes place is the following: an antigen-processing cell engulfs an antigen → enzymes in the antigen-processing cell break down the antigen into smaller fragments → these fragments are transported to the surface of the antigen-processing cell (APC), bound with class II MHC molecules → a T-cell receptor can now recognize the antigen linked with the MHC and thus binds to it (Fig. 1.9). Antigen-processing cells include: macrophages; dendritic cell; follicular dendritic cell; Langerhans' cell; Langerhans' cells are dendritic cells specific to the skin.

Dendritic cell. The interdigital dendritic cells are most important for the presentation of antigen to T-cells. Dendritic cells are the principle antigen-processing cell involved in primary immune responses. Their

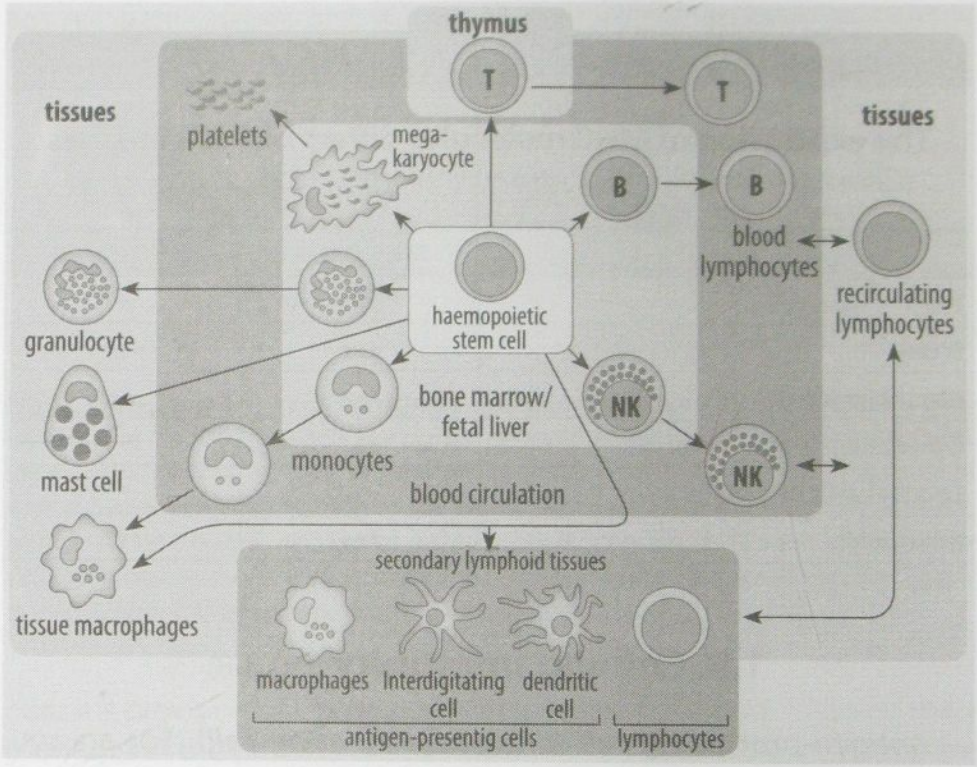

Fig. 1.8. All the cells shown arise from the haemopoietic stem cell. *Platelets produced by megakaryocytes are released into the circulation. Granulocytes and monocytes pass from the circulation into the tissues. Mast cells are identifiable in all tissues. B-cells mature in the fetal liver and bone marrow in mammals, whereas T-cells mature in the thymus. The origin of the large granular lymphocytes with NK activity is probably the bone marrow. Lymphocytes recirculate through secondary lymphoid tissues. Interdigitating cells and dendritic cells act as antigen-presenting cells in secondary lymphoid tissues. (I. Roitt et al., Immunology, 2001).*

major function is to obtain antigen in tissues, migrate to lymphoid organs and activate T-cells. (Fig. 1.10).

Leukocyte – White Blood Cell (WBC). White blood cells are the principal components of the immune system and function by destroying "foreign" substances such as bacteria and viruses. Like all blood cells,

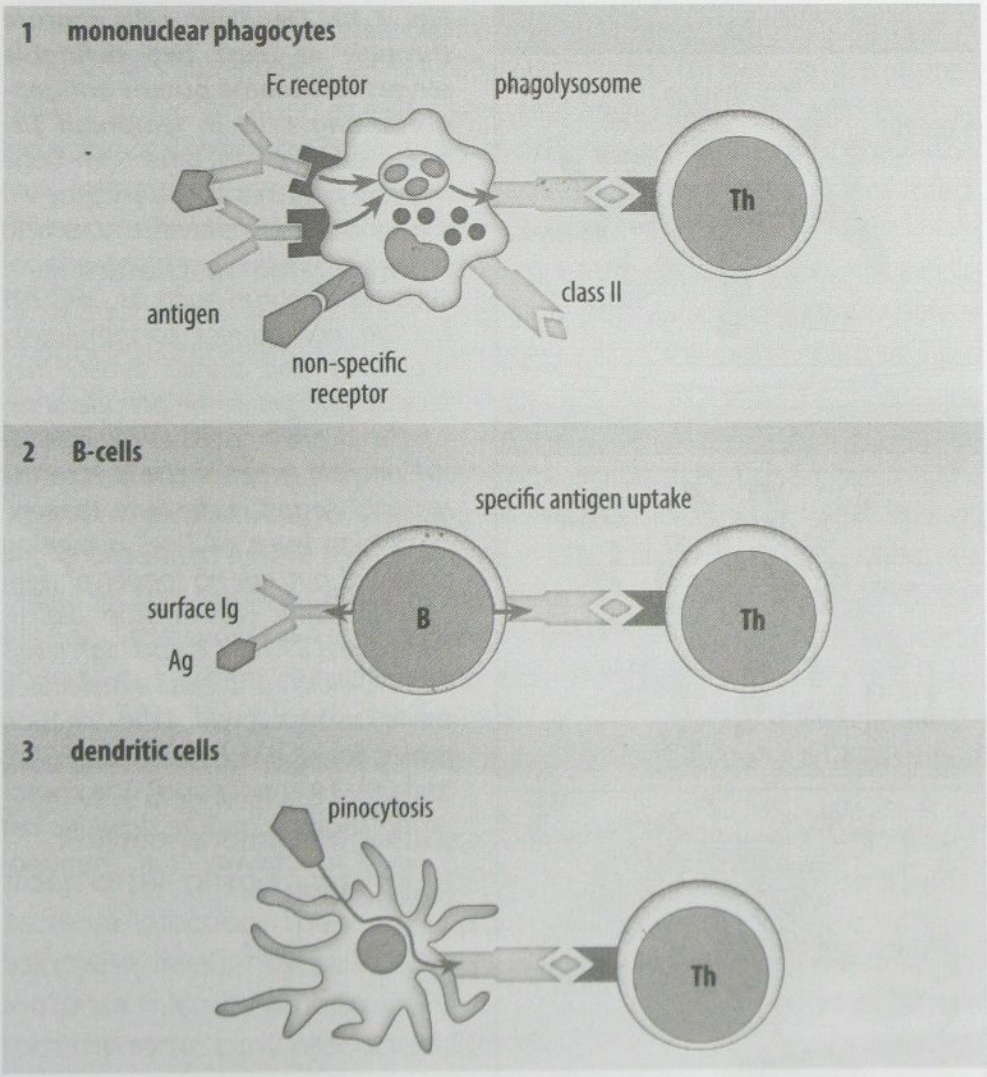

Fig. 1.9. Mononuclear phagocytes (1), B-cells (2) and dendritic cells (3) can all present antigen to MHC class II-restricted T-helper (Th) cells. *Macrophages take up bacteria or particulate antigen via non-specific receptors or as immune complexes, process it and return fragments to the cell surface in association with class II molecules. Activated B-cells can take up antigen via their surface immunoglobulin and present it to T-cells associated with their class II molecules. Dendritic cells constitutively express class II MHC molecules and take up antigen by macropinocytosis. (I. Roitt et al., Immunology, 2001).*

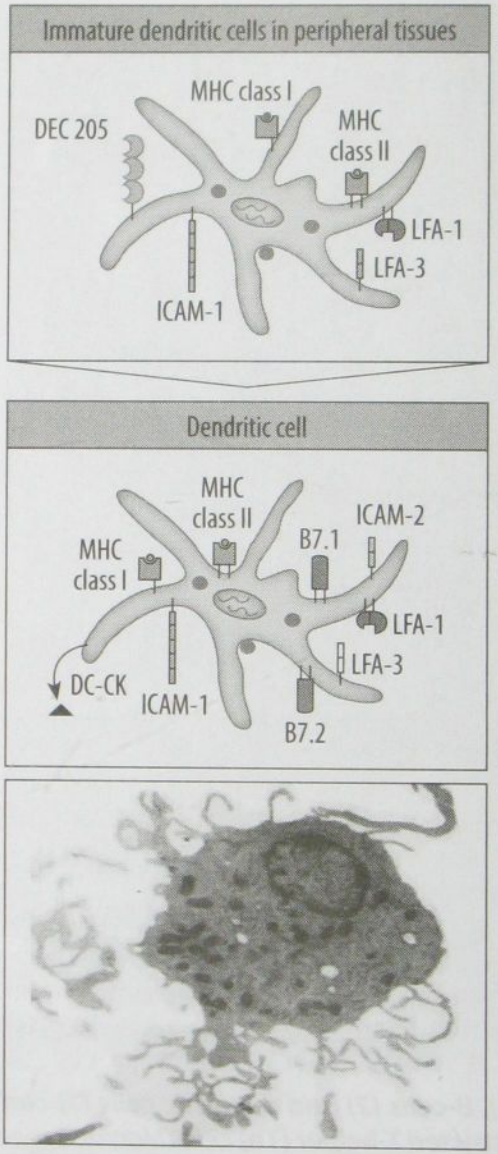

Fig. 1.10. Dendritic cells mature through at least two definable stages to become potent antigen-presenting cells in lymphoid tissue. *Dendritic cells arise from bone marrow progenitors and migrate via the blood to peripheral tissues and organs, where they are highly phagocytic via receptors such as DEC 205 but do not express co-stimulatory molecules (top panel). When they pick up antigen in the peripheral tissues, they are induced to migrate via the afferent lymphatic vessels to the regional lymph node. Here they express high levels of T-cell activating potential but are no longer phagocytic. Dendritic cells in lymphoid tissue express B7.1, B7.2, and high levels of MHC class I and class II molecules, as well as high levels of the adhesion molecules ICAM-1, ICAM-2, LFA-1, and LFA-3 (central panel). The photograph shows a mature dendritic cell (Charles A. Janeway et al., Immunobiology, 1999).*

they are produced in the bone marrow. There are 5 main types of white blood cells, subdivided into 2 main groups:

Polymorphonuclear leukocytes (granulocytes): neutrophils; eosinophils; basophils.

Mononuclear leukocytes: monocytes; lymphocytes.

Normal values range from 4100/ml to 10900/ml but can be altered greatly by factors such as exercise, stress and disease. A low WBC may indicate viral infection or toxic reactions. A high WBC count may indicate infection, leukemia, or tissue damage. An increased risk of infection occurs once the WBC drops below 100/ml.

Mononuclear phagocytes. The most important group capable of phagocytosis and long-life cells is a population of mononuclear phagocytes, proceeding from the stem cells of the bone marrow, having the function to capture particles, including those of infectious agents, with their absorption and destruction. Each macrophage contains packets of chemicals and enzymes, which digest the ingested antigen or microbe. Phagocytes are strategically located in those tissues of the organism where the penetration of such particles is possible. For example, the Kupffer cells line the blood sinusoidal capillaries of the liver, and the synovial A-cells cover the joint cavities. The mononuclear phagocytes, which circulate in the blood, are called monocytes. They migrate from the blood in the tissues where they are converted into the tissue macrophages, capable of presenting antigens to T-lymphocytes very effectively.

Polymorphonuclear neutrophils. It is the second significant group of the phagocytes. Neutrophils form a primary defense against bacterial infection. These are polymorphonuclear neutrophilic granulocytes, frequently called simply neutrophils or PMN. Neutrophils compose majority among the leukocytes of the blood and originate from the same early precursor cells as monocytes and macrophages. Similar to monocytes neutrophils migrate in the tissue reacting to the specific stimuli, but in contrast to the monocytes they are related to the short-lived cells, which perish after absorbing foreign material and destroying it.

The eosinophils are formed in the bone marrow, and all stages of differentiation to the mature cell occur there. The number of the eosinophils, which circulate in the blood flow, does not exceed 1 % of the total number of these cells in the organism; they circulate for

Resident tissue macrophage populations

(I. Roitt et al., Immunology, 2001)

organ	name/site	functions/properties
bone marrow	stromal macrophage	interacts with haematopoietic cells; removes erythroid nuclei
liver	Kupffer cells	clearance of cells and complexes from the blood
spleen	red pulp macrophages; white pulp tingible bodies; marginal zone macrophages	clearance of senescent blood cells phagocytosis of apoptotic B-cells interface with circulation and immune system
lymph node	subcapsular sinus macrophages; medullary macrophages	interface with afferent lymph; interface with efferent lymph
thymus	thymic macrophage	clearance of apoptotic cells
gut	lamina propria	endocytosis
lung	alveolar macrophage	clearance of particulates
brain	microglia in neutrophil choroid plexus	interacts with neurons' interface with cerebrospinal fluid
skin	Langerhans' cells	antigen capture
reproductive tract	ovary, testis	clearance of dying cells
endocrine organs	adrenal, thyroid, pancreas, etc.	metabolic homeostasis
bone	osteoclasts	bone re-modelling

10 hours in the blood, then they migrate in the tissue where they live for 48 hours and perish after degranulation. The eosinophils control release of the histamine and other biologically active materials, neutralizing their excessive quantity. This function is very important for the process of the organism protection in helminthiases and in development of the allergic reaction.

Causes of eosinophilia

Common	Uncommon
Atopic disease Asthma Adverse drug reactions Parasitic infestation (e.g. toxocara, strongyloides, filariasis)	Neoplasia (Hodgkin's lymphoma) Connective tissue diseases Systemic vasculitis (Churg – Strauss syndrome) Coccidioidomycosis (fungal infection) Eosinophilic pulmonary syndrome (including Loeffler's syndrome) Eosinophilic gastroenteritis Eosinophilic leukaemia Idiopathic hypereosinophilic syndrome Dermatitis herpetiformis

The basophils take an active part in development of the allergic reactions of the immediate type. Having penetrated into tissues basophils are converted into the mast cells, which contain the large number of histamine – biologically active material, which stimulates the development of allergy. Because of the basophils the poisons of insects or animals are immediately blocked in the tissues and are not spread throughout the whole body. Also basophils regulate the ability of the blood coagulation with the aid of heparin.

The lymphocytes are the cells having the ability to react only to a limited group of structurally similar antigens. This ability is determined by the presence of the corresponding membrane receptors in the lymphocyte, specific for the determinants of this or that antigen. Each lymphocyte possesses a population of receptors with the identical antigen-binding centers, and a separate group, or clone; the lymphocytes differ from another clone by the structure of the antigen-binding center of the receptors, capable of reacting only to the specific set of antigens.

The specific immunological recognition of the pathogenic organisms is completely the function of the lymphocytes; therefore they initiate the reactions of the acquired immunity. The lymphocytes are

distinguished between themselves not only by the specificity of their receptors, but also by their functional properties. The main lymphocyte sub-types are: ***B-cells*** (special ***B-cells produce specific antibodies***, proteins that help destroy foreign substances and, ***B-cells of immunological memory); T-cells*** (T-cells attack virus-infected cells, foreign tissue, and cancer cells. They also produce a number of substances that regulate the immune response); ***NK-cells*** (among other functions, natural killer cells destroy cancer cells and virus-infected cells through phagocytosis and by producing substances that can kill such cells); ***Zero-cells*** (an early population of lymphocytes bearing neither T-cell nor B-cell differentiation antigens).

Basic classes of T-lymphocytes: T-helpers, T-suppressors, T-killers, (cytotoxic T-lymphocytes – T-suppressors + T-killers), T-cells of the immunological memory.

T-lymphocytes fulfill two functions in the organism: effector and regulatory (Fig. 1.11).

Specific cytotoxicity regarding the foreign cells is the basic effector function of the T-lymphocytes; they attack their organism cells, infected with a virus or another foreign agent. ***T-killers*** destroy cells in the direct contact with the target (effector cell), the ejection of the oxidizing ferments occurs, which leads to lysis (dissolution) of the target cell and its destruction. T-killer (cytoxic T-cells) – cells that kill target cells bearing appropriate antigen within the groove of an MHC class I molecule that is identical to that of the T-cell (Fig. 1.12).

The regulatory function of the T-lymphocytes consists in the regulation of the specific immune response (Fig. 1.13). The recognition of the antigen by T-cells ***(T-helpers of the 1st type)*** occurs only under the condition of presentation in the association with the molecules of MNS (the main complex of histocompatibility).

The T-lymphocytes accomplish their functions of effect on other cells by the release of the dissoluble proteins – cytokines, which transfer signals to other cells or via straight intercellular contacts. The regulatory function of T-lymphocytes consists in regulation of the specific immune response. T-helpers of the 1st type initiate the specific cellular

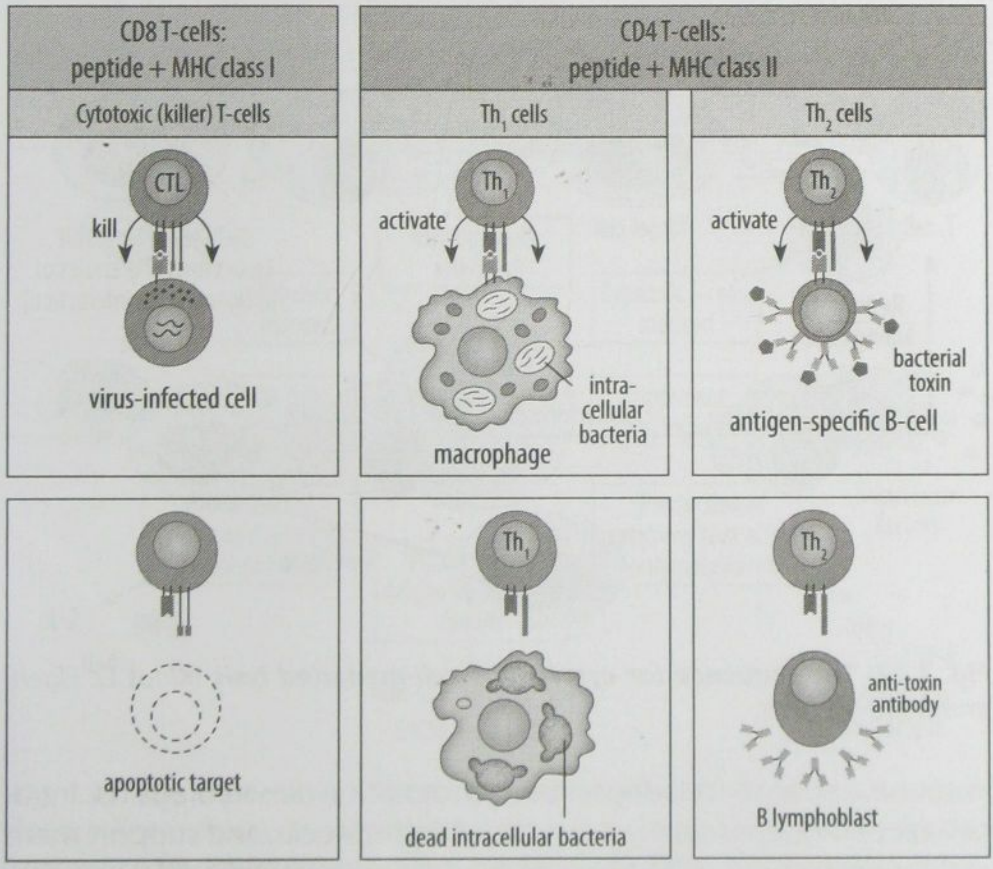

Fig. 1.11. There are three classes of effector T-cells, specialized to deal with three classes of pathogen. *CD8 cytotoxic cells (left panels) kill target cells that display antigenic fragments of cytosolic pathogens, most notably viruses, bound to MHC class I molecules at the cell surface. Th_1-cells (middle panels) and Th_2-cells (right panels) both express the CD4 co-receptor and recognize fragments of antigens degraded within intracellular vesicles, displayed at the cell surface by MHC class II molecules. The Th_1-cells, upon activation, activate macrophages, allowing them to destroy intracellular microorganisms more efficiently; they can also activate B-cells to produce strongly opsonizing antibodies belonging to certain IgG subclasses (IgG1 and IgG3 in humans, and their homologs IgG2a and IgG2b in the mouse). Th_2-cells, on the other hand, drive B-cells to differentiate and produce immunoglobulins of all other types, and are responsible for initiating B-cell responses by activating naive B-cells to proliferate and secrete IgM. The various types of immunoglobulin together make up the effector molecules of the humoral immune response. (Charles A. Janeway et al., Immunobiology, 1999).*

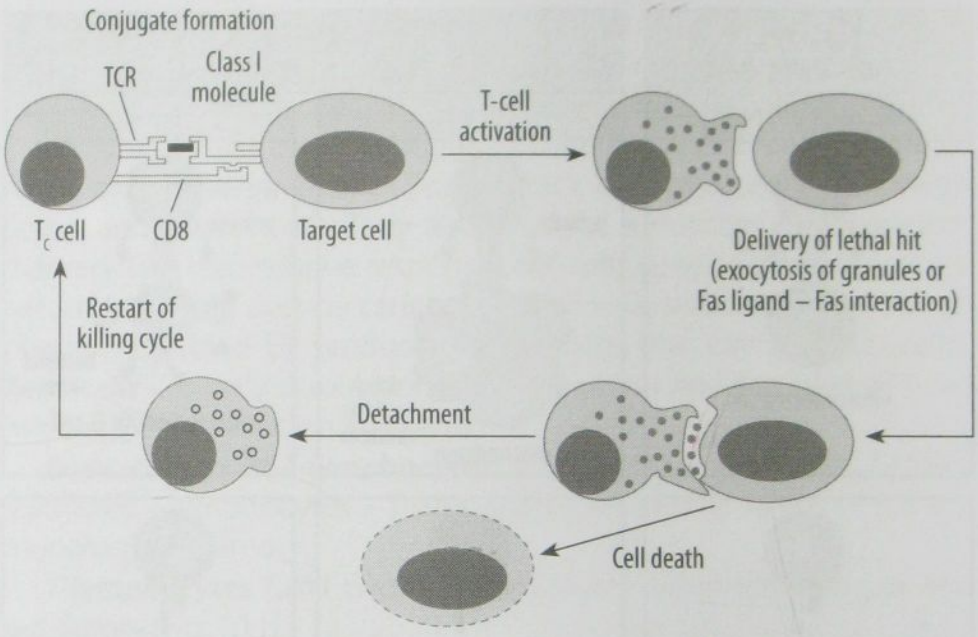

Fig. 1.12. The sequence for cytotoxic T-cell-mediated lysis *(Klaus D. Elgert, Immunology, 1996).*

immune response (cell-dependent cytotoxicity) directed against intracellular pathogens (mainly viruses) and atypical cells, and support them at the necessary level. T-helpers of the 2nd type stimulate differentiation of antibody forming cells (B-lymphocytes → plasmic cells) in response to antigen stimulus. They are responsible for regulation of the immune protection from the extracellular pathogens (gram-positive and gram-negative bacteria, fungi). T-helpers of the 17th type participate in protection from extracellular pathogens, which cannot be effectively eliminated by T-helpers of the 1st and 2nd type, participate in the autoimmune processes and regulation of the antitumor immune response.

T-suppressors (cytotoxic T-lymphocytes) are capable of suppressing the immune response. Activation of the T-suppressors passes a number of phases (the T-helpers participate in them) and it can be associated with the foreign antigen (specific) or cannot be associated (nonspecific). The basic concept of suppressor T-cells (cytotoxic T-lym-

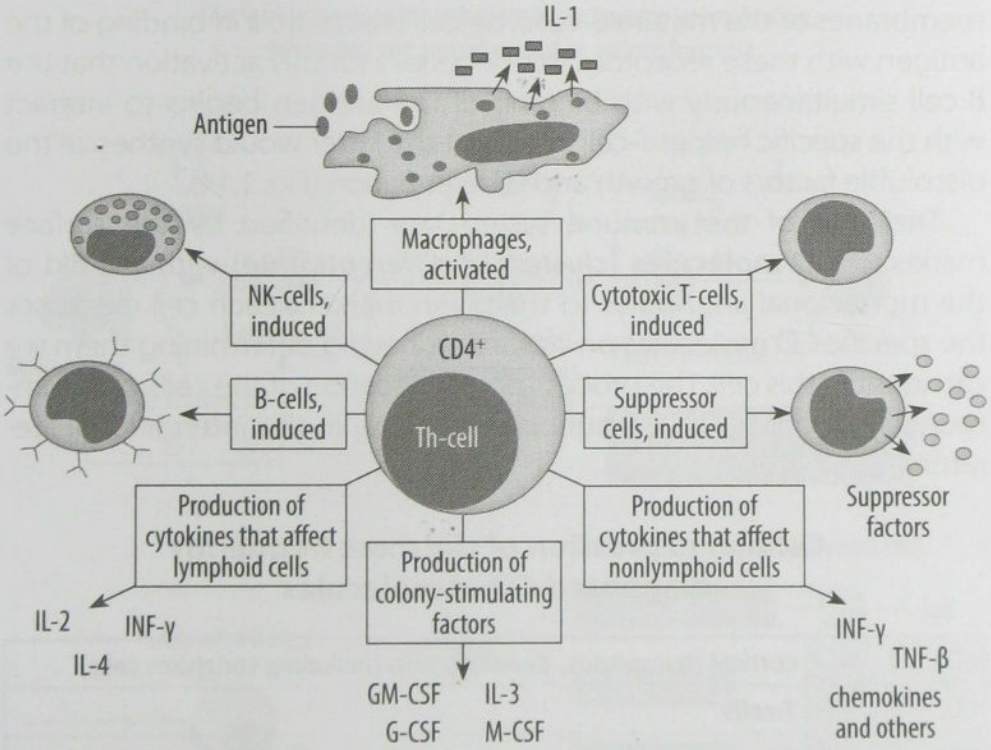

Fig. 1.13. The importance of CD4⁺ Th-lymphocytes in the immune response. *These cells are directly or indirectly responsible for many aspects of the immune response and for nonlymphoid cell functions (Klaus D. Elgert, Immunology, 1996).*

phocytes) is a cell-type that specifically suppresses the action of other cells in the immune system, notably B-cells and T-cells, thereby preventing the establishment of the immune response. Most suppressor T-cells are CD8 positive – like cytotoxic T-cells.

Thus, interaction of T-helpers – T-suppressors controls the intensity of development of the specific response of the immune system to a foreigner. The relationship of T-helpers – T-suppressors is called the index of ***immunoregulation or immunoregulatory index***.

B-lymphocytes originate from the precursor cells of the bone marrow. The B-lymphocytes are the antibody-forming cells or B-cells of immunological memory. There are receptors for the antigen on the

membranes of the mature B-cell. The cell is activated in binding of the antigen with these receptors. It is necessary for this activation that the B-cell simultaneously with binding of the antigen begins to interact with the specific helper T-cell or so that the latter would synthesize the dissoluble factors of growth and differentiation (Fig. 1.14).

The cells of the immune system are identified by the surface markers – ***CD* molecules (*clusters of differentiation*)** with the aid of the monoclonal antibodies to the given markers. Each cell expresses the specific CD molecules on its surface, having determining them we can identify this cell. The process of identification of the cells of the immune system is called immunophenotyping in the international literature.

Cellular expression of the most frequently diagnosed CD of molecules

CD1	cortical thymocytes, dendritic cells (including Langhans cells)
CD3	**T-cells**
CD4	T-helpers
CD8	T-cytotoxic (suppressors)
CD14	**monocytes**
CD15	**granulocytes**
CD16	**NK-cell**
CD19	predecessors of B-cells and B-cells
CD20	predecessors of B-cells, mature B-cells
CD21	mature B-cells
CD22	**B-cells**
CD25	regulatory cells – activated T-, B-cells and macrophages
CD45RA	"naive" T-cells
CD45RO	T-cells of memory
CD56	NK-cell

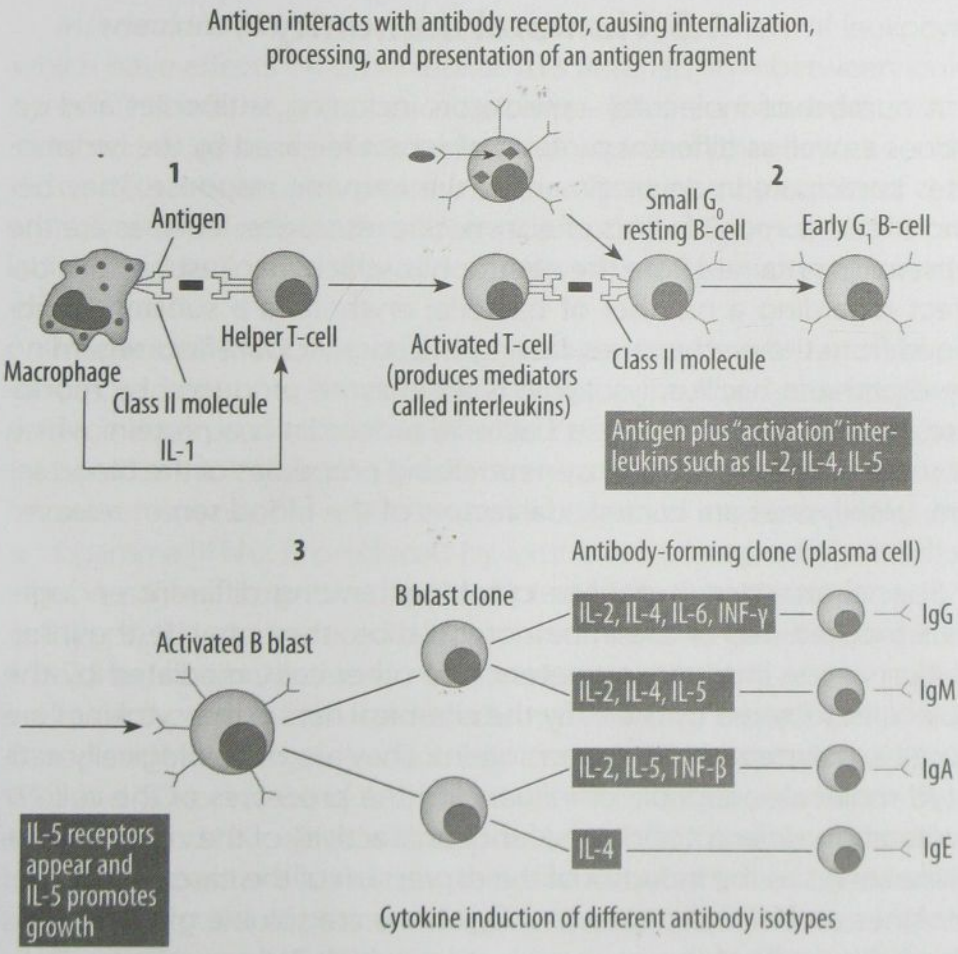

Fig. 1.14. The stages of B-cell activation. *The first step (1) involves uptake of antigen by macrophages, reappearance of processed antigen with class II MHG surface protein, and presentation of this antigen-class II MHC complex to CD4 Th-cells, leading to the production of mediators by macrophages and Th-cells. In the second step (2), the excitation of B-cells from their resting state (from G to C), phase of the cell cycle) is accomplished by the binding of antigen to a surface antibody and its final presentation in association with class II MHC molecules, and to the interaction of macrophage-derived IL-1 with Th-cells, which then interact with the B-cell. In the third step (3) the B-cell replicates and matures to a plasma cell capable of antibody secretion (Klaus D. Elgert, Immunology, 1996).*

1.5. Humoral immunity

A number of molecules – mediators including antibodies and cytokines as well as different proteins of serum released by the lymphocytes participate in development of the immune response. They belong to the humoral factors of nonspecific resistance: leukines are the substances obtained from the neutrophils, which manifest bactericidal effect regarding a number of bacteria; erythrite is a substance, obtained from the erythrocytes, having the bactericidal effect regarding the diphtheria bacillus; lysozyme is an enzyme produced by monocytes, macrophages, and lysis bacteria; properdin is a protein, which provides the bactericidal, virus-neutralizing properties of the blood serum; beta-lysines are bactericidal factors of the blood serum released by the thrombocytes.

Special attention is paid to **cytokines** among different endogenous mechanisms of the immunoregulation; they provide the interrelation of the immunocompetent and other cells, mediated by the molecules secreted by them. By the chemical nature the cytokines are proteins, polypeptides or glycoproteins. They are the biologically activated molecules, capable of influencing the processes of the cellular proliferation, differentiation and functional activity of the cells. Each cytokine serves as the inductor of the expression of the cascade of other cytokines and/or their receptors. Cytokines are soluble glycoproteins released by cells of the immune system, which act nonenzymatically through specific receptors to regulate immune responses. Cytokines resemble hormones they act at low concentrations bound with high affinity to a specific receptor.

Common cytokines include:

1) interleukins (IL-1 – IL-31);
2) colony stimulating factors (CSF), granulocytic-macrophage, granulocytic, monocytic-macrophage);
3) tumor necrosis factors (TNF), lifotoxin;
4) interferons (α, β, γ, κ, λ, τ, ω);
5) chemokines (CCL, CXCL, CL, CX4CL and others).

Interleukins (IL). Glycoproteins secreted by a variety of leukocytes which have effects on other leukocytes (Interleukin = between leukocytes). This is a large group of cytokines (about a hundred), synthesized mainly by the T-cells as well as mononuclear phagocytes or other tissue cells. Interleukins possess diverse functions, but most of them stimulate other cells for division or differentiation. Each interleukin acts on the separate, limited group of cells, which express receptors specific to this IL.

Interferons (IFN). A group of cytokine proteins with antiviral properties, capable of enhancing and modifying the immune response. Interferon is released to coat uninfected cells so that they don't become infected. Some interferons induce antiviral activity, others enhance the immune response. There are three main classes of interferon: alpha, beta and gamma. IFN-α is produced by virus-infected monocytes and lymphocytes. IFN-β is produced by virus-infected fibroblasts (and some other cell types). IFN-γ is produced by stimulated T- and NK-cells. IFN-γ increases MHC II expression, activates macrophages, neutrophils and NK-cells as well as activating vascular endothelium, promoting T- and B-cell differentiation and increasing IL-1 and IL-2 synthesis. IgG2a increases and IgE, G1, G2b and G3 (opposite of IL-4) decreases. All IFNs induce cell growth, activate CTL and NK-cells as well as increases MHC I expression. Interferons contribute to antiviral stability by the cells uncontaminated by a virus and create the first line of protection against the majority of viruses.

Tumor Necrosis Factor (TNF) – is a protein (cytokine) which mediates tumor cell necrosis and destroys cancer cells; it is found in two forms: TNF-α (cachetin) and TNF-β (lymphotoxin). Both forms of TNF bind to the same receptors and therefore have the same activities. TNF-α is produced by macrophages and some other cells. TNF-β is produced by T-cells. IL-1 and TNF act alone or together to induce systemic inflammation (e.g., fever). LPS (an endotoxin) from bacteria stimulates production of TNF-α. TNF is also chemotactic for neutrophils and monocytes, as well as increases neutrophil activity. TNF causes the symptoms associated with bacterial infections (septic shock, fever, lethargy, headache, nausea and inflammation) (Fig. 1.15).

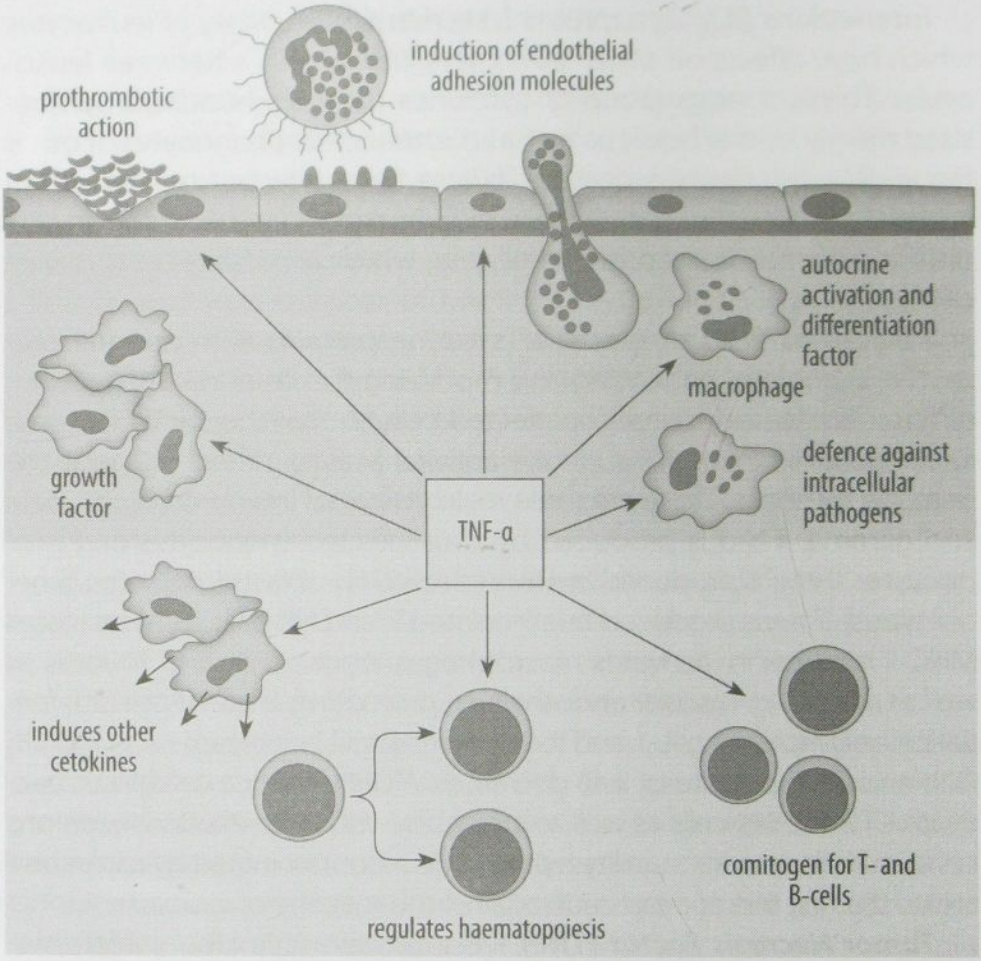

Fig. 1.15. TNF-α has several functions in inflammation. *It is prothrombotic and promotes leucocyte adhesion and migration (top). It has an important role in the regulation of macrophage activation and immune responses in tissues (centre) and also modulates haematopoiesis and lymphocyte development (bottom) (I. Roitt et al., Immunology, 2001).*

Colony Stimulating Factor (CSF). Granulocyte-Colony Stimulating Factor (G-CSF), Macrophage-Colony stimulating factor (M-CSF), Granulocyte-Macrophage-Colony stimulating factor (GM-CSF). These cytokines participate in the division regulation and differentiation of

the stem cells of the bone marrow and precursor cells of the blood leukocytes. Cytokine proteins that stimulate growth and reproduction of certain kinds of the blood cells in the bone marrow are also referred to as growth factors. The production of white blood cells is controlled by colony stimulating factors. The balance of different CSF is to a certain degree caused by the relationship between different types of the leukocytes formed in the bone marrow. Some CSFs stimulate further differentiation of cells out of the bone marrow.

Chemokines regulate chemotaxis of the cells of the immune system, ensuring their migration and partial activation.

The antibody-forming cells produce ***antibodies – the molecules of immunoglobulins (Ig),*** i.e., the proteins capable of interacting with the appropriate antigens. Each B-cell is programmed to produce, carry on itself and secrete the antibodies only of one specificity.

Each immunoglobulin unit is made up of two heavy chains and two light chains and has two antigen-binding sites. Antibodies are diverse, with more than 1010 possible variations, yet each antibody is designed to recognize only a specfic antigen. Initially bound to B-cells, upon encountering its specific antigen, an antibody/antigen complex stimulates the B-cell to produce copies of the antibody with the aid of helper T-cells. The new antibodies, which are all designed to recognize the infecting antigen, are released into the intercellular fluid where they bind to the infecting antigen, identifying it for destruction by phagocytes and the complement system.

There are 5 classes of Ig – IgA, IgM, IgG, IgD, IgE, each of which possesses the specific effector functions.

Immunoglobulins A make 10–15 % of all Ig of the blood serum; they are predominant Ig of secretions (mucous discharge of the respiratory tract, gastrointestinal tract, saliva, tears, colostrum and female milk). Secretory IgA is like a dimer, consisting of two molecules of IgA and is called secretory component. IgA, leaving the blood flow, penetrates through the epithelial layer and is bound with the secretory component (it is formed in the epithelial cells). The formed ***secretory IgA*** remains either on the surface of the epithelial cell or in the layer of the

The main interleukins

Name	Cells source	Targets	Functions
IL-1α IL-1β	macrophages, B-cells, T-cells, endothelium, LGLs, fibroblasts	T-cells, B-cells, macrophages endothelium, tissue cells	simulates activities of T-cells, B-cells, macrophages, leucocyte adhesion
IL-2	T-cells	T-cells	stimulates T-cytotoxic cells, T-cell growth, costimulates B-cell differentiation
IL-3	T-cells, stem cells	–	simulates multipotenlial hemopoietic cell growth
IL-4	T-cells	B-cells, T-cells	stimulates production and growth B-cells, mast cell growth, class II MHC molecule expression on B-cells and macrophages, enhances IgGl and IgE production
IL-5	T-cells	B-cells	activates eosinophils and B-cells, stimulates growth of B-cells, enhances IgA production
IL-6	T-cells, B-cells, fibroblasts, macrophages	B-cells, hepatocytes	promotes B-cell differentiation into plasma cells and secretion of antibody; increases production of acute phase proteins by hepatocytes
IL-7	monocytes, bone marrow stromal cells	pre-B-cells, T-cells	stimulates growth and differentiation of B-cells, certain mature T-cells, inhibits migration of phagocytic neutrophils away from the site of infection
IL-8	monocytes	neutrophils, T-cells, basophils, keratinocytes	stimulates chemotaxis of neutrophils and T-cells, stimulates granulocyte activity
IL-9	T-cells	–	attracts phagocytic neutrophils to the site of infection, T-cell growth factor
IL-10	T-cells	Th_1-cells	inhibits cytokine synthesis by Th_1-cells

Name	Cells source	Targets	Functions
IL-11	bone marrow stromal cells, fibroblasts	haemopoietic progenitors osteoclasts	stimulates the maturation of hematopoietic cells, colony stimulating factor, inhibits pro-inflammatory cytokine production
IL-12	monocytes	T-cells	stimulates differentiation of $CD4^+$ T-cells to Th_1-cells
IL-13	activated T-cells	monocytes, B-cells	inhibits inflammatory monokine production, stimulates growth and differentiation of B-cells
IL-14	T-cells	–	B-cell growth factor, inhibits Ig secretion
IL-15	monocytes, epithelium, muscle	T-cells, activated B-cells	shares IL-2 bioactivities, stimulates proliferation of activated B-cells and T-cells
IL-16	eosinophils, $CD8^+$ T-cells	$CD4^+$ T-cells	chemoattraction of CD4 cells
IL-17	$CD4^+$ T-cells	epithelium, fibroblasts, endothelium	$CD34^+$ progenitors, simulates secretion of IL-6, IL-8, G-CSF, PGE2, enhances expression ICAM-1
IL-18	macrophages, hepatocytes keratinocytes	co-factor in Th_1-cells induction	enhances NK-cells activity, induces IFN-γ production
IL-21	T-cells, mast cells	T-cells, B-cells, mast cells, eosinophils, hepatocytes	induces acute phase reactants
IL-22	activated cells	Th_2-cells	inhibits IL-4 production
IL-23	activated dendritic cells	memory T-cells	induced proliferation of memory T-cells and moderate levels of IFN-γ production
IL-27	activated APCs, activated dendritic cells	NK-cells, naive $CD4^+$ T-cells, mast cells, monocytes	initial activator of Th_1 responses, potent antitumor activity

Other main cytokines

Name	Cells source	Targets	Functions
Migration inhibition factor (MIF)	T-cells, macrophages	macrophages	inhibits migration of macrophages away from the site of inflammation
Macrophage activating factor (MAF)	T-cells, macrophages	macrophages	stimulates phagocytic activity of macrophages by enhancing their lysosomal activities
Interferon-α (IFN-α)	leucocytes epithelia, fibroblasts	tissue cells	antiviral state, stimulation of NK-cells, MHC class I induction, antiproliferative
Interferon-β (IFN-β)	leucocytes epithelia, fibroblasts	tissue cells, leucocytes	antiviral state, MHC class I induction, antiproliferative, stimulation of NK-ells
Interferon-γ (IFN-γ)	T-cells, NK-cells epithelia, fibroblasts	leucocytes, tissue cells, Th_2-cells	activates antiviral proteins, T-cells, and macrophages; induces class II MHC molecule expression on macrophages
Colony-stimulating factors (CSF)	macrophages, fibroblasts	stem cells	stimulates the growth and differentiation of colonies of granulocytes and macrophages from bone marrow progenitor cells
Transforming growth factor-β (TGF-β)	–	cells of other types	inhibits and stimulates extracellular matrix formation; inhibits B-cells, T-cells and NK-cell activity
Tumor necrosis factor-α (TNF-α)	macrophages, mast cells, lymphocytes	macrophages, granulocytes, tissue cells	regulates other immune cells, endothelial activation, causes lysis of target cells
Tumor necrosis factor-β (TNF-β)	lymphocytes	–	regulates other immune cells, endothelial activation, causes lysis of target cells

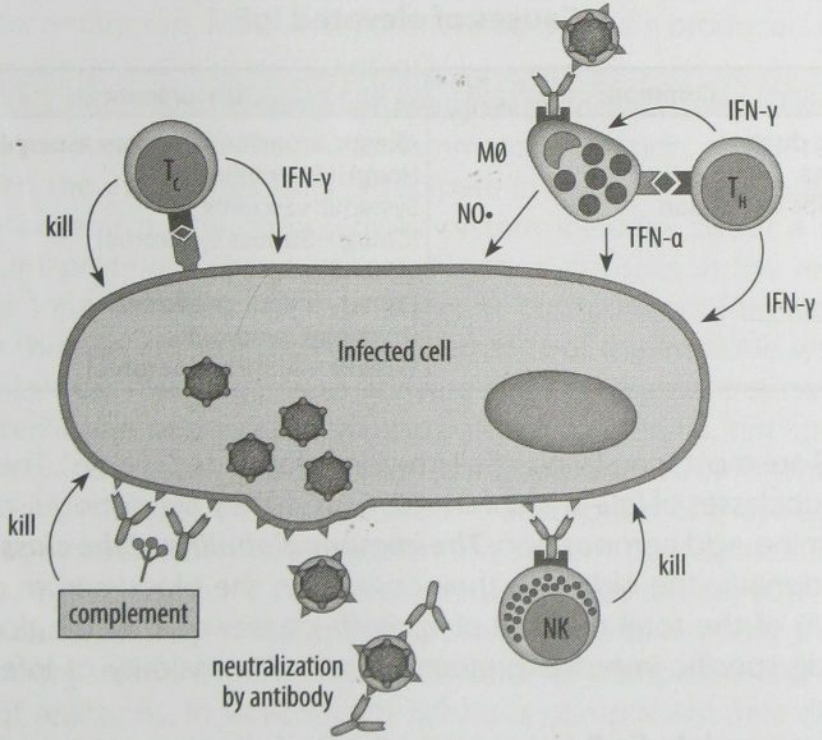

Fig. 1.16. Penetration of virus into mucosal surfaces is inhibited by IgA.
Following the initial infection, the virus may spread to other tissues via the blood stream. Interferons produced by the innate (IFN-α and IFN-β) and adaptive (IFN-γ) immune responses make neighbouring cells resistant to infection by spreading virus. Antibodies are important in controlling free virus, whereas. T-cells and NK-cells are effective at killing infected cells (I. Roitt et al., Immunology, 2001).

mucus above the epithelium and fulfills its basic nonspecific protective function. The newborns get the secretory IgA during the first days of life through the mother colostrum, protecting their bronchopulmonary and gastrointestinal tract until they form their own mechanisms of formation of the secretory IgA and their own microflora (Fig. 1.16).

The immunoglobulins of the class G make 75 % of all Ig of the blood serum of man. The molecular weight provides the possibility of penetration through the placenta from a mother to a fetus. The molecules

Causes of elevated IgE

Common	Uncommon
Atopic disease Asthma Parasitic infestation	Allergic bronchopulmonary aspergillosis Hodgkin's lymphoma Systemic vasculitis (Churg – Strauss syndrome) IgE myeloma Primary immunodeficiencies (Hyper IgE syndrome, Wiskott – Aldrich syndrome)

of IgG are most long living of all immunoglobulins (23 days). There are four subclasses of IgG in man: G1, G2, G3, G4, they are distinguished by the amino-acid composition. ***The immunoglobulins of the class M*** are evolutionarily the oldest Ig, their content in the blood serum makes 5–10 % of the total number of Ig. Both classes of immunoglobulins provide specific immune protection from the majority of infectious agents.

Immunoglobulin E makes about 0.2 % of all serum immunoglobulins. It is accumulated predominantly in the tissues, mucous and skin membranes, where it is gathered on the surface of the mast cells, basophils, eosinophils. Its life cycle is 2.5 days. The main function seems to protect the host against invading parasites. The antigen-specific IgE interacts with mast cells and eosinophils to protect the host against the invading parasite. IgE-antibodies play a basic role in the formation of the hypersensitivity response of the immediate type, i.e. anaphylaxis.

Immunoglobulins D – represents about 0.25 % of the total serum immunoglobulins and its half-life is 2.8 days. Little is known about humoral functions of this immunoglobulin. IgD is the major antigen receptor isotype on the surface of most peripheral B-cells. Serum IgD was considered as an early marker of B-cell activation. IgD can have a regulatory role, e.g., to enhance a protective antibody response of the IgM, IgG, or IgA isotype, or to interfere with viral replication. IgD is a potent inducer of tumor necrosis factor alpha (TNF-α), IL-1, and IL-1

receptor antagonist. Monocytes seem to be the main producers of cytokines *in vitro* in the presence of IgD.

The proteins of the ***complement system*** serve as the mediators of phagocytosis, regulate the inflammatory reaction, and interacting with the antibodies they participate in the immune protection of the organism. The complement system includes about a score of serum proteins, whose general function consists in the regulation of inflammation. The complement components interact between themselves and with other elements of the immune system. A number of microorganisms activate the complement system by ***the alternative way*** – a mechanism of the congenital, nonspecific immunity. As a result the complement components bind with the surface of microbes, which leads to the absorption of these agents by phagocytes. When the complement system participates in the reactions of the specific acquired immunity, they usually activate it by ***the classical way*** of the antibody having bound with the surface of microorganisms (Fig. 1.17). The activation of complement is a cascade of reactions, in which each previous component acts on the following one.

The complement system is activated by the complex antigen-antibody in such sequence: C1, C4, C2 and C3, C5, C6, C7, C8, C9. The complement causes bacteriolysis, stimulates phagocytosis, and causes changes in the membrane, activation of the factors, which participate in the inflammatory reactions, leading to damage of the cells. The biological properties of the complement (C) include the following:

- C1 – C4 – neutralization of viruses;
- C1 – C5 – formation of the histamine-releasing factors (anaphylatoxins), intensification of the phagocytosis reactions;
- C5 – C9 – participation in the reactions of cytolysis, bacteriolysis, in the reactions of the transplant rejection.

Lysozyme is a specific enzyme, which destroys the walls of bacteria. There is lysozyme in a large amount in the saliva; it explains its antibacterial properties.

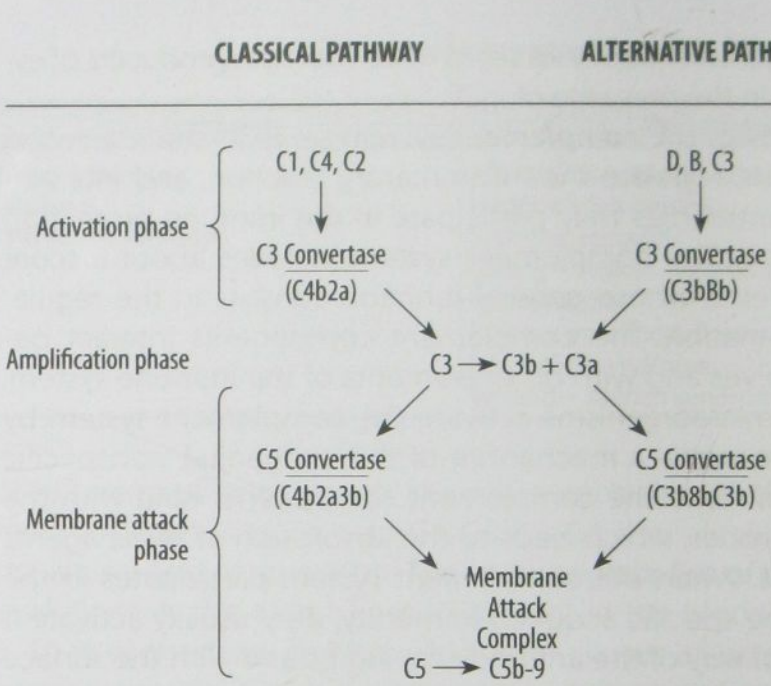

Fig. 1.17. The two complement activation pathways *(Klaus D. Elgert, Immunology, 1996).*

Transferin is the protein, which competes with the bacteria for capture of the specific substances (for example, iron), necessary for their development. As a result the growth and multiplication of the bacteria is slowed down.

C-reactive protein is activated similarly to the compliment on penetration into the blood of foreign structures. Addition of this protein to the bacteria makes them vulnerable for the cells of the immune system. C-reactive protein (CRP) is named for the ability to bind with the C-protein of pneumococci. Because of this binding phagocytes begin to absorb pneumococci more actively – the process is called opsonization. Antibodies and complement components mainly act as the opsonin, i.e. opsonizing molecules.

1.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Tasks of clinical immunology and allergology. Structure and function of the immune system.
2. Differences between the specific and nonspecific immune response. Apoptosis (a concept and role in functioning of the organism).
3. Antigen presentation: a role in the formation of the immune response. Antigen presentation cells.
4. Phagocytosis: a role in the realization of the nonspecific and specific immune response.
5. Humoral factors of the nonspecific immune protection of the organism.
6. Cells-killers: basic types, their functions and peculiarities.
7. Granulocytes: functions and role in the immune response. Diagnostic significance in different pathological states.
8. Agranulocytes: functions and role in the response. Diagnostic significance in different pathological states.
9. A system of complement. Biological consequences of activation of the complement system. Ways of activation.
10. B-lymphocytes: markers and functions. Diagnostic significance in different pathological states.
11. T-lymphocytes: forms and basic markers. Diagnostic significance in different pathological states.
12. Regulated T-lymphocytes, basic functions. Significance of the functional balance between the T-helpers (Th_1/Th_2).
13. Immunoglobulins: structure, functions, classes. Diagnostic significance in different pathological states.
14. Cytokines: basic classes and their functions.
15. Main complex of the histocompatibility of man. Classes of antigens and their role in forming the immune response.

Practical skills

1. Estimation of the organism state on the basis of data of leukograms and immunograms.
2. Quantitative and functional immunological tests.
3. Immunogram: basic indices.
4. Methods of determining a quantity and functional ability of T- and B-lymphocytes.
5. Methods of determining the state of phagocytic-macrophage system.

Tests

1. One of the differences between the specific and nonspecific immune response is:
   A. Participation of macrophages
   B. Participation of the complement system
   C. Production of interferons
   D. Production of specific antibodies
   E. Production of acute phase proteins

2. Specific immune response regulates:
   A. B-cell
   B. T-cell
   C. Antigen presentation cells
   D. Neutrophils
   E. Cells of the spleen

3. Humoral specific immune response:
   A. Is a result of the production of antibodies by plasma cells
   B. Is a result of activation of T-killers
   C. Is a result of activation of neutrophils
   D. Is a part of the congenital immune response
   E. Occurs in the thymus

4. Antigenic determinants of T-cells are areas, which are connected with:
   A. IgM and IgD on the surface of the B-cells
   B. Fc receptors on the macrophages
   C. MHC antigens, presented by T-cells and for which there are the specific receptors of the T-cell
   D. Receptors on polynuclear leukocytes
   E. "The domain of death" of the lymphocytes

5. Central organs of the immune system:
   A. The thymus and liver
   B. The bone marrow and thymus
   C. The thymus and spleen
   D. The spleen and bone marrow
   E. The thymus and lymph nodes

6. Natural killer (NK) cells are produced in:
   A. The bone marrow
   B. The spleen
   C. The mucous membranes
   D. The liver
   E. The skin

7. Main function of the dendritic cells:
   A. Phagocytosis
   B. Opsonization
   C. Production of interferons
   D. Antigen presentation
   E. Chemotaxis

8. T-helpers have:
   A. CD1 determinants on the surface
   B. CD4 determinants on the surface
   C. CD8 determinants on the surface

D. CD14 determinants on the surface
E. CD19 determinants on the surface

9. T-lymphocytes have:
   A. CD56 determinants on the surface
   B. CD3 of the determinants on the surface
   C. CD18 of the determinants on the surface
   D. CD15 of the determinants on the surface
   E. CD22 of the determinants on the surface

10. What immunoglobulin is the secretory component connected with:
    A. IgA
    B. IgM
    C. IgG
    D. IgE
    E. IgD

CHAPTER II

METHODS OF IMMUNOLOGICAL INVESTIGATION. THE CONCEPT OF IMMUNOGRAM. BASIC RULES FOR ESTIMATION OF THE IMMUNE STATUS

The immune system functions according to certain biological laws, which have been formed during phylogenesis. The important principles of its functioning are: close cooperation with other systems of an organism, first of all, with nervous and endocrine systems (neuro-immune-endocrine regulation); self-organization and expediency (a choice of priority mechanism of the immune response, its expressiveness and duration); synchronism and coordination of work of different parts and mechanisms; possibility of duplication of functions of defectively functioning links or mechanisms; presence of functional reserves. Therefore, the estimation of the condition of the immune status should be made in complex, using all arsenals of methods of examination: anamnesis; clinical, instrumental (diagnostic tests), and laboratory methods.

2.1. Immunologic anamnesis

Immunologic anamnesis is made to reveal possible dysfunctions of the immune system and differentiated depending on patient' complaints and prospective presence of specific pathology (allergic, oncological and autoimmune diseases, immunodeficient conditions, immunopathology of reproduction, etc.).

The general plan of taking the immunologic anamnesis includes:

1) detection of peculiarities of the family anamnesis: presence of allergic, autoimmune, oncological, lymphoproliferative diseases,

pathology of the endocrine system, predisposition to frequent respiratory viral diseases, recurrent bacterial and fungal infections in close relatives (parents, brothers and sisters, grandmothers and grandfathers, aunts and uncles);

2) presence of chronic diseases of various organs and systems, peculiarities of their course in a patient;
3) infectious diseases having been before, pathological conditions and surgical interventions (traumas, burns, poisonings with chemical substances, tonsillectomy, etc.), peculiarities of their course and efficacy of the therapy;
4) contacts with harmful ecological factors (industrial hazards, living conditions);
5) peculiarities of the vital activity, capability of influencing formation of the immunopathological conditions (stress, physical overexertion, frequent changes of climatic zones, bad habits, hypodynamia, poor nutrition, etc.);
6) peculiarities of temperature reaction in respiratory viral infections (absence of rise in the temperature or long-term postinfectious subfebrile temperature);
7) presence of pathological syndromes and conditions (stable anemia, fever of unclear etiology, long-term diarrhea, loss of the body weight of unclear genesis, frequent manifestations of herpetic infections, lymphadenopathy, etc.);
8) frequent acute bacterial/fungal infections or aggravations of the foci of chronic infection (sinusites, otitises, osteomyelitis, furunculosis, etc.);
9) peculiarities of the long-term course therapy (radiation and chemotherapy, intake of glucocorticosteroids, antibiotics, contraceptives, anti-inflammatory and immunotropic preparations, homeopathic medicines and biologically active additives);
10) pathological responses (allergy, hyperthermia, complications) to medicines and vaccination.

2.2. Clinical and instrumental methods of estimation of the immune system condition

The following methods of examination are used differentially depending on prospective presence of various pathologies.

They include:

1) physical and instrumental examination (ultrasonic diagnostics) of organs of the immune system (spleen, thymus, lymph nodes), examination of the tonsils;
2) examination of the condition of integuments and visible mucous membranes (eruptions, hemorrhages, etc.);
3) examination of the condition of the nervous, cardiovascular, broncho-pulmonary, digestive, endocrine, secretory systems and locomotor apparatus (depending on prospective diseases and pathological conditions);
4) detection of malignant neoplasms and foci of chronic infection;
5) diagnostic tests in patient (elimination, provocation, tests of passive carriage).

Diagnostic tests are made to establish the allergen or factor exerting negative influence on the immune system.

Elimination tests. A patient is excluded contact with prospective causative-significant factors (allergens, chemical substances, household chemicals, cosmetics, etc.) for some time, and then changes in his/her condition are estimated.

Provocation tests. They are used for identification of causative-significant allergen (it is introduced directly into the "shock organ" – the skin, mucosa of the nose, conjunctiva or bronchial tubes). The response is estimated in presence of clinical manifestations, by means of clinical-functional (rhinopneumotaxometry, spirography) and laboratory tests (Yasinovsky test – the test of inhibition of leukocyte migration in the oral cavity, leuco- and thrombopenia in food allergy). A version of provocation tests are tests with tuberculin (Mantoux test), brucellin, etc.

2.3. Syndromes of immune system dysfunction

In presence of dysfunction of the immune system four syndromes of the immune disturbances are distinguished: infectious, lymphoproliferative, allergic and autoimmune.

Infectious syndrome is usually diagnosed in the following diseases: *chronic recurrent infections (bacterial infections of ENT, broncho-pulmonary system, skin, genitals, etc.; viruses of herpetic group), chronic hepatites, parasitic infections.*

Clinically it is characterized by presence of: long-tern subfebrile period; fever of unclear etiology; lymphadenopathy; frequent diseases of acute respiratory viral infections (more than 4 per year in adults) with predisposition to development of bacterial complications; predisposition to generalized course of infectious diseases; long-term diarrhea; vegeto-vascular dystonia with prevalence of vagotonia; disturbance of microbiocenosis of the intestines, nasopharynx and vagina with prevalence of mycotic manifestations.

Lymphoproliferative syndrome is diagnosed in presence of: ***X-linked lymphoproliferative syndrome – Duncan's disease*** (splenomegaly, hyperplasia of all groups of lymph nodes with phenomena of lymphadenitis, frequent bacterial infections, mononucleosis); ***tumors of the immune system*** (T-and B-cellular lymphomas, lymphosarcoma, Hodgkin's disease, Kaposi's sarcoma); ***lympholeucosis, lymphadenopathies of the inflammatory nature*** (lymphadenitis, lymphangitis); ***malignant metastases in the lymph nodes.***

The basic clinical symptom is enlargement of the lymph nodes. However, enlargement of the lymph nodes is also probable ***in the active immune response to an antigen*** (substantial increase of the lymphocyte number and macrophages in the regional lymph nodes). The basic laboratory criterion is essential increase of lymphocyte quantity against the background of leucocytosis or leucopenia.

Due the peculiarities of etiopathogenesis two forms of lymphoproliferative syndrome are distinguished – lymphadenopathy of the inflammatory or tumorous nature. ***Signs of inflammatory lymphade-***

nopathies: lymph nodes (are enlarged slightly or moderately; of insignificant or moderate density; painful to palpation; are usually mobile and not adhered); the skin above them is frequently hyperemic; sometimes lymphangitis is observed; regional lymph nodes are more often enlarged. ***Signs of tumorous lymphadenopathies:*** lymph nodes (are more often considerably enlarged, of moderate density or dense, painless to palpation; are usually adhered and form packages); the skin above them is not hyperemic.

Additional methods of diagnosis: biopsy of the lymph node (with histological identification); determination of alkaline phosphatase activity in the blood; ultrasound investigation of the abdominal organs; roentgenography of the lungs and bones of the skeleton; lymphography. ***Due to indications the following methods are used:*** computer tomography of the chest and abdominal organs; diagnostic laparotomy with splenectomy or laparoscopy with biopsy of the intraabdominal lymph nodes; scintigraphy of the bone marrow, liver, and spleen.

Allergic syndrome is characterized by presence of: ***bronchial asthma; allergic rhinitis and rhinosinusitis; pollinosis; urticaria; Quincke's edema; atopic or contact dermatitis; medicinal allergy; allergic response to chemical substances).***

Autoimmune syndrome is diagnosed in presence of: autoimmune diseases (rheumatoid arthritis, systemic lupus erythematosus, a syndrome of scleroderma, Sjogren syndrome, Wegener's granulomatosis, nodular periarteriitis, autoimmune abortive disease, Addison's disease, myasternia gravis, multiple sclerosis, etc.); immune leucopenias and thrombocytopenias; hemolytic anemias, schizophrenia, severe course of a climacteric syndrome.

2.4. Laboratory methods of estimation of the immune system

There are some approaches to differentiation of the immunogram indices. There are immunologic tests of the 1st and 2nd level; indices

of the cellular and humoral immunity; quantitative (determination of the absolute and relative contents of cells of the immune system) and functional (determination of the functional activity of cells of the immune system) indices.

The basic indices of the immunogram

I level test:

Cellular link. Quantitative: determination of the relative and absolute number of leukocytes, basophils, eosinophils, neutrophils, lymphocytes and monocytes in the peripheral blood; determination of the relative and absolute amount of T (CD3, E-RFC erythrocyte rosette-forming cells) – and B (CD19 – CD22, EAC-RFC) lymphocytes.

Functional: determination of the phagocytic activity of leukocytes: phagocytic index (PI); amount of active phagocytes (PA).

Humoral link: determination of serum antibodies concentration of the basic classes (Ig A, Ig M, Ig G).

II level test:

Cellular link. Quantitative: determination of subpopulations of regulatory T-lymphocytes; T-helpers (CD4); T-cytotoxic, suppressors (CD8); NK-cells (natural killers), K- and NK-cells;

Functional: reaction of inhibition of leukocytes migration (RILM); reaction of blast transformation of lymphocytes (RBTL); determination of oxidation-reduction activity of neutrophils- the test of restoration of nitro blue tetrazolium (NBT); estimation of the receptor apparatus of phagocytes (MCAB); estimation of various stages of phagocytosis (and its completeness); estimation of adhesive abilities of neutrophils; determination of migration and chemotaxis of phagocytic cells.

Humoral link: determination of the level of general IgE; determination of the level of circulating immune complexes (CIC); determination of the contents of complement – general hemolytic activity (and its various components); determination of the contents of cytokines; determination of levels of autoantibodies to various organs and tissues.

The characteristic of some methods of determination

Cellular link:

Determination of lymphocyte subpopulations: 1) the use of monoclonal antibodies for detection of clusters of differentiations – CD (a method of membranous immunofluorescence on flow cytofluorimeters); 2) a method of cytochemical staining with the use of streptavidin – biotin enzyme mark in visual estimation in the light microscope, etc.); 3) methods of rosette-formation with erythrocytes of the ram and mouse.

Reaction of inhibition of leukocyte migration in the blood allows to estimate the ability of T-lymphocytes to produce lymphokines in response to antigenic stimulation. It allows to determine a sensitization of human cells to various allergens *in vitro*.

Reaction of spontaneous blast transformations of lymphocytes is an ability of lymphocytes to transformation without stimulation. The study is carried out for estimation of the functional activity of T- lymphocytes.

Stimulated reaction of blast transformations of lymphocytes with mitogens (PHG – phytohemogglutinin, ConA – concanavalin A) characterizes the functional ability T-lymphocytes to transformation and duplication under the influence of antigens, allergens and mitogens.

Spontaneous test with NBT (nitro blue tetrazolium) allows to estimate a condition of oxygen-depending mechanism of the bacterial action of phagocytes (granulocytes) of the blood *in vitro*. It characterizes a condition and degree of activation of endocellular NADP-H-oxidase antibacterial system.

Activated test with NBT (nitro blue tetrazolium) allows to estimate a functional reserve of oxygen-depending mechanism of the bacterial action of phagocytes. The test is used for revealing reserve possibilities of the endocellular systems of phagocytes.

Determination of the phagocytic activity of neutrophils: 1) phagocytic number (PN) – average amount of the microbes absorbed by one neutrophils of the blood; 2) phagocytic index – relative amount of

neutrophils (expressed in percentage), participating in phagocytosis; 3) the amount of active phagocytes – absolute amount of phagocytizing neutrophils in 1 l of the blood; 4) the index of phagocytosis completeness – reflects digesting ability of phagocytes.

Humoral link:

Determination of contents of antibodies (a method of radial immunodiffusion by Mancini; IEA; radioimmunologic method; turbodimetric method; chemoluminiscence) estimates the contents of different classes of antibodies in the blood serum.

Determination of the titer of the complementary activity (by 50 % or 100 % hemolysis) estimates activity of complement components in its activation by a classical and alternative way.

Possibilities of modern diagnostic equipment for laboratory research

1. Flow cytofluorometer:

1) immunephenotyping (determination of subpopulations of cells in the peripheral blood, samples of the tissue and cellular suspensions);
2) analysis of reticulocytes (automatic calculation of reticulocytes in the integral blood);
3) analysis of hemopoietic cells (differentiation of hemoblastoses). Calculation of $CD34^+$ HCP (hemopietic cells-predecessors) – "the gold standard" in estimation of amount of the hemopietic cells;
4) analysis of parameters of the cellular cycle (DNA-cytometry) – measurement of the amount of DNA in the cell, S-phases and discrimination of doublets;
5) analysis of the endocellular cytokines;
6) analysis of apoptosis (amount of the cells, being at different stages of apoptosis);
7) mobilization of endocellular calcium (determination of endocellular Ca^{2+});

8) estimation of activity of the endocellular enzymes and pH.
9) typing of tumors (including solid tumors) using tissue-specific markers.

2. Immunoenzymatic analyzer (automatic, semi-automatic, flatbed):

1) diagnosis of infections: hepatitis A (antibodies to VHA), hepatitis B (HBsAg, HBeAg, anti-HBsAg, anti-HBeAg, anti-HBcAg), hepatitis C (antibodies to VHC), a clamidiosis (antigens, IgG, IgA, IgM), trichomoniasis (IgM), toxoplasmosis (IgG, IgM), lambliasis, adenovirus (IgG, IgM), candidiases, aspergillosis, Lime disease, cytomegalovirus (IgG, IgM), Epstein – Barr virus (IgG, IgM), human immunodeficiency virus 1, 2 (antibodies), a virus of herpes simplex (IgG, IgM), virus of zoster (IgG, IgM), virus reproach of measles (IgA, IgG, IgM), a virus of parotitis (IgA, IgG, IgM), a virus of rubella (IgG, IgM), campilobacteriosis (IgG, IgM), poliomyelitis, diphtheria (IgG), echinococcosis (IgG), syphilis (IgG, IgM), yersiniosis, tetanus;
2) determination of tumorous markers (an alpha-fetoprotein, carcinoembryonic antigen, prostatospecific antigen, CA-125, CA-15-3, CA-19-9, CA-242, ferritin, tissue polypeptide antigen, beta-2-microglobulin, UBC (cancer of the bladder);
3) determination of functioning of the glands of internal secretion:
   - the thyroid gland (triiodothyronine (T3), thyroxin (T4), thyrotropic hormone, thyroglobulin, antibodies to thyroglobulin, antibodies to thyroperoxidase, antibodies to mictosomal antigens of thyrocytes);
   - cortisol, testosterone, androstendion, progesterone, adrenaline, melatonin, serotonin, somatotropic hormone, estradiol, estriol, calcitonin;
4) determination of the reproductive function (prolactin, luteinizing hormone (LH), follicle-stimulating hormone (FSH), chorionic gonadotropin of human, antibodies to sperm;
5) allergologic studies (general IgE, antibodies to allergens, histamine);
6) diagnosis of the autoimmune and systemic diseases (rheumatoid factor, C-reactive protein, anti-streptolysin O (ASLO), antibodies

to DNA and RNA (dsDNA, histones, SS-A, SS-B, RNP, Scl-70, Sm, Jo-1);

7) estimation of hemostasis (protein C, PAI-1, D-dimer, tissue activator of plasminogen, a complex t-PA-PA-1, plaminogen glycated, u-plasminogen, scu-plasminogen, plasmin, α-antiplasmin, fibrinonectin, vitronectin,).

3. Chemiluminescence Immunoassay Analyzer:

1) diagnosis of the autoimmune and rheumatic diseases (determination of antibodies to native DNA; anti-streptolysin O; rheumatoid factor; C-reactive protein; determination of antibodies to extracted nuclear antigens);
2) determination of the complement components (C1 inhibitor, C3, C4 in the blood serum);
3) oncomarkers (CEA-cancer-embryonic antigen, AFP, PSA, PSAfree, 3rd generation of PCA, CA 125, CA 15-3, CA 19-9, PAP-prostatic acid phosphotase, cytokeratin);
4) cardiovascular markers (creatinine kinase-MB, creatinine kinase-MB turbo, troponin I, troponin I turbo, myoglobin, myoglobin turbo);
5) cytokines (IL-6, IL-8, IL-1b, IL2R, TNF-α-factor of tumor necrosis, LBP-lipopolysaccharide binding protein);
6) function of the thyroid gland (general T3, free T3, general T4, free T4, TBG-thyroxin-binding globulin, thyrotropic hormone, 3rd generation AT-TH, AT-TBG-antibodies to peroxidase of thyrocytes, the test of absorption of thyroid hormones);
7) reproductive hormones (estradiol, unconjugated estradiol, progesterone, general testosterone, LH, FSH, CGH-chorionic gonadotropin of human, CGH turbo, prolactin, DHEA-SO4, GBSH-globulin binding sexual hormones, free beta-CGH);
8) infectious diseases (CMV IgG, Rubella IgG, Rubella IgM, Toxoplasma IgG, Toxoplasma IgM, H. Pilori IgG, Anti-HBs, HbsAg, HbsAg confirming, Anti-HBc, Anti-HBc IgM, Lyme IgG (IgG to Lime disease));
9) diabetes (C-peptide, insulin);

10) metabolites of the bone tissue (parathyroid hormone, parathyroid hormone turbo, a marker of bone resorption);
11) allergology (total IgE, specific IgE to various allergens)
12) medicinal monitoring (theophilin, digitoxin, digoxin, carbamazepine, phenitoin, valporic acid, phenobarbital);
13) anemia (B12, folic acid, ferritin, erythropoietin);
14) narcotic substances (cocaine, opiates, cannibinoids, metabolites of nicotine, amfetamin, metamfetamin);
15) other analites (ACTH, b-2microglobulin, C-reactive protein, albumin, STH, cortisol).

2.5. Immunogram: indications for administration and interpretation of results

Indications for estimation of the immune status of a patient

1. Suspicion on primary or secondary immunodeficiency.
2. Frequent viral respiratory infections with constant development of bacterial complications.
3. Recurrent bacterial infections (otitises, sinusites, pneumonias, furunculosis, osteomyelitis).
4. Septic conditions.
5. Frequently recurrent chronic somatic diseases.
6. Long subfebrile period of obscure etiology.
7. Lymphadenopathy.
8. Long or atypical course of the inflammatory and infectious diseases, which are resistant to antibioticotherapy.
9. Frequently recurrent mycoses with a long course, resistant to antifungal therapy.
10. Frequent relapses of herpetic infections of the skin and mucous membranes.
11. Autoimmune diseases.

12. Allergic diseases.
13. Suspicion on Acquired Immune Deficiency Syndrome (AIDS) or HIV-infection.
14. Oncological diseases.
15. Examination of recipients before transplantation of organs.
16. Examination of patients before serious surgical interventions or in complicated course of the postoperative period.
17. Examination of patients with the reproductive dysfunction.
18. The control of therapy with cytostatics, immunodepressants and immunomodulators.

Recommendations for interpretation of immunogram results (Kazmirchuk V. E., Maltsev D. V.):

1. The high-grade clinical analysis of the immunogram can be made only in complex with estimation of the clinical picture of the a disease in a patient and data of his/her anamnesis. It is impossible to make a clinical conclusion only on the basis of immunogram, as the same changes of immunogram indices can be observed in essentially different pathological processes.
2. The complex analysis of immunograms is more informative than estimation of any index separately. Identical changes of the certain index in different phases of acute inflammatory process can be considered both as a favorable and unfavorable sign.
3. The real information on changes of the immunogram is given by significant index disturbances in the immunogram (40–50 % of the norm and more). Due to lability of the immunogram indices their insignificant fluctuations are possible in absolutely healthy persons.
4. Clinical data play a decisive role, and the immunogram carries auxiliary diagnostic and prognostic value. Absence of shifts in the immunogram in presence of the clinical picture of pathology demands studying of the component function of separate links of the immune system.

5. The analysis of the immunogram in dynamics (especially in comparison with clinical dynamics) is more informative from the point of view of both diagnostics, and the prognosis of the disease course, helps to avoid erroneous interpretation.
6. Individual indices of the norm in the patient have diagnostic and prognostic value (taking into account age and presence of concomitant chronic diseases, actions of harmful factors, medicine therapy).
7. A ratio of the immunogram indices but not their absolute values is of primary value in estimation of the immunogram.
8. While estimating the immunogram indices it is necessary to consider a possibility of their fluctuations because of meal, physical activities, sensation of fear, time of the day.
9. Discrepancy in shifts of the immunogram indices and clinical picture of the disease (a syndrome of dissociation) is evidence of the unfavorable development of the process.
10. The higher antigenicity and bigger the zone of its penetration, the brighter will be the inflammatory process. Therefore, shifts in the immunogram should be more expressed, and it will be the evidence in favor of adequacy of the response of the immune system. Absence of the specified changes in the leuco-and immunogram is an unfavorable symptom, which is evidence of inadequacy of the immune system work.

2.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Immunologic anamnesis: the basic points.
2. Clinical and instrumental methods of investigation of the immune system. Kinds of diagnostic tests.
3. Characteristics of syndromes of the immune dysfunctions.

4. Quantitative and functional immunologic tests.
5. Modern methods of estimation of the immune status. Basic determined indices.
6. Basic indications for immunologic examination.

Practical skills

1. To be able to take the immunologic anamnesis.
2. To estimate the condition of the immune system by results of clinical and instrumental examination.
3. To make the differential diagnosis of the basic syndromes of the immune dysfunctions.
4. To know basic rules of the immunogram interpretation.

Tests

1. Which of the following complaints do not speak about suspicion on presence of dysfunction of the immune system?
   A. Long-term diarrhea
   B. Pain in the muscles after physical activity
   C. Long-term postinfectious subfebrile period
   D. Stable anemia
   E. Lymphadenopathy

2. Frequency of acute respiratory viral infections of more than... may be the evidence of presence of the infectious syndrome in adults:
   A. Two times a year
   B. Three times a year
   S. Four times a year
   D. Five times a year
   E. Six times a year

3. What of the syndromes is not related to the syndromes of the immune dysfunctions?

A. Autoimmune syndromes
B. Infectious syndromes
C. Lymphoproliferative syndromes
D. Gastrointestinal syndromes
E. Allergic syndromes

4. A sign of tumorous lymphadenopathy is:
A. Painlessness to palpation
B. Hyperemia of integuments above the lymph nodes
C. Mobility of the lymph nodes
D. Lymphangitis
E. Insignificant density of the lymph nodes

5. Functional immunologic tests of the first level include the determination of:
A. CD3-cells
B. B-lymphocytes
C. Phagocytic index
D. Antibodies
E. Monocytes

6. Quantitative immunological test of the first level is the determination of:
A. Circulating immune complexes
B. CD8 cells
C. The deceleration reaction of leukocyte migration
D. The reaction of blast transformation of lymphocytes
E. CD22 cells

7. Estimation of the functional reserve of the oxygen-depending mechanism of the bacterial action of phagocytes is made by:
A. Spontaneous RBTL
B. Spontaneous NBT-test
C. RILM

D. Stimulated RBTL
E. Activated NBT-test

8. For differentiation of hemoblasts, "gold standard" in estimation of the quantity of hemopoietic cell is calculation of ... cells used:
   A. $CD15^+$
   B. $CD21^+$
   C. $CD26^+$
   D. $CD34^+$ HPC
   E. $CD56^+$

9. What criterion is the indication for administration of the immunogram?
   A. Osteoporosis
   B. Autoimmune diseases
   C. Recurrent bacterial infections
   D. Long subfebrile period of unclear etiology
   E. Frequent relapses of herpetic infections of the skin and mucous membranes

10. What index reflects digesting ability of phagocytes?
    A. Phagocytic index
    B. Amount of active phagocytes
    C. Index of completeness of phagocytosis
    D. NBT-test
    E. Quantity of active phagocytes

CHAPTER III

IMMUNITY AND INFECTIOUS PROCESS. AIDS/HIV

3.1. Characteristics of the course and stages of the infectious process pathogenesis

The infectious process is a complex of physiological and pathological responses of an organism of the host, developed in reply to penetration of pathogenic microorganisms.

The character of the course of an infectious disease is determined by a virulent ability of a microorganism and the condition of the immune system of the host's organism. Pathogenic microorganisms discharge a number of factors, which are capable of providing their existence in the host's organism: enzymes *hyaluronidase* and *neuraminidase* provide penetration of the pathogen and its distribution in an organism; *adhesins* (fimbriae, fibrilles and other structures of the bacteria adhesion or proteins of capside virus) provide an attachment to cells of the host's organism tissues; *exo- and endotoxins* disturb homeostasis of the host's organism (reduce ability of the organism to resistance); *peptidoglicanes* (one of the basic structural elements of the microbic wall) excessively activate immunocompetent cells in a large quantity → inhibition of the immune responses; *capsules and capsule-like structures* of microorganisms prevent their phagocytosis; *molecules inhibiting cytokine action*.

There are several stages of pathogenesis of the infectious process (Ignatov P. E., 2002):

- colonization and penetration (attachment of the causative agent to the cells-targets and penetration through natural barriers);

- protection against immunity responses (a complex of genetically determined mechanisms, by means of which the causative agent neutralizes (or suppresses) protective responses of the organism);
- distribution (break of the regional lymphoid barriers and distribution of the causative agent in the internal environment of the hosts's organism);
- inflammation (nonspecific responses of inflammation, adaptive responses of the organism (fever, diarrhea, cough), reactions of the adaptive immunity, which develop in response to toxins and antigens of the causative agent or to its own damaged proteins);
- recovery or synchronization of an infectious disease (intensification of the specific immune response and reduction in intensity (or full discontinuance) of nonspecific inflammatory reactions).

3.2. Antibacterial immunity

Antibacterial immunity provides protection of the organism by destruction of microbic – pathogenic and conditionally-pathogenic agents (Fig. 3.1). Factors of nonspecific protection are natural biological barriers (the skin, mucous membranes) and system of the local immune protection of mucous membranes (secretory IgA, lysozyme, neutrophils, tissue macrophages, etc.).

Great bulk of bacteria, which have got through the natural barriers of an organism into its internal environment, are destroyed by phagocytes (by phagocytosis, less often by extracellular and contact cytolysis) and system of complement (cytolysis) (Fig. 3.2). NK-cells also participate in antibacterial protection, especially in infection by the endocellular causative agents.

Neutralization of bacteria in the organism is also provided by ***antibacterial antibodies*** (Fig. 3.3 and 3.4). Antibodies cause dissolution or

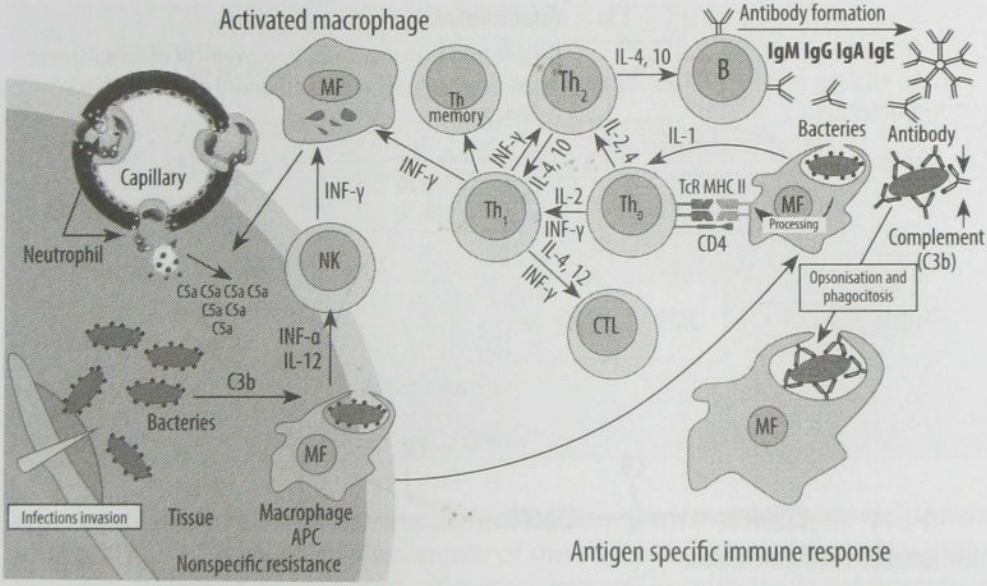

Fig. 3.1. Scheme of the antibacterial immune response.
(Source: http://nsau.edu.ru/images/vetfac/images/ebooks/microbiology/index.htm).

agglutination of bacteria. Transition of the virulent forms of microbes into nonvirulent ones also occurs in their presence. A test for determination of IgG avidity is made for more exact establishment of the infection period in increased contents of IgM and IgG. Avidity is a degree of durability of binding of the antigen and antibodies. Low avidity antibodies are encountered in primary infection, high avidity antibodies – in the infection being in the past or reinfection. The basic bacterial antigens are shown on Fig. 3.5.

In protection of the organism against infections with endocellular persistency of the causative agent the important role is played by **cytotoxic T-lymphocytes.** Infragamma radiation can also suppress growth of the causative agent inside of the organism cells. The mechanism of antibacterial immunity differs in struggle against different infectious causative agent.

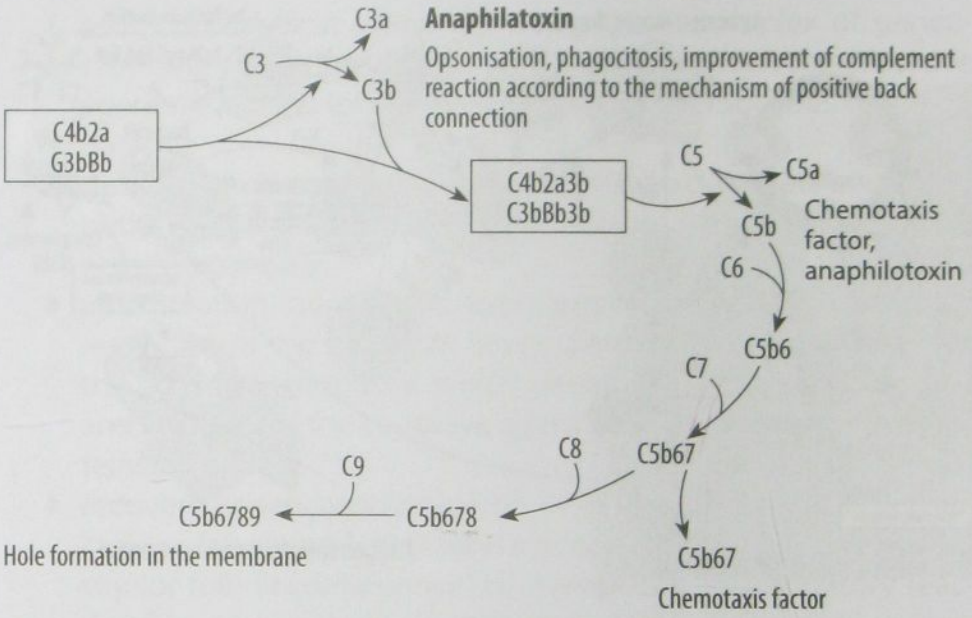

Fig. 3.2. Formation of the membrane attacking complement complex. *N. A. Soter, K. F. Austen. Effactor system of inflammation / I. B. Fitzerald et al. (eds.) // Dermatology in general medicine. – McGraw-Hill Book company, 1979. (Source: http://www.medicum.nnov.ru/doctor/library/immunology/Lolor/).*

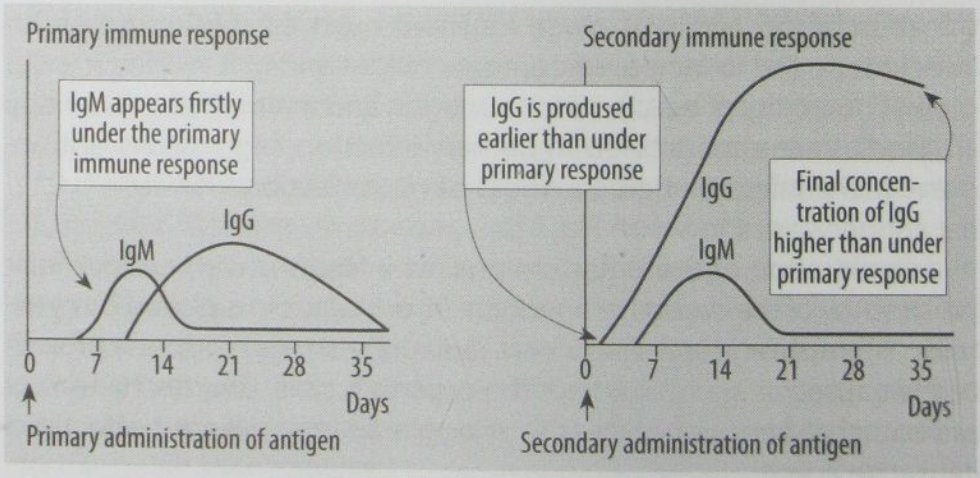

Fig. 3.3. Formation of antibodies during the primary and secondary immune response. *(Sourse: http://collegemicrob.narod.ru/immunology).*

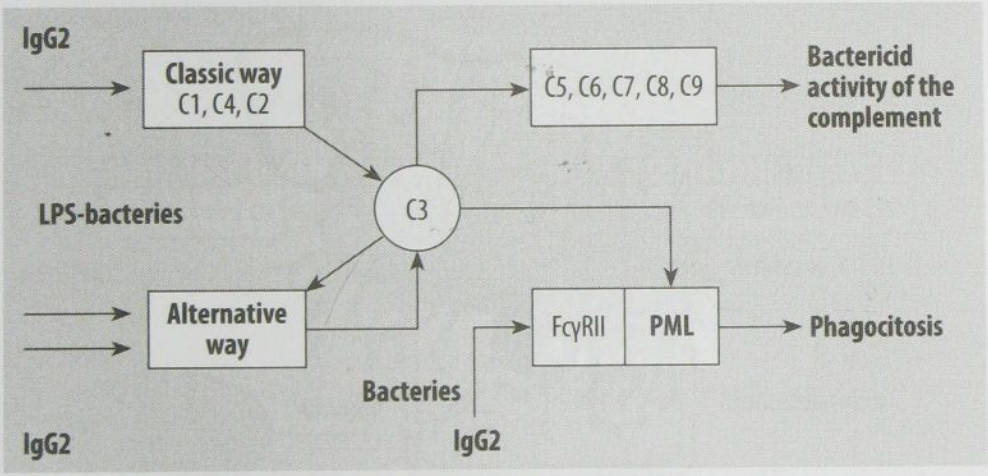

Fig. 3.4. Inactivation of incapsulated microorganisms with participation of IgG2. *(C1 – C9 are the components of the complement system; PMN – polymorphonuclear lymphocytes) (Sourse: http://il.ks.ua/base_files/arcive/IgG.htm).*

3.3. Antiviral immunity and protection against multicellular parasites

Antiviral immunity

It is provided by a number of mechanisms of neutralization of viruses by antibodies, phagocytosis of the viral particles, suppression of multipcation of a virus in an organism, etc. (Fig. 3.6). Of great value in processes of the antiviral immune response is antiviral antibodies, mainly the antibodies to superficial antigens of **viriones** (Fig. 3.7) or membranes of the cell infected by a virus, which block distribution of viral particles to an organism. Adsorption of antibodies on viriones interferes with their attachment to a cell of the host's organism and penetration into it. It occurs on the mucous membranes by the secretory IgA, IgM – in the blood, and IgG – in the extracellular fluid.

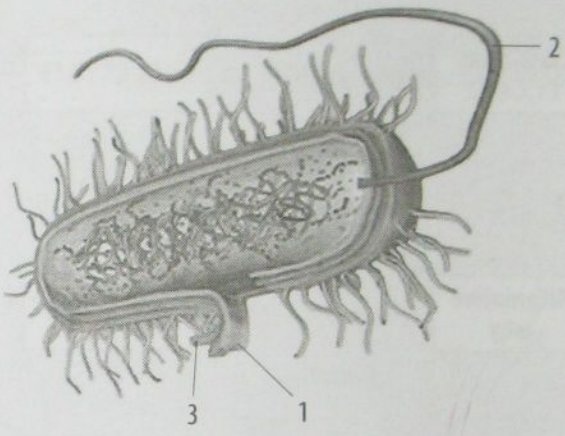

Fig. 3.5. Basic antigens of bacteria: *O-antigen (3 – cellular wall); H-antigen (2 – flagellum); K-antigen (1 – capsule). (Source: http://www.collegemicrob.narod.ru/immunology/antigen.html).*

Viral infections cause formation of ***interferon*** in the lymphoid cells inhibiting multiplication of a virus. Usually interferon starts to be produced intensively on the 3rd–4th day after development of the viral disease. The important factor of intensification of its formation is rise in the body temperature above 38° C.

Complement-mediated lysis and complement-dependent intensification of phagocytosis of viral agents are also important mechanisms of the antiviral immune response.

Activated macrophages and T-killers destroy the cells of an organism infected by viruses, interfering with further multiplication of a virus.

Phagocytes are also capable of destroying viral agents, however, according to modern notions, phagocytosis is not the leading mechanism of protection of an organism from viral infections.

Lately researches have been actively made on studying the nature and mechanisms of functioning of the endocellular factors of suppression of virus multiplication of the infected cell.

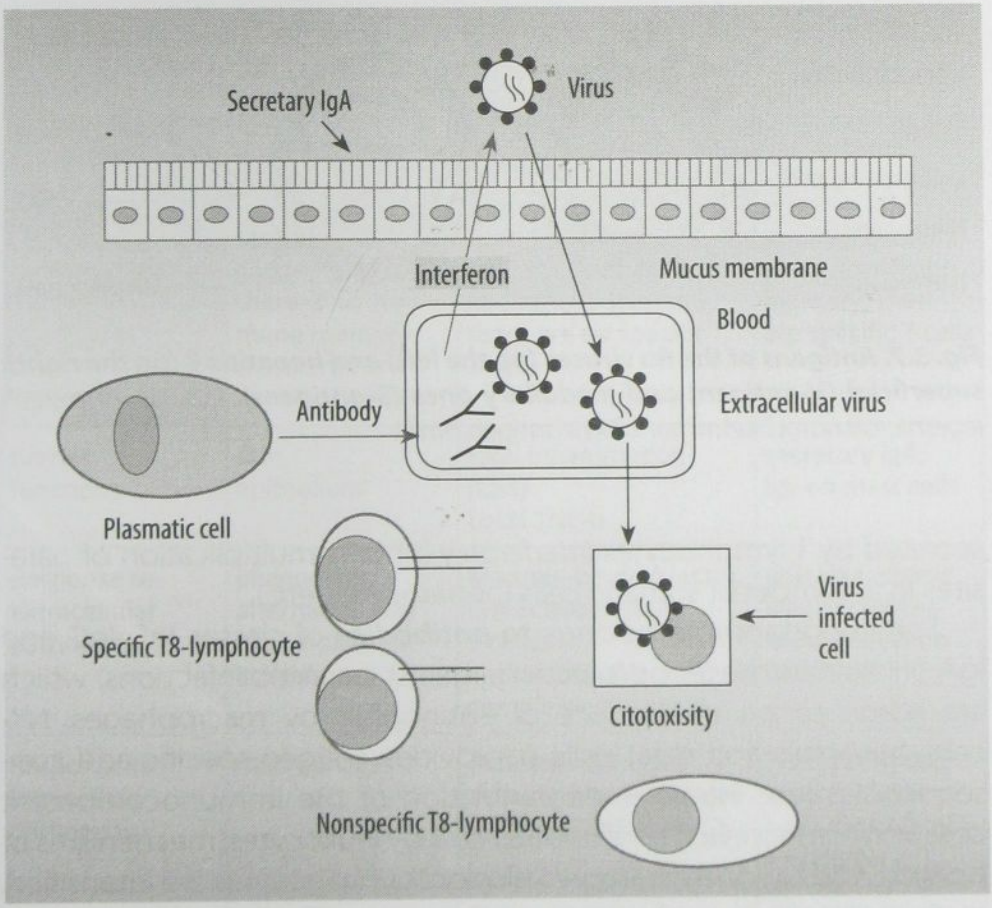

Fig. 3.6. Scheme of antiviral immune response. *(Source: http://nsau.edu.ru/images/vetfac/images/ebooks/microbiology/index.htm).*

Protection against multicellular parasites

It is provided by ***eosinophils,*** dischargng MBP (main basic protein), and ***neutrophils,*** discharging defensins in immediate proximity from parasites (*extracellular cytolysis*). Lysis of the parasitic cells and their opsonization is carry out ***by complement components*** (C3b). ***Macrophages*** destroy fine and endocellular parasites by phagocytosis

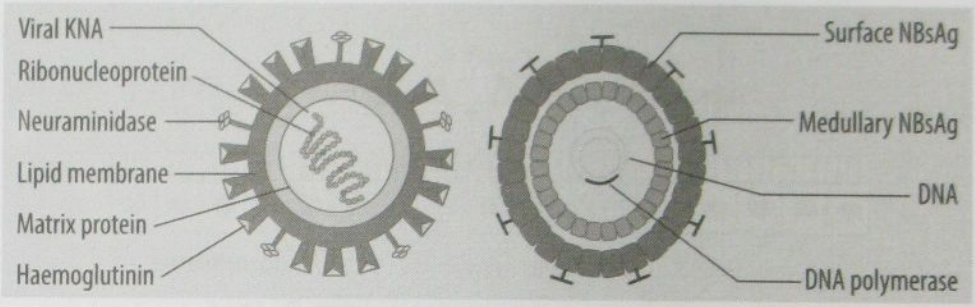

Fig. 3.7. Antigens of the flu viruses (on the left) and hepatitis B (on the right): superficial (V-antigen) and medullary ones (S-antigens). *(Sourse: http://collegemicrob.narod.ru/immunology/antigen.html).*

secreted by T-lymphocytes *interferon γ* inhibits multiplication of parasites in a number of somatic cells (hepatocytes, etc.).

The important role belongs ***to antibodies*** of classes IgE, IgG and IgA (in helmintoses) in protection against parasitic infections, which are adsorbed on the surface of eosinophils by macrophages, NK-cells, basophils and mast cells. It provides antigen-specific and consequently more intensive degranulation of the immunocompetent cells and *contact cytolysis of affected cells* by leukocytes; mechanisms of phagocytosis and "respiratory explosion" of phagocytes are intensified, there is an activation of complement by a classical way on the surface of the parasite and neutralization of the toxins produced by helminths.

Macrophages, neutrophils and NK-cells are capable of contact cytolysis of the cell affected by the parasite. Cytotoxic T-lymphocytes are capable of destroying the cells infected by endocellular parasites.

3.4. AIDS/HIV

AIDS (acquired immune deficiency syndrome) is a final stage of HIV disease, which causes severe damage to the immune system. Human immunodeficiency virus (HIV) causes AIDS. The virus attacks the

Phases of the immune response

(Charles A. Janeway et al., Immunology, 1999, as amended)

Immune response	Immediate (0–4 hours)	Early (4–96 hours)	Late (after 96 hours)
	nonspecific, congenital; there is no immune memory; there are no specific T-cells	nonspecific and specific; induced; there is no immune memory; there are no specific T-cells	specific; induced; there is immune memory; there are specific T-cells
Barrier functions	skin, epithelium	local inflammation (C5a) Local TNF-α	secretory IgA; IgE on mast cells
Response to extracellular pathogens	phagocytes; alternative way of complement activation	Mannan-binding lectin; C-reactive protein; T-cells; antibodies of B-cells; complement system	IgG; IgM; classic way of complement activation
Response to intracellular pathogens	macrophages	Activated NK-cells; activated macrophages IL-1, IL-6, TNF-α, IL-12	T-cells acivating macrophages with the help of Interferon γ
Response to cells infected by a virus	NK-cells	TNF-α, TNF-β; NK-cells activated by IL-12	Cytotoxic T-cells; interferon γ

immune system and leaves the body vulnerable to a variety of life-threatening infections and cancers. Common bacteria, yeast, parasites, and viruses that usually do not cause serious disease in people with healthy immune system can cause fatal illnesses in people with AIDS.

HIV has been found in saliva, tears, nervous system tissue and spinal fluid, blood, semen, vaginal fluid and breast milk. Blood, semen, vaginal secretions and breast milk have been shown to transmit infection to others. The virus can be transmitted:

- Through sexual contact – including oral, vaginal, and anal sex.
- Through the blood – via the blood transfusions or needle sharing.

From mother to a child – a pregnant woman can transmit the virus to her fetus through their shared the blood circulation, or a nursing mother can transmit it to her baby in her breast milk.

People who are infected with HIV may have no symptoms for 10 years or longer, but they can still transmit the infection to others during this symptom-free period. If the infection is not detected and treated, the immune system gradually weakens and AIDS develops. Acute HIV infection progresses over time (usually a few weeks to months) to asymptomatic HIV infection and then to early symptomatic HIV infection. Later, it progresses to AIDS (advanced HIV infection with CD4 T-cell count below 200 cells/mm^3).People with AIDS have had their immune system damaged by HIV and are very susceptible to these opportunistic infections. Common symptoms are: chills, fever, rash, sweats (particularly at night), swollen lymph glands, weakness, weight loss.

Infection with HIV may produce no symptoms. Some people experience flu-like symptoms with fever, rash, sore throat, and swollen lymph nodes, usually 2–4 weeks after contracting the virus. This is called the acute retroviral syndrome. Some people with HIV infection stay symptom-free for years between the time when they are exposed to the virus and when they develop AIDS.

According to the Center for Disease Control and Prevention, persons may be diagnosed with AIDS if they are HIV-positive and have a CD4 cell count below 200 cells/mm^3, even if they don't have an opportunistic infection. AIDS may also be diagnosed if a person develops one of the opportunistic infections and cancers that occur more commonly in people with HIV infection. These infections are unusual in people with a healthy immune system. The following illnesses are common with a CD4 count below 350 cells/mm^3: herpes simplex virus, herpes zoster, Kaposi's sarcoma, non-Hodgkin's lymphoma, tuberculosis.

Common with CD4 count below 200 cells/mm^3: bacillary angiomatosis, candida esophagitis, Pneumocystis carinii pneumonia, caused by a fungus.

Common with CD4 count below 100 cells/mm^3: AIDS dementia, Cryptococcal meningitis, Cryptosporidium diarrhea, Progressive multifocal leukoencephalopathy, Toxoplasma encephalitis, wasting syndrome.

Common with CD4 count below 50 cells/mm^3: cytomegalovirus infection, mycobacteriumavium.

Treatment: There is no cure for AIDS at this time. Treatment consists of highly active antiretroviral therapy (HAART) which slows progression of the disease. Treatment also includes preventive and active treatment of opportunistic infections. Antiretroviral therapy suppresses the replication of the HIV virus in the body. A combination of several antiretroviral drugs, called highly active antiretroviral therapy (HAART), has been very effective in reducing the number of HIV particles in the bloodstream. Treatment is typically a non-nucleoside reverse transcriptase inhibitor (NNRTI) plus two nucleoside analogue reverse transcriptase inhibitors (NRTIs). Typical NRTIs include: zidovudine (AZT) or tenofovir (TDF) and lamivudine (3TC) or emtricitabine (FTC). Combinations of agents which include a protease inhibitors (PI) are used if the above regimen loses effectiveness.

HAART is not a cure for HIV, but it has been very effective for the past 12 years. People on HAART with suppressed levels of HIV can still transmit the virus to others through sex or by sharing needles. There is good evidence that if the levels of HIV remain suppressed and the CD4 count remains high (above 200 cells/mm^3), life can be significantly prolonged and improved.

Any doctor prescribing HAART should carefully follow up the patient for possible side effects. In addition, blood tests measuring CD4 counts and HIV viral load should be taken every 3 months. The goal is to get the CD4 count as close to normal as possible, and to suppress the amount of HIV virus in the blood to a level where it cannot be detected.

Other antiviral medications are being investigated. In addition, growth factors that stimulate cell growth, such as erythropoetin (Epogen, Procrit, and Recomon) and filgrastim (G-CSF or Neupogen) are sometimes used to treat AIDS-associated anemia and low white blood cell counts.

There is no cure for AIDS. It is always fatal without treatment. Most patients survive many years after the diagnosis because of the availability of HAART. HAART has increased the amount of people with HIV remain alive.

3.5. Criteria of the laboratory estimation of the course of an infectious inflammatory process

As antiinfectious protection of an organism is predominantly provided by the immune system, dynamics of its indices in the infectious inflammatory process is a major criterion of the patient's condition, reflects efficacy of the given therapy and prognosis of the disease. The most essential indices of infectious inflammation are change of the cells contents in the blood, shift of the nuclear formula of neutrophils (leucocytic formulas) and acceleration of the erythrocyte sedimentation rate. Index estimation (a ratio of cells of certain types)is used for more profound characteristic of an organism's condition, course and prognosis of the disease.

Changing the content of immunocompetent cells in the blood

Leucocytosis. It is an early sign of the inflammatory disease. Increased contents of leukocytes often precede development of fever by 12–24 hours. Expressiveness of leucocytosis depends on pathogenicity of the causative agent, severity of the inflammatory process and reactivity of an organism. The normal contents of leukocytes or leucopenia in infectious diseases, which are usually accompanied by leucocytosis, are infavorable signs, especially leucopenia with a nuclear shift to the left in the period of height of the infectious disease. Leucocytosis in infections, which are usually accompanied by leucopenia is evidence of development of complications. ***Leucopenia:*** the flu, measles, rubella, brucellosis, typhoid fever, aggravation of viral hepatitis.

Neutrophilia: infections caused by bacteria, fungi, protozoa, rickettsia, some viruses, spirochetes. Neutrophilia, especially in the nuclear shift to the left, against the background of the normal contents of leukocytes or leucopenia is a sign of the severe process. ***Neutropenia:*** the flu, chicken pox, measles, rubella, a virus of hepatitis B, typhoid fever, malaria, paratyphoid, tularemia, brucellosis, tuberculosis. Neutropenia is often observed in the severe course of the inflammatory process.

Eosinophilia: parasitic, helminthic and protozoan invasions (labliosis, echinococcosis, ascaridosis, trichinosis, strongyloidiasis, opisthorchiasis, toxocariasis, etc); the acute period of some infectious diseases (scarlet fever, chicken pox, tuberculosis, infectious mononucleosis, gonorrhoea). ***Eosinopenia:*** initial phase of the inflammatory process; severe purulent infections. Eosinopenia in a combination with leucopenia is evidence of low resistance of an organism. Eosinopenia in the postoperative period is characteristic of patients in a severe condition.

Basophilia: chicken pox (sometimes).

Lymphocytosis: infectious mononucleosis, viral hepatitis, cytomegaloviral infection, whooping cough, respiratory viral infections, toxoplasmosis, herpes, rubella, HIV-infection. ***Lymphopenia:*** the acute period of infectious inflammation; milliar tuberculosis; AIDS. Lymphopenia in tuberculosis is considered to be an unfavorable sign.

Monocytosis: infections (of viral, fungal, protozoan and rickettsia etiology) as well as the period of convalescence after acute infections; tuberculosis; syphilis; brucellosis. Monocytosis in tuberculosis is evidence of active distribution of the process. For estimation of the tuberculosis course a ratio monocytes/lymphocytes is used (0.3–1.0 in the norm), which can exceed 1.0 in the active phase of the disease, and decreases during recovery. ***Monocytopenia:*** pyogenic infections.

Nuclear shift of the leucocytic formula is a change of the normal percentage ratio of leukocytes of neutrophil series: segmented, stab; metamyelocytes (young); myelocytes; promyelocytes; myeloblasts. Depending on the direction of the shift there are shifts of the leucocytic formula to the left (in increase in the number of young and immature

forms of neutrophils) and to the right (basically in the increase due to the amount of segmented neutrophils).

Shift of the leucocytic formula to the left is marked in acute infectious diseases and inflammatory processes. However, it is also characteristic of acidosis and comas; intoxications; physical overstrain; initial stage of chronic myeloleucosis; metastases of malignant neoplasms; myeloproliferative diseases (chronic myeloleucosis, erythremia, myelofibrosis; acute leucosis).

There are ***regenerative and degenerate types*** of the shift of the leucocytic formula to the left.

Regenerative shift is characterized by the increase in the amount of stab and young neutrophils against the background of leucocytosis that is usually observed in infectious and inflammatory diseases. There are also ***leucemoid reactions*** (apppearance of myelocytes, promyelocytes and sometimes myeloblasts in the blood except for metamyelocytes, as a rule, it is against the background of sharply expressed leucocytosis). Leucemoid reactions can be observed in severe course of the infectious process (sepsis, peritonitis, bacterial pneumonia, etc.) against the background of a high level of resistance of an organism; tuberculosis; exhaustion of the myeloid germ of the bone marrow (after long expressed leucocytosis); acute hemolysis; malignant tumors, especially with metastases in the bone marrow.

Degenerate shift is characterized by increased (a various degree of expressiveness) contents of stab neutrophils, metamyelocytes and myelocytes usually in reduction of the number of segmented neutrophils against the background of signs of the degenerate changes of cytolemma, cytoplasms and nucleus of the cells that is evidence of suppression of hemopoiesis. It is marked in severe infectious diseases and purulent-septic processes with expressed intoxication. It can be accompanied by leucocytosis (salmonellosis; dysentery; peritonitis; uraemic and diabetic coma) or leucopenia (viral infections; typhoparatyphoid diseases).

Shift of the leucocytic formuls to the right (when mature forms with 5-6 segments, instead of three prevail among neutrophils, the

shift index is less than 0.04) in infectious diseases, as a rule, is marked in favorable course of the disease. At the same time, it can be marked in healthy people (about 20 %) as well as in megaloblast anemias; diseases of the kidneys and liver; condition after the blood transfusion; radiation sickness; polycetemia; B12-deficient anemias.

Nonspecific index of the inflammatory process is **erythrocyte sedimentation rate (ESR)** – the index of division rate of the blood into layers for 1 hour: the upper transparent (plasma of the blood) is estimated in mm; the lower (sedimentary erythrocytes and other blood cells). The value of ESR depends on an electrostatic charge of erythrocytes, which decreases because of adsorption of proteins in the acute phase of inflammation on their surfaces (C-reactive protein, gaptoglobin, alpha-1-antitripsin, etc.), leading to agglutination of erythrocytes and accelerated sedimentation. ***Accelerated ESR:*** inflammatory and infectious diseases; intoxications; traumas; a condition after surgical interventions; anemias; malignant neoplasms; diseases of the kidneys (a nephrotic syndrome); intake of medicines (glucocorticoids, estrogen); pregnancy, menstruation, the postnatal period. ***Decelerated ESR:*** sickle – cell anemia, spherocytosis; erythremia; symptomatic erythrocytoses. Acceleration of ESR in the acute inflammatory and infectious diseases is registered later than leucocytosis (more often in a day).

Index estimation of the organism condition and course of the infectious inflammatory process

Load index (LI) (Lebedev K. A. et al., 1987) (index of loading tests of rosette-formation – *ratio of* ***E-POL\E-POM***) reflects intensity of functioning of the immune system (it is especially indicated in examination for revealing a chronic process in the stage of remission of the disease). The degree of reduction in the acute inflammatory process depends on severity of the inflammatory process and resistance of an organism (IL can go up↑ at a stage of the developed clinical manifestations in very severe cases in development of decompensation in the work of the immune system). It is frequently < 2 in patients with chronic diseases in the stage

of clinical remission. In the period of aggravation of the chronic disease in good resistance of an organism it remains at the same level; IL goes up↑ at the lowered efficacy of the immune system. Its values in healthy people: children of younger age – 70 % (1.6–2.3); 90 % (1.5–2.5); adults of middle age – 70 % (2.0); 90 % (1.9–3.0); 95 % (1.7–3.5 %); 95 % (1.4–2.9); over 70 – 70 % (2.0–3.2); 90 % (2.1–3.5); 95 % (2.2–4.0).

Index of the shift of neutrophils (ISN) (G. D. Dashtajants, 1982) (relation of all nonsegmented forms of neutrophils to segmented) – a criterion of the course severity of the acute infectious disease and prognosis of its outcome. It is calculated by the formula:

Index of the shift of neutrophils = (M + Y + SN) / S, in which M – amount of myelocytes, Y – amount of young neutrophils, SN – amount of stab neutrophils, S – amount of segmented neutrophils.

It makes 0.05–0.08 in the norm. A degree of severity of the disease is determined by the index of the shift: a severe degree – the index is from 1.0 and higher; moderate degree – the index is 0.3–1.0; mild degree – the index is not more than 0.3.

The leucocytic index of intoxication (LII) reflects acuteness of inflammation in an organism and response to endogenic intoxication, and is calculated by the formula of Ya. Ya. Kalf-Kalif (1941):

$$\text{LII} = \frac{(4 \times \text{myelocytes} + 3 \times \text{juvenile} + 2 \times \text{stab nuclear} + \text{segmentonuclear})(\text{plasmacytes} + 1)}{(\text{monocytes} + \text{lymphocytes}) \times (\text{eosinophils} + 1)}$$

Normal values of LII is 0.5–1.5. In viral infection LII decreases at the expense of lymphocytosis, and increases in the inflammatory processes of other etiology. The increase of LII takes place at the expense of reduction of lymphocytes and eosinophils and increase in the number of segmented neutrophils and plasmatic cells. The increase of LII of up to 4–9 is evidence of significant endogenic intoxications. High leucocytosis and increased LII of 10–20 and higher is a sign of septic shock. Leucopenia with high LII is a bad prognostic sign. In emergency conditions the index can be used on the 1st–2nd day of the disease.

Dynamics of the immunogram in normal development of the inflammatory process (Lebedev K. A., Ponjakina I. D., 2002, reduced):

The incubation period: often ↓percentage of T-lymphocytes

The prodromal period: ↓ of the relative contents of eosinophils; ↓ absolute and relative quantity of basophils; ↓ of the relative amount of T-lymphocytes; ↑ contents of 0-cells; essential ↓ of the index of loading.

Initial manifestations of the clinical picture of the disease: leucocytosis; relative neutrophilia; shift of the nuclear formula of neutrophils to the left; further ↓ of the number of eosinophils and T-lymphocytes and ↑ of 0-cells; some increase of the relative contents of T-helpers at the expense of ↓ of T-cytotoxic lymphocytes (correlates with severity of the process); ↑ of the phagocytic activity of neutrophils; the lowest values of the index of loading.

The developed clinical picture of the disease: leucocytosis; ↑ of the amounts of monocytes (in the middle of the stage or on the 5^{th}–7^{th} day from the beginning of the clinical manifestations); closer to the end of the stage the relative amount of neutrophils is normalized (or ↓), ↑ of the relative contents of lymphocytes; shift of the nuclear formula of neutrophils persists (or ↑); at the beginning or middle of the stage ↑ of ESR); ↑ of the phagocytic activity of neutrophils (a major criterion of the favorably developing inflammatory process).

Crisis of the disease with the subsequent regress of the clinical manifestations: normalization of the relative contents of eosinophils (an early favorable sign); ↑ of the relative amount of B-lymphocytes; ↑ of the number of T-cytotoxic lymphocytes in relation to T-helpers; normalization of T-lymphocytes and 0-cells; ↓ of the amount of leukocytes; shift of the nuclear formula of neutrophils is normalized; accelerated ESR persists; the index of loading begins ↑.

Convalescence: decreased amount of T-lymphocytes and increased contents of 0-cells (a criterion of uncompleteness of the process); the increased level of B-lymphocytes; ↓ of ESR; a constant tendency to ↑ of the index of loading.

3.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Factors discharged by pathogenic microorganisms in the host's organism: kinds and characteristic of their influence on functioning of a macroorganism.
2. Stages of pathogenesis of the infectious process. Factors of the antibacterial immune protection of an organism.
3. Antiviral immune response.
4. Mechanisms of protection of the organism from multicellular parasites.
5. Basic laboratory indices describing a course of the infectious inflammatory process.
6. Dynamics of the immunogram in the normal course of the inflammatory process.
7. Types of the nuclear shift of the leucocyte formula to the left: the characteristics and causes.

Practical skills

1. Interpretation of change of the granulocyte and agranulocyte contents in the infectious inflammatory process. Differential diagnostics with other pathological conditions.
2. The estimation of expressiveness of the nuclear shift of the leucocyte formula in the infectious inflammatory process. Differential diagnostics with other pathological conditions.
3. The estimation of erythrocyte sedimentation rate in the infectious inflammatory process.
4. The index estimation of the organism's condition and course of the infectious disease: kinds of the basic indices, their calculation and interpretation of changes.

Tests

1. Adhesins discharged by pathogenic microorganisms provide:
   A. Penetration through protective barriers of a macroorganism
   B. Adhesion of erythrocytes
   C. Inhibition of production of specific antibodies
   D. Attachment of pathogens to cells of the microorganism tissues
   E. Inhibition of the protective immune responses

2. Specific immune protection against pathogenic microorganisms is provided by:
   A. System of complement
   B. Macrophages
   C. Natural killers
   D. Antibodies of classes E and A
   E. Antibodies of classes of M and G

3. ... destroy cells of the organism infected by a virus, interfering with its further multiplication:
   A. B-lymphocytes
   B. Tissue macrophages
   C. T-killers
   D. Components of the complements system
   E. T-helpers

4. Protection of the organism from parasitic infections is mainly provided by:
   A. Basophils
   B. Eosinophils
   C. T-helpers of the second type
   D. Lymphocytes
   E. Monocytes

5. Presence of leucocytosis is usually characteristic of:
   A. Measles
   B. Rubella
   C. Meningococcal infection
   D. Brucellosis
   E. Typhoid fever

6. Presence of leucopenia in the blood count in a combination with eosinopenia is a sign of:
   A. Height of the disease course
   B. Beginning of convalescence
   C. No significant value
   D. High resistance of the organism to a pathogen
   E. Low resistance of the organism to a pathogen

7. Neutrophilia with the expressed nuclear shift to the left against the background of leucopenia in the infectious inflammatory process is a sign of:
   A. Fast recovery
   B. Developments of complications
   C. Adequacy of the given therapy
   D. Severe course of the disease
   E. Favorable prognosis

8. Patients with whooping cough are characterized by presence of:
   A. Eosinophilia
   B. Lymphocytosis
   C. Basophslia
   D. Monocytosis
   E. Neutrophilia

9. What index of the blood count in the patient with presence of bacterial inflammatory process changes after others during the disease course?

A. Quantity of leukocytes
B. Quantity of neutrophils
C. Quantity of lymphocytes
D. Quantity of stab neutrophils
E. Erythrocyte sedimentation rate

10. Normalization of contents of what cells in the patient with active inflammatory process is an early favorable sign:
A. Basophils
B. Eosinophils
C. Neutrophils
D. Lymphocytes
E. Monocytes

CHAPTER IV

PRIMARY IMMUNODEFICIENCY

4.1. Definition of immunodeficiency

Immunodeficiency is a disturbance of the structure and function of any component of the integral immune system, loss of the ability to resist any infections by the organism and to restore the disturbances of its organs. Besides, in immunodeficiency the process of renovation of the organism is slowed down or generally stops.

Immunodeficiency is a congenital or acquired defect of the immune system, which is manifested by sharp reduction in the quantity of separate populations of immunocompetent cells or by disturbance of the synthesis of immunoglobulins (agammaglobulinemia).

All immunodeficient states are divided into two large groups:

- congenital (hereditary) immunodeficiency and
- acquired immunodeficiency

Hereditary immunodeficient state (primary immunological deficiency) is based on the genetically determined defects of the cells of the immune system.

4.2. Classification of primary immunodeficiencies

There are many immunodeficiencies caused by different causes. At present more than 70 congenital (primary) immunnodeficiencies are identified. Usually these are severe diseases in children, caused by defects of any component of the immune system (Fig. 4.1). Defects may affect different immunocompetent cells, including T- and B-lymphocytes and macrophages. Di George syndrome, which is accompanied by the underdevelopment of the thymus, may be an example of the

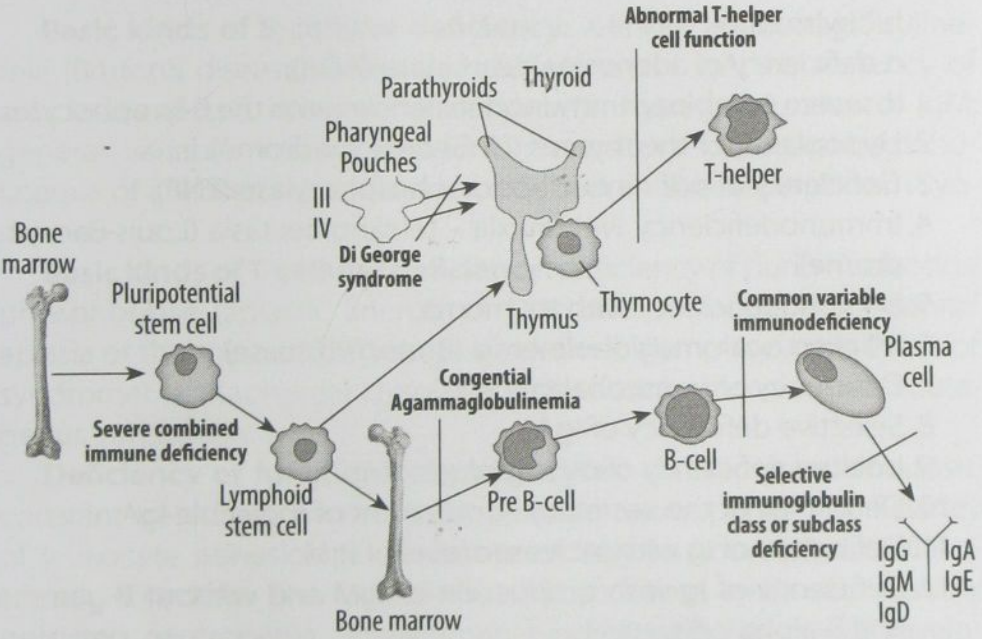

Fig. 4.1. Sites of developmental dysfunction in humoral and cell-mediated immunity immunodeficiencies. *(Klaus D. Elgert, Immunology, 1996).*

predominant affection of the T-cellular component of immunity. The defects of T-cells increase sensitivity of the organism to various microorganisms (from yeast(s) to the viruses), which are harmless in the normal conditions.

Impairments in the macrophages also lead to severe pathologies, for example, to chronic granulematosis. There are pathologies associated with the production of antibodies by B-lymphocytes. In this case the organism susceptibility to the repeated infections, caused by pyogenic bacteria, grows. The congenital immunodeficiencies are encountered sufficiently rarely (on the average 1 case per 25.000–100.000 people).

Classification of forms of primary specific immunological deficiencies (WHO, 1980):

1. Severe combined immunodeficiencies:
   a. reticular disgenesia;

b. "Swiss type";
c. deficiency of adenosine deaminase (ADA);
d. severe combined immunodeficiencies with the B-lymphocytes.

2. Hypoplasias of the thymus (Di George syndrome).
3. Deficiency of purine nucleoside phosphorylase (PNP).
4. Immunodeficiency with ataxia – teleangioectasia (Louis-Bar syndrome).
5. Immunodeficiency with thymoma.
6. X-linked agammaglobulinemia (Bruton's disease).
7. Deficiency of transcobalamine II.
8. Selective deficiency of IgA.
9. Isolated deficiency of other classes of Ig.
10. Deficiency of the secretory component of molecule IgA.
11. Deficiency of Ig with increased level of IgM.
12. Deficiency of Ig with production of IgM and without B-gamma and B-alpha cells.
13. Transitory hypogammaglobulinemia of infancy.
14. Deficiency of antibodies with the normal or increased level of immunoglobulines in the blood.
15. Deficiency of kappa-chains.
16. Wiskott – Aldrich syndrome.
17. Variable forms of immunological deficiency (general and nonclassified):
    a. predominant deficiency of Ig;
    b. predominant deficiency of T-cells.

Primary immunodeficient states are divided into 5 groups:

1. Deficiency of the humoral B-cellular component of immunity.
2. Deficiency of T-cellular component.
3. Deficiency of the phagocyte functions: polynuclears and monocyte-macrophages.
4. Deficiency of the complement factors.
5. Combined immunodeficient states, which include deficiency of several components of the immunological reactivity and stem cells.

Basic kinds of B-cellular deficiency: X-linked agammaglobulinemia (Bruton's disease); selective insufficiency of IgA; insufficiency of subclasses of IgG, immunodeficiency with hyperproduction of IgM; general variable immunodeficiency; transitory hypogammaglobulinemia of children's age; disease of Gedeon – Sheideger; Bloom syndrome.

Basic kinds of T-cellular deficiency: deficiency of purin nucleoside phosphorilase (aplastic anemia of Diamond – Blackfan); congenital aplasia of the thymus (Di George syndrome); alymphocytosis (Nezelof syndrome); metaphyseal dysplasia of McKusick; chronic muco-cutaneous candidiasis.

Deficiency of functions of phagocytes: chronic granulomatosis; constant hereditary neutropenia; periodic neutropenia; insufficiency of leukocyte adhesion (a syndrome of "lazy leukocytes"); Chediak – Higashi (Chediak – Steinbrink – Higashi) syndrome; cyclic neutropenia; persisted neutropenia; Shwachman syndrome; deficiency of myeloperoxidase; a syndrome of hypergammaimmunoglobulinemia E (Job syndrome).

Deficiency of complement components is clinically manifested in the form of night hemoglobinuria, angioedema (congenital angioneuroitic edema) – vascular edema (deficiency of C1-inhibitor), high morbidity with systemic lupus erythematosus (deficiency of components C1g, C1r, C1s, C4, C2), lowered resistance to neisseria (deficiency of components C5, C6, C7, C8, factor D and properdin), to activators of purulent infectious diseases (deficiency of component C3, factor B or I).

The basic kinds of combined immunodeficiencies: severe combined immunodeficiency (reticular dysgenesia; lymphocytophtisis ("Swiss type"); deficiency of adenosine deminase; severe combined immunodeficiencies with B-lymphocytes); Wiskott – Aldrich syndrome; immunodeficiency with thymoma (Goodpasture syndrome); ataxia and teleangioectasia (Louis-Bar syndrome).

Human immunodeficiency syndromes

(Charles A. Janeway et al., Immunobiology, 1999)

Deficiency syndrome	Specific abnormality	Immune defect	Susceptibility
Severe combined immunedeficiency	ADA deficiency	No T- or B-cells	General
	PNP deficiency	No T- or B-cells	General
	X-linked scid, γc chain deficiency	No T-cells	General
	Autosomal scid DNA repair defect	No T- or B-cells	General
Di George syndrome	Thymic aplasia	Variable numbers of T- and B-cells	General
MHC class I deficiency	TAP mutations	No CD8 T-cells	Viruses
MHC class II deficiency	Lack of expression of MHC class II	No CD4 T-cells	General
Wiskott – Aldrich syndrome	X-linked; defective WASP gene	Defective polysaccharide antibody responses	Encapsulated extracellular bacteria
Common variable immunodeficiency	Unknown; MHC-linked	Defective antibody production	Extracellular bacteria
X-linked agammaglobulinemia	Loss of Btk (Bruton's tyrosine kinase	No B-cells	Extracellular bacteria, viruses
X-linked hyper-IgM syndrome	Defective CD40 ligand	No isotype switching	Extracellular bacteria
Selective IgA and/or IgG deficiency	Unknown; MHC-linked	No IgA synthesis	Respiratory infections
Phagocyte deficiencies	Many different ones	Loss of phagocyte function	Extracellular bacteria and fungi
Complement deficiencies	Many different ones	Loss of specific complement components	Extracellular bacteria especially Neisseria spp.

Deficiency syndrome	Specific abnormality	Immune defect	Susceptibility
Natural killer (NK) cell defect	Unknown	Loss of NK function	Herpes viruses
X-linked lympho-proliferative syn-drome	SAP mutant	Inability to control B-cell growth	EBV-driven B-cell tumors
Ataxia telangiectasia	Gene with PI-3 kinase homology	reduced number of T-cells	Respiratory infections
Bloom syndrome	Defective DNA helicase	reduced number of T-cells; reduced antibody levels	Respiratory infections

4.3. Clinical manifestations in the immunodeficient states

Suppositional dysfunction of the T-cellular system:

1. Systemic disease after the immunization by any living virus or vaccine BCG; the uncommon, life threatening complications after the infections caused by the common, not dangerous viruses (gigantocellular pneumonia in rubella, pneumonia in chicken-pox).
2. Chronic candidiasis of the oral cavity persisting after a child has reached the age of 6 months and not yielded to the action of adequate chemotherapeutic drugs.
3. Chronic candidiasis of the skin and mucous membranes.
4. Characteristic signs (soft thin hair, dwarfism due to shortening of the extremities, typical roentgenological changes) of the syndrome of hypoplasia of the hair and cartilage.
5. Intrauterine reaction of the transplant against the host – erythroderma and total baldness (absence of eyebrows).
6. Reaction of the transplant against the host after the blood transfusion.

7. Hypocalciemia of newborns (Di George syndrome, especially in combination with the characteristic anomalies of the face, pinnae, and heart).
8. Small size of (the diameter less than 10 μm) lymphocytes, their number constantly comprises less than 1500 in 1 μl; it is necessary to exclude their loss through the gastrointestinal tract and lymphatic system.

Suppositional dysfunction of the B-cellular system:

- relapsing bacterial pneumonia, sepsis, meningitis;
- main lymphoid hyperplasia.

Suppositional dysfunction of the B- and T-cellular systems: (combined immunodeficiency):

1. All enumerated manifestations except chronic candidiasis of the skin and mucous membranes as well as main lymphoid hyperplasia.
2. Signs of Wiskott – Aldrich syndrome (purulent otitis, thrombocytopenia and eczema).
3. Signs of ataxia and teleangioectasia (Louis-Bar syndrome).

Signs that indicate the state of immunodeficiency, but without clear evidence of T-or B-cell defect:

1. Pneumonia caused by *Pneumocystis carinii*.
2. Eczema, which is resistant to medication.
3. Ulcerous colitis in children under 1 year.
4. Diarrhea, which can not be corrected.
5. Inexplicable hematologic deficiency (deficiency of erythrocytes, leukocytes, thrombocytes).
6. Severe generalized seborrheic dermatitis (Leiner – Moussous disease) can be evidence of the C5 insufficiency; seborrhea frequently accompanies the combined immunodeficiency.
7. Relapsing purulent infections are observed in the C3 insufficiency.

Presumptive biochemical defect:

1. Signs of the combined immunodeficiency with characteristic damages of the skeletal system (insufficiency of adenosine deaminase).
2. Signs of the aplastic anemia of Diamond – Blackfan (insufficiency of nucleoside phosphorylase).

Supposed dysfunction of the polymorphonuclear leukocytes.

1. Skin infections (if they are combined with bronchial asthma, eczema, rough features of the face, it is possible to think about Buckley syndrome).
2. Chronic osteomyelitis, caused by Klebsiella or Serratia purulent lymphadenitis (chronic granulomatous disease).

4.4. Principles of treatment of primary immunodeficiencies

1. Replaceable therapy by preparations of thymic hormones (synthetic analogues) and antibodies.
2. Therapy of colony-stimulating factors (synthetic analogues) and myelopides.
3. Transplantation of the bone marrow and fetal thymus.
4. Treatment of concomitant infectious diseases.

4.5. Diagnosis and treatment of some primary immunodeficiencies

Selective deficiency of IgA. Selective deficiency of IgA is the most common immune deficiency disorder. Persons with this disorder have low or absent levels of the blood protein called immunoglobulin A. IgA deficiency is usually inherited; that means it is passed down through families. It may be inherited as an autosomal dominant

or autosomal recessive trait. It is found in approximately 1 in 500–700 individuals of the European origin. Symptoms include frequent episodes of: bronchitis; chronic diarrhea; conjunctivitis; mouth infection; otitis media; pneumonia; sinusitis; skin infections; bronchiectasis; unexplained asthma. *Diagnosis:* there may be a family history of IgA deficiency; detected IgA, IgG, IgG subclass measurements, IgM, quantitative immunoglobulins, serum immunoelectrophoresis. *Treatment:* No specific treatment is available. Some people gradually develop normal levels of IgA without treatment. Infections should be treated with antibiotics. Those with selective IgA deficiency who also have IgG subclass deficiencies can benefit from immunoglobulin (IVIG) treatments given through the vein. Drugs: ginseng; Echinacea; drugs of zinc; probiotics; vitamin regimen (A, B6, E).

Transient hypogammaglobulinemia of infancy. In healthy babies, antibody levels in the blood reach a natural low point when they are between three and four months of age. In children who have Transient hypogammaglobulinemia of infancy, the levels of IgG and IgA levels remain low after six months of age because not enough immunoglobulin is produced. The disorder is temporary. It usually resolves between the ages of two and four years old. In rare cases, the disorder can persist until the children reaches six year old. *Causes:* The cause for B-cell developmental abnormalities is unknown. There may be a hereditary component to Transient hypogammaglobulinemia of infancy, which causes problems with the conversion of B-cells to plasma cells. Common symptoms of Transient hypogammaglobulinemia of infancy include, recurrent ear infections or non-infectious inflammation of the middle ear, recurrent bronchitis, frequent sinusitis (infection or inflammation of the sinuses), bacterial infection (pneumonia) and infections of the skin or meningitis (infection of the membranes that cover the spinal cord and brain). *Diagnosis:* Transient hypogammaglobulinemia of infancy is usually suspected if a child experiences recurrent infections over the age of six months. A blood test can indicate low levels of antibodies in the bloodstream. If the antibody levels are less than what is considered normal for children of the same age, the child may

have this disease. However, low levels of antibodies are nonspecific because it can be the result of any other immunodeficiency disorder. *Treatment:* Transient hypogammaglobulinemia of infancy will resolve on its own, without treatment. Antibiotics. Intravenous immunoglobulin therapy is debated. A conjugated heptavalent pneumococcal vaccine is recommended for routine immunization in children who are two months old. Probiotics. Hydrotherapy. Propolis.

Di George syndrome is an inherited condition that lies at the more severe end of a spectrum of syndromes (also known as CATCH22 or 22q11.2 deletion syndrome) that occurs when a part of the DNA on chromosome 22 is missing. The parathyroid glands in the neck may have failed to develop, leading to low levels of calcium in the blood. This can result in muscle spasms (tetany) and seizures. The thymus gland may also be underdeveloped or absent, resulting in a deficiency of an important type of the immune cell known as T-lymphocyte. Infections (seldom life-threatening) and autoimmune diseases (such as haemolytic anaemia, inflammatory bowel disease and juvenile rheumatoid arthritis) are common. There are often heart defects, particularly affecting large vessels that come out of the heart. There may be a typical facial appearance with features such as a small jaw, small, low-set ears with abnormal folds, unusual eyes, small mouth, a rather bulbous nose and square nasal tip, and hypernasal speech with cleft palate. Short stature and learning difficulties which may range from mild to moderate are also common. Sometimes the syndrome isn't detected until later in infancy, especially when problems are mild. The symptoms most frequently observed are: abnormality of the palate and the ears, arousing difficulties of speaking and understanding, or deficiency of the hearing (audition); cardiac deformation; disturbance of the immune systems; hypotension; risks of schizophrenia which begins in the childhood or the adolescence. Antenatal *diagnosis*, usually using CVS or amniocentesis, is possible especially when there's a family history or abnormalities on ultrasound scanning. Di George syndrome can't be cured, but treatment of problems such as low calcium, surgery for heart problems and thymus cell transplants to restore the immune

system can reduce complications. *Treatment* of Di George syndrome aims at correcting the abnormalities of organs or affected tissues. The treatment thus depends on the nature and on the severity of the abnormality. The treatment of hypocalcemia and hypoparathyroidin can require a treatment by the calcium and administration of hormone parathyroidin of replacement. An abnormality of the heart can require the administration of medicines to improve the cardiac function or surgical correction. The type of surgical operation depends on the nature of the cardiac defect. New techniques of transplant of the thymus also improve the long-term results. The future of a child with Di George syndrome depends on the level of gravity of the present abnormalities. The gravity of the cardiac disorder is generally the most determining factor. In case of severe deficiency, a correction treatment is necessary.

Louis-Bar syndrome is an autosomal-recessive disease with hypoplasia of the thymus. *Clinical course:* progressive cerebellum ataxia since early childhood; progressive telangiectasia of the conjunctiva and face area; peculiarity of the eye movements (of ophthalmoplegia type); gradual formation of visible dilatation of the peripheral vessels; recurrent infections of the bronchial-pulmonary system with formation of bronchoectases; delayed puberty; high risk of development of lymphomas. *Diagnosis:* genetic investigations; ultrasonic investigation of the thymus; determination of the indices of T-cellular immunity and level of IgA in the blood (it is absent or lowered in 70 %). *Treatment:* immunoglobulin (in presence of indications); exercise therapy; logopedics; vitamin therapy in high doses.

Bruton's disease. Mutations in the gene for BTK (located at Xq21.3–22) are responsible for the disease. X-linked agammaglobulinemia (XLA) is a genetic disorder in which the development of B-cells stops during differentiation. It has been shown to be caused by a variety of mutations in the gene encoding Bruton's tyrosine kinase (BTK, also known as BPK or ATK) – enzyme needed for maturation of B-cells. XLA is often characterized by recurrent bacterial infections due to a decrease in the number of B-cells and a subsequent reduction in the level of

serum immunoglobulin. Bruton's agammaglobulinemia is an X-linked genetic condition caused by an abnormality in a key enzyme needed for proper function of the immune system. People who have this disorder have low levels of protective antibodies and are vulnerable to repeated and potentially fatal infections. Bruton's agammaglobulinemia is inherited in an X-linked recessive manner; thus, almost all persons with the disorder are males. Females have two X chromosomes, which means they have two copies of the BTK gene, whereas males only have one X chromosome and one copy of the BTK gene. If a male has an altered BTK gene, he will have Bruton's agammaglobulinemia. If a female has one altered BTK gene, she will be a carrier and will be at risk to pass the altered gene on to her children. Symptoms: children with Bruton's agammaglobulinemia are born healthy and usually begin to show signs of infection in the first three to nine months of life, when antibodies that came from the mother during pregnancy and early breast-feeding disappear. Patients with Bruton's agammaglobulinemia can have infections that involve the skin, bone, brain, gastrointestinal tract, sinuses, eyes, ears, nose, airways to the lung, or lung itself. Besides signs of recurrent infections, other physical findings in patients with Bruton's agammaglobulinemia include slow growth, wheezing, small tonsils, and abnormal levels of tooth decay. Children may also develop unusual symptoms such as joint disease, destruction of red blood cells, kidney damage, and skin and muscle inflammation. *Diagnosis:* recurrent infections or infections that fail to respond completely or quickly to antibiotics; the presence of unusually small lymph nodes and tonsils; patients do not have periods of well-being between bouts of illness. Detect level of immunoglobulins. Make genetic testing of the BTK gene. *Treatment:* replacing immunoglobulin; antibiotics; bone marrow transplantation.

Chediak – Higashi syndrome is inherited by the autosomal-recessive type. Clinical course: albinism of the skin and hair; inexplicable attacks of fever; recurrent viral and bacterial infections with a high fever; photophobia. *Diagnosis:* genetic investigations; thrombocytopenia; neutropenia; lymphohisterocytic proliferation in the liver, spleen and

bone marrow. *Treatment:* transplantation of the bone marrow; antibiotic therapy of the infectious diseases.

Angioedema (congenital angioneurotic hypostasis) is inherited by autosomal-dominant type. *Clinical course:* edema of hypodermic tissues (of the face – but not of the periorbital or perioral parts; head, extremities, buttocks, anterior abdominal wall, genitals for 2–5 days); edema of the mucous membrane – organs of the abdominal cavity (abdominal pains) and larynx (duration of 2–3 days). Edemas are characterized by absence of inflammation signs. *Diagnosis:* genetic investigations; the family and individual anamnesis; reduction in contents of C1-inhibitor, sometimes C2- and C4-components. *Treatment:* introduction of the plasmic C1-inhibitor (recombinant C1-ING) and fresh frozen plasma; preparations of androgens (danasol, oximetalon); ε-aminocapronic acid.

Wiskott – Aldrich syndrome is caused by a defect (mutation) of a gene, which is located on the short shoulder of X chromosomes (Xp11.22 – p11.23) and codes the protein of the Wiskott – Aldrich syndrome – a component of lymphocytes and thrombocytes. Men fall ill with this syndrome more often. *Clinical course:* frequent viral and bacterial infections (otites, pneumonias, meningitis); eczema; thrombocytopenia; predisposition to bleedings; high risk of leukaemia or lymphoma. Sometimes anemia and splenomegaly is marked. Diagnosis: determination of the protein of Wiskott – Aldrich syndrome in the blood; thrombocytopenia; low levels of Ig in the blood. *Treatment:* preparations of immunoglobulins; transplantation of the bone marrow; sometimes splenectomy; administration of antibiotics in infections; transfusion of thrombocytes in bleedings.

Severe combined immunodeficiency *(reticular dysgenesia; "Swiss type"; deficiency of adenasine desaminase; severe combined immunodeficiencies with B-lymphocytes)* is genetically determined diseases, with serious cellular defects. The rate is of 1:20000–1:50000. Clinical course: diarrhea; recurrent candidiasis of the fauces and skin; recurrent, resistant to treatment pneumonias; exhaustion and retarda-

tion of physical development; lymph nodes are small (or absent); tonsils are not found; absence of the thymus. *Treatment:* transplantation of the thymus; preparations of immunoglobulins.

4.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Basic principles of classification of immunodeficiencies.
2. Congenital immunodeficiencies of B-cell link: the mechanisms of development, peculiarities of the clinical course, immunodiagnosis and treatment.
3. Congenital immunodeficiencies of T-cell link: the mechanisms of development, peculiarities of clinical course, immunodiagnosis and treatment.
4. Congenital combined immunodeficiencies and immunodeficiencies of B-, T-cellular links: the mechanisms of development, peculiarities of the clinical course, immunodiagnosis and treatment.
5. Congenital immunodeficiencies of the phagocytic link of the immune system: the mechanisms of development, peculiarities of the clinical course, immunodiagnosis and treatment.
6. Congenital immunodeficiencies of the complement system: the mechanisms of development, peculiarities of the clinical course, immunodiagnostis and treatment.

Practical skills

1. To carry out questioning and physical examination of patients with immunodeficiency states (to take immunological anamnesis, to determine hereditary tendency to development of immunodeficiencies).

2. To know how to fill in the immunological questionnaire of the patient, on the basis of what data to determine "the risk group" as to development of immunopathology.
3. To master the skills of determination of the necessary spectrum of immunological tests for investigation of patients with immunodependent pathology.
4. To reveal the presence of basic clinical symptoms and syndromes of immune disturbances.
5. To make a differential diagnosis, substantiate and make the diagnosis with the basic immunodeficiency syndromes on the basis of the data analysis of laboratory and instrumental investigation
6. To carry out clinical and immunological differential diagnostics of congenital and acquired immunodeficiencies.
7. Selective deficiency of IgA (clinical course, diagnosis, principles of treatment).
8. Transient hypogammaglobulinemia of infancy (clinical course, diagnosis, principles of treatment).
9. Wiskott – Aldrich syndrome (clinical course, diagnosis, principles of treatment).
10. Di George syndrome (clinical course, diagnosis, principles of treatment).
11. Louis-Bar syndrome (clinical course, diagnosis, principles of treatment).
12. Bruton's disease (clinical course, diagnosis, principles of treatment).

Tests

1. Alarming sign of primary immunodeficiency in children is inefficiency of the antibacterial therapy of the bacterial infections given during:
   A. 2 weeks
   B. 1 month
   C. 1.5 months

D. 2 months
E. 2.5 months

2. Alarming sign of primary immunodeficiency in children is the amount of acute otitis media for ... over a year:
A. More than 4 times
B. More than 5 times
C. More than 6 times
D. More than 7 times
E. More than 8 times

3. Alarming sign of primary immunodeficiency in children is having ... more than twice within a year:
A. Acute bronchitis
B. Acute obstructive bronchitis
C. Pneumonia
D. Laryngotracheitis
E. Tonsillitis

4. What disease is related to primary immunodeficiency with disturbance in a humoral link of the immune response?
A. Di George syndrome
B. Leukocyte adhesion deficiency
C. Wiskott – Aldrich syndrome
D. Common variable immune deficiency
E. Louis-Bar syndrome

5. What disease is related to primary immunodeficiency with disturbances in the T-cellular link of the immune response?
A. Common variable immune deficiency
B. Leukocyte adhesion deficiency
C. Di George syndrome
D. Wiskott – Aldrich syndrome
E. Louis-Bar syndrome

6. What disease is related to primary immunodeficiencies with combined disturbances?
   A. X-linked agammaglubulinemia
   B. Wiskott – Aldrich syndrome
   C. Common variable immune deficiency
   D. Di George syndrome
   E. Leukocyte adhesion deficiency

7. What disease is characterized by presence of hypocalcaemia of newborns and congenital anomalies of face, auricle and heart?
   A. Buckley syndrome
   B. Wiskott – Aldrich syndrome
   C. Bruton's disease
   D. Louis-Bar syndrome
   E. Di George syndrome

8. Alarming sign of primary immunodeficiency in adults is atypical course of:
   A. Endocrine diseases
   B. Allergic diseases
   C. Nephrologic diseases
   D. Autoimmune diseases
   E. Cardiologic diseases

9. Presence of what sign is not characteristic of immunodeficient conditions:
   A. Recurrent purulent infections
   B. Recurrent candidiasis
   C. Reduction of the amount of basophils in the blood
   D. Uncorrected diarrhea
   E. Reduction of the amount of lymphocytes in the blood

10. Presence recurrent bacterial infections, eczema and thrombocytopenia is characteristic of:
    A. Buckley syndrome
    E. Di George syndrome
    C. Bruton's disease
    D. Louis-Bar syndrome
    E. Wiskott – Aldrich syndrome

CHAPTER V

ACQUIRED (SECONDARY) IMMUNODEFICIENCY

5.1. Definition of the acquired (secondary) immunodeficiency

The acquired (secondary) immunodeficiency arises in the course of the patients' life and it is the result of influence of a number of chemical, radioactive, drug and other substances on the organism as well as influence of viral infections, chronic inflammatory processes, difficult surgery, injuries, stress.

The acquired immunodeficiencies are a group of diseases, the basis of which are disturbances either of separate components of immunity or the complex damage of this system under the effect of the factors of environment or pathologic processes, not associated in their etiology with the immune system, but exerting a suppressing effect on it.

The immunodeficient state may be caused by irradiation, glucocorticoid therapy, application of pharmacological medicines but according to the data of world statistics, the emaciation as a result of underfeeding – is the most frequent cause of the immunodeficient states. Furthermore, immunodeficiency appears as an associated phenomenon in such pathologies as diseases of the gastrointestinal tract, nephrotic disturbances, multiple myelomas and others.

Viral infections frequently exert an immunodepressive influence. Lymphoproliferative diseases (chronic lymphoid leucosis, myeloma and macroglobulinemia of Waldenstrom) are responsible for the general suppression of the cellular immunity. Many effects, such as X-ray irradiation, introduction of the cytotoxic agents and corticosteroids can also suppress immunoreactivity. Secondary immunodeficiencies

are observed in the malignant neoplasms, including hemoblastoses, viral infections, for example, HIV-infection or the infection, caused by Epstein – Barr virus, immunosuppressive therapy, aging, emaciation, loss of immunoglobulins, for example, in the nephrotic syndrome or exudative enteropathy. HIV-infection is the leading cause for secondary immunodeficiency today. It is manifested by chronic infections including those caused by conditionally pathogenic microorganisms, and by malignant neoplasms, first of all, lymphomas and Kaposi's sarcoma.

Many factors can decrease immunoreactivity nonspecifically. In particular, the reactions of the cellular immunity are impaired in malnutrition, deficiency of iron is especially important in this respect.

Acquired immunodeficient states are divided into 5 groups (as primary immunodeficiency):

1. Deficiency of the humoral B-cellular component of immunity;
2. Deficiency of T-cellular component;
3. Deficiency of the phagocyte functions: polynuclears and monocyte-macrophages;
4. Deficiency of the complement factors;
5. Combined immunodeficient states, which include deficiency of several components of the immunological reactivity and stem cells.

5.2. Criteria of diagnostics of secondary immunodeficiency conditions

Disturbances in various links of the immune system are accompanied by predominant susceptibility to different infectious agents. The inflammatory processes of the respiratory tracts, skin, bones, joints, caused by bacterial causative agent (staphylococcus, streptococcus, pneumococcus) are more often encountered in defects of *humoral immunity*, and predisposition to viral, parasitic and fungal diseases and

affections by mycobacteria of tuberculosis – *in deficiency of the cellular immunity* (D. V. Stephanie, Yu. E. Veltishchev, 1992). One of the signs of defficiency of B-cellular link of the immune system is hyperplasia of the lymphoid tissues. Recurrent bacterial infections are characteristic of patients with *deficiency of phagocyte functions*. Severe complications after children's infections; chronic persisting candidiasis of the skin and mucous membranes resistant to therapy; reduction in quantity of lymphocytes in the leucogram in presence of lymphocytes of small size (smaller than 10 microns) are evidence of *deficiency of the T-cellular link*.

Probability of the immunodeficiency state is more often manifested by:

1. Recurrent purulent inflammatory diseases (furunculosis, lymphadenites, erysipelatous inflammation, pneumonias, otitises, sinusites, etc.).
2. Recurrent eczema or candidiasis of the skin and mucous membranes resistant to treatment.
3. Parodoxic temperature reactions of an organism (absence of rise in the body temperature in viral and bacterial infections; feverish conditions of unspecified etiology in absence of clinical manifestations and changes of laboratory indices; long preserved subfebrile period after respiratory viral infections).
4. Increased morbidity with respiratory viral infections, frequent complications and clinical manifestations of herpetic infections.
5. Atypical character of the course of infectious and somatic diseases (prolonged, chronization, multiple complications, inefficacy of therapy).
6. Severe inflammatory diseases caused by a number of pathogenic pneumonias of hemophilic, pseudomonas, legionnaires, pneumocystic etiology) or conditionally-pathogenic (colon bacillus, proteus) microorganisms.
7. Lymphoadenopathy (regional or generalized) of unspecified etiology, persisting for 2–3 months or longer.
8. Long-term diarrhea resistant to correction.

In secondary immunodeficiencies the specified clinical manifestations are quite often accompanied by clinical semiology of the vegetative dysfunctions: general and muscular weakness; decrease of intellectual and physical capacity for work; undue fatiguability; increased perspiration; sleep disorders; increased irritability; periodic headaches, myalgias; predisposition to hypotension and faints.

The inadequate response of an organism to vaccination (especially alive vaccines) is characteristic of patients, mainly children.

In the leucogram reduction in the contents of separate cells – leucopenia, neutropenia, lymphopenia can be marked. In inflammatory diseases there is often discrepancy between character (or the period) of the disease course and dynamics of the leucogram.

In presence of the given clinical signs in the patient it is necessary to carry out investigation of the immune status condition. The grounds for diagnostics of the immunodeficient conditions is presence of stable essential reduction in the contents of immunocompetent cells (T-lymphocytes, B-lymphocytes, active phagocytic cells, etc.), immunoglobulins of various classes (more often A and G) or complement components as well as functional indices (phagocytic index, NBT-test, etc.) in immunpgrams (made in dynamics – at least twice, with an interval of one month).

5.3. Basic principles of immunotropic therapy prescription. Immunorehabilitation and immunoprophylaxis

During all history of the mankind there are preparations influencing functioning of the immune system (phytodecoctions and tinctures, the use of products of beekeeping and animal origin in food, etc.). Progress of medical science and pharmacy has essentially expanded an arsenal of the highly effective preparations used in the clinical practice regulating a condition of the immune homeostasis (cytokines, monoclonal antibodies, vaccines, serums, etc.). At the same time, approaches

to prescription of the immunotropic therapy essentially differ and are widely discussed today. Classifications and approaches to prescription of immunotropic therapy considerably differ depending on traditions of national medicine and various scientific medical schools. The terminology used is interpreted in different ways (therapy: immunotropic; immunopharmacologic; immunomodulating; immunocorrecting; immunoactivating, etc). Unfortunately, this situation has led to groundless, frequently inadequate prescribing immunotropic medicines in the clinical practice by doctors of different specialities. Development of the standardized approaches to prescribing this therapy is very important.

Kinds of therapeutic influences on the immune system

Immunotropic therapy is the application of the medical products influencing a functional condition of the immune system for the therapeutic or preventive purposes.

Immunostimulation is the influence on the immune system to activate its functional condition and stimulate the immune responses.

Immunosuppression is the influence on the immune system with the purpose of oppression of its functional condition and inhibition of pathological immune responses.

Immunoprevention is the influence on the immune system to prevent development of the disease, aggravation of the chronic pathological processes and development of complications.

Immunorehabilitation is the influence (medicamentous or non-medicamentous) on the immune system with the purpose of restoration of the disturbed functional activity.

There are several kinds of immunotherapy (immunoprevention):

- ***specific*** – change of intensity of the specific immune response to a concrete antigen (pathogen, allergen);
- ***non-specific*** – change of intensity of the nonspecific immune response to an antigen due to a change of the functional condition of the immune system.

There are also the following immunotherapy (immunoprevention) types:

1) ***Active:***
   - specific – the use of various schemes of introduction of an antigen or recombinant antigen modifications in an organism;
   - *non-specific* – the use of chemical preparations, adjuvants (Freind, BCG, etc.), etc.

2) ***Passive:***
   - *specific* – the use of specific (including monoclonal) antibodies;
   - *non-specific* – the use of preparations of immunoglogulins, cytokines, thymic factors, transplantation of the bone marrow.

5.4. Systematization of immunotropic therapy and groups of immunotropic preparations in Ukraine

Professor G. N. Drannik (2006), depending on the effect of the influence on the immune system distinguishes: 1) immunostimulating therapy (in primary and secondary immunodeficiencies, which are accompanied by recurrent bacterial and viral infections); 2) immunosuppressive therapy (treatment of the autoimmune and lymphoproliferative diseases and in transplantation of the organs and tissues); 3) immunomodulating therapy (it is indicated for healthy people who have had psychoemotional exertion or maximal physical activities).

Professor V. V. Chopyak (2009) divides the prescribing the immunotropic therapy depending on the stage of rendering medical aid into: 1) immunotherapy – target aid; 2) immunorehabilitation – general aid; 3) immunoprevention – target and general aid, assigning the following groups of preparations:

1. Target immunological preparations for immunotherapy:
   - serum;
   - immunoglobulins, especially intravenous forms;

- interferons;
- preparations obtained by gene therapy;
- monoclonal antibodies;
- inhibitors of kinases (protein, lipid, tyrosine, serinkinases – Syk, STAT, Jak, Ras, Zap, MAPK);
- synthetic drugs (inhibitors of the synthesis of nucleotides – methotrexate; calcineurin – cyclosporine; cytokines – azathioprine; antimetabolites – thalidomide);
- microRNA oligonucleotides;
- anti-viral drugs.

2. Target immunological preparations for immunotherapy and immunorehabilitation:
   - anticytokine herbal preparations (zinaxin).
3. Target immunological preparations for immunoprevention and immunotherapy:
   - vaccines, including adjuvants, stabilizers, liposomes, immunostimulating complexes, virosomes, cytokines, antagonists TLR nucleotide vaccines, TLR antagonists, nucleotide vaccines, nuclein-acid vaccines, pox-, adeno-, alpha-viruses;
   - allergovaccines (recombinant, T-peptide, the Th_1-stimulating, antigen-stimulating complexes, anti-IgE, adjuvants).
4. General immunological preparations for immunotherapy and immunorehabilitation:
   - glucocorticoids;
   - nonsteroid anti-inflammatory drugs;
   - ACE inhibitors;
   - inhibitors of histamine;
   - adjuvants (ligands of TLR, polyelectrolytes);
   - physiological proteins, polypeptides, peptides;
   - stimulators of interferon;
   - interferon inducers.
5. General immunological preparations for immunorehabilitation and immunoprevention:
   - metabolites (vitamins, minerals);

- adaptogens and biostimulators (synthetic and vegetative);
- clearance preparations (enterosorbents);
- probiotics and eubiotics.

Systematization of groups of the immunotropic preparations is given in the State Form of medical products (see Appendix 2), which is annually updated and supplemented.

5.5. General recommendations for immunotropic therapy prescription

Uniform recommendations for prescribing the immunotropic therapy are absent and are discussed that is caused by a low level of efficacy of many preparations (as a rule, widely administered by practising doctors). Mainly, it concerns prescribing the immunostimulating preparations used in the secondary immunodeficient conditions or in patients for immunorehabilitation and nonspecific immunoprevention. Whereas application of immunosuppressive (autoimmune, oncological, allergic diseases) or substitutional therapy (primary immunodeficiencies), antiviral preparations (HIV, herpes, cytomegalovirus, Epstein – Barr virus) as well as active specific immunoprevention is regulated enough.

In 2000 the working commission of experts from Russia and CIS countries offered recommendations on the use of the immunotropic preparations (Sepiashvili R. I, 2000):

1. Not to prescribe immunotropic preparations without investigation of the immune status.
2. Prescribing immunotherapy is contraindicated in case of revealing deviations in the immune status and in absence of clinical manifestations of immunopathology (dynamic monitoring of parameters of the immune status is recommended in this case).
3. Application of the immunotropic preparations without estimation of the immune status is possible exclusively for prevention: in prognosis of epidemic of any infectious disease (for example,

the flu), before the scheduled surgical intervention, in AIDS and HIV-infected patients, severe oncological patients.

In addition to these recommendations it is necessary to clarify few points:

1. It is not always the existence of disorders in immunograms allow you to prescribe adequate immunotropic therapy. During assessment of immunograms it should be considered: the period of the disease (acute, recovery, remission), the degree of severity, the therapy (antibiotics, etc.), patients age and sex, seasonal changes, etc. Frequently routine indices of single investigation allow to make only rough diagnostic conclusions. Fuller information for prescribing immunotherapy gives the immunogram made in dynamics (especially in the period of remission of the disease) in presence of the stable expressed changes of the indices.
2. Immunostimulating therapy (immunorehabilitation) should not be opposed to a causal treatment, but only to supplement it in the presence of clinical and laboratory evidence.
3. Immunostimulative therapy is generally not prescribed when the patient has an allergic or autoimmune diseases, or for the corresponding changes in immunograms.
4. Choosing the drug depends on the mechanism of its action, of gravity of the patient's state of disease, severity of the immune system disorders, and must be substantiated with the indications and possible side effects.
5. It is possible to use several immunostimulators influencing on different links of the immune system, however, taking into consideration a principle of "immunologic balance". The combination of specific and nonspecific immunotherapy as well as prescription of preparations with various mechanism of action (stimulation of the phagocytic link, antiviral protection, etc.) is more justified.
6. Regulations on early prescribing immunotherapy during the acute period of infectious diseases in complex with etiotropic

therapy are debatable. Though efficacy of immunotherapy is often higher in the acute period than in the period of remission. It is necessary to take into account severity of the patient's condition and intensity of treatment (immunotropic effects of other preparations) in order to prevent inadequate response of the immune system (in case of tension of its functional reserves) and polyprogmasia. Active immunotherapy is contraindicated in the severe course of acute diseases

7. Immunoprevention should be given taking into account seasonal aggravations of the disease or the most unfavorable periods of the year for diseases with acute respiratory viral infections (October – November and February – March).
8. The use of the immunotropic preparations can cause side-effects and allergic reactions, especially in their long and ungrounded application.
9. The effectiveness of immunotherapy significantly increases in combination with detoxification therapy (including sorbents).
10. Immunotherapy is administered in complex with recommendations on normalization of life activity of the patient (nutrition regimen and full value diet, exclusion of stress situations, etc), vitamin therapy, pro- and eubiotics, etc.
11. During immunotherapy it is recommended to make monitoring of changes in the immunological indices, as clinical efficacy not always corresponds to normalization of the immunological indices.
12. Choosing of a preparation (combinations of preparations) for repeated courses of the immunostimulating therapy is made taking into account efficacy of previous treatment of the patient, as the individual reaction is often marked during taking immunotropic preparations.

In the conclusion it should be noted that adequate and effective prescription of the immunotropic therapy provides knowledge of laws of the immune system functioning and understanding necessary care and precaution of the use of the immunotropic preparations. Unfor-

tunately, modern methods of diagnosis do not allow to estimate fully the condition of adaptable reserves of the immune system in a definite patient. Whereas it is possible to achieve normalization of indices of the immune system by sanation of the foci of chronic infection in an organism or correctly chosen etiotropic therapy of acute and chronic diseases in many patients.

5.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Secondary immunodeficient conditions: causes, classification.
2. Secondary immunodeficient conditions: peculiarities of the clinical course, diagnostics, principles of therapy.
3. Concepts: immunotherapy, immunostimulation, immunosuppression, immunorehabilitation, immunoprevention. Differences between them.
4. Active and passive immunotherapy and immunoprevention.
5. General recommendations for administration of immunotherapy.
6. The mechanism of action of preparations on the basis of monoclonal antibodies.

Practical skills

1. Indications for prescription of interferon preparations.
2. Indications for prescription of natural and synthetic interferon inducers.
3. Indications for prescription of preparations of a bacterial origin.
4. Indications for prescription of herbal adaptogens.
5. Prescribing the course of immunprevention to a child who often falls ill with respiratory viral infections, with the lowered contents of IgA and IgG in the blood.

6. Prescribing the course of immunotherapy to a patient with the expressed disturbance in the phagocytic link of the immune system.
7. Prescribing the course of immunotherapy to a patient after severe pneumonia without expressed disturbances of the immune status.
8. Prescribing the course of immunotherapy to a patient with disturbances in the T-cellular link of the immune response (absolute and relative reduction in the contents of T-lymphocytes at the expense of T-helpers).

Tests

1. Purulent inflammatory processes in an organism caused by bacterial coccal flora, is mainly encountered in:
   A. Deficiency of the humoral B-cellular link of immunity
   B. Deficiency of the T-cellular link
   C. Deficiency of the phagocyte function
   D. Deficiency of the complement factors
   E. Combined immunodeficiency states

2. Predisposition to viral, parasitic and fungal diseases and affections with mycobacteria of tuberculosis is usually observed in:
   A. Deficiency of the humoral B-cellular link of immunity
   B. Deficiency of the T-cellular link
   C. Deficiency of the phagocyte function
   D. Deficiency of the complement factors
   E. Combined immunodeficiency states

3. The lowered resistance to Neisseria is more often observed in:
   A. Deficiency of the humoral B-cellular link of immunity
   B. Deficiency of the T-cellular link
   C. Deficiency of the phagocyte function
   D. Deficiency of the complement factors
   E. Combined immunodeficiency states

4. Influence on the immune system to prevent development of the disease, aggravation of the chronic pathological processes and development of complications is called:
   A. Immunostimulation
   B. Immunomodulation
   C. Immunorehabilitation
   D. Immunoprevention
   E. Immunosuppression

5. Healthy people who have had psychoemotional exertion or maximal physical activities are indicated the administration of preparations for:
   A. Immunosuppression
   B. immunomodulation
   C. Immunostimulation
   D. Immunoprevention
   E. Immunoactivation

6. The use of immunotropic preparations without estimation of the immune status is possible exclusively:
   A. In patients with pneumonia
   B. For prevention of deterioration of a condition of patients with dysentery
   C. In prognosis of the flu epidemic
   D. In patients with chronic cholecystitis
   E. For those who have had operation on the bladder

7. To the group of what immunotropic preparations is viferon related to:
   A. Immunoglobulins
   B. Interferons
   C. Interleukins
   D. Interferon inducers
   E. herbal adaptogens

8. What preparation is not related to preparations of the bacterial origin?
   A. Respibron
   B. Liasten
   C. Immudon
   D. Immunofan
   E. Ribomunil

9. What preparation exerts a direct stimulating influence on the phagocytic cells and natural killers, and has detoxication action?
   A. Polyoxidonium
   B. Laferobion
   C. Tirolon
   D. Timalin
   E. Kagocel

10. What preparation inhibits formation and release of proinflammatory cytokines by T-cells and corpulent cells?
    A. Infliximab
    B. Pimecrolimus
    C. Everolimus
    D. Tacrolimus
    E. Azatioprin

CHAPTER VI

FUNDAMENTALS OF TRANSPLANTATION IMMUNITY

6.1. The concept of transplantation immunity

In 1945 the English immunologist P. Medawar revealed antibodies in the recipient, specific to the antigens of the donor while transplanting the skin flap between the rabbits. It was the starting point for forming one of the sections of the immunological studies – transplantation immunology.

Lately together with the transplantation of the bone marrow, kidney, liver and heart there have been used the transplantations of the small intestine, portion and segments of the liver, lung, bones, pancreas and cells of pancreatic islets as well as other organs and tissues.

Transplantation immunity is a complex of hyperimmune responses, which develops in response to the transplantation of an organ or tissue from the genetically different individual. It is caused by the presence of a number of the antigens:

- MHC antigens;
- Erythrocyte antigens of the system AB0 and Rh;
- A small complex of antigens of histocompatibility, coded by Y- chromosome.

6.2. Types of transplants (grafts) (Fig. 6.1)

Autotransplant(autograft) is tissues of the donor transplanted to him/her.

Isotransplant (syngraft) is an organ or tissues transplanted to the syngenic, i.e. having the same genotype individual (monozygotic twins or animal of the same inbred line).

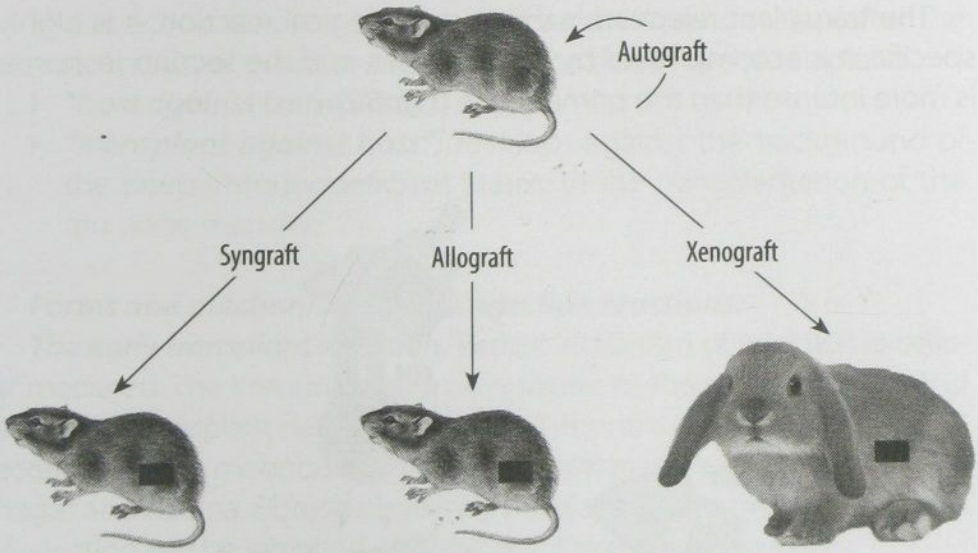

Fig. 6.1. Types of transplants. *These four types of grafts are represented by the direction of the arrows (Klaus D. Elgert, Immunology, 1996).*

Allotransplant (allograft) (the old term – homograft) is an organ or tissue transplanted between the allogenic individuals, i.e., between the representatives of one and the same form who have different genotype. For example, transplantation from one person to another. The most frequent procedure of allotransplantation is the blood transfusion.

Xenotransplantat (xenograft) (heterotransplant) is an organ or tissue transplanted from the representative of one biological form to the representative of another form.

6.3. Forms and mechanisms of the transplant rejection

The increased sensitivity to the transplanted tissue occurs in approximately 1–2 weeks after transplantation and it persists from 1 month to several years.

The transplant rejection is the immunological reaction: it is highly specific, it is accomplished by lymphocytes, and the second response is more intense than the primary one (Fig. 6.2).

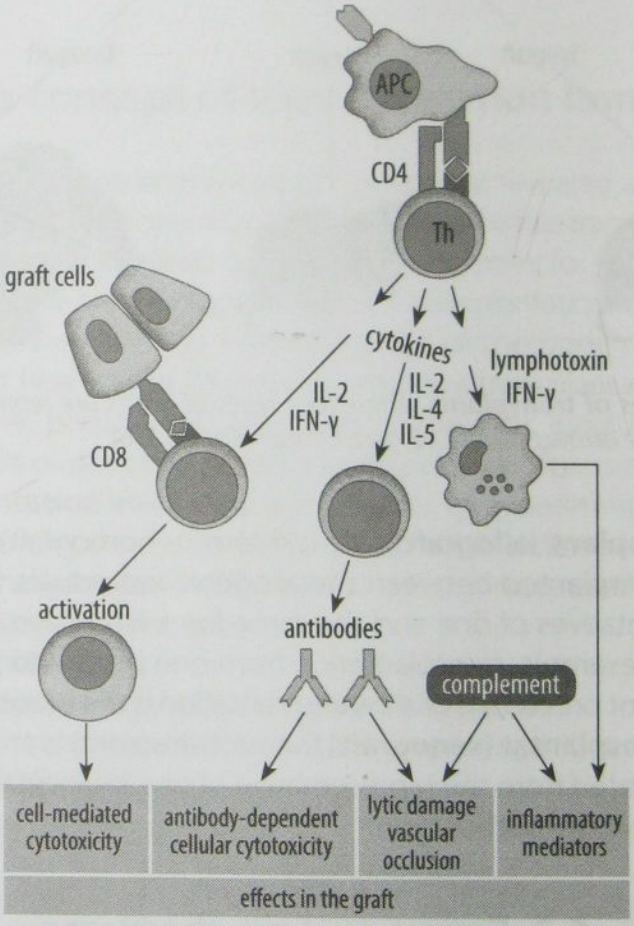

Fig. 6.2. Th-cells are activated by APCs to release lymphokines. *IL-2 and IFN-γ are required for Tc-cell activation; IL-2, IL-4 and IL-5 are involved in B-cell activation; a mixture of lymphotoxin and IFN-γ acts as macrophage-activating factor (MAF). These cells reject the graft by specific cell-mediated and antibody-mediated immune pathways, or by non-specific inflammatory reactions (I. Roitt et al., Immunology, 2001).*

The rejection reaction by two mechanisms may develop after the transplantation of tissue or organ from the donor to the recipient:

- ***"host against transplant";***
- ***"transplant against host"*** develops against the background of the deep immunodeficient states, in the transplantation of the red bone marrow.

Forms and mechanisms of the rejection reactions:

The early transplant rejection. Basic mechanism of rejection is cellular mediated. The immune response is similar to the tuberculin test, and causes the transplant destruction for days–months. It is histologically characterized by the mononuclear cellular infiltration of the transplant, hemorrhages and edema. Fibrosis frequently develops due to hypoxia. This form of rejection can be inhibited with the aid of immunosuppressors.

Late transplant rejection. It is mainly manifested in patients with the immunodeficient state. Pathomorphology of it differs from early rejection regarding the fact that the endothelium of vessels is involved; there is its proliferation with the subsequent contraction of the vessel lumen, which leads to ischemia and necrosis of the transplant.

Hyperimmune (superacute) transplant rejection (by the type of "white transplant"). It results from the presence of the preexisting antibodies. The antibodies interact with the HLA antigens of the donor, which are located on the transplant endothelium. The formed complexes activate the complement, which damages the endothelium and thrombocytes, leading to thrombosis of the transplant vessels – vascularizations of the transplant does not occur. The process of the tissue dying off begins immediately after transplantation. The process is irreversible and is not prevented by any of the known methods of immunosuppression.

The development of the reaction of transplantation immunity consists of three stages:

***Stage I:* recognition.** The process of recognition includes the predecessors of the cytotoxic T-lymphocytes and the predecessors of helper and inflammatory T-cells (Th_0). After the recognition of the an-

tigen the cells of these types migrate in the nearest lymphoid tissue, more frequently in the regional lymph node.

***Stage II:* maturation and accumulation.** Maturation and accumulation of the cells of different types – effectors of the rejection reaction – occurs in the peripheral lymphoid tissue. In the lymphoid tissue the antigen provides the accumulation of T-cells of inflammation (Th_1) after assimilation by macrophages and coming on the cellular surface in the immunogenic form. While expressed on the surface of the B -cells it turns on the helper T-cells (Th_2), it provides the accumulation of specific antibodies. The secreting antibodies can be sorbed on the surface of the natural killers (NK-cells) – as a result the NK-cells binding immunoglobulin acquire capability for antibody dependant cytolysis of the transplant cells.

Stage III:* destruction.** There are ***specific participants in destruction and rejection of the transplant: CD8 of the T-cell, CD4 of the T-cell of inflammation (Th_1), specific and ***nonspecific*** immunoglobulins: activated macrophages and natural killers.

6.4. Pre-transplantation monitoring

It is very difficult to select the donor completely compatible with the recipient by the HLA antigens, since the number of combinations, composed of more than 100 antigens of this family is extremely great. The probability to find the completely compatible donor makes from 1 : 1.000 to 1 : 1.000 000 depending on the prevalence of this or that HLA antigen. The probability of selecting the completely compatible donor among the native brothers and sisters makes 1 : 4, since HLA genes are inherited by Mendel's laws.

Estimation of the compatibility of the donor and recipient by the HLA antigens:

- HLA antigens of the recipient and donor are determined;
- antigens of AB0 system of recipient and donor are determinated;

- sensitization of the recipient by the HLA antigens is excluded;
- test for the individual compatibility is carried out.

Besides the donor is selected he/she coincides with the recipient in the antigens of the system AB0. This is especially important for the kidney transplantation.

Determination of the HLA antigens of the recipient

1. The serological methods

The lymphocytotoxic test (a basic method): the sera against different HLA antigens are added to the culture of the investigated lymphocytes; after incubation the complement is added – the lymphocytes carrying the antigen are destroyed under the effect of the complement – then the dye is added to the lymphocytes, which stains only the living cells. The result is evaluated according to the relative number of the killed lymphocytes.

2. Molecular-genetic methods – study of DNA. They are used only for typing the HLA genes of the class II.

a. Analysis of polymorphism of the lengths of restriction fragments. The method is based on the ability of bacterial endonucleases to split DNA in those sections, in which the sequences of nucleotides are concentrated – sites of restriction specific of endonuclease. The application of endonucleases allowed to re-

Evaluation of the results of the lymphocytotoxic test

(A. Zachary, G. Teresi. ASHI Laboratory Manual, 1990)

Number of those been killed cells, %	Mark	Result
0–10	1	Negative
11–20	2	Doubtful
21–50	4	Weakly positive
51–80	6	Positive
81–100	8	Sharply positive

veal polymorphism of the lengths of the restriction fragments of DNA, similar to polymorphism of HLA, determined serologically.

b. ***Determination of specific oligonucleotide sequences.*** Single-chain oligonucleotide probes are used completely complementary to the unique sequences of each known allele of the HLA gene. To determine the unknown allele it is possible to use consecutively a series of the probes of different specificity.

c. ***PCR*** – obtaining of the large number of copies of the DNA fragments with the specific nucleotide sequence.

3. Cellular methods. After recognition of the foreign antigen the T-lymphocytes proliferation begins. This process can be reproduced *in vitro* in the mixed culture of lymphocytes, which consists of the donor and recipient lymphocytes. If the donor and recipient have different HLA antigens of the class II, proliferation is noted in the mixed culture.

Development of the recipient sensitization by HLA antigens

Determination of antibodies to HLA antigens

1. Coefficient of seropositivity is a relation of the number of lymphocyte models, which cause positive reaction, to the total number of models in the panel, expressed in the percentage. The investigation is carried out with the aid of the lymphocytotoxic test. The coefficient of seropositivity reflects the risk of the superacute rejection of the transplant, taken from the random donor. The higher this coefficient, the more difficult is to select the compatible donor. If the coefficient of seropositivity exceeds 80 %, transplantation is possible only from the donor, completely compatible with the recipient by the HLA antigens.

2. Serological methods reveal the following antibodies in the recipient serum:

1) antibody to the HLA antigens of the class I: HLA-A, HLA-B and HLA-C. These antigens are present on the surface of the lymphocytes and monocytes.

2) antibody to the HLA antigens of the class II: HLA-DR, HLA-DQ and HLA-DP. These antigens are present on the surface of the monocytes and B-lymphocytes. They are absent on the surface of the nonsensitized T-lymphocytes.

Test for individual compatibility

The final stage of the donor selection is a test for the individual compatibility. The lymphocytotoxic test is based on the test for individual compatibility. The recipient serum is added to the donor lymphocytes. The purpose of the study is to reveal any antibodies, which can react with the HLA antigens of the donor and cause the superacute rejection of the transplant. Positive lymphocytotoxic test is evidence of the high risk not only of the superacute, but also acute and chronic rejection of the transplant.

The group of patients with the high risk of the transplant rejection include the recipients, previously immunized by the HLA antigens (coefficient of seropositivity exceeds 15 % and recipients who withstood transplantation earlier, independent of the coefficient of seropositivity at the present moment.

6.5. Immunosuppressive therapy and posttransplantation monitoring

Immunosuppressive therapy is carried out in all patients before and after transplantation. The exceptions are those cases when the donor and recipient are monozygotic twins.

Modern approaches to the immunosuppressive therapy provide for the simultaneous use of several immunodepressants and their prescription before and after of transplantation for prevention and treatment of the transplant rejection. At present corticosteroids, azathioprine, cyclosporin, mono- and polyclonal antibodies are used as the immunodepressants. These remedies prevent activation of the immune response or block the effector mechanisms of immunity.

Immunodepressive drugs (immunosuppressors, immunodepressants): Azathioprine, anti-lympholin-KR, Batriden, Crizanol, Cyclo-

sporine, Auranofin, Tacrolimus, Muromonab-CD3, Sirolimus, polyclonal antibodies to the lymphocytes (antilymphocytic immunoglobulin and anti-thymocyte immunoglobulin).

Polyclonal antibodies to the lymphocytes (antilymphocyte immunoglobulin and anti-thymocyte immunoglobulin) are obtained from the serum of rabbits and other animals after immunization by the lymphocytes or cells of the man thymus. The mechanism of the action of polyclonal antibodies consists in destruction of the lymphocytes and reduction in their number in the blood. These remedies are used both with the preventive and with the therapeutic purposes. However, they increase the risk of infections.

The heterologous antibodies are applied against separate subpopulations of the T-lymphocytes. Muromonab-CD3 is a drug of mouse monoclonal antibodies to CD3 closely related to the antigen-recognizing receptor of the T-lymphocytes of man. After binding with the antibody CD3 disappears from the surface of the T-lymphocytes for some time, which makes their activation impossible. After some time CD3 again reappears on the surface of the T-lymphocytes; however, it remains blocked by muromonab-CD3. The drug is used in the transplant rejection when corticosteroids are ineffective. It is used both for the prevention and treatment of the transplant rejection.

At present the clinical tests are conducted of the humanized monoclonal antibodies, created by the methods of genetic engineering. Theoretically such antibodies must destroy lymphocyte-targets more effectively and decrease the risk of the antibody production to the mouse antigens.

Important component of immunosuppressive therapy is glucocorticoids. Prednisolone is usually used, because its side- and therapeutic effects are simpler to trace, and it is effective in the transplant rejection in the high doses. As a rule, the drug is administered in the dose of 200–300 mg/day directly before or during the transplantation, and then the dose is reduced to 30 mg/week.

Immunological tolerance to the transplant. The induction of the recipient tolerance to the donor's antigens is the ideal method, with

the aid of which it is possible to prevent the transplant rejection as well as avoid the immunosuppressive therapy.

At present for induction of the tolerance to the donor's antigens before the transplantation we use:

1) transfusion of the donor's whole blood to the recipient;
2) transfusion of the donor's leukocyte mass and irradiation of the recipient lymphoid organs (nonspecific immunosuppression);
3) combination of these methods with the immunosuppressive therapy.

The schemes of treatment described above allow to use more sparing immunosuppressive therapy after transplantation.

Immunological studies after the transplantation

Diagnostics of the transplant rejection is made regularly in all patients who underwent transplantation. There are no reliable methods of immunological diagnostics of the transplant rejection. Biopsy remains the only reliable method of diagnostics of the transplant rejection today.

Determination of the absolute number of the T-lymphocytes in the blood is the best method of assessment of the effectiveness of muromonab-CD3, anti-thymocyte and antilymphocyte immunoglobulins. The T-lymphocytes number in the blood is determined by the method of flowing cytofluorimetry. The determination of the T-lymphocytes number in the blood allows to select a dose and to establish the duration of application of the mono- and polyclonal antibodies.

6.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Modern concept about the structure and functions of the main complex of histocompatibility. The structure of HLA antigens.
2. Immunological indications and contraindications to the transplantation of organs and tissues. Selection of the pair donor – recipient.

3. Antilymphotcytotoxic antibodies, their prognostic value.
4. Peculiarities of pre- and posttransplantation immunological monitoring.
5. Types of rejection crisis, their clinico-immunological characteristics and prognostication value.
6. Kinds of transplants.
7. Mechanisms of the transplant rejection.
8. Forms and mechanisms of the rejection reactions.
9. The development of the reaction of transplantation immunity.

Practical skills

1. To interpret data of tests on the selection of donor and recipient for conducting the transplantation.
2. To master the skills to diagnose supraacute, acute and chronic crises of rejection during the transplantation of organs and tissues.
3. To make differential diagnostics of the crisis of rejection and infectious complications in patients after transplantation.
4. To administer immunosuppressive therapy and to evaluate its effectiveness after the transplantation of organs and tissues.

Tests

1. Transplantation of the organ or tissue from the donor of one species to the recipient of the same species is called:
   A. Autotransplant
   B. Isotransplant
   C. Syngraft
   D. Allotransplant
   E. Xenograft

2. Autotransplant is transplantation of an organ or tissue:
   A. Within one species
   B. To the same donor

C. From genetically homogeneous person
D. From one species to another
E. From the monogerminal twin

3. What type of the transplant rejection proceeds by the mechanism of the 4th type of the hypersensitivity reaction?
A. Early
B. Delayed
C. Late
D. Acute
E. Superacute

4. Presence of the preexisting antibodies in the recipient to the tissue antigens of the donor results in development of the transplant rejection:
A. Late
B. Delayed
C. Early
D. Acute
E. Hyperimmune

5. The mechanism of the transplant rejection – transplant vs host may develop in the recipient in presence of:
A. Autoimmune disease
B. Normal functioning of the immune system
C. Hereditary allergic predisposition
D. Expressed immunodeficiency condition
E. Compete discordance with donor's HLA antigens

6. While choosing the donor the main thing is the concordance of antigens in the donor and recipient according to:
A. MHC
B. AB0
C. Rh-factor

D. AB0 and Rh-factor
E. Small complex of histocompatibility

7. Pre-transplantation preparation consists in prescription of the following therapy:
A. Immunostimulating
B. Immunomodulating
C. Immunosuppressive
D. Immunoprophylactic
E. Immunoactivating

8. The best indices of lymphocytotoxic test for making transplantion:
A. Negative
B. Doubtful
C. Weakly positive
D. Positive
E. Sharply positive

9. What is the sequence of transplant rejection?
A. Accumulation and maturation; recognition; destruction
B. Accumulation and maturation; destruction; recognition
C. Destruction; accumulation and maturation; recognition
D. Recognition; accumulation and maturation; destruction
E. Recognition; destruction; accumulation and maturation;

10. What is the most reliable method of the diagnosis of transplant rejection after transplantation?
A. Determination of the T-lymphocyte number
B. Determination of antigens
C. Ultrasonic study
D. Biopsy
E. X-ray examination

CHAPTER VII

REPRODUCTIVE IMMUNOLOGY

The immunology of reproduction deals with the study of the immune mechanisms, which participate in the development of sex cells of men and women, fertilization, pregnancy, childbirths, postpartum period as well as gynecological diseases.

7.1. Immune interrelations in the system "father – mother – fetus"

Fetus inherits mother's cells only by half – the remaining proteins and genes are paternal, they are foreign. Therefore, the immune system of mother may "turn on" active rejection, and immunological tolerance (compatibility). The phenomenon of immunological paradox is nonrejection of genetically foreign fetus. It allows to preserve fetus.

The immunological individuality of man determines the main complex of tissue compatibility – a system of leukocyte antigens (HLA).

For the normal course of pregnancy the antigens, which the fetus obtained from the father, should be recognized by the immune system of the mother. The more children in the married couple are, the higher is the content of the antibodies in the blood of a pregnant woman to HLA of her husband. If there are few such antibodies (for example, in case of the closely-related marriage, when HLA of the husband is similar to personal HLA of the wife), then the probability of miscarriage grows.

In normal development of pregnancy the paternal antigen circulating immunocomplexes with the paternal antigens and free antibodies to the paternal antigens are determined in the early periods of pregnancy. The alloimmune response of the mother is directed against some, but not against all noncoincident HLA antigens of the fetus.

The women who are completely compatible with their husbands in the HLA antigens do not produce sufficient amount of the antibodies to the fetus antigens and suffer from habitual noncarrying of pregnancy.

Basic mechanisms of the fetus protection are:

- suppressor cells of the endometrium;
- immunosorption properties of the placent;.
- humoral factors of the mother facilitating development of immunological tolerances:

1) immunomodulating protein TJ6 plays a large part in the development of immunological tolerance, in particular, during pregnancy. TJ6 is expressed in the uterine tissues. Secreted TJ6 binds with the receptors of cytotoxic NK-cells at the site of implantation and induces apoptosis of these cells. Progesterone stimulates production of TJ6;

2) spermine in the amnion and fetuin – glycoprotein of the fetus plasma suppresses the production of proinflammatory cytokines;

3) early pregnancy factor (EPF) is the protein, which appears in the blood serum of the pregnant women in 48 hours after fertilization and is not revealed in case of the loss of the fertilized egg. This is a sensitive marker, which reflects the embryo viability.

Immune status of the pregnant females

The cellular component:

- The content of leukocytes does not change, of lymphocytes – decreases (especially in the 3rd trimester), which contributes to better survival of the fetus. The total number of T-lymphocytes is reduced in the 3rd trimester. Reduction in the number of B-lymphocytes is observed.
- The number of T-suppressors rises.
- The number of T-helpers does not change, but the functional activity of helpers is reduced.

- The content of the natural killer cells is reduced by the 3rd months of pregnancy, and a quantity of O-lymphocytes increases 2 times (in multipara women it reliably rises in the development of pregnancy and increases depending on the number of deliveries).
- Reduction in the killer activity and the bactericidal function of the macrophages.

The humoral component:

- The cellular-mediated immune response is reduced – production IL-2 and IL-8 is suppressed.
- The activity of the complement system is reduced.
- The hormonal mediators, which block AB are synthesized.
- The levels of CIC are changed: increase in the first trimester and reduction by the end of pregnancy.
- The level of C3 rises (in gestosis it is lowered).
- Increase of IL-1 and TNF (more than 6 times) and levels of IL-4, IL-10
- The level of IgG and A in women in the second-half of pregnancy is sharply reduced, and IgM is increased.

7.2. Miscarriage (noncarrying of pregnancy)

Classical habitual noncarrying of pregnancy is accepted to be the situation of three or more sequential noncarrying of pregnancy; however, many practicing doctors mean noncarrying of more than one pregnancy by this term. Noncarrying of pregnancy is the integrated response of the female organism to any expressed trouble in the status of the health of the pregnant female, fetus, environment as well as many other factors. The rate of noncarrying of pregnancy makes 15–20 %, more than 70 % abortions of them occur in the 1st term. Spontaneous abortions are encountered in 2 cases per 400 couples.

Causes of habitual noncarrying of pregnancy: infection (1 %), anatomical defects (5–10 %), insufficiency of the lutein phase (5–20 %), chromosomal anomalies (7–50 %), **immune mechanisms (50 %)**, unknown

causes (15 %). In the immune cause of noncarrying of pregnancy the chance to bear at full term makes 30 % after 3 abortions (without medical interventions), 25 % after 4 abortions, and 5 % after 5 abortions. In the adequate therapy the chance of full term pregnancy makes 70–85 %.

Causes of the spontaneous abortion

1. Biochemical:

- Incapability of the pregnant woman to produce the blocking factors.
- High immunoreactivity of the maternal organism to the feto-placental complex.

2. The subcellular level (R, AG):

- In the anti-phospholipid syndrome (APS).
- In the presence of chromosomal anomalies.
- Incompatibility by the blood groups (AB0).
- Inadequate expression of the HLA AG of I class on the placenta;
- In defects of the local protection.
- In 5 incompatibilities by the antigens of histocompatibility, inherited by the embryo from the father.
- In complete identity of histocompatibility AG of the mother and father. Spontaneous abortions occur more frequently when parents are relatively compatible by the HLA antigens of the loci A, B and especially D – the development of the trophoblast is weakly stimulated.
- The inherent defect of the cellular recognition, which is characterized by complete or partial absence of the allogenic response to the lymphocytes of the father.
- The hereditary predisposition: women are predisposed to noncarrying of pregnancy having HLA A1; A3; A9; A29; V5; V7; V12; DR3; DR7; the phenotypes of A1.9; A3.9; A9.23; V5.12; V7.52; DR1.7; the haplotype of A9; V5; DR2 and A3; V7; DR1.

3. The cellular level

- Reduction in the quantity of Ts in the blood; in absence of Ts in the decidual membrane.

- High proliferating activity of lymphocytes.

4. Tissue/organ – in the defects of organs.

5. The organism level:

- As a result of application of some medicines.
- After the embryo death in the uterus.
- In disturbance of the immunological tolerance in the mother.

Three groups of immunological habitual noncarrying of pregnancy:

1. The autoimmune mechanisms

Autoimmune abortive disease (AIAD) – a separate nosologic form of the autoimmune diseases. The basic sign of AIAD – association of repeated abortions, frequently at the late term, and the circulating lupous anticoagulant (with presence or absence of other clinical-serological markers of autoimmunity).

2. The excessive alloimmunne responses

Three forms of antibodies are important for bearing: maternal antibodies against the paternal leukocytes (APLA, blocking antibodies), anti-phospholipid antibodies (APA), antinuclear antibodies (ANA).

1) Antibodies against paternal leukocytes

APLA are antibodies, which block the HLA antigens of the father, expressed by the fetus, from the effector cells of the mother immune system. In conceiving the endometrium lymphocytes produce APLA-antibodies against the paternal HLA antigens. APLA are already revealed on the 5th week of gestation, they protect the fetus from the maternal natural killers that facilitate the embryo rejection. In the multipara women APLA circulation is also revealed out of pregnancy, while in women with habitual noncarrying of pregnancy the level of antibodies is low or is not determined at all.

2) Anti-phospholipid antibodies

Anti-phospholipid syndrome when antibodies attack the "native" phospholipids is one of the main structural elements of the cellular membranes. Phospholipids of the vascular walls are one of the key points in the process of the blood coagulation, formation of the

three-dimensional net of the clot. Antibodies to the phospholipids deregulate coagulation/anti-coagulation. It results in the danger of micro-thromboses, impairment of the placenta formation and abortion. Anti-phospholipid antibodies are revealed in 22 % of women with the habitual noncarrying of pregnancy. Frequency of APS rises by 15 % with each following abortion. *Clinical course:* 1) tendency to the thromboses, thromboembolisms, relapses of the thromboses; 2) thrombocytopenia; 3) repeated abortions; 4) pulmonary hypertension; 5) affection of the valve apparatus of the heart (in majority of cases of the mitral one) by the type of insufficiency; 6) neurological disturbances (epilepsy, chorea, migraine); 7) affection of the skin (ulcer, necroses). *Treatment:* small doses of aspirin and heparin correct the coagulation properties of the blood, and this frequently leads to good results – pregnancy develops normally.

3) Antinuclear antibodies

An increase in the titer of the antinuclear antibodies is revealed in 22 % of women with the habitual noncarrying of pregnancy and in 50 % of women with infertility and IVF failure. Antibodies to DNA may be directed at the native DNA, denatured DNA, polynucleotides and histones. The increased titers of the autoantibody to DNA can produce inflammatory changes in the placenta and start the reaction of the fetus rejection. *Treatment:* when presence of ANA is combined with the habitual noncarrying of pregnancy *prednisolone* is administered for suppression of the inflammation and stabilization of the cellular membranes. In presence of indications prednisolone is administered before conception. Adequate treatment allows to bear pregnancy in 80–85 %.

3. Insufficient protective reactions – gestosis

The origin of gestosis is not in strengthening, but in weakening of the recognition of the fetus alloantigens by the mother. In gestosis the maternal organism loses the ability to recognize foreign AG of the fetus and produce blocking AB. The disturbance of the uteroplacental blood circulation is caused by the damaging effect of the cellular and humoral cytotoxic reactions on the placenta facilitating dysfunction of the placenta as the immune barrier. *Treatment:* immunodepressants,

antioxidants; the small doses of aspirin; the termination of pregnancy (the basic method)

4. In recent years it is reliably established that the most frequent cause of the habitual noncarrying of pregnancy is coincidence of the mother and father in two and more loci of the system HLA.

7.3. Immunodependent forms of infertility in marriage

Infertility may have immunological causes. 20 % of cases of infertility are not associated with the organic disturbances of the sexual sphere of husbands among 3 million infertile marriages in the USA. Only 2–5 % of infertile women are revealed to have AB to the sperm, although it is assumed that the immunological infertility is encountered in 10–20 % of cases. Titers of AB over 1:16 are correlated with infertility.

Causes of sterility in men:

- 80 % of men with the infertile marriage are revealed to have autoAB to the sperm.
- Absence of the suppressor factors in the seminal fluid of the men, which decrease the expression of AG on the spermatozoids in the normal conditions and suppress the immune response.

Causes of sterility in women:

- ABs to the spermatozoids are revealed in 40 % of women among the infertile couples. ABs are mainly concentrated in the mucus extract of the cervix of the uterus (in 50 % of the infertile women). ABs of the woman may block the mobility of the particles of the spermatozoid membrane and disturb the processes of adhesion and confluence with the ovule.
- Increased phagocytosis of spermatozoids. The peritoneal macrophages of man are capable of phagocytizing normal sperm.

Diagnostics – determination of anti-sperm AB in the cervical mucus.

Anti-sperm antibodies in men

The immune system usually recognizes only proteins as "its own", which were in the organism at the moment of birth. The proteins, which are synthesized more lately (for example, in the testicle tissue) are already recognized as foreign.

Different mechanisms prevent the formation of anti-sperm antibodies in the different parts of the man reproductive system:

1. Protection of spermatozoids and cells of spermatogenesis is achieved by the hematotesticular barrier (HTB) in the testicle, which isolates the cells of spermatogenesis from the immunocompetent cells of the organism. The cells of Sertoli and their branches compose the basis of this barrier.
2. After release of spermatozoid from the testicle spermatozoids are capable of mimicry (adjust) in the environment, i.e. dropping of the previously sorbed antigens from the membrane and adsorbing new ones – from other media, including those from the female reproductive tract. This ability is expressed more strongly in viable spermia.
3. The spermoplasm contains the local regulatory factors, which impede the formation of anti-sperm antibodies and development of the cellular anti-sperm sensitization (for example, the immunosuppressive factor of spermoplasm). These factors are secreted in the accessory glands of the man's reproductive system.

The failure of the protection mechanisms at any level leads to the development of anti-sperm antibodies. Damage of HTB opens access to the testicle tissue for the immune system, being the carrier of antigens, to which the immunological tolerance is not produced in the organism. For example, surgical interventions, injuries of the scrotum, inflammatory diseases of the man's reproductive system of different etiology frequently result in disturbance of the HTB integrity and development of autoimmunity to the antigens of spermatozoids.

The insufficiency of the mimicry mechanism leads to the contact of sperm antigens with the immune system and production of the anti-sperm antibodies both in the male and female organism. The disturbances of the processes of spermatogenesis (in the ovule) or the functional maturation of spermia (in epididymis) caused by different causes (inflammatory diseases, varicocele, cryptorchism, endocrine pathology) lead to reduction in the viability of spermatozoids and disturbance of the mimicry mechanism.

Anti-sperm antibodies in women

The sex organs of woman contain the large number of immunocompetent cells. The natural entry of sperm into the genital tracts of a woman may cause the immune response. However, different mechanisms, which decrease the immune response, prevent the formation of antibodies in the female organism. The balance of the T-lymphocytes is changed during ovulation: the level of T-helpers is reduced, and T-suppressors rises as well as the concentration of immunoglobulins and C3 of the component of the complement system decreases. The male mechanisms of protection play an important role in reduction in the immune response to spermatozoids: mimicry – absorption and desorption of the superficial antigens in change of the media and the immunosuppressive factor of spermoplasm. Furthermore, only the small number of genetically selected spermatozoids penetrate into the uterine tubes that are immunologically differ from the majority, whereas the rest are destructed blocking local immunity.

The disturbance of fertility is accompanied by the disturbance of protective mechanisms, so the immune response to the antigens of spermatozoids can arise in the inflammatory diseases of the vagina.

Methods of determining the anti-sperm antibodies:

To exclude the role of the anti-sperm antibodies in the genesis of infertility the tests of interaction of spermatozoids and cervical mucus are made **(postcoital test, Kurzrock – Miller test)** as well as determination of the anti-sperm antibodies in the blood serum, sperm and

cervical mucus. Negative postcoital test is an evidence of the presence of the anti-sperm antibodies, either in the sperm or in the cervical mucus. A study of the anti-sperm antibodies is required for the married couples with a negative postcoital test.

The simplest and informative test is a mixed antiglobuline test (MAR-test), recommended by WHO (World Health Organization) for the routine screening of the samples of the seminal fluid. One drop of fresh sperm is placed on the microscope slide, one drop of the latex particles, covered with IgA, then one drop of antiserum to the human IgG, and they are intermixed. % the spermatozoids is calculated under the microscope, glued with the latex particles, covered with IgG (this is completely comprehensive study since it is established that IgA practically always develop together with IgG).

Determination of the anti-sperm antibodies by latex agglutination. The seminal fluid or cervical mucus is diluted by buffer and centrifuged, the dilutions are 1:100, 1:200 and so forth are prepared from the supernatant liquid. The corresponding dilutions of the sample and latex particles are added on a special plane-table, and then the appearance of agglutination is evaluated. If it appears in the dilution 1:100, the test is considered positive.

Determination of the anti-sperm antibodies in the blood serum is made by the method of indirect solid-phase immunoenzyme analysis. The normal values are 0–60 U/mL; the intermediate values – 61–100 U/mL; increased – more than 100 U/mL.

Flow cytometry (determination of anti-sperm antibodies and their concentration on the membrane of spermatozoids), radio-immune, immunofluorescence and others.

7.4. Immune aspects of contraception

Main methods:

- contraceptive vaccines;
- immunization against one or several sex hormones (realizing-hormone of luteinizing hormone, LH itself, FSH, gonadal ste-

roids). A successful regulation of fertility is accompanied by the phenomena of deficiency in the sex hormones. The neutralization of hormones causes the hyperactivation of the stimulating hormone and may lead to hypertrophy of the gland;

- the synthetic peptide presenting C-terminal region of β-CE hormone of chorio-gonadotropin of man. Immunization of CHM gives a good effect. The basic problem is individual variability of the immune response;
- immunization by lactate dehydrogenase (LDG-x) of the sperm, which differs from LDG of other body parts of man; this protein is coded by Y-chromosome (women must not have this protein). Data of antibodies were revealed in the serum of some patients with infertility. Drawback – there are created titers of AB, insufficient for complete sterilization;
- immunization of the ovule by antibodies (AB) or by the antigens of the glistening membrane, which surrounds the ovule. ABs to them prevent the attachment of the spermatozoid.

7.5. Diagnostics of reproduction immunopathology

The outcome of pregnancy (normal delivery or abortion) may be prognosticated according to a change in the indices of the total number of T-lymphocytes, Th and Ts. Favorable indices are an increase in the total number of T-lymphocytes and Ts.

Indications to the determination of the immune status:

a. two abortions or two unsuccessful attempts in the artificial fertilization at the age over 35 or three abortions/three failures in the artificial fertilization at the age of up to 35;
b. the insufficient response of the ovaries to the stimulation of the ovulation (less than 6 egg cells);
c. loss of the zygote;
d. infertility of the obscure genesis;

e. diagnosed immunological problems (antinuclear antibodies, rheumatoid arthritis and/or systemic lupus erythematosis);
f. complicated obstetrical anamnesis – delay of the fetus development during the previous pregnancies;
g. one living child and repeated abortions in an attempt to bear the following pregnancies.

Immunogenetic analysis: determination of the agreements of DNA sequence in the married couple (DQ alpha, DQ alfa 2, DQ alpha 4, determination of the leukocyte antibodies).

Reproductive immunophenotype (immunogram, immune status):

1. Determination of the level: CD3, CD4, CD8, CD19, CD56, CD3/IL2-R, CD19/CD5, $CD19^+/CD5^+$. The higher their level is, the greater is production of antibodies to such hormones as estradiol, progesterone, CGM as well as to the serotonin and endorphins.
2. Determination of antibodies to progesterone.
3. Antinuclear antibodies.
4. Antibody to DNA and histones.
5. Antibody to phospholipids (anti-phospholipid antibodies).
6. Determination of a quantity of natural killers.
7. TJ6 protein.
8. Other studies: the determination of lupous anticoagulant, antibodies to the hormones, antibodies to the thyroid gland.

7.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Immune status of pregnant females.
2. Immunodependent forms of infertility in the married couples. Causes and mechanisms of the formation of autoantibodies to sex cells in men and women.

3. Immunopathogenesis of infertility, its diagnosis. Immunological approaches to treatment of infertility.
4. Immune interrelations in the systems "father – mother", "mother – fetus".
5. Causes and immunological mechanisms of formation of anti-sperm autoantibodies in men and women.
6. Autoantibodies for spermatozoids and oocytes, antibodies to them, the methods of determination.
7. Immune conflicts in the system "mother – fetus": diagnosis, treatment, prevention.
8. Immune aspects of contraception.

Practical skills

1. To master the skills of determination the necessity of conducting clinico-immunological study of the married couple on suspicion of immunodependent infertility.
2. To master the principles of immunodiagnosis and immunotherapy of infertility, caused by immunodeficiency diseases in woman.
3. To master the principles of immunodiagnosis and immunotherapy of infertility, caused by anti-gamete immune conflict.
4. To master the principles of immunodiagnosis and immunotherapy of infertility, caused by increased histocompatibility of the married couple.
5. Methods of determining the anti-sperm antibodies.
6. Methods of diagnostics of the reproduction immunopathology.

Tests

1. What of the enumerated factors does not provide fetus protection in mother's organism?
   A. Humoral factors of the mother contributing to development of the immunologic tolerance
   B. Suppressor cells of the endometrium

C. Immunosorption properties of the placenta
D. Complete concordance of the father's and mother's MHC in antigens
E. Changes in the immune system of the pregnant woman

2. Association of repeated miscarriages at late terms of pregnancy and circulating lupus anticoagulant is a diagnostic criterion of:
A. Antiphospholipid syndrome
B. Gestosis
C. Autoimmune abortion disease
D. Excessive alloimmune reactions
E. Inflammatory changes of the placenta caused by increased titer of the antinuclear antibodies

3. Mother's organism loses ability to recognize foreign AG of the fetus and produce blocking AB in:
A. Excessive alloimmune reactions
B. Increased titer of the antinuclear antibodies
C. Antiphospholipid syndrome
D. APLA-antibodies against father's HLA antigens
E. Gestosis

4. Changes in the immune state of the pregnant woman are accompanied by the increase of:
A. The number of B-lymphocytes
B. The number of T-suppressors
C. Activity of T-killers
D. Production of IL-2 and IL-8
E. The number of active phagocytes

5. What infectious disease in men may cause damage of the hemato-testicular barrier and formation of the antisperm antibodies?

A. Rubella
B. Scarlet fever
C. Toxoplasmosis
D. Chlamydiosis
E. Epidemic parotitis

6. Exclusion of the role of antisperm antibodies in infertility genesis begins with:
   A. Determination of concordance of DNA sequence in married couple:
   B. Determination of the antiphospholipid antibodies
   C. Determination of the antinuclear antibodies
   D. Postcoitus test
   E. US examination of the testicles

7. To prognosticate the normal outcome of pregnancy the favorable changes are increased amount of:
   A. CD3 and CD8
   B. CD3 and CD4
   C. CD4 and CD8
   D. CD3 and CD19
   E. CD4 and CD19

8. What is the indication for determination of the immune status (reproductive phenotype) in women over 35?
   A. One miscarriage or one failure of the artificial fertilization
   B. Childless marriage for 6 months
   C. Two miscarriages or two failures of the artificial fertilization
   D. Three miscarriages or three failures of the artificial fertilization
   E. Childless marriage for 12 months

9. The immunologic tests while investigating the reproductive phenotype do not include the determination of:

A. Antibodies to progesterone
B. Antisperm antibodies
C. Antinuclear antibodies
D. Antibodies to DNA and histones
E. Antibodies to phospholipids (antiphospholipid antibodies)

10. As immune methods of contraception we do not use immunization against:
A. One or several sexual hormones
B. Synthetic peptide of the hormone of human choriogonadotropin
C. Spermatozoids
D. Lactate dehydrogenase of the sperm
E. Antigens of the ovocyte

CHAPTER VIII

ANTINEOPLASTIC IMMUNITY

8.1. Classification of oncogenes. Causes of tumors

About 10 million mutant cells are formed in the organism of a healthy human for twenty-four hours. Mutagenesis is accelerated under the effect of the ionizing radiation and toxic chemical substances.

Agents, which cause formation of any tumors, are **oncogens**.

Oncogens that facilitate malignant transformation are called **carcinogens**.

There are 4 groups of oncogens:

1. Chemical:

1) cancerogenic substances – compounds, which probably cause formation of the malignant tumor or increase in the rate of its development: they produce changes in DNA or induce chromosomal aberrations; the compounds of the epigenetic effect produce changes in the proteins, which regulate cell growth; the compounds, which act synergistically with the viruses (derepression of oncogens) or are promotor for the cancerogenic substances;
2) food oncogens;
3) oncogens – hormones (estrogens, glucocorticoids).

2. Physical:

1) ultraviolet radiation;
2) radioactive radiation (X-ray, radioisotopes).

3. Viral:

1) oncogenous of RNA-viruses (retroviruses, oncornaviruses);
2) oncogenous DNA-viruses (viruses of papilloma, EBV, hepatitis C).

4. Genetic (hereditary loss of one or several genes of the tumor suppression of).

Causes of tumors:

- the immunological tolerance;
- the absence of the protective tumor antigens;
- the genetically determined weak reaction to the tumor antigens;
- the insufficiency of the immune supervision of the thymus;
- induction of the peripheral selection of T-lymphocytes by the tumor and dysfunctions of the immunocompetent cells in the tumor focus;
- the disbalance of cytokines, which are produced by infiltrating tumor lymphocytes.

In cases of the expressed cellular infiltration of the tumor stroma – development of the neoplasm is comparatively slow. Tumors with the complete absence of the immunocompetent cells in the stroma grow more rapidly and give metastases early. If at the initial stages of tumor development the signs of antigenic stimulation are noted in the regional lymph nodes – sinuses of histiocytosis – the manifestations of the antineoplastic protection and favourable prognostic sign. They are the clinical manifestation of the tumors is possible in case of consistency of the immune response.

By the sensitivity to the response of the immune system the tumors are conditionally divided into 3 groups:

1. ***Highly immunosensitive tumors*** (melanoma, cancer of the kidney and bladder).
2. ***Moderately immunosensitive tumors*** (lymphoma, cancer of the large intestine).
3. ***Low immunosensitive tumors*** (lung cancer, breast cancer).

8.2. Antineoplastic protection

The immune response consists of the cellular and humoral responses:

- the mononuclear cellular infiltration of tumors;
- the production of antibodies and development of cytotoxic T-lymphocytes;

- the activation of natural killers and macrophages.

The mechanism of primary recognition of the tumor antigens is the least studied. The primary recognition is achieved by antigen-presenting cells. They are activated in response to development of antigens of the tumor cells. These molecules are in the surface structures of the organism cells, but they are screened from recognition. The part of the screened molecules is lost in the malignant transformation. Antigens are released and react with the macrophagereceptors. It is established that the tumor cells stop the expression of the sialic acids, which leads to the release of the remaining carbohydrates (galactose, N-acetylglucosamine, mannose), and they will be actively recognized by the macrophages.

The ***cytotoxic T-lymphocytes (T-killers)***, which distinguish the specific antigens of tumors, are the ***main element of active antineoplastic protection***. The ability of the T-killers to react to the tumor cells depends on the expression of the HLA molecules of class I.

Mechanisms of destruction of the tumor cells:

1) the T-killer binds with the receptor of antigenic recognition with the complex peptide – HLA of class I of tumor cell for which the ions of Mg are necessary → the isolation of the damaging protein – perforin → the monomers of perforin are incorporated in the tumor cell membrane and in presence of Ca ions they are polymerized → they form pores ***("perforin pores")*** → the enzymes of T-killers – granzimes enter the cell through them → induction of apoptosis of the tumor cell;
2) excessive water gets into the cells through "perforie pores" → osmotic lysis;
3) the loss of active metabolites by the cell occurs through "perforin pores" and ionic asymmetry between the cytoplasm and the tissue fluid is impaired → the vital activity of the tumor cell is suppressed;
4) interaction of Fas – ligand of cytolemma of the T-killer with the molecule of Fas of the tumor cell → induction of apoptosis of the tumor cell.

One T-killer is capable of destroying only several tumor cells, after which the reserves of energy and perforins are wasted in it and it perishes.

The T-helpers of type I → synthesis of the factors of the tumor necrosis → interaction with the receptors of the tumors (receptor 55) → apoptosis of the tumor cell is induced, or release of γ-interferon → natural killers and macrophages are stimulated + suppression of angiogenesis of new formation + expression by the tumor of HLA molecules – A is strengthened (the cell becomes accessible for the immunological recognition by the T-killer.

Natural killers. Because of the nonspecificity of recognition they are capable of killing tumor cells without the preliminary sensitization to the tumor antigens. They have the lectine receptors, which recognize the carbohydrate remnants, which are released in the tumor cells. They manifest their effect only under the condition of excited expression of the HLA molecules of class I in the target cells.

LAK-cell (lymphokine activated killers). Tumor cells are destroyed without preliminary recognition of the tumor cells by specific antigens, and absence of the HLA molecules of class I on the cell – target is not obligatory for them. They are formed from "zero" lymphocytes under the effect of IL-2.

Specific antibodies. 4–5 days pass for the antigenic response. The tumor antigens interact with the B-lymphocytes → the activation of the B-lymphocyte → presentation to the immunogenic peptide (fragment of the antigen on the plasmatic membrane in the composition of the HLA molecules of class II) → release of IL-2 by the T-helper (having passed antigen specific activation by the macrophage) → B-lymphocyte is differentiated into the plasma cell → secretion of the antigen specific antibodies → they bind with the tumor cell antigens and visualize them for the cells of the immune system + formation of the immune complexes with the complement activation by the classical way → formation of the membrane attacking complexes with the subsequent osmotic lysis of the cell (however, the background level of the complement of man is insufficient for the development of the antibody dependant lysis of a significant quantity of the tumor cells).

8.3. Factors of immune resistance of tumor cells

1. Low immunogenicity of the tumor antigens (they originate from the organism's own cells – immunological tolerance persists).
2. A change of the antigens in the tumor progression. In the tumor growth genetic differences are accumulated in the tumor cells, which are under different conditions of proliferation → constant development of the new surface antigens, which do not manage to recognize the T-killers.
3. Disbalance between the rate of the tumor proliferation and immunocompetent cells (intensity of the tumor proliferation frequently exceeds the rate of accumulation of the antineoplastic immune factors). The high rate of tumor proliferation is caused by the presence of the large number of receptors to different growth factors (thrombocytic, epidermal, fibroblastic) and stimulating the increase of cytokines (IL-1β, IL-2).
4. Selection of the immunoresistant cells. While the period of the tumor existence increases, the immune response to it decreases (cells that are most sensitive to the immune response are destroyed in the early stages) → the tumor cells are capable of suppressing the HLA expression of class I for avoiding recognition by T-killers. But they become the targets of the natural killers in this case.
5. Development of the dissoluble antigens, which are associated with the tumor (malignantly degenerated cells release substances, which cause induction of "negative" immune response in the organism → work of the organism's immune system is impaired).
6. Rapid catabolism of the antibodies on the tumor cell membrane. The tumor cell can detach its surface antigens in the attack of the antibodies → complex of the antigen-antibody leaves the membrane of the tumor cell earlier than the activation of the complement occurs → it considerably decreases the effectiveness of the immune response + formation of CIC not only neutralizes the antineoplastic antibodies, but also provides the blockade of the re-

ceptors of the antigenic recognition of the immunocompetent cells → it protects the tumor cells from T-killers.

7. Production of the suppressor substances by the tumor (example – transforming growth factor β), which suppresses the reactions of the cellular immunity (Fig. 8.1).
8. Ability to induce apoptosis of the cytotoxic T-lymphocytes (some tumors express FasL, which is capable of inducing apoptosis in

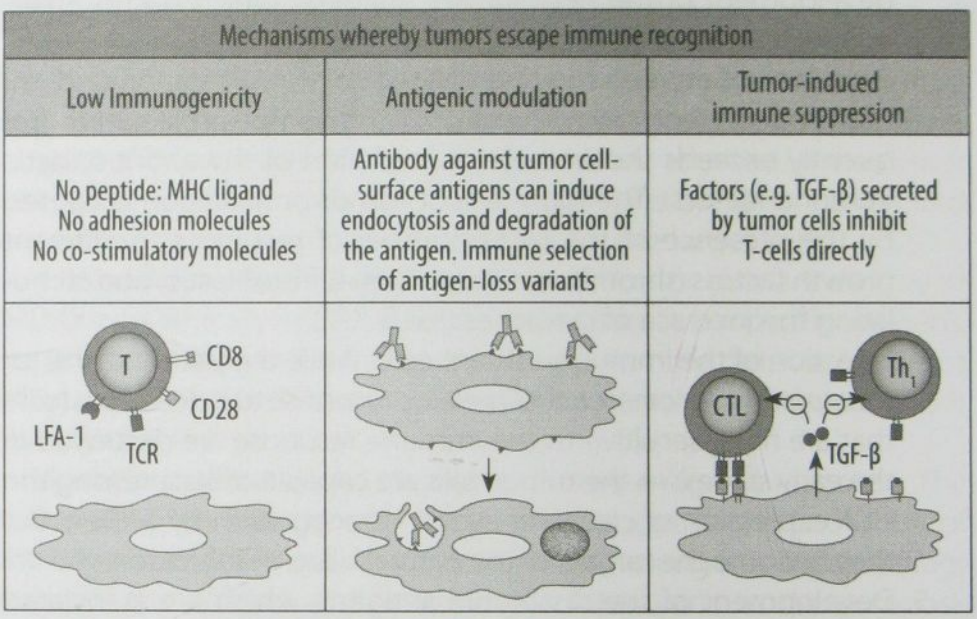

Fig. 8.1. Tumors can escape immune surveillance in a variety of ways. *First, tumors can have low immunogenicity (left panel). Some tumors do not have peptides of novel proteins that can be presented by MHC molecules, and therefore appear normal to the immune system. Others have lost one or more MHC molecules, and most do not express co-stimulatory proteins, which are required to activate naive T-cells. Second, tumors can initially express antigens to which the immune system responds but lose them by antibody-induced internalization or antigenic variation. When tumors are attacked by cells responding to a particular antigen, any tumor that does not express that antigen will have a selective advantage (center panel). Third, tumors often produce substances, such as TGF-β, that suppress immune responses directly (right panel) (Charles A. Janeway et al., Immunobiology, 1999).*

the Fas – positive cells (T-killers) → they perish during interaction with the tumor cells.

9. Expression "of receptor-traps" (TRAIL-3, TRAIL-4 which they correspond to the molecules in the structure, which achieve apoptosis of the cell, but they do not have a domain of death) by the tumor cells. They are activated by lymphocytes for destruction of the tumor cells via apoptosis, but this leads only to strengthening of the protein synthesis, which stimulates the division of the tumor cells (proliferation of tumor).
10. Utilization of the macrophages by a tumor in the role of the peculiar "Trojan horse". Tumor cells synthesize the factor, which suppresses migration of the macrophages (migration inhibiting factor) → macrophages having entered the tumor lose their mobility → they are deprived of the ability to transmit information of the discovered tumor → thanks to which tumor cells obtain the possibility to penetrate the blood stream and to extend in the organism.

8.4. Antigens of superficial structures of tumor cells

1. ***Oncofetal antigens*** (they are present in the fetal cells, but they are absent in adults). They are present in the spontaneous, induced by chemical substances and virus induced tumors (Fig. 8.2). They provide the increased metabolism of the tumor cells. In the tumor cells the genes are activated repeatedly, they function only in the fetal tissue – however, oncofetal antigens are glycoproteins.

Screening method of diagnostics is used: for determination of α-fetoprotein (hepatocellular carcinoma) and cancerous-embryonic antigen (malignant tumors of the large intestine).

2. ***Specific tumor antigens*** (they are present only in the tumor cells). The antigen of the genetic families MAGE and BAGE.

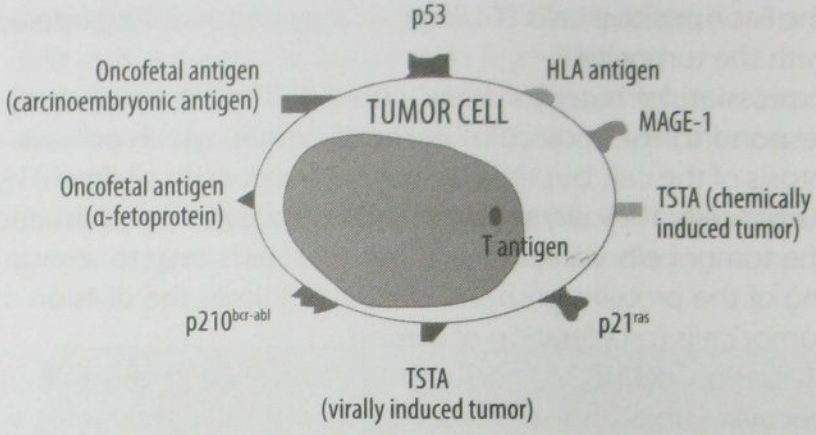

Fig. 8.2. Tumor-induced cell-surface antigens. *The cancerous transformation of cells leads to the expression of different cell-surface antigens The chemically and virally induced tumors express TSTA but also are considered to be TSA (such as MAGE-1, p21^{ras}, p210$^{bcr-abl}$, p53, and T antigen). In contrast, oncofetal antigens are considered to be TATA and TAA (Klaus D. Elgert, Immunology, 1996).*

3. ***Dissoluble tumor antigens*** (they circulate in the organism and they "distract" the immune system – the unjustified use of the immune factors).
4. ***Carbohydrates*** (a set of the carbohydrates of cancerous cells sharply differs from that in normal cells and is a tumor marker).

8.5. Immunodiagnostics and immunotherapy of tumors

- immunophenotyping of hemoblasts,
- immunohistochemical diagnosis,
- radio-immunolocalization of metastases (with the aid of the monoclonal antibodies),
- determination of the oncomarkers (OM).

Oncomarkers

In the opinion of the overwhelming majority of oncologists, the determination of oncomarker in the biological fluids of organism are the most acceptable method for early diagnostics of a primary tumor as well as monitoring of the effectiveness of radiation and chemotherapy. Different tumors discharge different proteins (markers). As a rule, the level of oncomarker does not exceed the normal values of concentration in the blood of a patient with non-oncologic pathology. At the same time, nonspecific, more frequently insignificant increase in the level of the oncomarker of the specific type is sometimes encountered in such pathologic states as the inflammatory diseases of the liver, pancreas, lungs, etc.

The oncomarker allows to differentiate a malignant tumor from a benign one on the basis of quantitative differences in the content of the corresponding antigen – tumor marker in the blood serum independently of the localization of the tumor focus. The tumor cell is capable of releasing 1 picogram (10–12 g) of the oncomarker in the blood of 1 mg of antigen, which concentration is about 200 ng/ml. The procedures of testing very frequently exceed this concentration by their sensitivity. Consequently, the increased level of markers is revealed already in a small size of the tumor.

However, it was impossible so far to develop the strictly tumor-specific serological diagnosticum, capable of detecting only malignant tumor of this histological type and revealing its localization in the earliest possible stages of formation. To a certain extent it is possible to increase the effectiveness of diagnostics by using combinations of different oncomarkers in the process of testing.

Most frequently determined oncomarkers:

1. Alpha-fetoprotein (AFP). It is recommended for detection and monitoring of the course and effectiveness of therapy of primary hepatocellular carcinoma, germinomas, detection of the defects of the fetus development and monitoring of the fetus state during pregnan-

cy. The increased level of AFP is observed in teratocarcinomas of the yolk bag of the ovary or testicles.

Its increased levels are observed in patients with the malignant tumor diseases: primary liver cancer, metastases of the malignant tumors in the liver, cancer of the ovary, cancer of the testicle, cancer of the stomach, cancer of the large intestine, cancer of the pancreas, breast cancer, and bronchial tumors. Its level is increased in the benign diseases: cirrhosis of the liver, acute viral hepatitis, chronic hepatitis, and chronic hepatic insufficiency

2. Neuron-specific enolase (NSE). An increase in its level is noted in patients with small cell carcinoma of the lungs, tumors of the nervous system.

3. Mucin-like carcinoma-associated antigen (CA 15-3). It is used for detection and monitoring of the course of breast cancer, control of effectiveness of its treatment as well as detection of the disease relapse.

The increased level of this marker is observed in approximately 80 % of women in metastasizing CMG, and relapse is accompanied by a substantial increase in its level long before the clinical manifestations.

Its increased levels are also noted in patients with the malignant tumor diseases: cancer of the stomach, liver, pancreas, ovaries, endometrium, and uterus. Increased levels are observed in the benign diseases: benign diseases of the mammary glands, benign diseases of the GIT, chronic non-specific diseases of the lungs (bronchitis and others), physiologically in pregnancy.

4. Prostate-specific antigen (PSA). It is the most sensitive and specific marker. It is used for diagnostics and monitoring of the treatment of the prostate gland cancer.

Its increased levels are noted in patients with the malignant tumor diseases: cancer of the prostate, cancer of the rectum and sigmoid, hepatocellular carcinoma, cancer of the kidneys. Its increased levels are observed in patients with the benign diseases: benign hyperplasia of the prostate, prostatitis, mechanical irritation of the prostate.

5. Chorionic gonadotropin of man (CGM). It is practically an "ideal" tumor marker for detection of some tumors – the sensitivity of this

marker in detection of the carcinoma of the testicle and placenta (chorionepithelioma) is 100 %. In tumors of the uterus there are practically no observed falsely positive results of determining CGM.

In men and non-pregnant women an increase in the CGM concentration is a reliable sign of malignant growth. The CGM determination is recommended for diagnostics, monitoring of the effectiveness of therapy and early detection of the relapses of trophoblastic tumors, chorioncarcinoma of the ovary or placenta, chorionadenomas, seminomas. It manifests the greatest sensitivity to carcinoma of the ovary or placenta.

Its level is increased in patients with malignant diseases: cancer of the testicles, cancer of the ovaries, chorioncarcinoma, cystic mole, cancer of the stomach, liver cancer, cancer of the small and large intestine, cancer of the kidneys, cancer of the ovaries, cancer of the uterus. Its increased indices are noted in benign diseases: physiologically in pregnancy, in women in the menopause with myoma or cyst of the ovary.

6. CA 125. A glycoprotein manufactured by cells of the serous malignant tumors of the ovaries.

It is the marker of monitoring the course and effectiveness of therapy in different types of cancer of the ovaries (serous, endometrial, clear-cell). The test allows to reveal the relapse of the disease 3–4 months prior to its clinical manifestation.

Its level is increased in patients with the malignant tumor diseases: cancer of the ovaries, cancer of the uterus, cancer of the endometrium, breast cancer, cancer of the pancreas, cancer of the rectum and sigmoid, cancer of the stomach. Its increased levels are in benign diseases: benign diseases of the ovaries and endometrium, kidney failure, acute pancreatitis, acute hepatitis, cirrhosis of the liver, physiologically in pregnancy, endometriosis.

7. CEA (cancer-embryonic antigen). An increase in the concentration of CEA is observed in different carcinomas of the digestive tract as well as lung cancer, cancer of the mammary gland, head and neck, malignant neoplasms of the connective-tissue origin. In development of the tumors of different localization the CEA level rises and sufficiently precisely reflects the state of the malignant process.

The CEA index grows slowly, steadily in presence of the oncologic disease.

8. CA 19–9. It is used for diagnostics and monitoring of treatment as well as early detection of cancer of the pancreas, stomach, large bowels, and rectum.

Its level is increased in patients with the malignant diseases: cancer of the pancreas, cancer of the gall bladder and bile ducts, primary cancer of the liver, cancer of the stomach, cancer of the rectum and sigmoid, breast cancer, cancer of the ovary, cancer of the uterus. Its increased levels are observed in the benign diseases: cirrhosis of the liver, acute, toxic and chronic hepatites, cholelithiasis.

9. CA 242. At present it is one of the basic markers utilized for diagnostics and monitoring cancer of the pancreas, cancer of the large intestine and rectum. Specificity is considerably higher than in CA 19–9, it allows to make diagnostics already at the early stages of the disease. According to different data this test helps to prognose development of the relapses of colorectal cancer in 5–6 months.

10. TPA of cyk (cytokeratin) is used for the differentiation of the stable and progressive stages of the disease as well as for the prognostication and follow-up the course of the disease during treatment of patients with epithelial-cellular carcinoma.

11. UBC 11 (cytokeratin) markers of cancer of the bladder. The sensitive indicator of proliferation of the tumor cells in the bladder can be used for evaluation of prognosis of the disease, monitoring and control of the treatment.

12. TPS. TPS level is determined in patients with epithelial-cellular carcinomas, for example, in cancer of the breast, prostate, ovaries and in gastrointestinal carcinoma. It is detected in especially high concentrations in patients with a rapid metastastic spreading. There is a level of prognostic significance before the operation, a high level after chemotherapy correlates with one-year survival.

13. Tumor M2-PK, a metabolic oncomarker. The majority of the human tumors are characterized by production of the isomeric form of pyruvate kinase of tumor M2-PK. Its concentration indicates switching

of cells from the normal type of metabolism to the tumor one, a high correlation is noted with the degree of malignancy (stage of the tumor).

It is a marker of the degree of aggressiveness of the malignant tumor. It reveals the specific type of metabolism of the tumor cells independent of their origin and localization. In contrast to other oncomarkers, it is utilized in the clinical practice, it is not an accumulative, but a metabolic marker, and enters the blood flow early and in a quantity sufficient for the determination. Determination of the content of Tu M2-PK gives the possibility of early diagnostics of metastasis development or relapses of the tumor. (cancer of the kidney, lung, mammary gland, malignant tumors of the gullet, stomach, pancreas, colorectal cancer).

A scheme of administration of oncomarker studies:

- to determine the OM level before the treatment and subsequently to investigate those oncomarkers, which were increased;

Combination of tumor markers

Form of the disease	Marker
the stomach	CA 72–4, CEA
the rectum and sigmoid	CEA, CA 19–9
the lungs	CA 19–9, CEA, AFP
the ovary, the cervix of the uterus	CA 72–4, CA 125, b-CGM
the uterus	AFP, b-CGM
breast cancer	CEA, CA 15–3
the pancreas	CA 125, CA 19–9
liver cancer	AFP, CA 19–9
cystic mole	CA 15–3, b-CGM
the prostate gland: chronic prostatitis, adenoma, cancer	PSA free

- after the course of treatment (operation) to investigate in 2–10 days (according to the period of the half lifetime of the marker) for establishment of the initial level for further monitoring;
- for the estimation of effectiveness in the treatment (operation) to conduct a study in 1 month;
- further study of the OM level in the blood to be made with 3 month interval for 1–2 years, further with the interval of 6 months for 3–5 years;
- to make an OM study before any change in the treatment;
- to determine OM level on suspicion of the relapse and metastastic spreading;
- to determine OM level in 3–4 weeks after the first detection of the increased concentration.

Principles of immunotherapy and immunoprophylaxis of tumors

There are two forms of the immunotherapy of the tumors:

- ***specific*** – induction of development of the specific antitneoplastic reactions;
- ***non-specific*** – restoration of the impaired quantitative and functional indices of the immune system and as a result an increase of the antineioplastic resistance of the organism.

There are several scientific directions of the development of drugs for the immunotherapy of tumors.

Cytokinotherapy is a leading method of the tumor immunotherapy (Fig. 8.3).

Preparations of interferons (α and β) – activate macrophages, natural killers and cytotoxic T-lymphocytes. They strengthen the expression of HLA molecules of class I on the surface of the tumor cells.

Laferon (recombinant IFN-α_2b); **Velferon** (IFN-α_2); **Reaferon** and **Realdiron** (IFN-α_2b); **Roferon A** (IFN-α_2); **Intron A** (IFN-α_2b).

Preparations of interleukin 2 (growth factor of the T-lymphocytes). **Roncoleukin.** Indications: cancer of the kidney, melanoma. It is used

Cytokine therapy for tumors
(I. Roitt et al., Immunology, 2001)

cytokine	tumor type and results	cytokine effects and possible anti-tumor mechanisms
IFN-α	prolonged remissions of hairy-cell leukaemia weak effects on some carcinomas	possible cytostatic effect on tumor; increased expression of MHC class I; cytostasis
IFN-γ	ineffective systemically, remissions of peritoneal carcinoma of the ovary	increased MHC class I and II macrophage activation; Tc activation; cytostasis
IL-2	remissions in renal cancer and melanoma	T-cell activation and proliferation; NK-cell activation
TNF-α	malignant ascites can reduce	increased tumor cell adhesion; macrophage and lymphocyte activation

separately and in combination with *lymph-activated killers* or with *the lymphocytes, which infiltrate a tumor.*

Tumor necrosis factor α causes apoptosis of the tumor cells by interaction with the membrane receptor p55. It is also a powerful activator of macrophages and natural killers. Indications: melanoma – regional perfusions or local introduction.

Application of the monoclonal antibodies. Some antigens of the tumor cellular surface (cancerous-embryonic antigen in tumors of the large intestine, the idiotine antigenic determinants of immunoglobulins in B-cellular lymphomas) may serve as specific targets for the antibodies (Fig. 8.4). **Bevacizumab** (Avastin) – contains monoclonal antibodies to the receptors of the endothelial growth factor, which is produced by the tumor cells for the realization of effective neoangiogenesis (ischemia of tumor). **Rituximab** and **Trastuzumab** are approved for the clinical application in the USA and **Adrecolomab** – in Germany.

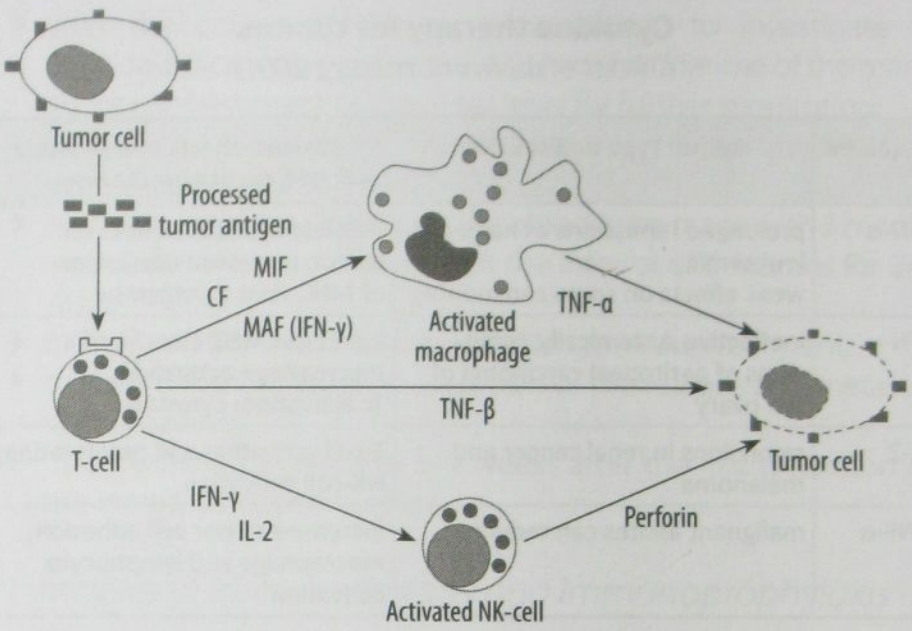

Fig. 8.3. Cytokine involvement in tumor cell killing. *During tumor growth. T-cells are activated by tumor antigens that induce them to release cytokines. T-cells release IFN-γ, lymphotoxin (LT), chemotactic factors (CF) migration inhibition factor (MIF) and macrophage-activating factor (MAF). IFN-γ activates NK-cells, which, in turn, kill tumor cells, while TNF directly destroys tumor cells. The other three cytokines (CF, MIF and MAF) attract macrophages to the tumor site, keep them there, and activate them. Activated macrophages either inhibit tumor cell proliferation or kill (using TNF-α) the tumor cells (Klaus D. Elgert, Immunology, 1996).*

Active specific immunotherapy (genetic therapy) of tumor

The purpose is overcoming of the tolerance of the antineoplastic T-lymphocytes or increase in the activity of the T-cells, which possess a low affinity to the antigens, expressed by the tumor cells. One of the priority tasks of antineoplastic vaccination is provision of the antigen-presenting cells with tumor-specific antigens.

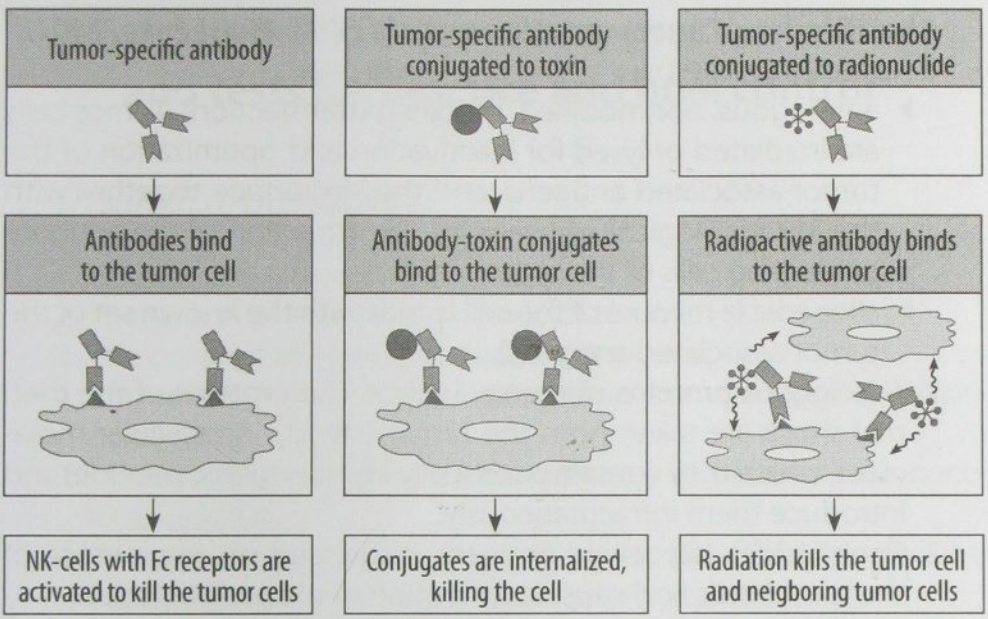

Fig. 8.4. Ways in which a monoclonal antibody or antibody conjugate could eliminate or reduce a tumor. *Monoclonal antibodies that recognize tumor-specific antigens might be used in a variety of ways to help eliminate tumors. Tumor-specific antibodies of the correct isotypes might be able to direct the lysis of the tumor cells by NK-cells, activating the NK-cells via their Fc receptors (left panels). A more useful strategy might be to couple the antibody to a powerful toxin (center panels). When the antibody binds to the tumor cell and is endocytosed, the toxin is released from the antibody and can kill the tumor cell. If the antibody is coupled to a radionuclide (right panels), binding of the antibody to a tumor cell will deliver a dose of radiation sufficient to kill the tumor cell. In addition, nearby tumor cells could also receive a lethal radiation dose, even though they did not bind the antibody (Charles A. Janeway et al., Immunobiology, 1999).*

There were created antineoplastic vaccines with the aid of the introduction of the new genes in the genome of the tumor cell:

- the genes of antineoplastic cytokines or their receptors;
- the tumor-associated antigens;
- suicide genes; foreign antigens or viruses;
- hybrid proteins (tumor-associated antigens and cytokines).

Classification of antineoplastic vaccines (V. M. Moiseenko, 2001).

1. Vaccines on the basis of the one-piece cells:
   - autologous: not modified, modified (transfection). Tumor cells are irradiated or lysed for inactivation and optimization of the tumor-associated antigens, and they introduce together with the immunological adjuvant for the attraction of the antigen-presenting cells of the organism;
   - allogenic (a mixture of the cell tumor with the known set of the tumor-associated antigens).
2. Autologous proteins of thermal shock. The proteins of the thermal shock are taken from the tumor (steady intracellular molecules, which they contain potentially immunogenic peptide) and introduce them intracutaneously.
3. Gangliosides (glycolipid antigens, expressed on the surface of the tumor cells and causing formation of the antibodies).
4. Synthetic tumor-associated peptides.
5. DNA.
6. Recombinant viruses.
7. Vaccines on the basis of the dendritic cells.

Lately considerable attention has been paid to the use of the dendritic cells. The dendritic cells present a system of the heterogeneous antigen-presenting cells, morphology, phenotype and functions of which undergo significant changes during the process of differentiation from hematopoietic $CD34^+$ cells precursors of the bone marrow. The usual or genetically modified dendritic cells (secreting different cytokines) are used.

Vaccination *(immunoprophylaxis)* is made against the virus of hepatitis B, human papilloma virus, virus of Epstein – Barr.

8.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Antitumorous factors.
2. The factors of immunoresistance of tumors.
3. The concept of tumour-associated antigens.
4. Immune changes in oncologic patients. Immunodiagnosis in oncology.
5. Modern approaches to immunotherapy of patients with oncologic diseases.
6. To interpret indices of immunograms in the oncologic patients with estimation of anti-blastoma factors of protection.
7. The main principles of immunodiagnosis of tumors.
8. Oncomarkers

Practical skills

1. To evaluate the results of detection of tumour-associated antigens in the early of immunodiagnosis of tumors and early detection of relapses.
2. Immunodiagnosis, including differential one as to CD phenotype of the tumor cells.
3. To master the principles of immunotherapy of tumors.
4. To master the principles of immunoprophylaxis of tumors.

Tests

1. All of the following components of the immune response are naturally occurring to cancer cells except
   A. NK-cell effect
   B. Specific cellular immunity via lymphocytes

C. Blocking factors that interfere with the immunologic attack on tumor cells
D. Heightened ("turned on") general immune responsiveness
E. Macrophage killing of tumor cells
F. None of these

2. A cancer caused by a virus is called:
A. Hepatoma
B. Rous sarcoma
C. Ataxia telangiectasia
D. All of these
E. None of these

3. What of the following factors favor tumor growth?
A. Cellular immunity and cytotoxic antibodies
B. Cytotoxic antibodies and NK-cells
C. Cellular immunity and activated macrophages
D. Blocking factors
E. None of these

4. Examples of cancer-associated antigens that probably arise from tissue dedifferentiation include:
A. Carcinoembryonic antigen
B. α-fetoprotein
C. A and B are correct
D. Neither A nor B are correct
E. None of these

5. To classify a tumor antigen as an oncofetal antigen, it must be present in regenerating cells or:
A. In fetal or embryonic cells
B. In plasma or serum
C. In cells of endodermal origin
D. In virus-transformed cells
E. None of these

6. Tumor-specific transplantation antigens (TSTA):
A. Are internal antigens

B. Are similar to T antigens
C. Really stands for limo-thy-stuart-thomas-allen antigen
D. Are surface antigens
E. None of these

7. A raised level of *a*-fetoprotein in the blood is associated with:
A. Primary liver cancer
B. Secondary liver cancer
C. Viral hepatitis
D. All of these
E. None of these

8. In treating a cancer patient, you are asked to selectively destroy only tumor cells. What approach is probably the best:
A. Subject the patient to high doses of radiation (radiation therapy)
B. Treat the patient with tumoricidal drugs (chemotherapy)
C. Treat the patient with immunotoxins
D. Treat the patient with cytokines
E. None of these

9. One of the following characteristic is not a characteristic of natural killer (NK) cells:
A. They exhibit immunologic specificity and memory, like T-cells
B. They can kill tumor cells
C. They are lymphocyte-like cells
D. They are similar to plasma cells
E. None of these

10. Lymphokine-activated killer (LAK) cells destroy:
A. Allografts
B. Tumors
C. Virus-infected cells
D. Cells causing autoimmunity
E. None of these

CHAPTER IX

AUTOIMMUNE DISEASES

9.1. The concept of autoimmunity reaction and autoimmune disease

The autoimmune process (autoimmune response) is a form of the immune response, induced by the autoantigenic determinants under the normal conditions and pathology; it is one of the mechanisms of homeostasis maintenance.

The autoantibodies in comparatively low titers are revealed in healthy people, the frequency of positive results constantly increases with age, approximately up to 60–70. The formation of autoantibodies and development of the autoimmune diseases is more frequently observed in women than in men.

The autoimmune disease is a disease of the immune system associated with the disturbance of formation or maintenance of the immunological tolerance, which is manifested in the form the clinically manifested immune-causing self-destruction of the organs and tissues of organism. It is based on the organism loss of the immunological tolerance to the antigens of its own tissues. If the autoantibodies react with the components of one organ, then the pathologic process is of the local nature. In the systemic processes they react with the components of many tissues of organism.

Signs, by which this or that disease may be referred to the autoimmune one, are formulated by L. Vitebsky (1961):

1. Presence of the autoantibodies or cytotoxic T-lymphocytes, directed at the antigen, associated with the present disease.
2. Identification of the autoantigen, at which the immune response is directed.

3. Transfer of the autoimmune process with the aid of the serum, which contains antibodies or cytotoxic T-lymphocytes.
4. Possibility of creation of the experimental model of the disease with the aid of introduction of the autoantigen with development of the corresponding morphological disturbances, characteristic of the disease.

9.2. Etiology and pathogenesis of autoimmune diseases

Basic theories of the pathogenesis of the autoimmune diseases:

1. Theory of "forbidden" clones (most popular today) – at some stages of maturation of the immune system elimination of T- and B-lymphocytes occurs, which possess autoreactivity (producing autoAB). However, if total elimination does not occur, tolerance failure is possible in future.
2. Theory of the sequestered antigens – the definite tissues are protected by the histohematic barriers (sexual glands, the eye tissues, brain, thyroid gland and others). During damage of the histohematic barrier these tissues will be recognized as foreign.
3. Theory of disorder of the immunological regulation: reduction in the function of T-suppressors; an increase in the function of T-helpers.
4. Theory of the polyclonal activation of the B-lymphocytes – in polyclonal activation of the B-lymphocytes autoreactive B-lymphocytes are activated.
5. Theory of development of autoimmunity under the effect of superantigens – a number of bacteria produce super-AG (enterotoxins A, B, C for *Staphylococcus aureus*, erythrogenic toxin for streptococcus, etc.), which can activate the autoreactive T-l and B-l or antigen-presenting cells.
6. Theory of genetic predisposition – there is a the genetically determined predisposition to development of the autoimmune

diseases, controlled by minimum six genes on different chromosomes (their major portion is in the main complex of the HLA histocompatibility of man); majority of the autoimmune diseases are associated with the presence of AG DR2, DR3, DR4 and DR5 in HLA-phenotype of man.

7. Theory of molecular mimicry – the similarity of AG of some infectious agents and autoAG may lead to development of the autoimmune diseases (classical poststreptococcal glomerulonephritis and others).

Mechanisms and causes

The autoimmune disease is a pathologic process, and the autoantibodies and/or cellular autoimmune response play an important role in pathogenesis of it. In the autoimmune response the B-lymphocytes develop autoantibodies against the background of increase in the activity of T- helpers. The factors providing the development of these clones of lymphocytes include: genetic factors, interfering microbial antigens, disturbance in the cytokine network of regulation and factors of the environment. The probability of failure of the mechanisms of the immunological tolerance grows with age.

There are 8 possible versions of development of the autoimmune response (there may be observed 2 of them or more simultaneously):

1. ***Intracellular virus infection*** (virus of Epstein – Barr and others) – "its own" cell, which carries foreign antigens, may be destroyed together with them.
2. ***Medicines and other factors attached to the cells*** (penicillin; malarial agent and others) – "its own" cell, which carries foreign antigens, may be destroyed together with them.
3. ***Interfering antigens*** (streptococci of group A, B; spirochaeta; tripanosoms and others) – in presence of the autoreactive B-lymphocytes the invasive microorganisms having antigenic determinants common with the host, are capable of causing the production of autoantibodies to "their" antigens.

4. ***Interfering idiotops*** – in presence of the autoreactive B-lymphocytes the invasive microorganisms having antigenic determinants common with the host, are capable of causing the production of autoantibodies to "their" antigens.
5. ***Late developed or sequestered antigens*** (lens; sperm) – the antigens, which arose at the later stage of development or which were released from the sequestered tissues, in contact with the immune system are perceived as "strangers".
6. ***Anomalous representation of the antigen*** (thyroid and pancreases) – the presentation of the antigen by the cells, which are not specialized for this function, may lead to autoreactivity.
7. ***Polyclonal activation*** (virus of Epstein – Barr; malaria; tripanosoms; "the reaction: transplant against the host") – Autoreactive B-lymphocytes may be stimulated by directly polyclonal activators bypassing the normal conditions for activation.
8. ***Insufficiency of regulation*** – disturbance of regulation in the idiotype-anti-idiotypic network, in the system of cells – suppressors can lead to the fact that the autoimmune reaction becomes the cause of the disease.

Genetic factors

There is a strict correlation between the autoimmune diseases and specific HLA (Fig. 9.1).

Antigen HLA-B27: the only antigen, which plays a role in diagnostics of the autoimmune diseases. It is frequently revealed in juvenile rheumatoid arthritis, chronic inflammatory diseases of the bowels, reactive arthritis.

Antigen HLA-DR4: in carriers of this antigen rheumatoid arthritis is more frequently accompanied by severe affection of the joints and extraarticular manifestations and has less favorable outcome than in the remaining patients with rheumatoid arthritis.

Autoimmune diseases classified by the mechanism of tissue damage

(Charles A. Janeway et al., Immunobiology, 1999)

Some common autoimmune diseases classified by immunopathogenic mechanism		
Syndrome	**Autoantigen**	**Consequence**
Type II antibody to cell-surface or matrix antigens		
Autoimmune hemolytic anemia	Rh blood group antigens. I antigen	Destruction of red blood cells by complement and phagocytes, anemia
Autoimmune Trombocytopenic purpura	Platelet integrin GpIIb IIIa	Abnormal bleeding
Goodpasture syndrome	Non-collagenous domain of basement membrane collagen type IV	Glomerulonephritis, pulmonary hemorrhage
Pemphigus vulgaris	Epidermal cadherin	Blistering of skin
Acute rheumatic fever	Streptococcal cell-wall antigens. Antibodies cross-react with cardiac muscle	Arthritis, myocarditis, late scarring of heart valves
Type III immune-complex disease		
Mixed essential cryoglobulinemia	Rheumatoid factor IgG complexes (with or without hepatitis C antigens)	Systemic vasculitis
Systemic lupus erythematosus	DNA, histones, ribosomes, snRNP, scRNP	Glomerulonephritis, vasculitis, arthritis
Type IV T-cell mediated disease		
Insulin-dependent diabetes mellitus	Pancreatic β-cell antigen	β-cell destruction
Rheumatoid arthritis	Unknown synovial joint antigen	Joint inflammation and destruction
Experimental autoimmune encephalomyelitis (EAE), multiple sclerosis	Myelin basic protein, proteolipid protein, myelin oligodendrocyte glycoprotein	Brain invasion by CD4 T-cells, paralysis

Associations of HLA serotype with susceptibility to autoimmune disease			
Disease	**HLA allele**	**Relative risk**	**Sex ratio (♀:♂)**
Ankylosing spondylitis	B27	87.4	0.3
Acute anterior uveitis	B27	10.04	< 0.5
Goodpasture syndrome	DR2	15.9	~ 1
Multiple sclerosis	DR2	4.8	10
Graves' disease	DR3	3.7	4–5
Myasthenia gravis	DR3	2.5	~ 1
Systemic lupus erythematosus	DR3	5.8	10–20
Insulin-dependent diabetes mellitus	DR3/DR4 heterozygote	~ 25	~ 1
Rheumatoid arthritis	DR4	4.2	3
Pemphigus vulgaris	DR4	14.4	~ 1
Hashimoto's thyroiditis	DR5	3.2	4–5

Fig. 9.1. Associations of HLA serotype and of sex with susceptibility to autoimmune disease. *The relative risk for an HLA allele in an autoimmune disease is calculated by comparing the observed number of patients carrying the HLA allele with the number that would be expected, given the prevalence of the HLA allele in the general population. For insulin-dependent diabetes mellitus, the association is in fact with the HLA-DQ gene, which is tightly linked to the DR genes but is not detectable by serotyping. Some diseases show a significant bias in the sex ratio; this is taken to imply that sex hormones are involved in pathogenesis. Consistent with this, the difference in the sex ratio in these diseases is the greatest between the menarche and the menopause, when levels of such hormones are the highest (Charles A. Janeway et al., Immunobiology, 1999).*

9.3. Classification of autoimmune diseases

At present the autoimmune diseases include all diseases associated with formation of the autoantibodies, with exception of those cases, when it is possible to assert that the immunological phenomena bear the clearly expressed second nature.

The autoimmune diseases are divided into two groups:

1) ***organ-specific*** – for example, severe myasthenia, Hashimoto's thyroiditis, Grave's disease (thyrotoxicosis with diffuse goiter) and others;
2) ***systemic*** – for example, systemic lupus erythematosis, rheumatoid arthritis and others.

Autoimmune diseases are also divided by the types of the autoreactive T-helpers:

1) ***predominant T-helpers of the 1st type*** (rheumatoid arthritis, reactive arthritis, Wegener's granulematosis, Lyme arthritis, gigantocellular arthritis);
2) ***T-helpers of the 2nd type*** (syndrome of Chaurg – Straus);
3) ***predominant T-helpers of the 2nd type*** (SLE, dermatomyositis, systemic scleroderma, Sjogren syndrome).

Basic autoimmune diseases:

1. Collagenoses (SKV, rheumatoid arthritis, scleroderma, dermatomyositis).
2. Diseases of the skin (Sjogren syndrome, psoriasis, vitiligo, herpetiform dermatitis, pemphigus, bullous pemphigoid).
3. Neurologic diseases (Bekhterev's disease, multiple sclerosis, acute postinfection polyneuritis (Guillain – Barre syndrome), myasthenia).
4. Pathology of the endocrine system (Hashimoto's thyroiditis, Grave's disease (thyrotoxicosis with diffuse goiter), insulin-dependant diabetes mellitus (type I), the autoimmune affection of the adrenal glands (Addison's disease), autoimmune polyendocrinopathy).

5. Sarcoidosis.
6. Idiopathic pulmonary fibrosis.
7. Diseases of the digestion organs (nonspecific ulcerous colitis, Crohn's disease, autoimmune gastritis, primary billiary cirrhosis, chronic active hepatitis, autoimmune enteropathy, celiacia).
8. Diseases of the kidneys (glomerulonephritis, Goodpasture syndrome).
9. Diseases of the genital system (autoimmune orchitis, autoimmune infertility, primary syndrome of antiphospholipid antibodies).
10. Diseases of the eyes (autoimmune uveitis, sympathetic ophthalmia, autoimmune conjunctivitis).
11. Diseases of the blood vessels (nodular periarteritis, gigantocellular granulomatous arteritis).
12. Diseases of the blood (autoimmune hemolytic anemia, autoimmune thrombocytopenia, autoimmune neutropenia and others).

9.4. Principles of diagnostics and treatment of autoimmune diseases

Most significant for making a diagnosis is development of the autoantibodies to the tissue antigens (SLE – to native DNA, RNA, mitochodria; multiple sclerosis – to the myeline; arthritis, vasculitis – to IgG; thyreoditis – to the thyroglobulin; glomerulonephritis, Goodpasture syndrome – to the basal lamina of the renal glomerules; insulin-dependant diabetes mellitus – to insulin or receptors of insulin, etc.).

Immunolaboratory diagnostics:

1. Presence of specific autoantibodies.
2. Presence of the specific cellular sensitization (it is revealed with the aid of the reaction and test of the inhibition of the leukocyte migration in presence of the corresponding autoantigen).
3. Increase in the level of gamma globulin and/or IgG.

4. Change in the quantity of T-helpers and T-suppressors, which leads to an increase in the immunoregulator index.
5. Reduction in the level of C3 and C4 in the affected tissues (IgG, IgM, C3, C4 and fibrin).
6. Lymphoid-cellular infiltration of the affected tissues.
7. Determination of HLA-phenotype.

The determination of cryoglobulins – immunoglobulins of the sera, which reversibly precipitate at a temperature below 37 °C.

The precipitates, which contain both monoclonal (for example, the rheumatoid factor) and polyclonal (for example, IgG) antibodies, are called mixed cryoglobulins. Mixed cryoglobulinaemia is usually manifested by vasculites of the skin. In this case the areas of body subjected to the action of cold, are most frequently affected. Mixed cryoglobulinaemia is characteristic of the autoimmune diseases. It is observed in SLE, nodular periarteritis, Sjogren syndrome and Kawasaki disease. Hepatitis A, B and C are always accompanied by cryoglobulinaemia. Cryoglobulins are also revealed in hemoblastoses, chronic infections and sarcoidosis. When cryo-precipitates contain only monoclonal antibodies, myelomatosis and macroglobulinemia of Waldenstrem are excluded.

Studying of the synovial fluid. On suspicion of rheumatoid arthritis and SLE the hemolytic activity of complement and the rheumatoid factor are determined in the synovial fluid. In rheumatoid arthritis and LE the complement hemolytic activity in the synovial fluid of the affected joint is usually reduced and makes less than 30 % of the normal level in the serum. In majority of other diseases with affection of the joints, the hemolytic activity of complement corresponds to the normal value in the serum or exceeds it.

The rheumatoid factor. Any particles, covered with IgG, can be agglutinated by the rheumatoid factor. Initially the erythrocytes of the ram covered with antibodies were used for detection of the rheumatoid factor. Recently, nephelometry has been used as the method of determination of the rheumatoid factor (it evaluates an increase in the serum turbidity after the addition of IgG to it). The rheumatoid factor is revealed in the autoimmune diseases, which are accompanied by af-

fection of the joints, infectious endocarditis and some chronic diseases of the liver. Predominantly IgM to IgG are revealed with the aid of the nephelometric reaction. Besides IgG and IgA to IgG can also be revealed in the serum. In certain cases the rheumatoid factor is determined only in the synovial fluid, and it is absent in the serum. The development of the rheumatoid factor in the synovial fluid of the affected joints allows to confirm the diagnosis of seronegative rheumatoid arthritis.

The study of the ***antinuclear antibodies*** by the method of immunofluorescence is observed to give the spotty staining of the tissue sections. In some patients there are signs of several autoimmune diseases, but, in contrast to the patients with the mixed disease of the

Some of the autoantibodies to the tissue antigens

Autoantibodies	Disease
IgG; collagen	Rheumatoid arthritis
Native DNA, RNA, mitochodria nucleoprotein	Systemic lupus erythematosus
Thyroglobulin	Hashimoto's thyroiditis; primary myxedema
Ducts, mitochondria, nuclei thyroid; IgG	Sjogren syndrome
Thyroid-stimulating hormone receptors	Thyrotoxicosis (Grave's disease)
Cytoplasm of adrenal cells	Addison's disease
Spermatozoa	Male infertility
Cytoplasm of steroid-producing cells	Premature onset of menopause
Intrinsic factor; parietal cell	Pernicious anemia
Cytoplasm and surface of islet cells	Juvenile diabetes
Glomerular and lung basement membrane	Goodpasture syndrome
Skeletal and heart muscle; acetylcholine receptor	Myasthenia gravis
Colon "LPS"	Ulcerative colitis

connective tissue, there are no antibodies to the ribonucleoprotein. In this case, if the existing signs meet the criteria of several autoimmune diseases, the diagnosis of the cross syndrome is made, but if there are no signs the diagnosis of the undifferentiated disease of the connective tissue is made. The signs, which allow to make the diagnosis of this or that disease, usually develop subsequently: rheumatoid arthritis, SLE, systemic scleroderma, etc.

Principles of treatment of the autoimmune diseases

The methods of modern therapy consist in: directed change in various stages of the immune response (injection of immunoglobulin preparations; the preparations of monoclonal antibodies to CD4, CD25, IL-1, TNF-α and others); include the directed effect on the metabolic reaction of a number of organ-specific diseases; the use of anti-inflammatory drugs (corticosteroids, salicylates, D penicillamine, auric salts, antimalarial preparations); selective immunodepressants (cyclosporine, lobensarit, subrium, tenidap, mycophenolate mofeit, a group of azospiranes); intravenule immunoglobulins; monoclonal antibodies (inhibitors of TNF-α, IL-1, IL-6); immunoadsorption therapy (selesob, immunosorb – Tr50, prosorb, Ig – terasorb) (Fig. 9.2). Plasmaferesis gives a temporary improvement in the state of patients.

9.5. Diagnosis and treatment of some autoimmune diseases

Rheumatoid arthritis is an autoimmune disease that causes chronic inflammation of the joints. Rheumatoid arthritis can also cause inflammation of the tissue around the joints, as well as in other organs in the body. While rheumatoid arthritis is a chronic illness, meaning it can last for years, patients may experience long periods without symptoms. However, rheumatoid arthritis is typically a progressive illness that has the potential to cause joint destruction and functional disability. The

cause of rheumatoid arthritis is unknown. It is believed that the tendency to develop rheumatoid arthritis may be genetically inherited. It is also suspected that certain infections or factors in the environment might trigger the activation of the immune system in susceptible individuals. The symptoms of rheumatoid arthritis come and go, depending on the degree of tissue inflammation. During remissions, symptoms of the disease disappear, and people generally feel well. When the disease becomes active again (relapse), symptoms return. When the disease is active, symptoms can include fatigue, loss of energy, lack of appetite, low-grade fever, muscle and joint aches, and stiffness. Muscle and joint stiffness is usually most notable in the morning and after periods of inactivity. During attacks, joints frequently become red, swollen, painful, and tender. In rheumatoid arthritis, multiple joints are usually inflamed in a symmetrical pattern (both sides of the body affected). The small joints of both hands and wrists are often involved. Since rheumatoid arthritis is a systemic disease, its inflammation can affect organs and areas of the body other than the joints. Inflammation of the glands of the eyes and mouth can cause dryness of these areas and is referred to as Sjogren syndrome. Rheumatoid inflammation of the lung lining (pleuritis) causes chest pain with deep breathing, shortness of breath, or cough. The lung tissue itself can also become inflamed, scarred, and sometimes nodules of inflammation (rheumatoid nodules) develop within the lungs. Inflammation of the tissue (pericardium) surrounding the heart, called pericarditis, can cause a chest pain that typically changes in intensity when lying down or leaning forward.

The diagnosis will be based on the pattern of symptoms, the distribution of the inflamed joints, and the blood and X-ray findings. Blood tests: rheumatoid factor; citrulline antibody (also referred to as anticitrulline antibody, anticyclic citrullinated peptide antibody, and anti-CCP); antinuclear antibody (ANA); tests called the sedimentation rate and C-reactive protein tests. Joint X-rays may be normal or only show swelling of the soft tissues early in the disease. As the disease progresses, X-rays can show bony erosions typical of rheumatoid arthritis in the joints.

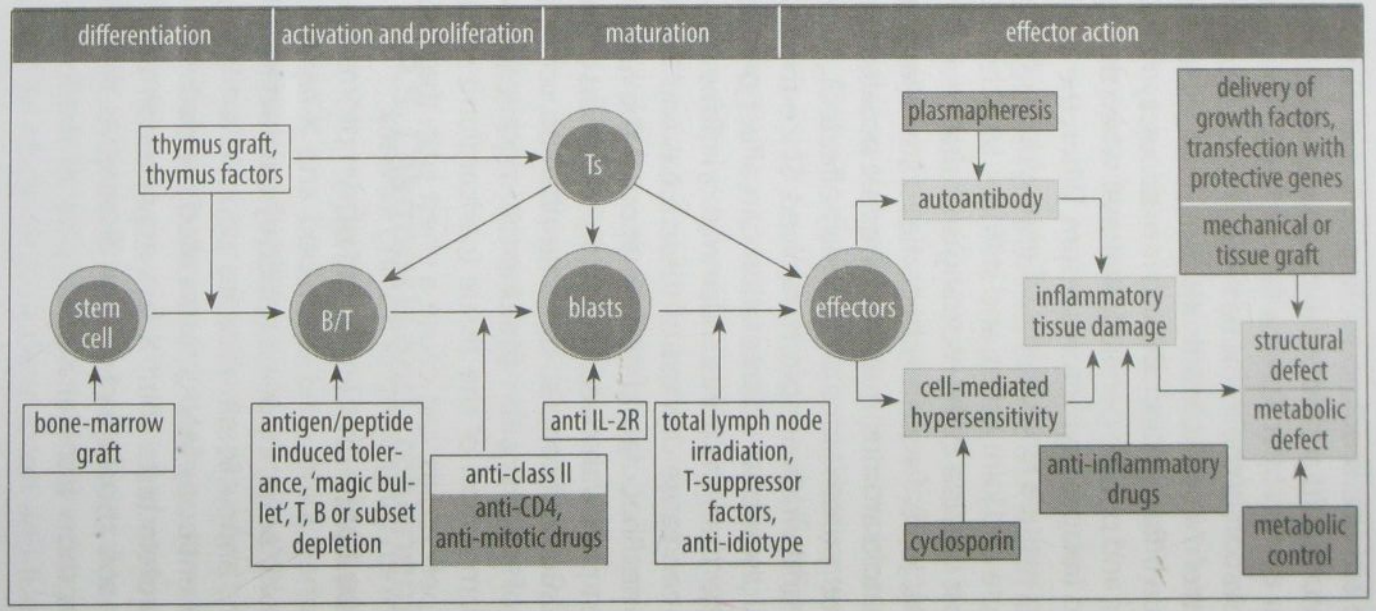

Fig. 9.2. Current and potential treatment of autoimmune disease. *Current treatments for arresting the pathological developments in autoimmune disease are given in grey boxes, and those that may become practicable – in white boxes. Antimitotic drugs are given in severe cases of SLE or chronic active hepatitis, and anti-inflammatory drugs are widely prescribed in rheumatoid arthritis. Organ-specific disorders (e.g. primary myxoedema) can be treated by supplying the defective component (e.g. thyroid hormone). When a live graft is necessary, immunosuppressive therapy can protect the tissue from damage (I. Roitt et al., Immunology, 2001).*

Treatment: "first-line" medications (nonsteroidal anti-inflammatory drugs and corticosteroids); "second-line" or "slow-acting" drugs – disease-modifying anti-rheumatic drugs or DMARDs (hydroxychloroquine, sulfasalazine, gold salts – gold thioglucose, gold thiomalate, auranofin, D-penicillamine, immunosuppressive medicines); the types of joint surgery range from arthroscopy to partial and complete replacement of the joint.

Systemic lupus erythematosis is an autoimmune disease that is characterized by acute and chronic inflammation of various tissues of the body. Both discoid and systemic lupus are more common in women than men (about eight times more common). The disease can affect all ages but it most commonly begins from 20–45 years of age. The precise cause for the abnormal autoimmunity that causes lupus is not known. Inherited genes, viruses, ultraviolet light, and certain medications may all play some role. More than 90 % of cases of "drug-induced lupus" occurs as a side effect of one of the following six drugs: hydralazine; quinidine and procainamide; phenytoin; isoniazid and d-penicillamine. But drug-induced Systemic lupus erythematosis usually resolves when the medications are discontinued. Common complaints and symptoms include fatigue, low-grade fever, loss of appetite, muscle aches, arthritis, ulcers of the mouth and nose, facial rash ("butterfly rash"), unusual sensitivity to sunlight (photosensitivity), inflammation of the membrane that surrounds the lungs (pleuritis) and the heart (pericarditis), and poor circulation to the fingers and toes to cold in exposure (Raynaud's phenomenon). The skin rash in discoid lupus is often found on the face and scalp. It is usually red and may have raised borders. Discoid lupus rashes are usually painless and do not itch, but scarring can cause permanent hair loss (alopecia). The hair loss can be patchy or diffuse and appear to be more like hair thinning. Over half of the people with Systemic lupus erythematosis have a characteristic red, flat facial rash over the bridge of their nose. Most people with Systemic lupus erythematosis will develop arthritis during the course of their illness. Arthritis in Systemic lupus erythematosis commonly includes swelling, pain, stiffness, and even deformity of the small joints of the hands, wrists, and feet. More

serious organ involvement with inflammation occurs in the brain, liver, and kidneys. Inflammation of muscles (myositis) can cause muscle pain and weakness. Vasculitis is characterized by inflammation with damage to the walls of various blood vessels. Inflammation of the membrane of the lungs (pleuritis) and of the heart (pericarditis) can cause sharp chest pain. The chest pain is aggravated by coughing, deep breathing, and certain changes in the body position. Kidney inflammation in SLE can cause leakage of protein into the urine, fluid retention, high blood pressure, and even renal failure. Involvement of the brain can cause personality changes, thought disorders (psychosis), seizures, and even coma. Damage to nerves can cause numbness, tingling, and weakness of the involved body parts or extremities. Brain involvement is referred to as lupus cerebritis.

The diagnosis. 11 criteria are used for diagnosing systemic lupus erythematosus (American Rheumatism Association): malar (over the cheeks of the face) "butterfly" rash; discoid skin rash (patchy redness with hyperpigmentation and hypopigmentation that can cause scarring); photosensitivity (skin rash in reaction to sunlight (ultraviolet light) exposure); mucous membrane ulcers (spontaneous ulcers of the mouth membrane, nose, or throat); arthritis (two or more swollen, tender joints of the extremities); pleuritis or pericarditis (inflammation of the tissue around the heart or lungs, usually associated with chest pain upon breathing or changes of the body position); kidney abnormalities (abnormal amounts of urine protein or clumps of cellular elements called casts detectable in urinalysis); brain irritation (manifested by seizures and/or psychosis); blood-count abnormalities (low counts of white or red blood cells, or platelets in routine blood test); immunologic disorder (abnormal immune tests include anti-DNA or anti-Smith antibodies, falsely positive blood test for syphilis, anticardiolipin antibodies, lupus anticoagulant, or positive LE prep test); antinuclear antibody (positive ANA antibody test – antinuclear antibodies in the blood). Additional criteria: tests called the sedimentation rate and C-reactive protein, blood-chemistry test, direct analysis of internal body fluids, and tissue biopsies.

Ttreatment: nonsteroidal anti-inflammatory drugs; corticosteroids hydroxychloroquine; chloroquine or quinacrine; dapsone and retinoic acid; immunosuppressive medicines; mycophenolate mofetil; plasmapheresis; rituximab.

Myasthenia gravis is an autoimmune disease that causes muscle weakness. The disease affects the neuromuscular junction, interrupting the communication between the nerve and muscle, and thus causing weakness. A person with Myasthenia gravis may have difficulty moving his/her eyes, walking, speaking clearly, swallowing, and even breathing, depending on the severity and distribution of weakness. Increased weakness with exertion, and improvement with rest is a characteristic feature of Myasthenia gravis. The earliest symptoms of Myasthenia gravis often result from weakness of the extraocular muscles, which control eye movements. Symptoms involving the eye (ocular symptoms) include double vision (diplopia), especially when not gazing straight ahead, and difficulty in raising the eyelids (ptosis). A person with ptosis may need to tilt their head back to see. Eye-related symptoms remain the only symptoms in about 15 % of Myasthenia gravis patients. Another common early symptom is difficulty of chewing and swallowing due to weakness in the bulbar muscles, which are in the mouth and throat. Choking becomes more likely, especially with food that requires extensive chewing. Weakness usually becomes more widespread within several months of the first symptoms, reaching their maximum within a year in two-thirds of patients. Weakness may involve muscles of the arms, legs, neck, trunk, and face, and affect the ability to lift objects, walk, hold the head up, and speak. Myasthenia gravis is often diagnosed accurately by a careful medical history and a neuromuscular examinatin, but several tests are used to confirm the diagnosis.

The diagnosis. Thorough physical examination includes: looking upward and sidewards for 30 seconds: ptosis and diplopia; looking at the feet while lying on the back for 60 seconds; keeping the arms stretched forward for 60 seconds; 10 deep knee bends; walking 30 steps on both the toes and heels; 5 situps, lying down and sitting up

completely; "Peek sign": after complete initial apposition of the lid margins, they quickly (within 30 seconds) start to separate and the sclera starts to show. Blood tests: test for antibodies against the acetylcholine receptor and the MuSK protein; spirometry; electromyogram; a chest CT-scan showing thymoma.

Treatment: edrophonium; acetylcholinesterase inhibitors (neostigmine and pyridostigmine); corticosteroids; azathioprine and cyclosporine; thymectomy.

9.6. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. Determination of the concept of autoimmunity, autoimmune disease or syndrome.
2. Mechanisms of tolerance break, the genetic predisposition to development of autoimmune diseases.
3. Classification of autoimmune diseases.
4. Basic theories of the pathogenesis of the autoimmune diseases.
5. Mechanisms and causes of the autoimmune disease.
6. Possible versions of development of the autoimmune response.
7. General principles of immunolaboratory diagnosis of autoimmune diseases.
8. Modern approaches to the application of immunotropic drugs in treatment of autoimmune diseases.

Practical skills

1. Laboratory criteria of immunodiagnosis of autoimmune diseases.
2. To master the skills of evaluation of laboratory results, determination of the immunological criteria of autoimmune pathology.

3. Principles of treatment of the autoimmune diseases.
4. Rheumatoid arthritis (clinical presentations, course, diagnosis, principles of treatment).
5. Systemic lupus erythematosis (clinical presentations, course, diagnosis, principles of treatment).
6. Myasthenia gravis (clinical presentations, course, diagnosis, principles of treatment).

Tests

1. What theory of development of the autoimmune diseases is the most popular today?
   A. Sequestered antigens
   B. Disorders of the immune regulation
   C. "Forbidden" clones
   D. Polyclonal activation of B-lymphocytes
   E. Development of autoimmunity under the influence of superantigens

2. In diagnosis of the genetic predisposition to the autoimmune diseases the main part is determination of HLA antigen:
   A. A3
   B. A9
   C. B7
   D. B27
   E. DR7

3. Systemic autoimmune diseases include:
   A. Nodular periarteriitis
   B. Myasthenia gravis
   C. Sjogren syndrome
   D. Autoimmune uveitis
   E. Addison's disease

4. Pathogenesis of the autoimmune diseases is based on the following types of hypersensitivity reactions:
   A. All 5 types
   B. Only the 2nd and 3rd types
   C. Only the 3rd type
   D. The first 4 types
   E. The 2nd, 3rd and 4th types

5. The main criterion of making a diagnosis of the autoimmune diseases is:
   A. Immunologic methods of investigation
   B. C-reactive protein
   C. Rheumatic tests
   D. Functional methods of investigation
   E. Determination of the blood protein fractions

6. The main criterion of immunodiagnosis of the autoimmune diseases is:
   A. Determination of the rheumatoid factor
   B. Disbalance of the subpopulations of the regulatory T-cells
   C. Determination of the specific autoantibodies
   D. Determination of the HLA-phenotype
   E. Reduction of the level of C3 and C4 complement components

7. Determination of the native DNA, RNA, mitochodria nucleoprotein is made for diagnosis of:
   A. Rheumatoid arthritis
   B. Sjogren syndrome
   C. Pernicious anemia
   D. Systemic lupus erythematosus
   E. Goodpasture syndrome

8. What of the enumerated symptoms is characteristic of rheumatoid arthritis?
   A. Morning stiffness in the joints for over one hour
   B. Asymmetric character of the joint affection
   C. Rheumatoid nodules (subcutaneous nodules on the protruded areas of the bones and periarticular areas
   D. Arthritis of the hand joints
   E. Detection of the rheumatoid factor in the blood serum

9. What of the enumerated symptoms is usually noncharacteristic of systemic lupus erythematosus?
   A. Discoid eruptions
   B. Serosites (pericarditis, pleurisy)
   C. Photosensitization
   D. Nephritis (proteinuria, cylindruria)
   E. Hematologic disturbances (leucocytosis and lymphocytosis)

10. Determination of IgG; collagen is made to diagnose:
    A. Rheumatoid arthritis
    B. Dermatomyositis
    C. Myasthenia gravis
    D. Systemic lupus erythematosus
    E. Thyrotoxicosis (Grave's disease)

CHAPTER X

REACTIONS OF HYPERSENSITIVITY. ALLERGIC (NON-ATOPIC) DISEASES

10.1. Classifications of hypersensitivity reactions

Hypersensitivity response is pathological immune responses, which are based the excessive (hyperimmune) response to exo- and endogenic antigenes (allergens). Because of failure of functioning, the immune system of an organism recognizes antigenes (exogenic or own tissues) not only as foreign but also as serious threat to vital activity of the whole organism. As a result nechanisms of immunosuppression are switched off or inhibited and the hyperimmune process develops, which is uncontrollable (or not fully controllable) by the immune system. Last decade it was considered that the diseases caused by responses of hypersensitivity, arose on the basis of hereditary predisposition in combination with certain conditions of the life style: long excessive antigenic loading; unfavorable influence of various factors of environment or severe infectious diseases on the immune system; unjustified administration of a plenty of medicines, etc.

While characterising ***the antigenes,*** which have caused hypersensitivity response in an organism, it is accepted to use the term ***"allergen".***

The classification according to Gell and Coombs (1968)

The first three types are related to the immediate type of allergic reactions, their influence after repeated contact with the allergen develops from 15-20 minutes to several hours. **The fourth type of allergy** (delayed-type hypersensitivity) develops in 1–2 days.

10.2. Pathogenesis of hypersensitivity reactions

The first type of hypersensitivity reactions

The first type of the reaction – allergic reaction or the reaction of the anaphylactic type of hypersensitivity. It is based on the reagin mechanism of the tissue damage, which usually takes place with the participation of immunoglobulins E, or rarely of immunoglobulins G_4 on the surface of the membranes and mast cells (Fig. 10.1).

Anaphylactic type: *anaphylactic shock, hives, pollinoses, atopic bronchial asthma, Quincke's edema, allergic rhinitis, atopic dermatitis (neurodermatitis).*

Anaphylaxis (from the Greek. *ana-* – again and *aphylaxía* – unprotectedness) is a pathologic process, which develops in man or mammal during the introduction of foreign substances of protein nature – antigens (anaphylactogens) – into the organism, usually bypassing the digestive tract. The first entry of anaphylactogen into the blood causes production of specific antibodies to it, which occurs without visible clinical manifestations (Fig. 10.2). The repeated introduction of the

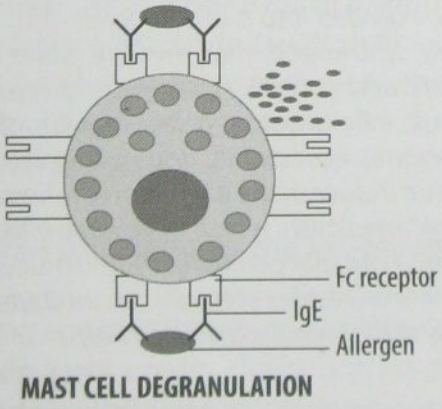

Fig. 10.1 Immune malfunctions leading to tissue damage: type I – immediate (anaphylactic) hypersensitivity. *On primary exposure to allergen (antigen), excess amounts of IgE are produced which bind to mast cells by their Fc receptors. Secondary exposure of allergen leads to cross-linked IgE molecules, causing degranulation and vasoactive mediator release (Klaus D. Elgert, Immunology, 1996).*

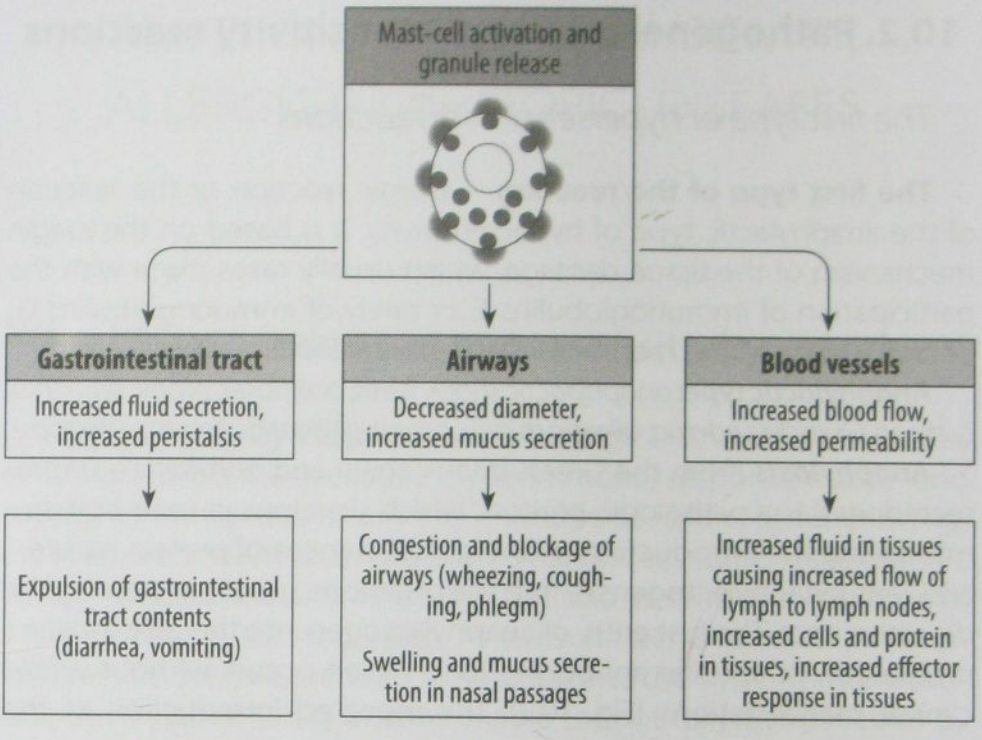

Fig. 10.2. Mast-cell products have different effects on different tissues. *Mast-cell products can be divided into two categories: first, those molecules, both preformed and rapidly synthesized, that mediate acute inflammatory events after mast-cell activation; and second, cytokines and lipid mediators, which induce a late-phase chronic inflammatory response with influx and activation of Th_2-lymphocytes, monocytes, eosinophils, and neutrophils. There is some overlap between mediators that induce acute and chronic Inflammatory responses, particularly among the lipid mediators, which have rapid effects causing smooth muscle contraction, increased vascular permeability, and mucus secretion, and also induce the influx and activation of leukocytes, which contribute to the late-phase response (Charles A. Janeway et al., Immunobiology, 1999).*

same anaphylactogen after the establishment of sensitization leads to rapid (in several seconds or minutes) development of the anaphylactic reaction. The development of anaphylaxis both in man and animals consists of three sequential processes: sensitization (preparation); anaphylactic shock (resolution); desensitization.

Most frequently anaphylaxis is mediated by the immune mechanisms with the participation of the antibodies of immunoglobulin (Ig)E or by the complex antigen-antibody. If the reaction is not caused by interaction of the antigen-antibody, then it is considered to be anaphylactoid. Idiopathic anaphylaxis may be also observed.

Most frequent causes of anaphylaxis, mediated by the IgE-mechanisms are:

- Drugs (antibiotics of the penicillin series, aminoglucosides, streptomycin, nitrofurans, sulfanilamides, tetracycline, amphotericin B).
- Hormones (insulin, adrenocoritcotropic hormone (ACTH), parathyroid hormone, corticotropin, progesterone).
- Enzymes (trypsin, streptokinase, chymotrypsin, penicillinase).
- Antiserum (tetanus, diphtherial, antilymphocytic globulin).
- Poison and saliva (hymenopterous, snakes, ants).
- Vaccine (tetanus, containing the egg white (influenza), allergovaccines).
- Food products (nuts, fish, egg, bean, etc.).
- Others (latex, proteins of man or animals, polysaccharides).

The complement-mediated mechanisms of anaphylaxis:

- the transfusion reactions associated with deficiency of IgA;
- cytotoxic (cellular-fixed antigens, transfusion reactions to the cellular elements, IgG, IgM);
- aggregative (intravenous Ig).

Causes of anaphylaxis mediated by the IgE-independent mechanisms:

- Histamine releasing agents (opiods, muscular relation drugs, vancomicin, ciproflaxin, pentamidin, radiocontrast preparations, the inhibitors of the angiotensinconverting enzyme, dextran).
- Aspirin and nonsteroid antipyretic remedies (mediated through the arachidonic way of metabolism).
- The physical factors: physical load; temperature (cold, heat).
- Idiopathic factors.

- Undifferentiated somatic idiopathic anaphylaxis (mono-organ symptoms imitating anaphylaxis).

The clinical manifestations of anaphylaxis are associated with a definite "shock organ", in which the immune responses occur; the level of chemical mediators released from the effector cells; increased sensitivity to these substances.

Anaphylactoid reaction does not have the clinical signs, which allows to distinguish it from anaphylaxis.

The second type of hypersensitivity reactions

The second type of the reaction is characterized by the reaction of the antibody with the antigen on the surface of the host cell, which causes the destruction of this cell.

There may be simple chemical substances, components of the cellular membrane, the noncellular structures – ***haptens*** (Fig. 10.3). The condition of development is excess of the antigen, which is attached to the cells (for example, to the erythrocytes). Shock cells – most frequently they are cells of the blood, endothelium of the vessels, hepatocytes, epithelium of the kidneys. When the allergen is attached to the cells, their surface becomes antigenically foreign, resulting in humoral immune response; immunoglobulins G are synthesized, cells are destroyed, and the allergen disappears. This phenomenon was called the antibody-dependant cytotoxicity (Fig. 10.4).

Example: analgin-induced agranulocytosis. In some people the introduction of analgin leads to the fixation of its molecules on the cellular membrane of the precursor cells of leukocytes. It causes change in the structure of the cellular membrane and formation of the complex antigen, which becomes foreign for the organism, and habitual immune response develops to it with the formation of the antibodies related to IgG. The antibodies bind with the antigen with the formation of the immune complex, which leads to the activation of the complement system, phagocytosis, antibody-dependant cytotoxicity is induced, and finally the process comes to the end with the destruction of the target cell.

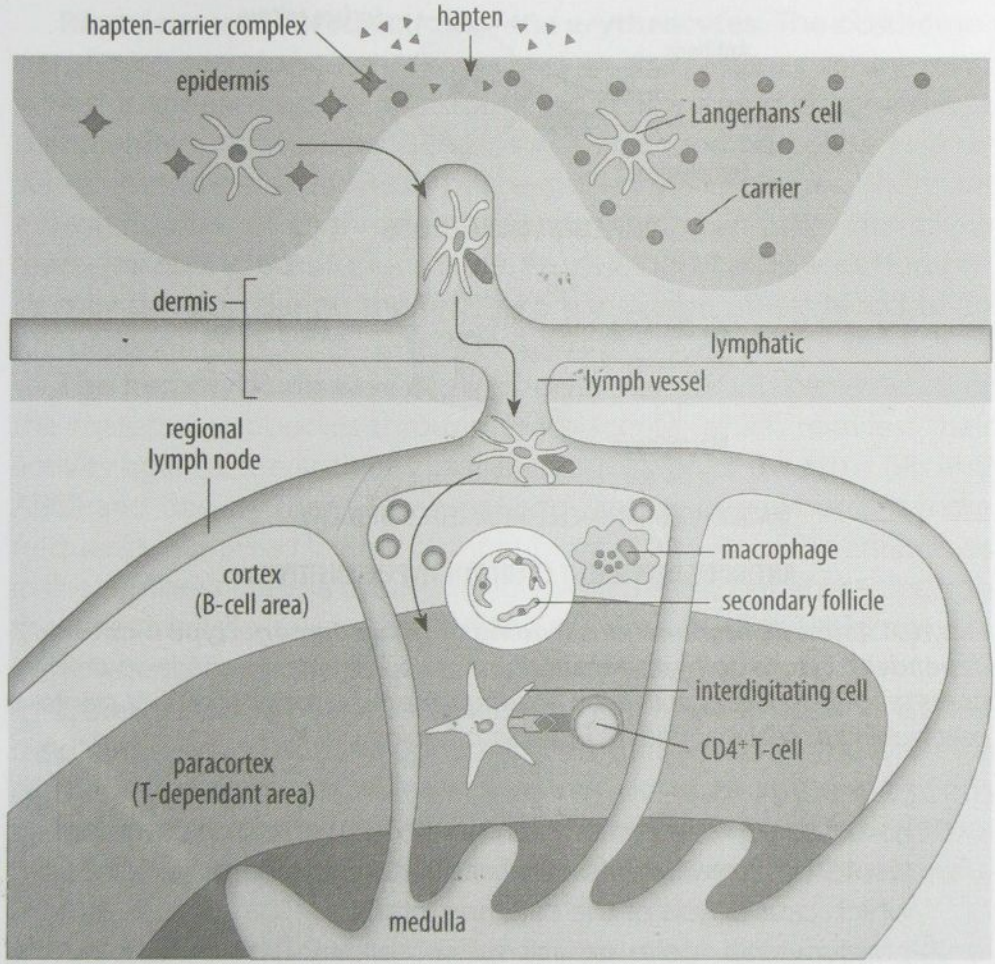

Fig. 10.3. The hapten forms a hapten-carrier complex in the epidermis. *Langerhans' cells internalize the antigen, undergo maturation, and migrate via afferent lymphatics to the paracortical area of the regional lymph node where peptide/MHC complexes on the surface of the Langerhans' cell can also be directly haptenated. As interdigitating cells, they present antigen to CD4+ T-cells (I. Roitt et al., Immunology, 2001).*

The specific antibody, usually IgG or IgM, synthesized against the antigen, interacts with it on the surface of the cell and causes damage of the cell in several ways:

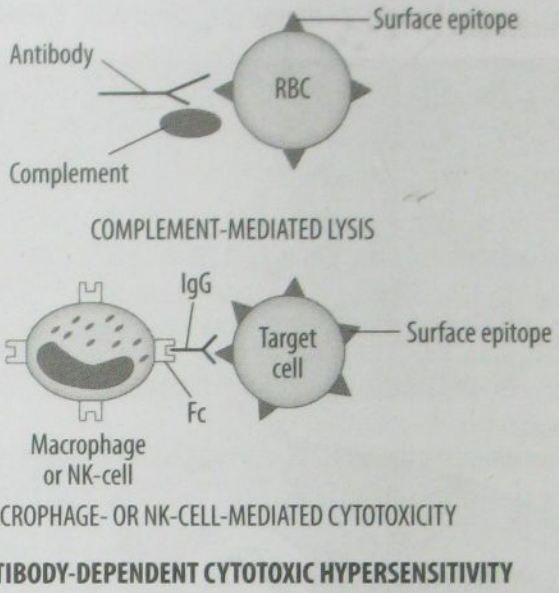

Fig. 10.4. Immune malfunctions leading to tissue damage: type II-antibody-dependent cytotoxic hypersensitivity. *Antibodies bind to antigenic determinants on cells, inducing activation of complement, phagocytic cells, or cytotoxic cells, leading to cell lysis (Klaus D. Elgert, Immunology, 1996).*

1) lysis of the cell – the activation of the cascade of the complement results in formation of "membrane attacking complex" of C5b-9, which causes lysis of the cell membrane;
2) phagocytosis – the cell carrying antigen is absorbed by the phagocytizing macrophages, lymphocyte-killers, cytotoxic lymphocytes, which have Fc or C3b receptors; it allows them to recognize complexes of the antigen-antibody on the cell.

The manifestations of the reaction of type II hypersensitivity depend on the type of the cell, which carries antigen.

Cytotoxic type: *medicinal allergy, immune leucopenia; the hemolytic disease of newborns with the rhesus-conflict, the autoimmune hemolytic anemia and thrombcytopenia, Goodpasture syndrome, chronic active hepatitis, ulcerous colitis, Sjogren syndrome, myasthenia, pemphigus, some cases of juvenile diabetes mellitus and other diseases.*

Reactions with destruction of the erythrocytes. The posthemotransfusion reactions – the antibodies in the patient's serum react with the antigens on the transfused red cells, causing either indirect complement intravascular hemolysis or extended hemolysis as a result of immune phagocytosis by the spleen macrophages. There are a large number of erythrocyte antigens, which can cause hemolytic reactions during transfusions (ABO, Rh, Kell, Kidd, Lewis, etc). Hemolysis may develop during the repeated transfusion of Rh^+ blood to an Rh^- patient.

The hemolytic disease of newborns develops in penetration of the maternal antibodies through the placenta, which manifest their activity against the antigens of the erythrocytes of the fetus (Rh and ABO) and destroy them. The hemolytic disease of newborns is more frequently observed in Rh-incompatibility, as anti-Rh of the antibodies in the mother's plasma usually are IgG, which easily penetrate through the placenta. IgM is usually anti-A and anti-B antibodies, which cannot penetrate through the placenta in the normal conditions.

Other hemolytic reactions – hemolysis may be caused by the medicines, which act as haptens in combination with the proteins of the erythrocyte membrane or it may develop in the infectious diseases, associated with development of antierythrocyte antibodies, for example, in infectious mononucleosis, mycoplasmal pneumonia.

Reactions with destruction of the neutrophils – maternal antibodies to the antigens of the fetus, neutrophils may cause neonatal leucopenia, if they penetrate through the placenta. Sometimes the posttransfusion reactions arise because of the activity of the host serum against the leukocyte HLA of the donor's antigens.

Reactions with destruction of the thrombocytes – the posttransfusion feverish reactions and neonatal thrombocytopenia can arise as a result of the factors, described above for the leukocytes. Idiopathic thrombocytopenic purpura is a frequent autoimmune disease, in which the antibodies are formed against their own antigens of the thrombocyte membrane.

Reactions on the basal membrane – antibodies against the antigens of the basal membranes in the renal glomerules and the pulmonary alveoli develop in the Goodpasture syndrome.

Inhibition – the inhibiting antibodies play a key role in severe myasthenia (*myasthenia gravis*) – the disease, which is characterized by the disturbance of the neuromuscular transmission and development of muscular weakness. The disease is caused by the antibodies (IgG), directed at the acetylcholine receptors on the motor endplate. Antibodies compete with the acetylcholine for the place of binding on the receptor, thus blocking the transmission of the nerve impulse. The mechanism of inhibition is also the basis of pernicious anemia, in which the antibodies bind with the internal factor, and they inhibit the absorption of the vitamin B_{12}.

The third type of hypersensitivity reactions (Arthus reaction)

The third type of the reaction (immunocomplex reaction) is a reaction of hypersensitivity, caused by the formation of the precipitating complexes antigen-antibody in the small excess of antigens. Complexes are deposited on the vessel walls; they activate the complement system and cause the inflammatory processes (for example, serum disease, immunocomplex nephritis) (Fig. 10.5). The reaction mechanism is associated with the damage of tissues by the immune complexes circulating in the bloodstream, with the participation of immunoglobulins G and M (Fig. 10.6).

Arthus reaction – damage by the immune complex: *exogenous allergic alveolitis (farmer's lung); exogenous allergic conjunctivitis, Serum sickness; immunocomplex glomerulonephritis, rheumatoid arthritis; systemic lupus erythematosis and others.*

Type III of hypersensitivity develops in formation of the large number of the immune complexes or during impairment of their elimination by the reticuloendothelial system. Quite large complexes are assimilated by phagocytes after interaction with the complement and

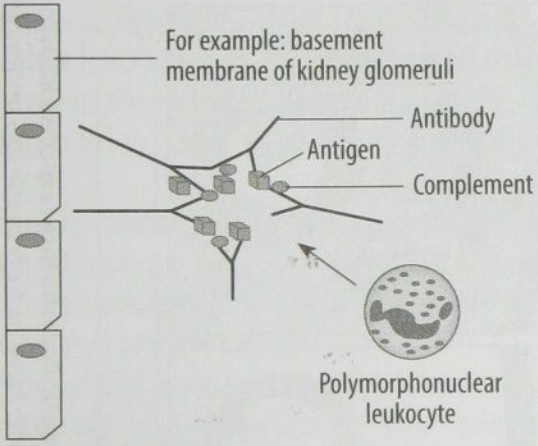

Fig. 10.5. Immune malfunctions leading to tissue damage: type III – immune complex-mediated hypersensitivity. *Interactions between antibodies and antigens can lead to immune complexes that are deposited in the tissue. Complement is often fixed, inducing inflammatory reactions because of neutrophil attraction to the site of deposition, causing tissue damage (Klaus D. Elgert, Immunology, 1996).*

then they are excreted from the organism. At the same time small complexes, which are formed under the conditions of antigen excess, may be sorbed in different organs and tissues. The resulted damage intermediated by the complement and effector cells are called the disease of the immune complexes or reactions of hypersensitivity of type III. The large weakly dissoluble complexes can also be deposited in the tissues in deficiency of the complement.

Thus, hypersensitivity of type III is realized under the conditions when the soluble antigens of serum interact with their antibodies with formation of the aggregates (immune complexes) in the definite tissues areas.

The diseases caused by formation of the immune complexes may be divided into three big groups:

- associated with the persistent infection;
- associated with the autoimmune diseases;
- associated with inhalation of the antigenic material.

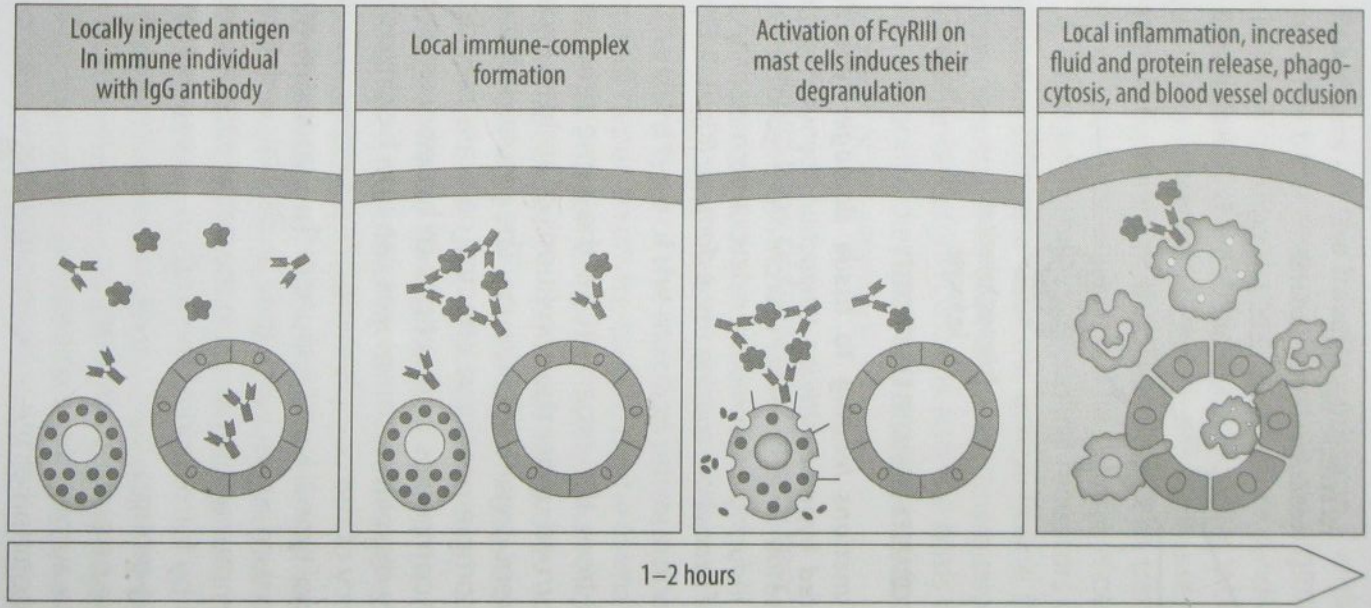

Fig. 10.6. The deposition of immune complexes in local tissues causes a local inflammatory response known as an Arthus reaction (type III hypersensitivity reaction). *In individuals who have already made IgG antibody against an antigen, the same antigen injected into the skin forms immune complexes with IgG antibody that has diffused out of the capillaries. Because the dose of antigen is low, the immune complexes are only formed close to the site of injection, where they activate Fc receptor-bearing mast cells. As a result of mast-cell activation, inflammatory cells invade the site, and blood vessel permeability and blood flow are increased. Platelets also accumulate inside the vessel at the site, ultimately leading to vessel occlusion (Charles A. Janeway et al., Immunobiology, 1999).*

In the first case the combination of chronic infection with the weak humoral response leads to a constant formation of the immune complexes and in the end to their deposit in the tissues (in leprosy, malaria, dengue hemorrhagic fever, virus hepatitis and staphylococcal endocarditis).

In the autoimmune diseases the disease of the immune complexes is caused by the continuous production of antibodies to the autoantigens (diseases with this etiology include rheumatoid arthritis, systemic lupus erythematosis and polymyositis).

During inhalation of the antigenic material the immune complexes can be formed on the surface of cavities (for example, in the lungs during the repeated inhalation of the antigenic components of actinomycetes as well as the antigens of plant or animal origin; the diseases with this etiology include pulmonary disease of farmers, pulmonary disease of the pigeon breeder).

The fourth type of hypersensitivity reactions – delayed-type hypersensitivity (DTH)

The fourth type of the reaction is the reaction of the cell-dependent type of hypersensitivity (the cellular reaction or hypersensitivity of the retarded type). The reaction caused by macrophages and Th_1-lymphocytes, which answer for the stimulation of the cellular immunity (Fig. 10.7). It develops mainly in 1–3 days after the effect of the allergen: there is consolidation and inflammation of the tissue, resulting in its infiltration by T-lymphocytes and macrophages. Any organs and tissues may be involved in the process. Skin, gastrointestinal tract, respiratory organs are more frequently affected in development of the fourth type of allergic reactions.

Three examples of type IV reaction:

Contact hypersensitivity is characterized by a reaction at the site of contact with the allergen. It is an epidermal response most often elicited by small molecules called haptens. The cell involved in antigen

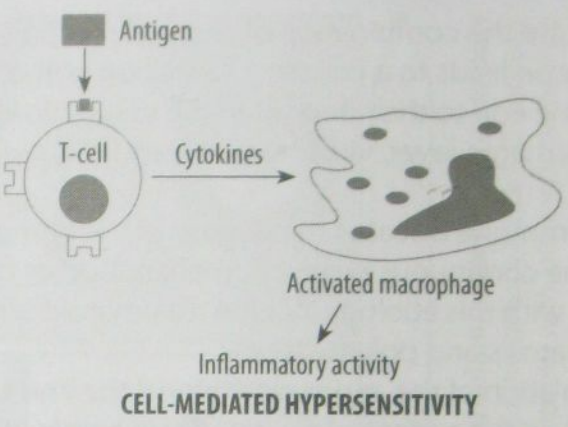

Fig. 10.7. Immune malfunctions leading to tissue damage: type IV – cell-mediated (delayed-type) hypersensitivity. *Following secondary exposure to the same antigen, this tissue damage is mediated by antigen-sensitized T-cells through release of cytokines. The cytokines attract and activate macrophages that release mediators, leading to inflammatory reactions (Klaus D. Elgert, Immunology, 1996).*

presentation at this site is the Langerhans' cell. The pathway from initial exposure involves sensitization (with Langerhans' cells presenting allergen to $CD4^+$ helper T-cells) and exposure, followed by aggregation of mononuclear cells around the blood vessels and glands in the epidermis and edema. A variety of cytokines are involved in this process, including IL-2, IL-3, IFN and GM-CSF. The reaction decreases 48–72 hours following exposure.

Tuberculin hypersensitivity was first observed when soluble antigens from organisms such as mycobacteria were administered subcutaneously. In these individuals fever, general malaise, plus an area of red, hard swelling was observed. The skin test for tuberculosis is of this nature. This reaction is induced by a series of cellular migrations and activations: T-cell migration from capillaries; disruption of collagen in dermis; macrophage infiltration; lymphocytes and macrophages express HLA-DR; granulomatous appearance, no edema, self-limitation.

Granulomatous hypersensitivity is characterized by persistence of the antigen within macrophages as well as of the lesion. Such antigens

are particulate matter such as talc and silica but also mycobacteria. The characteristic cells found in the lesion are epitheloid cells (probably macrophages) and giant-cells (multi-nucleated macrophages). The granuloma consists of a hard core of cells sometimes with a necrotic core. This is surrounded by lymphocytes with a deposition of collagen fibres.

Delayed-type hypersensitivity is the basis of the following allergic reactions: *contact dermatitis; brucellosis; infectious-allergic rhinitis; rejection reaction of the transplant and others.*

The most important diseases with the granulomatous reactions of the delayed-type hypersensitivity are leprosy, tuberculosis, shistosomosis, sarcoidosis, Crohn's disease.

Infectious diseases (brucellosis, glanders, tularemia, tuberculosis, leprosy, toxoplasmosis, many mycoses and others) are accompanied by development of DTH; therefore skin-allergic tests with the allergens of causative agents are used in diagnostics (by tuberculins, brucellin, malleinom, tularin, toxoplasmin and others).

Mechanisms: ***chemotaxis*** is the directed migration of the cells in response to the production of the specific chemotaxic factors; inhibition of macrophages – macrophages (phagocytizing cells) lose mobility and cannot leave the zone of reaction; the activation of phagocytes – activation of the cells, which are specialized in the absorption of the cellular material of endogenous and exogenous origin. The blastogenic effect is the stimulation of proliferation and differentiation of the organism cells. Delayed-type hypersensitivity is the immune response, which develops in 48–72 hours after contact with the antigen (Fig. 10.8).

Tuberculin test is the prototype of this form of response.

The fifth type of hypersensitivity reactions – stimulating type

Five types of allergic reactions (or the reactions of hypersensitivity) are distinguished today.

The fifth type of the reaction is a reaction of hypersensitivity, in which the antibodies exert the stimulating influence on the function

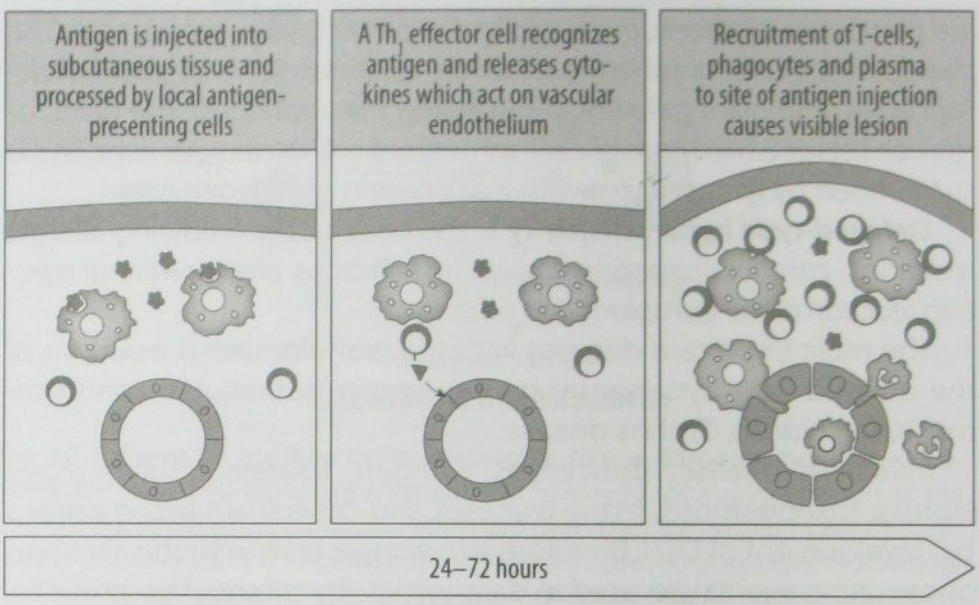

Fig. 10.8. The stages of a delayed-type hypersensitivity reaction. *The first phase involves uptake, processing, and presentation of the antigen by local antigen-presenting cells. In the second phase. Th_1-cells that were primed by a previous exposure to the antigen migrate into the site of injection and become activated. Because these specific cells are rare, and because there is no inflammation to attract cells into the site, it can take several hours for a T-cell of the correct specificity to arrive. These cells release mediators that activate local endothelial cells, recruiting an inflammatory cell infiltrate dominated by macrophages and causing the accumulation of fluid and protein. At this point, the lesion becomes apparent (Charles A. Janeway et al., Immunobiology, 1999).*

of cells. The example of this reaction is thyrotoxicosis related to the autoimmune diseases, in which the overproduction of thyroxin occurs due to the activity of specific antibodies. ABs participate in the process in this type, directed at the different components of the cellular surface, for example, the receptors of the physiological mediators (acetylcholine, adrenoreceptors, receptors for the hormones, etc). AB in the type V of the allergic reactions is more frequently related to the immunoglobulins of the class IgG. Bronchial asthma, diabetes mellitus,

autoimmune diseases of the endocrine glands and others may have a course of the type V.

10.3. Diagnostics and treatment of some allergic (non-atopic diseases)

Goodpasture syndrome is an autoimmune disease that can damage the lungs and kidneys. Why this happens – it is not fully understood. Researchers have identified a number of possible causes, among them the presence of an inherited component: exposure to certain chemicals, including hydrocarbon solvents and viral infections. First signs may be vague, such as fatigue, nausea, difficult breathing, or paleness. These signs are followed by kidney involvement, represented first by small amounts of the blood and protein in the urine, and other clinical and laboratory findings. Goodpasture syndrome may last only a few weeks or as long as 2 years. Bleeding in the lungs can be very serious and even fatal in some cases. But Goodpasture syndrome does not usually lead to permanent lung damage. Damage to the kidneys, however, may be long-lasting. To diagnose Goodpasture syndrome use a blood test, but a kidney or lung biopsy may be necessary to check for the presence of the harmful antibodies. Goodpasture syndrome is treated with oral im-

Form of delayed-type hypersensitivity

Form of DTH	Reaction time	Histology	Histology
Contact	48–72 hrs	Lymphocytes, later macrophages	Eczema. Edemas
Tuberculin	48–72 hrs	Lymphocytes, monocytes, macrophages	Local induration
Granulomatous	21–28 days	Macrophages, epithelioid cells, giant cells. Fibrosis	Consolidation in the skin, lungs and others

munosuppressive drugs-cyclophosphamide and corticosteroids, plasmapheresis.

Sjogren syndrome is a chronic, slowly progressive, inflammatory autoimmune disorder characterized by the infiltration of specialized cells of the immune system called lymphocytes (T-cells in the majority of cases), monocytes, and plasma cells into the parotid (salivary) glands and lacrimal (tear) glands. Two distinct forms of Sjogren syndrome have been recognized. Primary Sjogren syndrome – defined as dry eye and dry mouth that occurs by itself and is not associated with another autoimmune disorder. Secondary Sjogren syndrome – characterized by dry eye and dry mouth that occurs in the presence of a major underlying autoimmune connective tissue disease such as rheumatoid arthritis, systemic lupus erythematosus, or scleroderma. The classic symptoms of Sjogren syndrome affecting the oral cavity and the eyes. There are also systemic or extraglandular symptoms of Sjogren syndrome: sinus symptoms; cutaneous (skin) symptoms; vasculitis; pulmonary symptoms; kidney/bladder symptoms; gastrointestinal symptoms; neurological symptoms; gynecological symptoms. Sjogren syndrome is diagnosed based on factors such as symptoms, patient's history, physical examination, and examination of the oral cavity and eyes. Special laboratory tests that are used to confirm the diagnosis of Sjogren syndrome are designed to measure abnormalities in the production of saliva and tears. At present there is no known cure for Sjogren syndrome. The major goals of treatment are to control the symptoms and prevent or limit the involvement of other organs of the body. The treatment that better manages the symptoms associated with dry mouth is: saliva substitutes; saliva stimulants; interferon-alpha; good oral hygiene practices. The treatment that better manages dry eye symptoms is: artificial tears; methylcellulose inserts; eye ointments; muscarinic agonist drugs; immunomodulatory drugs; autologous serum drops; punctual occlusion surgery. The treatment includes systemic or extraglandular symptoms of other body organs such as the skin, lungs, kidney, gastrointestinal tract or vagina.

Serum sickness is a type III hypersensitivity reaction that results from the injection of heterologous or foreign protein or serum.

Causes: heterologous serum used in the prophylaxis and/or treatment of botulism, diphtheria, gas gangrene, organ transplant rejection, and snake and spider bites; vaccines; blood products; hormones; allopurinol, arsenicals and mercurial derivatives, barbiturates, bupropion, captopril, cephalosporins, furazolidone, gold salts, griseofulvin, halothane, hydralazine, infliximab, methyldopa, omalizumab, para-aminosalicylic acid, penicillamine, penicillins, phenytoin, piperazine, procainamide, quinidine. Primary serum sickness occurs 6–21 days after the administration of the inciting antigen. The onset may be more rapid with subsequent exposures to the same antigen, with symptoms occurring 1–4 days after exposure. The classic clinical manifestations consist of fever, arthralgia, lymphadenopathy, and skin eruption. Pain, pruritus, and erythematous swelling at the injection site usually precede the onset of the disease. Physical examination may reveal cutaneous symptoms; fever; lymphadenopathy; arthritis or arthralgias; edema; and renal, cardiovascular, neurologic, or pulmonary manifestations. Cutaneous symptoms (95 %) may include the following: urticaria; scarlatiniform rash; maculopapular or purpuric lesions; erythema multiforme. Characteristic serpiginous, erythematous, and purpuric eruption at the junction of the palmar or plantar skin with the dorsolateral surface of the hands, feet, fingers, and toes. Fever is invariably present and may precede rash in 10–20 % of cases. Lymphadenopathy (10–20 %) may be generalized or may include tenderness in the nodes that drain the injection site; splenomegaly may occur. Arthritis or arthralgias (10–50 %) usually affects multiple large joints, but occasionally, small joints and joints of the spine and temporomandibular joint may be inflamed. Myalgias or myositis may occur. Edema may also occur, particularly of the face and neck. Renal manifestations include proteinuria, microscopic hematuria, and oliguria; however, significant disease usually does not result. Cardiovascular findings may include myocardial and pericardial inflammation. Generalized vasculitis occurs rarely. Neurologic manifestations include peripheral neuropathy, brachial plexus neuritis, optic neuritis, cranial nerve palsies, Guillain – Barre syndrome, and encephalomyelitis.

Treatment: antihistamines; antipyretics; corticosteroids. Serum sickness may be limited only by the local manifestations (edema, redness, itching, and the formation of ulcer and numbness of the skin at the site of the introduction of serum in the blood). However, anaphylactic shock may develop during the repeated introduction of serum. The course of the disease: as a rule, the symptoms disappear in 3–5 days; however, in certain cases there may be the prolonged and even relapsing course. The cause of this is sensitization (increase in sensitivity) of the organism by the serum components. In case of applying the remedies of the long-term effect (application of Bicillin (benzathine penicillin G)) the symptoms may persist for several weeks and even months.

Farmer's lung ("extrinsic allergic alveolitis", "hypersensitivity alveolitis" or more generally "hypersensitivity pneumonitis"). Spores from two types of bacteria, *Micropolyspora faeni* and *Thermoactinomyces vulgaris*, and certain types of moulds called *Aspergillus* are the major causes of farmer's lung. The disease causes shortness of breath and a feeling of general malaise either in a sudden attack or as a slow, progressive disease. The disease is most common in regions with wet weather at harvest time. Farmer's lung is also more common on dairy farms. The degree of risk depends on the amount of dust that has collected in the person's lungs. A person can inhale an extremely large amount of dust within a very short time while working indoors: poultry workers, attendants of zoo and circus animals and pet shop workers.

There are three different types of allergic responses: acute or intense attack, sub-acute or low-level response, and chronic or long-term response. Acute farmer's lung is easy to notice and occurs in about one in three cases. It starts as an intense attack about 4 to 8 hours after the person breathes in a large amount of dust from moldy crops. These are some of symptoms: shortness of breath; a dry irritating cough; a sudden general feeling of sickness; fever and chills; a fast heart rate and accelerated breathing. If the person avoids further exposure to moldy dust, the signs and symptoms usually decrease after 12 hours, but they can last up to two weeks. Serious attacks can last as long as 12 weeks. Sub-acute farmer's

lung develops slowly, responding to continual exposure to small amounts of moldy dust. The symptoms include: coughing; shortness of breath; a mild fever and occasional chills; a general feeling of sickness; aches and pains in the muscles and joints and a loss of appetite and loss of weight. Chronic farmer's lung develops after several acute attacks over a period of years. It afflicts people who have been continually exposed to large amounts of moldy dust. Sometimes, the illness lasts several months and is marked by increasing shortness of breath, an occasional mild fever, and often, a significant loss in weight and a general lack of energy. The symptoms are accompanied by permanent lung damage and gradually worsen as exposure to moldy dust continues.

Diagnosis: the most important evidence of farmer's lung is the history of exposure and the development of symptoms 4 to 8 hours later; a lung X-ray; a blood test for antibodies; a pulmonary lavage test; lung function tests; a lung allergy challenge test; a lung biopsy.

10.4. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. The classification according to Gell and Coombs.
2. Modern classification of hypersensitivity reactions.
3. Basic mediators of allergies of HNT and HDT.
4. Mechanisms of development of anaphylactic reactions. Most frequent causes of anaphylaxis. The diseases caused by anaphylactic reactions.
5. Mechanisms of development of humoral cytotoxic reactions. The diseases caused by humoral cytotoxic reactions.
6. Mechanisms of development of reactions of the immune complex formation. The diseases caused by formation of the immune complexes.

7. Mechanisms of development of the pathologic immune responses, mediated by the T-sensitized lymphocytes. The diseases caused by HDT.
8. Mechanisms of development of autosensitization, caused by antibodies. The diseases caused by autosensitization.

Practical skills

1. Idiopathic thrombocytopenic purpura (clinical presentations, diagnosis, principles of treatment).
2. Goodpasture syndrome (clinical presentations, diagnosis, principles of treatment).
3. Sjogren syndrome (clinical presentations, diagnosis, principles of treatment).
4. Serum sickness (clinical presentations, diagnosis, principles of treatment).
5. Farmer's lung ("extrinsic allergic alveolitis") (clinical presentations, diagnosis, principles of treatment).

Tests

1. According to the classification of Gell and Coombs the second type of hypersensitivity reaction is called:
   A. Anaphylactic
   B. Immunocomplex
   C. Cell-mediate
   D. Cytotoxic
   E. Stimulating

2. The third type of hypersensitivity is based on pathogenesis of the following disease:
   A. Bronchial asthma
   B. Serum disease
   C. Anaphylactic shock
   D. Thrombocytopenic purpura
   E. Contact dermatitis

3. Pathogenesis of atopic diseases is based on the following type of hypersensitivity:
   A. I
   B. II
   C. III
   D. IV
   E. V

4. The manifestation of clinical symptoms in 48–72 hours after a contact with allergen is characteristic of the following type of hypersensitivity:
   A. Anaphylactic
   B. Immunocomplex
   C. Stimulating
   D. Cytotoxic
   E. Cell-mediate

5. Usually immunoglobullins of the class E participate in realization of the following type of hypersensitivity:
   A. Anaphylactic
   B. Cytotoxic
   C. Stimulating
   D. Immunocomplex
   E. Cell-mediate

6. What group of the diseases is characterized by predominant participation of the 3rd type of hypersensitivity in their pathogenesis?
   A. Dermatologic
   B. Atopic
   C. Collagenoses
   D. Blood diseases
   E. Infectious

7. What type of hypersensitivity is characterized by the reaction of antibody to antigen on the surface of the host cell causing destruction of this cell?
   A. I
   B. II

C. III
D. IV
E. V

8. What cells participate in development of the cell-dependent type of the hypersensitivity reaction?
A. Macrophages and lymphocytes
B. B-lymphocytes
C. Eosinophils
D. Neutrophils
E. Basophiles

9. Development of what disease is caused by the 5th type of the hypersensitivity reaction?
A. Farmer's lung
B. Uticaria
C. Brucellosis
D. Immunocomplex glomerulonephritis
E.Autoimmune thyroiditis

10. What group of the diseases is characterized by predominant participation of the 5th type of hypersensitivity in their pathogenesis?
A. Gastroenterological
B. Pulmonary
C. Infectious
D. Endocrine
E. Hematological

CHAPTER XI

ALLERGIC (ATOPIC) DISEASES

11.1. Etiology and pathogenesis of atopic diseases. Pseudoallergy

Allergy is a distorted (excessive) immune response of the organism to the exogenous agent. Allergy arises as the failure of the normal functioning of the immune system. Antigen is taken as threat to existence of the organism, and it is considered as allergen.

Viennese paediatrician Baron Clemens von Pirquet coined the term "allergy" (from the Greek *"allos"* – changed or altered state and *"ergon"* – reaction or reactivity) in 1906. Von Pirquet used the term to describe an altered reaction he had observed in patients, whom he put down to the influence of external factors, an allergen, on the immune system.

The ***most significant (most common) allergens*** are:

- ***everyday*** (mites of household dust, library dust, spores of mold fungi);
- ***epidermal*** (epidermis of a cat, dog, sheep, feather of the pillow);
- ***food*** (citrus fruit, cocoa, honey, crab, food dyes, aromatizers and flavoring substances);
- ***pollen*** (pollen of trees, gramineae and weeds);
- ***vaccine, sera and drugs*** (antibiotics, nitrofuran preparations, antiseptics, etc).

Pathogenesis

Initially (in 1968) Gell and Coombs proposed their classification of the hypersensitivity reactions as the classification of the types of the allergic reactions.

At present of vital importance in the pathogenesis of allergic reactions is the 1st type and partially the 3rd and 4th types of reactions (chronic hives, urticarias). The 3rd type of reactions can participate in the pathogenesis of bronchial asthma.

By mechanisms of development there are distinguished **true allergic reactions and pseudo-allergic reactions.** Clinical manifestations of these reactions are identical and are determined by participation of the same mediators (Fig. 11.1), and depend on what "shock organ" is affected. However, whereas pathogenesis of the true allergic reactions

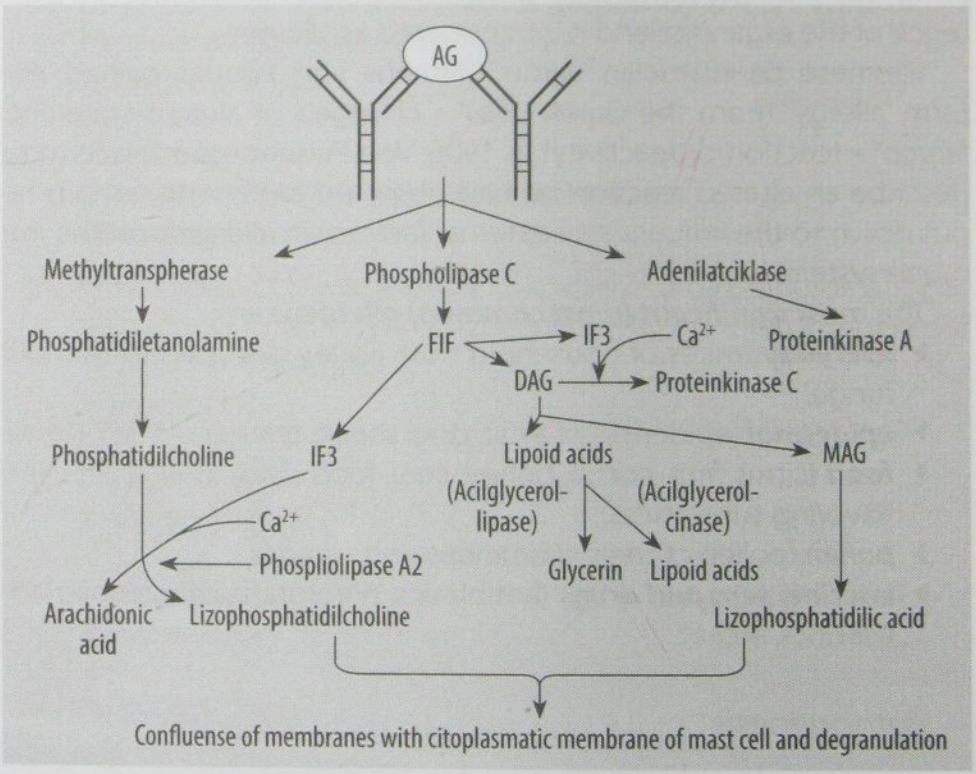

Fig. 11.1. Mediators synthesized in the process of mast cells activation. *AG – antigen, DAG – 1, 2 – diacylglycerol, IF – inositol – 1, 4, 5 – triphosphate, MAG – monoacylglycerin, FIF 2 – phosphatidylinositol – 4, 5 – diphosphate. (Source: http://www.medicum.nnov.ru/doctor/library/immunology/Lolor/23a.php).*

includes three stages: ***immune, pathochemical, pathophysiological,*** pathogenesis of the pseudo-allergic reactions has no immune stage (process begins at once with the pathochemical stage).

The causes of pseudo-allergic reactions:

- excessive intake of histamine by an organism (with a number of foodstuffs rich in histidine, dysbiocenosis of the intestines, disturbance of secretion of mucoproteins by the intestinal epithelium, etc.);
- histamine release from basophils and corpulent cells by the nonimmune way – under the influence of the blood substitutes, roentgen-contrast substances, local anestetics, food additives, by-products of helminths, physical factors (warmth and cold – ***thermal or cold urticaria,*** sunlight – ***photodermia***), etc.;
- disturbance of the processes of histamine inactivation in an organism (reduction in activity of the liver histaminase, dysfunctioning of histaminepexic mechanisms of the blood);
- **dysmetabolism** of the arachidonic acid – ***"aspirin bronchial asthma",*** characterized by presence of ***"aspirin triad"*** (severe continuous recurrent course of bronchial asthma, resistant to therapy; recurrent polyposis of the nose; intolerance of NSAIDs intolerance).

11.2. Diagnostics of allergic diseases

I. *Allergic anamnesis.* A detailed anamnesis is a basic information source, necessary for diagnostics and treatment of the allergic diseases. While examining patients with allergic diseases special attention should be paid to:

1. Variability of the symptoms (they develop and disappear rapidly, they develop in the specific place or in the specific season).
2. Individual allergic anamnesis.
3. Family allergic anamnesis.

II. *Physical examination.* Organs and systems, which are most frequently affected with the allergic diseases: the skin, eyes, respiratory organs are examined especially attentively.

Basic principles:

1. Not to fail to note the affection of the skin, it is necessary to investigate the entire skin. A patient can not mention about the skin manifestations, considering them insignificant, not related to the disease or feeling shy of them.
2. While examining the eyes it is possible to reveal hyperemia and edema of the conjunctiva, lacrymation and discharge from eyes.
3. It is compulsorily to examine the nose, and rhinoscopy should be carried out.
4. We may reveal wheezing auscultatively.

III. *Laboratory studies.* With their aid it is possible only to confirm or deny the diagnosis, based on the data of the anamnesis and physical examination as well as estimate the effectiveness of treatment and follow-up the state of patient.

1. The blood count:

1) an increase in the number of the eosinophils up to 5–15 %:
   a. moderate eosinophilia (15–40 % of the total number of leukocytes) is encountered not only in the allergic diseases, but also in the malignant neoplasms, for example, in lymphogranulomatosis, immunodeficiencies, congenital defects of the heart, cirrhosis of the liver, nodular periarteritis, herpetiform dermatitis, or during the radiation therapy, application of some medicines and peritoneal dialysis;
   b. expressed eosinophilia (50–90 % of the total number of leukocytes) is usually observed in helminthiases, for example, in the syndrome of larva migrans;
   c. the absolute number of eosinophils can be calculated, after determining the number of leukocytes and leukocyte formula;
2) absolute and relative lymphocytosis. A substantial change in the relationship of neutrophils/leukocytes.

2. Eosinophils in the smears. In exacerbation of the allergic diseases eosinophils predominate among the cells in the smears of the phlegm, discharge from the nose or eyes, in the concomitant infection there are neutrophils.

3. Total level of IgE in the serum. An increase in the total level of IgE in the serum confirms the diagnosis of the allergic disease, although the normal level of IgE does not exclude it. A radio-immunosorbent test (RAST) and solid-phase (IFA) allows to determine even the low concentrations of IgE (less than 50 IU/ml), ImmunoCAP (less than 80 KU/l). It is necessary to know the method of determining the level of IgE and the normal indices, accepted in this laboratory for the evaluation of the results of laboratory investigations. Approximately 70 % of adult patients with bronchial asthma and allergic rhinitis have the

IgE-mediated reactions to extrinsic antigens

(Charles A. Janeway et al., Immunobiology, 1999, as amended)

Syndrome	Common allergens	Route of entry	Response
Systemic anaphylaxis	Drugs Serum Venoms Peanuts	intravenous (either directly or following rapid absorption)	edema, increased vascular permeability, tracheal occlusion, circulatory, collapse, death
Acute urticaria (wheal-and-flare)	Insect bites Allergy testing	Subcutaneous	local increase in the blood flow and vascular permeability
Allergic rhinitis (hay fever)	Pollens (ragweed, timothy, birch) Dust-mite feces	Inhaled	Edema of nasal mucosa, irritation of nasal mucosa
Asthma	Pollens Dust-mite feces	Inhaled	bronchial constriction, increased mucus production, airway inflammation
Food allergy	shellfish, milk, eggs, fish, wheat	Oral	vomiting, diarrhea, pruritis (itching), urticaria (hives), anaphylaxis (rarely)

level of IgE exceeding the normal index by two standard deviations. More than 95 % children with a high level of IgE suffer from allergic diseases. The level of IgE may be especially high (more than 1000 IU/ml or 1000 KU/l) in diffuse neurodermatitis and atopic diseases of the respiratory organs.

Indications for determining the total level of IgE in the serum:

1) differential diagnostics of exogenous bronchial asthma and allergic rhinitis, especially in children of the young age;
2) differential diagnostics of the atopic diseases of the skin, especially in children;
3) the estimation of the risk of the allergic diseases of the lungs in children with bronchiolitis;
4) diagnostics and estimation of effectiveness in the treatment of allergic bronchopulmonary aspergillosis;
5) diagnostics of immunodeficiencies;
6) diagnostics of medicinal allergy;
7) diagnostics of myelomatosis.

In detection of the high level of IgE, first of all, helminthiases are excluded.

4. Determination of ***the levels of specific IgE*** in the blood serum (IFA diagnostics, ELISA, immunoblotting, MAST and ImmunoCAP) to different allergens.

5. Immunogram.

The signs of the allergic diseases:

- Absolute and relative lymphocytosis.
- Eosinophilia.
- An increase (more than 2.8) in the index of immunoregulation (Th/Tc) (CD4/CD8).
- An increase in the absolute and relative quantity of the B-lymphocytes.
- A reduction of the complement content.
- An increase in the levels of the circulating immune complexes.
- An increase in the autoantibodies to the tissues of the organs-targets (the skin, mucous membrane of the nose, bronchi, lungs).

IV. *Skin test.* There are cutaneous – puncture and scarification and intracutaneous tests. The positive results of skin tests (erythema and blister at the site of the allergen introduction) are of a diagnostic value only in combination with the data of the anamnesis, physical and laboratory investigations.

Indications and selection of the allergens. The basic indication for making the skin tests is development of the allergens, contact with which causes the disease. The diagnostic and therapeutic medicines of allergens are manufactured in the form of the concentrated or diluted extracts. There are diagnostic remedies of the allergens for the cutaneous and intracutaneous tests. The period of usability of the concentrated extracts of allergens is 2–3 years.

Technique of making:

1. ***Patch tests applied.*** The method is to apply an allergen to a patch which is then placed on the skin. This may be done to pinpoint a trigger of allergic contact dermatitis.
2. ***Scarification tests.*** Scarification scratches are made on the skin, on which the drops of the dilute solution of the allergen are placed.
3. ***The puncture tests (prick-test).*** **Skin-prick test remains the "gold standard" for identifying clinically relevant allergens**. The skin is cleaned by 70 % isopropyl or ethyl alcohol. So that the blisters would not merge, the distance between the adjacent punctures must be not less than 2 cm. A drop of the allergen extract is put on each selected point in the dilution 1 : 10, 1 : 20 or the undiluted standardized preparation is used, and the skin is pierced.
4. ***Intracutaneous tests*** are made in doubtful results of the puncture and scarification tests. The allergen extracts in the dilution of 1 : 100 are used for the intracutaneous tests. When less than five puncture or scarification tests are positive, intracutaneous tests can be carried out immediately. If the positive puncture or scarification tests are more, intracutaneous tests are carried out next day. Intracutaneous tests with the food allergens are not made.

Evaluation of the results:

1. One and the same method is always used for the evaluation of the results of skin tests, most habitual for the study.
2. Impairment of the technology of skin tests, the use of the allergen preparations with expired date, the tests made against the background of treatment with drugs decreasing skin sensitivity lead to pseudonegative results. The intake of H_1-blockers is withdrawn for 48 hrs, hydroxizine, terfenadine, loratadin and tricyclic antidepressants – for 96 hrs, and astemizole – 4 weeks prior to the study. Theophylline, adrenostimulators (inhalation and for the internal administration) and cromolin do not influence the skin sensitivity.
3. Introduction of the incorrectly prepared solutions of the allergens (incorrect selection of osmolarity and pH, presence of the irritating substances), impairment of the technology of making skin tests, for example, intracutaneous introduction of more than 0.02 ml of the allergen solution, urticate dermographism, introduction of the substances, which cause the release of histamine (for example, the extracts of food allergens), leads to the pseudopositive results.
4. Results of the skin tests are compulsorily compared with the data of anamnesis, physical and laboratory investigations.

During assessment of the skin tests it is necessary to consider:

1) Skin tests allow to determine rather precisely the cause of allergic rhino-conjunctivitis, but are less informative of exogenous bronchial asthma.
2) While making mass studies, especially in children of young age, the mixtures of allergens are used. Since during mixing of the allergen solutions their concentration is reduced, the skin tests with the mixtures of allergens are frequently negative. Therefore, if the data of anamnesis and clinical picture indicate allergy, and skin tests with the mixtures of allergens are negative, clean allergens are used for tests.

3) When food allergy is manifested by hives, Quincke's edema or by anaphylactic shock, skin tests with the food allergens are usually positive. However, their diagnostic significance is small, since together with the truly positive skin tests to the food allergens pseudopositive ones are frequently observed. However, negative skin tests have larger diagnostic value, since they indicate the absence of allergy to the specific food allergens with accuracy. If the results of skin tests are positive in food allergy, provocation food tests are made for confirmation of the diagnosis by the double blind method with the use of placebo as the control. If food allergy is accompanied by anaphylactic reactions, these tests are contraindicated.
4) Skin tests are not informative in diffuse neurodermatitis. Of large diagnostic significance in this disease are the provocative tests – inhalation, application and food. Provocation food tests are positive approximately in 30 % of children with diffuse neurodermatitis.
5) The value of skin tests is small in the medicinal allergy, since usually the allergy is caused not by the drug itself, but its metabolites, which cannot be determined. Skin tests are made only with the protein allergens, for example, with insulin, sera, and penicillins.

V. *Provocation test* is a method of development of sensitization, based on the introduction of the allergen in the target organ. There are sublingual, endonasal and inhalation provocation tests.

Allergen-induced bronchoconstriction

Inhalation of allergens in sensitized subjects develops into bronchorestriction within 10 minutes, reaches a maximum within 30 minutes, and usually resolves itself within one to three hours. In some subjects, the constriction does not return to normal, and recurs after 3 to 4 hrs, which may last up to a day or more. The first is named the early asthmatic response, and the latter – the late asthmatic response.

The major advantage of the provocation tests consists in the larger authenticity of their results.

The main disadvantages in the provocation tests consist in the following:

1) it is possible to make a test only with one allergen during one visit;
2) it is difficult to assess quantitatively the results of the study, especially in allergic rhinitis or conjunctivitis;
3) they yield badly to standardization;
4) they are combined with a high risk of severe allergic reactions, for example, bronchospasm, therefore, only an experienced doctor should make them. Provocation tests are contraindicated when there are indications of the immediate development of hives, Quincke's edema, bronchospasm or anaphylactic shock in contact with this allergen in anamnesis.

Provocation tests are frequently made in food allergy, since the skin tests are not informative in this case.

VI. *Functional diagnostics.*

Pulmonary function tests – study of the function of external respiration (spirometry and Peak flow) is used for differential diagnostics of the allergic and nonallergic diseases of the lungs, evaluation of reactivity of the bronchi, severity of these diseases and effectiveness of their treatment. Pulmonary function tests are a group of tests that measure how well the lungs take in and release air and how well they move gases such as oxygen from the atmosphere into the body's circulation.

The main tests to measure lung function include:

1) ***spirometry*** (this test measures the narrowing of your bronchial tubes by checking how much air you can exhale after a deep breathing in and how fast you can breathe out);
2) ***peak flow*** (a peak flow meter is a simple device that measures how hard you can breathe out);
3) ***bronchodilator reversibility test;***
4) ***bronchoconstriction test***;
5) ***nitric oxide test*** (this test is sometimes used to diagnose and monitor asthma. It measures the amount of gas called nitric oxide you have in your breath. If your airways are inflamed – a sign of

asthma – you may have higher than normal nitric oxide levels. This test isn't widely available).

Spirometry

Spirometry is most useful for: detection of the disease and its severity; identification of asthma triggers; progress/natural history monitoring; treatment response assessment; preoperative assessment (Fig. 11.2).

Normal Ventilatory Function: A person with Normal Spirometry will have lung volumes and flow rates within the normal range for people of their age, sex and height.

Obstructive Ventilatory Function: An obstructive disorder refers to any disease that affects the lumen of the airways. This could be due

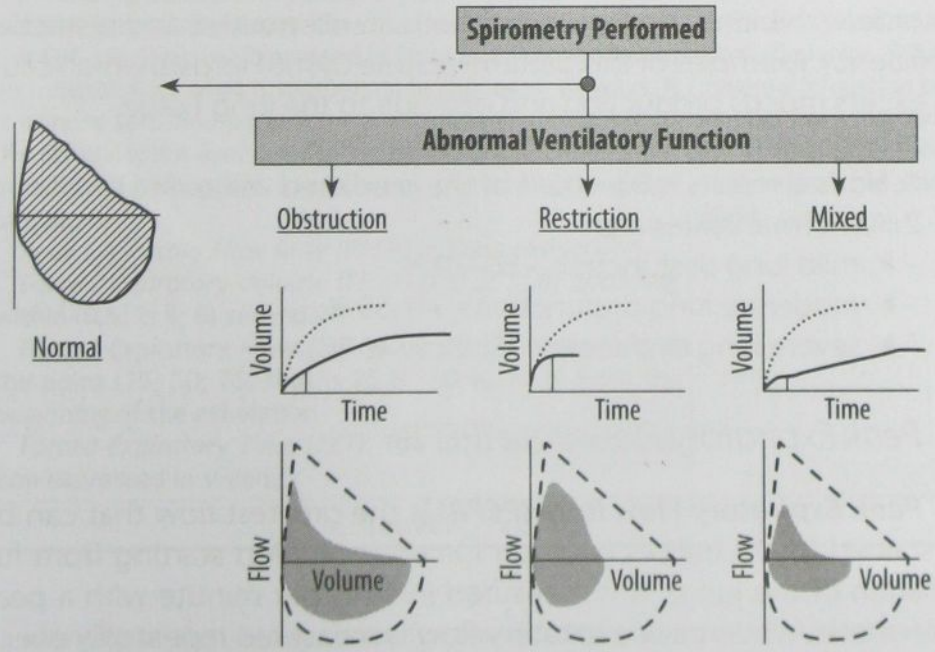

Fig. 11.2. Schematic diagram illustrating idealised shapes of flow-volume curves and spirograms for obstructing, restrictive and mixed ventilatory defects. *(David P. Johns, Rob Pierce, Spirometry, 2008).*

toexcessive mucus production, inflammation or bronchoconstriction. Asthma and Chronic Bronchitis are examples of obstructive disorders. In general terms the obstructive pattern presents itself as reduced flow rates and normal lung volumes (but with a reduced FEV_1) on the FVC manoeuvre.

Restrictive Ventilatory Function: A restrictive disorder is one that may affect the lung tissue itself or the capacity of the lungs to expand and hold predicted volumes of air. This could be due to fibrosis and scarring, or a physical deformity that restricts expansion. Someone who has had part of their lung removed would show a restrictive pattern and another example of a restrictive disease is pneumoconiosis. The restrictive pattern usually presents itself as reduced volumes and normal flow rates on the FVC manoeuvre.

Combined Ventilatory Function: A combined disorder is venffiatory disorder exhibiting the features of both an obstructive and restrictive deficiency. Examples of this pattern include Cystic Fibrosis, which causes excess mucus production and damage to the lung tissue.

Interpretation of Ventilatory Function Tests:

1. Normal results is 80–100 % of the predicted value.
2. Abnormal values are:
   - mild lung dysfunction – 60–79 %;
   - moderate lung dysfunction – 40–59 %;
   - severe lung dysfunction – below 40 %.

Peak Expiratory Flow Rate (PEFR)

Peak Expiratory Flow Rate (PEFR) is the greatest flow that can be sustained for 10 milliseconds on forced expiration starting from full inflation of the lungs. It is measured in litres per minute with a peak flow meter. When peak expiratory flow is measured repeatedly over a period and plotted against time, the pattern of the graph can be helpful in identifying particular aspects of the patient's disease. Isolated falls in PEF in relation to specific allergens or trigger factors can help

The main spirometry tests

<table>
<tr><td colspan="2">Static test (performed without regard to time)</td></tr>
<tr><td colspan="2">Vital Capacity (VC)
Forced Vital Capacity (FVC) the maximum volume of air in litres that can be forcibly and rapidly exhaled following a maximum inspiration. FVC is the basic manoeuvre in spirometry tests
Inspiratory Vital Capacity (IVC) the maximum volume of gas which can be inspired into the lungs during a relaxed inspiration from a position of full expiration</td></tr>
<tr><td colspan="2">Dynamic test (performed at forcible and maximum effort against time)</td></tr>
<tr><td>Forced Expiratory Volume in one second (FEV₁) is the volume of air expelled in the first second of a forced expiration starting from full inspiration (in normal lung function this should generally be over 75 %, i.e. the subject should get at least three quarters of their total air out in the first second)
Forced Expiratory Flow Rate (FEFR 25 % – 75 %) this is the average forced expiratory flow rate at the middle part of the FVC manoeuvre. Expressed in litres per second it gives an indication of what is happening in the lower airways. It is a more sensitive parameter and not as reproducible as the others. It is a useful serial measurement because it will be affected before FEV, so can act as an early warning sign of disease
Peak Expiratory Flow Rate (PEFR) in liters per second
Forced Expiratory Volume (FEV – 0.5; 1; 3; 6) occurring within (0.5; 1; 3; 6) seconds
Forced Expiratory Flow (FEF – 25; 50; 75) occurring at the point (25; 50; 75) that is 25 %, 50 %, 75 % from the beginning of the exhalation
Forced Expiratory Time (FET). The total time of Exhalation expressed in seconds</td><td>Forced Inspiratory Flow (FIF – 25; 50; 75) rate at the point (25; 50; 75) that is 25 %, 50 %, 75 % from the beginning of the inhalation
Forced Inspiratory Vital Capacity (FIVC) maximal inhalation beginning from the point of maximal exhalation
Flow Volume Loop (FVL)</td></tr>
<tr><td colspan="2">The FEV₁/FVC ratio</td></tr>
</table>

to identify and quantify these for the doctor and patient. A downward trend in PEF and an increase in its variability can identify worsening asthma and can be used by the doctor or patient to modify therapy. Peak flow meters are very helpful if you or your child have moderate to

severe asthma and require daily asthma medications. Even four-year children should be able to use a peak flow meter with good results. People with moderate-to-severe asthma should have a peak flow meter at home.

Technique:

- stand up or sit up straight;
- make sure the indicator is at the bottom of the meter (zero);
- take a deep breathing in, filling the lungs completely;
- place the mouthpiece in your mouth, lightly bite with your teeth and close your lips on it;
- be sure your tongue is away from the mouthpiece;
- blast the air out as hard and as fast as possible in a single blow;
- remove the meter from your mouth;
- record the number that appears on the meter and then repeat steps one through seven two times.

Record the highest of the three readings in an asthma diary. This reading is your peak expiratory flow (PEF). To ensure the results of your peak flow meter are comparable, be sure to use your meter the same way each time you take a reading.

Peak Flow Zones (recommended for patients):

The zones will help you check your asthma and take the right actions to keep it controlled. The colors used with each zone come from the traffic light.

Green Zone (80 to 100 % of your personal best) signals good control. Take your usual daily long-term-control medicines, if you take any. Keep taking these medicines even when you are in yellow or red zones.

Yellow Zone (50 to 79 % of your personal best) signals caution: your asthma is getting worse. Add quick-relief medicines. You have to increase other asthma medicines as directed by your doctor.

Red Zone (below 50 % of your personal best) signals medical alert! Add or increase quick-relief medicines.

Bronchodilator reversibility testing

This test uses spirometry and a type of drug called a bronchodilator (salbutamol, terbutaline, ipratropium bromide). Patients are given the drug to relax their breathing muscles so that they can breathe more easily. This will help you get the most accurate information about how the medicine affects breathing and how lungs work. Bronchodilator reversibility testing may help to differentiate COPD and asthma. There is presently no universal agreement on the definition of significant bronchodilator reversibility. According to the ATS/ERS the criteria for a significant response in adults is: > 12 % improvement in FEV_1 (or FVC) and an absolute improvement of > 0.2 L.

Normal subjects generally exhibit a smaller degree of reversibility (up to 8 % in most studies). The absence of reversibility does not exclude asthma because the asthmatic person's response can vary from time to time and at times airway calibre in asthmatic subjects is clearly normal and incapable of dramatic improvement.

Bronchoconstriction testing

- dosed exercise testing (a spirometry breathing test is done before and after you exercise on a treadmill);
- inhalation of cold air;
- inhalation of saline solution;
- inhalations of histamine and methacholine.

The X-ray examination

a. Roentgenography of the chest on primary examination is made in all patients with the allergic diseases of the lungs. The roentgenography of the chest allows to exclude pneumonia, atelectasis and pneumothorax, which can complicate the severe attack of bronchial asthma.

b. Investigation of the paranasal sinuses is made on suspicion of acute or chronic sinusitis – the frequent complication of the allergic diseases of the upper respiratory tract.

11.3. Treatment of allergic diseases

Glucocorticosteroids

They are the most effective and pathogenetically substantiated drugs for treatment of the allergic diseases. They act at all stages of pathogenesis (pathoimmune, pathochemical, pathophysiological).

The main effect at the pathophysiological stage on the epithelium of the blood vessels (they decrease the permeability) and the main symptom s of the disease are eliminated (edema, hypersecretion, hyperemia).

At the pathoimmune stage they block release of cytokines (interleukins and antibodies) by the immunocompetent cells, they contribute to the decrease of the eosinophil number. At the pathochemical stage they contribute to the stabilization of the membranes of mast cells and basophils.

They are used:

- in acute states – parenterally (prednisolone, hydrocortisone, dexamethasone);
- systematically – orally and intramuscularly prolonged forms – drugs-depot – effect for a month (polcortolon, diprospan);
- locally – inhalation forms, nasal sprays, ointments and creams.

Antihistamine drugs (blockers of H_1-histamine receptors)

They are effective only upon transfer from the pathochemical stage to the pathophysiological one.

Drugs of the first generation:

Dimedrol (Diphenhydramine), Tavegil (Clemastine), Phencarol (Quifenadine), Pipolphen, Diprazin (Promethazine hydrochloride), Diazoline (Mebhydrolin), Suprastin (chloropyramine), (Chloropyramine), Peritol (Cyproheptadine).

Peculiarities of pharmacokinetics and the mechanism of action:

- Competitive (with histamine) blockade of H_1-receptors – drugs should be taken frequently (3–4 times in a 24 hour period), and in large doses (risk of the toxic action);
- penetrate through the blood-brain barrier – sedative side-effect (sleepiness) + potentiate the effect of analgesics and antipyretics;
- Irritate the GIT mucosa – the side-effect is diarrhea, therefore the intake is after meal (adsorption velocity from the bowels is lowered);
- In the prolonged intake (more than 10 days) tachyphylaxis develops (addiction) – effectiveness is lowered;
- bind with the blood proteins (risk of the toxic effect during dehydration, cachexia and secretory dysfunction of the kidneys);
- muscarine-like effect (anticholinergic action) – they decrease the secretion of the mucous glands of the respiratory system – they are contraindicated in diseases of the respiration organs in presence of the thick phlegm in the bronchi (they reduce the drainage function of the bronchi).

Drugs of the second generation:

Hismanal, histalong (Astemizol), Claritin (Loratadine), Zyrtek (Cetirizine), Cestin (Ebastine), Trexyl (Terfenadine)

Anti-inflammatory effects of corticosteroid therapy

(Charles A. Janeway et al., Immunobiology, 1999, as amended)

Effect on	Physiological effects
↓ IL-1, TNF-α, GM-CSF, IL-3, IL-4, IL-5, IL-8	↓ Inflammation caused by cytokines
↓ NOS	↓ NO
↓ Phospholipase A2 ↓ Cyclo-oxygenase type 2 ↑ Lipocortin-1	↓ Prostaglandins ↓ Leukotrienes
↓ Adhesion molecules	Reduced emigration of leukocytes from vessels
↑ Endonucleases	Induction of apoptosis in lymphocytes and eosinophils

Peculiarities of pharmacokinetics and the mechanism of action:

They are noncompetitive blockade of H_1-histaminic receptors; they do not penetrate through the blood-brain barrier; they do not irritate the GIT mucosa; tachyphylaxis does not develop; do not bind with the blood proteins; anticholinergic action is absent.

Drawbacks:

1. Trexyl – there are cases of sudden death (it contributes to significant prolongation of P – Q interval) because of the cardiotoxic effect.
2. They are converted into active metabolites in the liver (they suppress the activity of the cytochromes of the hepatocytes).

Active metabolites of the drugs of the second generation: Telfast (Fexofenadine), Aerius (dezloratadine), Aleron (levocetirizine)

Their drawbacks are still under study.

According to the last recommendations of allergologists intake of the drugs of the first generation is indicated in the emergency allergic states, since they are in the injection forms, and the administration of the drugs of the second and third generation is indicated for the course therapy.

At the same time the drugs of the second and third generation do not exceed those of the first generation in the manifestation of the antiallergic effect. Furthermore, many people are noted to have individual sensitivity (selective) to the antihistamine drugs. There may be higher efficacy in intake of the drugs of the first generation and minimum reaction after the intake of the drug of the second or third generation.

Stabilizers of the mast cell membranes

They are effective at the pathochemical stage and ketotifen – upon transfer from the pathochemical stage to the pathophysiological one (it also possesses antihistaminic effect).

Cetotifen, sodium cromolin (intal, cromolin, cromogexal, cromoglin etc), Nedocromil of sodium (Tiled).

They are intended for the prolonged course (the minimum period is 2 months). Cetotifen is for internal administration (the main indication is food allergy – the skin forms of allergy). Cromolin and sodium nedocromil act only locally – in the form of inhalers (cromolin is also in the form of nasal spray and in the form of capsules. Nalcrom is used in the allergic diseases of GIT and food allergy).

Anti-leucotriene drugs (inhibitors of leucotriene metabolism)

They are effective at the pathoimmune stage and upon transfer from the pathoimmune stage to the pathochemical one. They are zafirlucast, montelucast. The indication is bronchial asthma. They are less effective than inhalation of corticosteroids.

Selectively binds to human immunoglobulin E

Omalizumab is an injectable drug that is used for treating asthma. Omalizumab is a monoclonal antibody targeting the high-affinity receptor binding site on human immunoglobulin IgE. Bound IgE is not available for basophil binding, degranulation is attenuated, and allergic symptoms are reduced. In asthma trials, omalizumab reduced inhaled corticosteroid and rescue medication requirements and improved asthma control and asthma quality of life in moderate to severe allergic asthmatics with disease poorly controlled by inhaled corticosteroids. In trials of patients with poorly controlled moderate to severe seasonal allergic rhinitis, omalizumab reduced the severity of exacerbations and rescue medication use, and improved rhinitis-related quality of life.

Allergen specific immunotherapy

It is based on the introduction of different dilutions of the causally significant allergen into the organism according to the specific scheme (with a strict observance of dosages and periods of introduction).

The mechanism of action:

- the production of the so-called blocking antibodies – IgG (development of sufficient amount of the B-lymphocytes pool in the

organism secreted the blocking antibodies instead of specific IgE and Ig G_4 to the introduction of concrete allergen);

- the production and release of many of the proinflammatory mediators (particularly cytokines) are diminished. This may be due to a direct effect on mast cells and eosinophils or an immunoregulatory effect mediated by specific populations of lymphocytes;
- after an initial rise, allergen-specific IgE levels in the plasma fall with allergen immunotherapy. This is thought to be due to active immunoregulatory mechanisms that alter when a specific individual responds to a particular allergen.

Not all mechanisms are likely to be active in every treated patient. It is the effective method of treatment of allergic reactions for: dust; pollen; mite; dander and insect venom.

11.4. Diagnostics and treatment of some allergic diseases

Anaphylactic shock (AS) is an acute allergic disease, characterized by life-threatening disturbances of the major systems of an organism. ***The most frequent causes:*** intake of medicines; stings of insects and snakes; foodstuffs. There are ***several variants of AS:*** hemodynamic (sharp decrease of AP, spasm or dilatation of the blood vessels); asphyxic (acute respiratory insufficiency due to bronchospasm and edema of mucous membranes of the bronchial tubes); cerebral (psychomotor excitation, spasms, disturbances of consciousness, edema of the brain); abdominal (severe abdominal pains, symptoms of peritoneum irritation – clinical course of "acute abdomen"). There are ***several variants of the course of AS:*** acute malignant; acute benign; prolonged; recurrent; abortive. ***Treatment:*** 1) discontinuance of the contact with allergen; 2) pharmacotherapy (dosage and way of introduction depends on severity of the condition): adrenaline drip/feed or i/v 0.3–1.0 ml to adults and 0.05 ml/year of life – to children; glucocorticoids i/v 60–120 mg to adult and 1–3 mg/kg – to children; antihistamine preparations i/v or

i/m by 1–2 ml to adults and 0.1 ml/years of life – to children. 3) symptomatic medicines (depending on the variant of AS).

Bronchial asthma (BA) is a chronic inflammatory disease of the respiratory tracts, caused by a significant amount of cells and mediators of inflammation. Diagnosis and treatment of BA is made according to the national protocols, which are substantially focused on the international agreements (GINA – the global initiative for asthma). ***The Order № 868 of 8/10/2013 of MH of Ukraine*** approved the protocol of rendering medical aid to patients with BA. Last revision of GINA was made in 2010 ***(GINA-2010). Clinical course and diagnosis. The Order № 868*** (it is practically identical to ***GINA-2010)***: incidental dyspnea with difficult exhalation; cough, especially at night or on physical exertion; incidental wheezing in the lungs; repeated constraint of the chest. These symptoms are more intensified at night and early in the morning (wake the patient), arise or worsen in: physical exertion; viral infection; influence of allergens; smoking; difference of external temperature; strong emotions (crying, laughter); action of chemical aerosols; intake of some medicines. Daily and seasonal variability of symptoms is characteristic. Values of PEFR and FEV_1 are < 80 % of the norm. The bronchodilatation test with β_2-agonist of the short action is > 12 % (or ≥ 200 ml). Daily variability of PEFR and FEV_1 is > 20 %. Allergologic individual and family anamnesis shows presence of atopic diseases. Skin tests with allergens are positive. There is an increased level of general and specific IgE. In patients with BA symptoms, but without impairments of respiratory function for specification of the diagnosis, provocative inhalation tests with histamine; metacholin; physical exertion are made.

Classification of BA. The Order № 868.

According to the degree of severity:

- *intermittent BA:* symptoms (episodes of cough, wheezing, dyspnea) are short-term, less often than 1 time a week within at least 3 months; short-term aggravations; night symptoms are not more often than 2 times/mon.; when there is no aggravation – absence of symptoms, PEFR or FEV_1 is ≥ 80 % of the norm, daily fluctuations of PEFR or FEV_1 are < 20 %;

- *mild persistent BA:* occurrence of symptoms minimum once a week but less often than once/day during 3 months; aggravations can disturb activity and sleep; presence of chronic symptoms, which demand symptomatic treatment almost daily; night symptoms more often than twice/mon; PEFR or FEV_1 is ≥ 80 % of the norm, daily fluctuations of PEFR or FEV_1 are 20–30 %;
- *moderate persistent BA:* daily symptoms; aggravations disturb activity and sleep; night symptoms are more often than once/mon; necessity for daily intake of β_2-agonists of the short action; PEFR or FEV_1 is within 60–80 % of the norm, daily fluctuations of PEFR or FEV_1 are > 30 %;
- *severe persistent BA:* presence of variable long symptoms; frequent night symptoms; restriction of physical activity caused by BA; frequent severe aggravations; absence of the appropriate control of the disease (despite treatment); constant presence of long daytime symptoms; frequent night symptoms; PEFR or FEV_1 is within < 60 % of the norm, daily fluctuations of PEFR or FEV_1 are > 30 %; control of BA can be impossible.

A concept of BA control: *a controllable course* (absence or minimal (≤ 2 times a week) daytime symptoms, absence of activity restriction, night symptoms, absence or minimal (≤ 2 times a week) requirement in broncholytics, absence of aggravations, normal indices of respiratory function); *a partial control* (any sign can be marked every week); *an uncontrollable course* (≥ 3 signs of the partial control are present every week).

Treatment. The Order № 868.

The step approach to pharmacotherapy:

- *intermittent BA*: treatment is symptomatic – inhalation of β_2-agonists of short action if necessary (not more than once a week for over 3 months) and preventive intake (before physical activity or possible contact with allergen);
- *mild persistent BA:* symptomatic treatment + 1 controling preparation (low doses of inhalation corticosteroids; alternative – cromones; modifiers of leucotriens or durable xanthines);

- *moderate persistent BA:* symptomatic treatment + 1–2 controlling preparations (simultaneous intake of low doses of inhalation corticosteroids and durable β_2-agonists; alternative – middle and high doses of inhalation corticosteroids or a combination of low doses of inhalation corticosteroids with modifiers of leucotriens or durable xanthines);
- *severe persistent BA:* symptomatic treatment + 2 and more controlling preparations (simultaneous intake of middle-high doses of inhalation corticosteroids and durable β_2-agonists, in inefficacy + modifiers of leucotriens or durable xanthines, or oral corticosteroids in minimally possible doses for a long time; in high level of IgE it is effective to administer anti-IgE-preparations; in inefficacy of treatment sparing – therapy with immunodepressants – metotrexate, cyclosporin A, preparations of gold) is administered.

The effectiveness of each treatment step is estimated within 3 months, after which it is possible to reconsider steps of therapy.

According to ***GINA-2010*** *there are 5 steps of the control over BA course:* I. β_2-agonists of short action; II. A choice between low doses of inhalation corticosteroids or modifiers of leucotriens; III. A choice between low doses of inhalation corticosteroids + durable β_2-agonists or middle doses of inhalation corticosteroids or low doses of inhalation corticosteroids + modifiers of leucotriens or low doses of inhalation corticosteroids + prolonged dimethylxanthines; IV. A step 3 + separately or in combination: middle doses of inhalation corticosteroids + durable β_2-agonists; modifiers of leucotriens; prolonged dimethylxantines. V. Step 4 + low doses of oral corticosteroids or anti-IgE-preparations.

The effectiveness of treatment is estimated within 1 month. Step-by-step decrease is recommended during the control over 3 months.

Treatment of aggravations of BA. According to the ***Order № 868*** there are 4 degrees of severity of aggravations: of mild and moderate severity (out-patient treatment); severe and threat of respiratory standstill (hospitalization for the majority of patients).

Basic modern bronchial dilators

Pharmacological group	Active factor	Proprietary name	Pharmaceutical form
β_2-short-acting agonists	salbutamol	Salbutamol Ventolin, Salbupart	metered aerosol, tablets
	fenoterol	Berotek, Partusisten	metered aerosol
	terbutaline	Terbutaline, Brikanil	metered aerosol, tablets, solution
	clenbuterol	Spiropent	syrup, solution
β_2-agonists of durable action	salmaterol	Serevent	metered aerosol
	formoterol	Foradil, Atimos, Oxis	metered aerosol
anticholinergic drugs	ipratropium bromide	Atrovent, Truvent	metered aerosol
	oxitropium	Oksivent, Ventilat	metered aerosol
long-acting xanthine	dimethylxanthine	Theothard, theopek etc.	tablets
β_2-short-acting agonists + anticholinergic drugs	ipratropium + fenoterol	Berodual	metered aerosol
	+ salbutamol, ipratropium	Combivent	metered aerosol

An out-patient stage. *Initial therapy* – inhaled β_2-agonists dose increases 2–4 inhales every 20 min/first hour. Then the dose is reconsidered depending on severity of aggravation and data of the peak-flow- meter (increase of PEFR >80 % of the norm or the best for the patient and lasts 3–4 hours – there is no need for taking other medicines). *In incomplete response:* to continue intake of inhaled β_2-agonists 6–10 inhales every 1–2 hrs + oral corticosteroids (prednisolone equivalent 0.5–1 mg/kg); use of the combined forms is possible: inhalation of cholinolytics + inhaled β_2-agonists). *In low effect:* to continue intake

of inhaled β_2-agonists up to 10 inhalations (it is better to do through the spacer) or a full dose through a nebulizer with intervals of less than an hour + inhalation of cholinolytics (it is possible to use combination forms with β_2-agonists) + peroral corticosteroids + emergency call for hospitalization.

An in-patient stage (a severe aggravation – hospitalization to the intensive care unit). *Initial therapy:* oxygen therapy; inhalation of β_2-agonists of the short action constantly for 1 hour (it is recommended to do it through a nebulizer). *In 1 hour estimation of the condition and correction of therapy is made:* 1) moderately severe degree of aggravation (oxygen therapy, inhalation of β_2-agonists + cholinolytics each hour, oral corticosteroids for 1–3 hrs till improvement of the condition); 2) in presence of risk factors of fatal BA in the anamnesis, PEFR < 60 % of the norm or the best for the patient, expressed symptoms of BA at rest, retraction of the chest, absence of clinical improvement (oxygen therapy, inhalation of β_2-agonists + cholinolytics, systemic corticosteroids). *A repeated estimation in 1–2 hrs:* 1) good effect within

Daily doses of inhaled corticosteroids for adults and children over 5 years (according to **GINA-2010**)

Preparation	Low doses (mg)	Medium doses (mg)	High doses (mg)
Beclomethasone dipropionate	200–500	> 500–1000	> 1000–2000
Budesonide	200–400	> 400–800	> 800–1600
Cyclesonide	80–160	> 160–320	> 320–1280
Flunisolide	500–1000	> 1000–2000	> 2000
Fluticasone propionate	100–250	> 250–500	> 500–1000
Mometasone furoate	200–400	> 400–800	> 800–1200
Triamcinolone acetonide	400–1000	> 1000–2000	> 2000

1–2 hrs after the last medical manipulation – the patient is discharged for continuation of treatment (inhalation of β_2-agonists, oral corticosteroids in most cases, recommendation to use combined inhalers, education of the patient to take medicines correctly, revision of the individual plan of treatment, active medical follow-up); 2) the incomplete response (oxygen therapy, inhalation of β_2-agonists + cholinolytics, systemic corticosteroids, i/v xanthines are recommended, monitoring of PEFR, SaO_2 in the arterial blood, pulse rate); 3) in ineffective therapy for 1–2 hours – transfer to the intensive care unit (oxygen therapy, inhalation of β_2-agonists + cholinolytics, i/v corticosteroids, β_2-agonists i/m, i/v, i/v xanthines, there may be intubation and ALV). According to **GINA-2010:** 1) we begin with inhalation of β_2-agonists, 2–4 inhalations every 20 min/first hour, then in mild aggravations 2–4 inhalations every 3–4 hrs and in moderate – 6–10 inhalations every 1–2 hrs; 2) oral corticosteroids (prednisolone equivalent 0.5–1 mg/kg a day), in cases of moderate or severe aggravation earlier intake is possible; 3) in the in-patient conditions – oxygen therapy (in saturation of $O_2 < 95$ %); 4) a combined intake of inhalation of β_2-agonists and cholinolytics (it is associated with low indices of hospitalization and greater increase of FEV_1 and PEFR) is recommended; 5) additional intake of methylxanthines is not recommended while using high doses of inhaled β_2-agonists (only in availability of inhalation of β_2-agonists, with calculation of a daily dose if the patient has already taken dimethylxanthine); 6) in severe aggravations with in inefficacy of bronchodilators and systemic glucocorticoids it is indicated to introduce 2 g of magnesium sulfate (for reduction of requirement for hospitalization).

Allergic rhinitis (AR) is IgE-mediated inflammation of the mucous membrane of the nose, caused by exposition to an allergen ***(ARIA, 2003)***. The International agreement on diagnosis, classification and treatment of allergic rhinitis was developed in 2003 (ARIA – allergic rhinitis and asthma). Last revision was ***ARIA, 2010. Clinical symptoms (ARIA, 2003):*** *Characteristic of AR* – two and more of the following: it lasts longer than 1 hour for many days; watery anterior rhinorrhea; sneezing, especially paroxysmal; nasal itching; ± conjunctivitis. *Un-*

characteristic of AR: unilateral symptoms; stuffiness in the nose without other symptoms; mucopurulent rrhinorrhea; posterior rrhinorrhea (postnasal leakage) with thickening of the mucous membrane and (or) with absence of anterior rrhinorrhea; pain; periodic nasal bleedings; anosmia. ***Classification:*** *by the character of the course: 1) intermittent* (≤4 days a week or ≤4 weeks) or seasonal; *2) persistent* (>4 days a week or >4 weeks) or all-the-year-round. *By a degree of severity: 1) mild persistent* (normal sleep; absence of disturbances of daily activity, playing sports, leisure; working capacity or study is not disturbed; symptoms do not trouble); *2) moderate-severe persistent* of one or several elements (disorder of sleep, daily activity, playing sports, leisure; reduction in working capacity; symptoms cause significant anxiety). ***Treatment:*** exclusion of contact with allergen; antihistamine preparations of 2 or 3 generations; corticosteroids intranasally and cromones (nasal sprays); specific immunotherapy; administration of intranasal antihistamine preparations (in seasonal AR – the conditional recommendation because of a low degree of evidencial standard – ***ARIA, 2010);*** modifiers of leucotriens (in seasonal AR, and in children of preschool age in all-the-year-round AR – the conditional recommendation because of a low degree of evidencial standard – ***ARIA, 2010).***

Insect allergy is the allergic reaction arising in contact with insects or their by-products. There are local and systemic reactions (Muller U. R.,1990). Treatment of acute allergic reaction (according to the protocol of rendering medical aid in insect allergies, approved ***by the Order № 432 of 7/3/2006 of MH of Ukraine)***. In local reaction: 1) to apply a tourniquet to the extremity above the site of a sting; 2) to remove a sting without damaging the sac with poison, which remains in the skin; 3) to apply ice to the site of a sting; 4) the site of a sting should be treated with 0.1 % solution of adrenaline in the dose of 0.3–0.5 ml in 4.5 ml of the physiological solution; 5) in significant local and general reaction adrenaline is given by drip/feed; 6) i/v antihistamine preparations of the first generation (in normal arterial pressure); 7) antihistamine preparations of the second and third generation per os for 2–3 days; 8) locally – ointments with glucocorti-

coids 2–4 times a day. In **systemic** reaction: 1) hospitalization within 5–10 days; 2) in reduction in the arterial pressure – i/v adrenaline or mesatone; 3) i/v glucocorticoids; 4) i/v antihistamine preparations (in normal arterial pressure); 5) antihistamine preparations perorally for 7 days; 6) in developemnt of the bronchoobstructive syndrome – i/v euphyllinum (dimethylxanthine). **Elimination measures:** to cover most part of the body with clothes and footwear; not to use food outside the premises; not to use the flavored soap or cosmetics; to keep away from apiaries; not to use products of beekeeping; in presence of insects not to make sharp movements; to protect windows with mosquito net; to have the first-aid set of first aid with an antishock set.

11.5. Theoretical questions, practical skills and tests for practice and final control

Theoretical questions

1. The causes of formation of allergic pathology. Stages of pathogenesis of allergic reactions. Classification of allergens.
2. Pseudo-allergy: concept and causes.
3. Allergologic anamnesis (components). Clinical manifestations of allergic diseases.
4. Laboratory methods of diagnostics of allergic diseases.
5. Skin allergic tests: kinds; a technique of making; interpretation of results.
6. Spirography: the basic parameters, types of disorder of ventilation of lungs. Peak-flow-meter: a technique of making, criteria of estimation.
7. Principles of antiallergic therapy: groups of preparations and the basic representatives. Difference of antihistamine preparations of the first generation from the second and the third ones.

Practical skills

1. Criteria of making the diagnosis of bronchial asthma.
2. Criteria of making the diagnosis of intermittent and mild persisting bronchial asthma.
3. Criteria of making the diagnosis of moderately severe and severe persisting bronchial asthma.
4. Concept of the control of bronchial asthma. Degrees of controlled course of bronchial asthma. Criteria.
5. Administer step therapy of bronchial asthma (steps I and II)
6. Administer step therapy of bronchial asthma (steps III and IV)
7. Administer treatment to the patient with aggravation of bronchial asthma at the out-patient stage.
8. Administer treatment to the patient with aggravation of bronchial asthma at the in-patient stage.
9. Administer examination and treatment to the patient with all-the-year-round allergic rhinitis (of mild and moderately severe degree).
10. Administer examination and treatment to the patient with seasonal allergic rhinitis (mild and moderately severe degree).
11. Algorithm of therapeutic measures in anaphylactic shock.

Tests

1. Most often the presence of allergy is indicated in the leukogram by the number increase of:
   A. Basophils, neutrophils
   B. Eosinophils, lymphocytes
   C. Red blood cells, eosinophils
   D. Neutrophils, eosinophils
   E. Monocytes, basophils

2. Which body part is more often "shocked" with food allergies:
   A. Nasal mucosa

B. Mucous of the eye
C. Bronchial mucosa
D. Skin
E. Vaginal mucosa

3. Which change of immunograms' parameter is not indicated as patient's atopic disease?
A. The number of B-lymphocytes
B. The nnumber of active phagocytes
C. The content of the complement
D. Immunoregulation index
E. Total IgE

4. Improving of what immunoglobulins class may indicate patient's atopic disease:
A. A
B. M
C. G
D. E
E. D

5. What criteria does not apply to the major differences in the pharmacological (pharmacokinetic) action of blockers N_1-histamine receptors of the first and second generation:
A. Penetration of the blood-brain barrier
B. Competitive blockade of N_1-histamine receptors
C. Effectiveness of using
D. Irritation of the mucous membranes
E. Tachyphylaxis

6. On the deterioration of which spirographic indicators are based the diagnosis of asthma:
A. FEV_1 and FVC
B. VC and PEFR

C. FVC and PEFR
D. VC and MEF_{25-75}
E. FEV_1 and PEFR

7. What differences between the range of the morning and evening values of the Peak flowmetry show that bronchial asthma is under control?
A. Up to 5 %
B. Up to 10 %
C. Up to 15 %
D. Up to 20 %
E. Up to 30 %

8. The emergency care in case of hemodynamic form of anaphylactic shock is administered intravenously as a priority:
A. Corticosteroids
B. Antihistamines
C. Adrenaline
D. Kordiamine (Corazol)
E. Eufilline

9. The emergency care in case of bronchial asthma episode is:
A. Intravenous administration of corticosteroids
B. Inhalation of β-agonist
C. Intravenous administration of antihistamine
D. Intravenous administration of aminophylline
E. Inhalation of corticosteroids

10. The status asthmaticus is considered an attack of asthma, which doesn't stop in:
A. 3 hours
B. 6 hours
C. 9 hours
D. 12 hours
E. 24 hours

Answers to the tests

Chapter I:	1. **D**	2. **B**	3. **A**	4. **C**	5. **B**	6. **A**	7. **D**	8. **B**	9. **B**	10. **A**
Chapter II:	1. **B**	2. **C**	3. **D**	4. **A**	5. **C**	6. **E**	7. **E**	8. **D**	9. **A**	10. **C**
Chapter III:	1. **D**	2. **E**	3. **C**	4. **B**	5. **C**	6. **E**	7. **D**	8. **B**	9. **E**	10. **B**
Chapter IV:	1. **D**	2. **E**	3. **C**	4. **D**	5. **C**	6. **B**	7. **E**	8. **D**	9. **C**	10. **E**
Chapter V:	1. **A**	2. **B**	3. **D**	4. **D**	5. **B**	6. **C**	7. **B**	8. **D**	9. **A**	10. **B**
Chapter VI:	1. **D**	2. **B**	3. **A**	4. **E**	5. **D**	6. **A**	7. **C**	8. **A**	9. **D**	10. **C**
Chapter VII:	1. **D**	2. **C**	3. **E**	4. **B**	5. **E**	6. **D**	7. **A**	8. **C**	9. **B**	10. **C**
Chapter VIII:	1. **D**	2. **B**	3. **D**	4. **C**	5. **A**	6. **D**	7. **A**	8. **C**	9. **A**	10. **B**
Chapter IX:	1. **C**	2. **D**	3. **A**	4. **E**	5. **A**	6. **C**	7. **D**	8. **B**	9. **E**	10. **A**
Chapter X:	1. **D**	2. **B**	3. **A**	4. **E**	5. **A**	6. **C**	7. **B**	8. **A**	9. **E**	10. **D**
Chapter XI:	1. **B**	2. **D**	3. **B**	4. **D**	5. **C**	6. **E**	7. **D**	8. **C**	9. **B**	10. **D**

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Glossary

ADCC (antibody-dependent cell-mediated cytotoxicity) – a cytotoxic reaction in which Fc receptor-bearing killer cells recognize target cells via specific antibodies.

Acquired immunity – protection (acquired immunity) developed through an immune response mediated by the person's own antibodies or sensitized T-cells following stimulation with antigen.

Allergen – any substance capable of inducing an allergic response. Allergens are protein molecules, such as plant pollens and animal danders.

Allergy – an immediate hypersensitivity state acquired through exposure to a specific allergen; for example, pollens lead to hay fever.

Allergic Rhinitis – an inflammation of the lining of the nose caused by inhaling an allergen such as dust mite, pollens or animal dander and also by eating certain foods. Symptoms may include itching, sneezing, blocked nose, runny nose and itchy/watery eyes. Allergic Rhinitis is often referred to as hay fever.

Allergen Immunotherapy – a series of injections (shots) or sublingual drops are administered which contain allergens such as bee venom, pollen, dust mite or animal dander to which the patient is allergic. At first the amount given is a low dose, then the amount is increased at regular intervals, over a period of 3–5 years. Allergen immunotherapy alters the way in which the immune system reacts to allergens, by switching off allergy.

Anaphylatoxins – a group of mediators involved in inflammation that are produced and released in serum during complement activation (derived from the third and fifth components of complement). The fragments C5a, C3a, and C4a are all anaphylatoxins.

Anaphylaxis – an antigen-specific immune reaction mediated primarily by IgE which results in vasodilation and constriction of smooth muscle, including those of the bronchus, and which may result in death. Anchor residues. Certain amino acid residues of antigenic peptides are required for interaction with sites in the binding pocket of MHC molecules.

Anaphylactic shock – allergic reaction to systemically administered antigen that causes circulatory collapseand suffocation due to

tracheal swelling. It results from binding ofantigen to IgE antibody on connective tissue mast cells throughout the body, leading to the disseminated release of inflammatory mediators.

Antibody – a molecule produced by animals in response to antigen which has the particular property of combining specifically with the antigen which induced its formation.

Antigen – a molecule which reacts with preformed antibody and the specific receptors on T and B-cells. Antigen receptors. The lymphocyte receptors for antigens including the T-cell receptor (TCR) and surface immunoglobulin on B-cells which acts as the B-cell's antigen receptor (BCR).

Antigen-presenting cells – highly specialized cells that can process antigens and display their peptide fragments on the cell surface together with molecules required for lymphocyte activation.

Apoptosis – programmed cell death, is a form of cell death in which the cell activates an internal death program.

Arthus reaction – a local inflammatory reaction, characterized by necrosis, occurring a few hours after intradermal inoculation of antigen into an animal previously immunized to the same antigen.

Ataxia-telangiectasia – an immunodeficiency disease involving both T and B-cells. It is characterized by thymic hypoplasia and serum immunoglobulin deficiencies.

Atopic dermatitis – an inflammation of the skin which is reddened, swollen, itchy and often weeping (also known as atopic eczema).

Asthma – an allergic inflammation of the airways producing swelling, narrowing and the build up of mucus within the airway, leading to difficulty in breathing.

Autoantibodies – antibodies that are formed against self-antigens.

Autoimmunity – immune recognition and reaction against the individual's own tissue.

Autoimmune hemolytic anemia – pathological condition with low levels of red blood cells (anemia), which is caused by autoantibodies that bind red blood cell surface antigens and target the red blood cell for destruction.

Autoimmune response – adaptive immune response directed at self antigens.

B-cells – lymphocytes which develop in the bone marrow in adults and produce antibody. They can be subdivided into two groups, B1 and B2. B1-cells use minimally mutated receptors which are close to the germline immunoglobulin sequences, whereas B2-cells are the major responding population in conventional immune responses to protein antigens.

Basophils – a multinucleated leukocyte with granules containing acid glycoproteins that bind basic dyes, resulting in a blue color. When these granules release their contents, anaphylactic reactions may result.

Blast cell – a cell, usually large, undergoing division, containing large amounts of RNA in the cytoplasm and actively synthesizing DNA. For example, the stage of T- or B-cells between resting cells and effector cells following antigen stimulation.

Bruton agammaglobulinemia – a disease in young boys inherited as an X-linked recessive character. These patients have normal T-cell function and cell-mediated immunity to viruses but have only low levels of immunoglobulins and do not exhibit antibody responses. Thus, they have great difficulty coping with bacterial infections.

Chediak – Higashi syndrome – defect in phagocytic cell function, of unknown causes, in which lysosomes fail to fuse properly with phagosomes and there is impaired killing of ingested bacteria.

Chemotaxis – the increased directional migration of cells to a site across a concentration gradient.

Clusters of differentiation (CD) – groups of monoclonal antibodies that identify the same cell-surface molecule. The cell-surface molecule is designated CD followed by a number (e.g. CD1, CD2, etc.).

Complement – a group of scrum proteins involved in the control of inflammation, the activation of phagocytes and the lytic attack on cell membranes.

Cytokines – proteins made by cells that affect the behavior of other cells. Cytokines made by lymphocytes are often called lymphokines or interleukins (abbreviated IL), but the generic term cytokine is used in this book and most of the literature. Cytokines act on specific cytokine receptors on the cells that they affect.

Cytotoxic T-cells – effector cells of cell-mediated immunity that can lyse allogeneic or virally infected cells. $CD8^+$ cytotoxic T-cells recognize these target cells by reacting with surface antigens associated with class I MHC molecules. Cytotoxic T-cells are important in host defense against cytosolic pathogens.

Delayed-type hypersensitivity (DTH) – a form of cell-mediated immunity that is mediated by T-cells and activated macrophages and takes 24 to 72 hrs to reach its peak after challenge of an immune (sensitized) individual. Contact dermatitis, seen following contact with the leaves of the poison ivy plant, is an example of delayed-type hypersensitivity.

Dendritic cells – interdigitating reticular cells, are found in T-cell areas of lymphoid tissues. They have a branched or dendritic morphology and are the most potent stimulators of T-cell responses. Non-lymphoid tissues also contain dendritic cells, but these do not seem to stimulate T-cell responses until they are activated and migrate to lymphoid tissues. The dendritic cell derives from bone marrow precursors. It is distinct from the follicular dendritic cell that presents antigen to B-cells.

Di George syndrome – recessive genetic immunodeficiency disease in which there is a failure to develop thymic epithelium, and is associated with absent parathyroid glands and large vessel anomalies. It seems to be due to a developmental defect in neural crest cells.

Drug-induced autoimmune hemolytic anemia – different drugs cause immunity by four different mechanisms: 1) immune complex formation; 2) drug adsorption to red cell membranes; 3) modification of red cell membranes; 4) idiopathic means.

Dysgammaglobulinemia – an immunodeficiency disease involving B-cells. It shows an imbalance mainly in IgA, but all combinations of immunoglobulin imbalance may occur.

Effector cells – lymphocytes that can mediate the removal of pathogens from the body without the need for further differentiation, as distinct from naive lymphocytes, which must proliferate and differentiate before they can mediate effector functions, and memory cells, which must differentiate and often proliferate before they become effector cells. They are also called armed effector cells in this book, to indicate that they can be triggered to effector function byantigen binding alone.

Endotoxins – bacterial toxins that are released only when the bacterial cell is damaged, as opposed to exotoxins, which are secreted bacterial toxins. The most important endotoxin is the lipopolysaccharide of Gram-negative bacteria, which is a potent inducer of cytokine synthesis.

Eosinophils – phagocytic cells that make up 2 to 5 % of the blood leukocytes. Eosinophils contain granules composed of a basic protein and histaminase. The basic protein is released by exocytosis, leading to damage to some pathogens. The histaminase can downreg-ulate inflammatory reactions.

Epstein – Barr virus (EBV) – causal agent of Burkitt's lymphoma and infectious mononucleosis, which has the ability to transform human B-cells into stable cell lines.

Farmer's lung – hypersensitivity disease caused by the interaction of IgG antibodies with large amounts of an inhaled allergen in the alveolar wall of the lung, causing alveolar wall inflammation and compromising gas exchange.

Forbidden clones – clones proposed by Burnet to explain that autoreactive immune cells are forbidden to react with self-constituents by being clonally deleted during ontogeny. It is now known that autoreactive immune cells exist but are in an inactive state.

Genotype – genetic constitution of an individual inherited from both parents; the genotype is not necessarily completely expressed in that individual.

Graves' disease – an antibody-mediated autoimmune disorder. Unlike other autoimmune disorders, it results directly from autoantibodies that stimulate thyroid cellular activity by displaying thyroid-stimulating hormone binding.

Graft rejection – a T-cell-mediated immune reaction elicited by grafting genetically dissimilar tissue to a recipient. The outcome of the reaction is the destruction and rejection of the transplanted tissue.

Goodpasture syndrome – autoimmune disease in which autoantibodies against basement membrane or type IV collagen are produced and cause extensive vasculitis.

Hapten – a substance that can combine with specific antibody but cannot initiate an immune response unless it is bound to a larger "carrier" molecule. In other words, a hapten is antigenic but not immunogenic by itself.

Hashimoto's thyroiditis – autoimmune disease characterized by persistent high levels of antibody against thyroid-specific antigens. These antibodies recruit NK-cells to the tissue, leading to damage and inflammation.

Hemolytic disease of the newborn – a disease that results from a mother's IgG antibodies crossing the placenta and destroying the fetus's red blood cells. The Rh^- mother becomes sensitized during the birth of the first child if the fetus's Rh^+ red cells enter the mother's circulation. The mother produces anti-Rh antibodies of the IgG class. These antibodies can cross the placenta during subsequent pregnancies, destroy fetal red blood cells, and result in the disease state.

Histocompatibility – sharing of transplantation antigens between two individuals. The genes coding for surface antigens that induce graft rejection. The MHC is the most important loci for these genes; however, the MHC contains genes that have other important roles in the immune response.

Humoral immunity – acquired immunity mediated by specific antibodies found in the blood and tissue fluids of the body.

Hyperacute graft rejection – the speed with which a graft such as kidney is rejected. In this case, the rejection occurs within minutes of the transplant due to the presence of high titers of preformed antibodies to the graft's antigens.

Hypersensitivity – the state, existing in a previously immunized (sensitized) individual, that leads to tissue damage on subsequent exposure to the antigen (allergen); for example, a form of allergy called bay fever.

Immunodeficiency diseases – group of inherited or acquired disorders in which some aspect or aspects of host defense are absent or functionally defective.

Immunologic memory – the faster and more powerful response of a host's immune system to an antigen on subsequent exposures. A heightened secondary immune response.

Immunotherapy – efforts to attack tumor cells through the immune system, which focus on stimulating or replenishing antitumor elements of the patient's immune response without further stimulating the suppressor cells.

Innate immunity – the defenses of a host that exist before, and function independently of, any exposure to foreign antigen. The immunity individuals have by being the individuals they are.

Insulin-dependent diabetes mellitus (IDDM) – β-cells of the pancreatic islets of Langerhans are destroyed so that no insulin is produced. The disease is believed to result from an autoimmune attack on the β-cells.

Interferons – cytokines that can induce cells to resist viral replication. **Interferon-α (IFN-α)** and **interferon-β (IFN-β)** are produced by leukocytes and fibroblasts respectively, as well as by other cells, whereas **interferon-γ (IFN-γ)** is a product of CD4 Th_1-cells, CD8 T-cells, and NK-cells. IFN-γ has as its primary action the activation of macrophages.

Interleukin – a generic name given to molecules secreted by lymphoid cells allowing them to communicate with each other.

K-cells – a group of lymphocytes which arc able to destroy their target by antibody-dependent cell-mediated cytotoxicity.

Langerhans' cells – macrophages in the skin that express large numbers of class II (Ia) molecules on their surface. They can pick up antigen and carry it to the regional lymph nodes. In the skin they are known as veiled cells and in the lymph nodes as dendritic cells.

LE-cells – neutrophils that have internalized lymphocytic nuclear material complexed with antibodies (lupus erythematosus [LE] factor). These cells are found in patients with the autoimmune disease systemic lupus erythematosus (SLE).

Leukemia – unrestrained proliferation of a malignant white blood cell characterized by very high numbers of the malignant cells in the blood. Leukemias can be lymphocytic, myelocytic, or monocytic.

Leukocyte – general term for a white blood cell. Leukocytes include lymphocytes, polymorphonuclear leukocytes, and monocytes.

Lymphocytes – spherical cells (7 to 10 μm in diameter) with a large, round nucleus surrounded by a small amount of cytoplasm. They are associated with all aspects of specific immunity. Lymphocytes are the principal constituents of lymphoid tissue.

Lysozyme – an enzyme found in tears, saliva, and nasal secretions lysing chiefly gram-positive cocci. It splits the muramic acid β(1–4)-N-acetylglucosamine linkage in the cell walls of the appropriate bacteria.

Macrophages – any of the diverse group of cells (not including granulocytes) characterized by the capacity to engulf (phagocytize) and destroy foreign substances. They play a pivotal role in nonspecific (removal of foreign material) and specific immunity (processing and presenting antigen to T-cells).

Mast cells – cells found distributed near the blood vessels in most tissues. These cells are full of granules containing inflammatory mediators.

Major histocompatibility complex (MHC) (HLA complex) – a region on a chromosome that contains genes controlling the expression of surface histocompatibility antigens and the capacity to generate an immune response.

Membrane attack complex – C5b-9 complex of the complement activation sequence responsible for lysis of target cells.

Memory cells – long-lived lymphocytes which have already been primed with their antigen, but have not undergone terminal differentiation into effector cells. They react more readily than naive lymphocytes when restimulated with the same antigen.

Monoclonal antibodies – antibodies produced by a single clone of cells. The resultant antibody molecules are identical in all aspects, such as affinity, binding specificity, isotype. allotype, idiotype. etc.

Monocytes – immature macrophages found in the blood. They differentiate into macrophages once they leave the circulation.

Myasthenia gravis – an antibody-mediated autoimmune disease characterized by muscular weakness and excessive fatigue. It is due to reduction in the number of acetylcholine receptors, which help trigger muscle contraction.

Natural killer cells (NK-cells) – large, usually granular, non-T-, non-B-lymphocytes, which kill certain tumor cells. NK-cells are important in innate immunity to viruses and other intracellular pathogens, as well as in antibody-dependent cell-mediated cytotoxicity (ADCC).

Neutrophils – polymorphonuclear granulocytes, which form the major population of the blood leucocytes.

Oncofetal antigens – tumor-associated antigens that normally appear on the surface of embryonic cells and are reexpressed on the surface of some adult neoplastic cells. They are a diagnostic tool for tumor load and recurrence of metastases after therapy.

Oncogenes – genes involved in regulating cell growth. When these genes are defective in structure or expression, they can cause cells to grow continuously to form a tumor.

Opsonization – alteration of the surface of a pathogen or other particle so that it can be ingested by phagocytes. Antibody and complement opsonize extracellular bacteria for destruction by neutrophils and macrophages.

Phagocytosis – internalization of particulate matter by cells. Usually, the phagocytic cells or phagocytes are macrophages or neutrophils, and the particles are bacteria that are taken up and destroyed.

Receptor – a cell surface molecule which binds specifically to particular extracellular molecules.

Regulatory T-cell – as part of the immune response, regulatory T-cells specialize in telling B-cells when to stop making antibodies. They also instruct T-cells to call off an assault at the end of an immune reaction.

Rheumatoid arthritis (RA) – an autoimmune disease exemplified by inflammation and deterioration of the joints.

Rheumatoid factor – anti-immunoglobulin antibody directed against IgG and found in the serum of some patients with rheumatoid arthritis.

Serum sickness – an adverse reaction to foreign antigen that occurs because the antigen was a heterologous protein. Serum sickness is an example of a type III hypersensitivity,.

Specific immunotherapy – cancer immunotherapy that relies on vaccines of tumor cells or tumor cell functions administered directly to the patient to increase or induce a host immune response to tumor-specific antigen.

Swiss-type agammaglobulinemia – an immunodeficiency disease involving both B- and T-cells. The thymus, T-cell immunity, and B-cell immunity are absent.

Systemic lupus erythematosus (SLE) – an autoimmune disease primarily of women, usually involving antinuclear antibodies.

T-lymphocytes (T-cells) – precommitted lymphocytes from the bone marrow that enter the thymus, briefly reside there, and leave as mature functional T-cells, hence the name thymus-derived or T-lymphocytes. T-cells are involved directly in cell-mediated immunity as effectors or indirectly in humoral and cell-mediated immunity as regulatory cells.

Thrombocytopenic purpura – autoimmune form of thrombocytopenia, caused by increased platelet destruction primarily following

coating of platelets with antibody. The antigenic stimulus for the disease is unknown.

Transforming growth factor-β (TGF-β) – a cytokine that may limit an immune response. Actually a family of molecules, it seems to be made by all cells. It helps to transform neoplastic cells and may spur tumor-mediated immunosuppression.

Transplantation (or grafting) – the placement of cells, tissues, or organs from a donor in a recipient (an exception is an autograft). Unless the donor and recipient are perfectly matched for histocompatibility antigens, the recipient will mount an immune response.

Tumor necrosis factor-α (TNF-α) – cytokine produced by macrophages and T-cells that has multiple functions in the immune response. It is the defining member of the TNF family of cytokines.

Vaccine – an immunogen-containing substance which, on introduction into an animal or individual, stimulates active immunity for future protection against infection by the appropriate organism.

Wiskott – Aldrich syndrome – an immunodeficiency disease involving both T- and B-cells. Cell-mediated immunity is absent, and antibody responses are defective. Patients cannot respond to polysaccharide antigens because of their low levels of serum IgM.

X-linked agammaglobulinemia (XLA) – genetic disorder in which B-cell development is arrested at the pre-B-cell stage and no mature B-cells or antibodies are formed. The disease is due to a defect in the gene encoding the protein tyrosine kinase.

X-linked hyper IgM syndrome – disease in which little or no IgG, IgE, or IgA antibody is produced and even IgM responses are deficient, but serum IgM levels are normal to high. It is due to a defect in the gene encoding the CD40 ligand.

Supplements

Supplement 1

Immunotropic preparations which are allowed to be applied on the territory of Ukraine (according to State List of Essential Medicines. 4th ed. – K., 2012. Approved by the Ministry of Health of Ukraine (from 28.03.2012. № 209 (with abridgements).

Immunomodulators

1. Immunoglobulins.

1) General:

- *human normal immunoglobulin* (increases nonspecific resistance of the organism) (Venoimmun, human normal immunoglobulin).

2) Specific:

- *anticytomegalovirus immunoglobulin* (antiviral action + raises nonspecific resistance of the organism) (human anticytomegalovirus immunoglobulin, Cytobiotect);
- *human immunoglobulin versus herpes simplex virus* (antiviral action + raises nonspecific resistance of the organism) (Gamolin, human immunoglobulin versus herpes simplex virus of the first type, human immunoglobulin versus herpes simplex virus of the second type);
- *human immunoglobulin versus Epstein – Barr virus* (virus-neutralizing action + raises a specific resistance of the organism) (human immunoglobulin versus Epstein – Barr virus);
- *immunoglobulin human antirhesus Rh0(D)* (prevents rhesus sensibilization of the rhesus-negative women as a result of entering rhesus positive (fetal blood) into the mother's blood stream) (immunoglobulin antirhesus Rh0(D) of the human).

2. Cytokines.

1) Interferons:

a) natural compounds;

- *human leukocyte interferon (wide range of antiviral action)* (human leukocyte interferon, dry).

b) recombinant compounds:

- *interferon alpha-2b* (immunoglobulin of antiproliferative and antiviral action) (Introbion, Laferon-PharmBiotek, Alpharekin, Laferobion, Heberon alfa R, Alpharon, Bioferon, Realdiron, Shanferon, Viferon, Intron, Nazoferon);
- *interferon beta-1a* (immunomodulation and antiviral action). (Betabioferon-1a, Imunoferon 1a, Avonex, Rebif);
- *interferon beta-1b* (antiviral action, increases suppressants activity of the mononuclear cells of the peripheral blood) (Betabioferon-1b, Imunoferon 1b, Betaferon);
- *peginterferon alpha-2a* (antiviral and antiproliferating action, inhibition of the hepatitis C virus replication (Pegasys, Peginterferon alpha 2a);
- *peginterferon alpha-2b* (a covalent conjugate of the recombinant interferon alpha-2b monometaxepolyethylenglycol) (Pegintron).

2) Growth factors:

- *filgrastim* (colony-stimulating factor – stimulates growth and differentiation of the cells of the functionally active neutrophils) (Granogen, Neutrogran, Grasalva, Grastim, Leukostim);
- *lenograstim* (colony-stimulating factor. It stimulates growth and differentiation of the functionally active neutrophils (Granocyte);
- *epoetin alpha* (stimulates erythropoiesis (Shanpoetin, Epoetal, Eprex);
- *epoetin beta* (favours erythrocytes creation of stem cells) (Veroepoetin, Erythrostim, Recormon).

3) Interleukins:

- *recombinant human Interleukin-2* (influences the growth, differentiation and activation of T- and B-lymphocytes, macrophages, oligodendroglial cells, development of the cytolytic activity of NK-cells, activazes the tumor – its infiltrating cells) (Bioleukin, Roncoleukin).

3. Interferon inductors.

1) Natural compounds:

- *kagocel* (the inductor of producing the mixture of α-, β- and γ-interferons) (Kagocel);
- *protfenolozidum* (direct antiviral action, stimulates the induction of α- and γ-interferons, strengthens the thymus endocrine function, normalizes quantitive composition of T-lymphocytes in the peripheral blood and strengthens the cytotoxic activity of natural killers (Protfenolozid).

2) Synthetic compounds:

- *inosine pranobex* (antiviral, immunomodulating action, influences the cell immune response, stimulates antiviral defense and suppresses virus reproduction) (Groprinosin, Isoprinosine);
- *tilorone* (direct antiviral action, stimulates the creations of α-, β-, γ-interferons; stimulates stem cells of the bone marrow) (Amixin, Lavomax);
- *cicloferon* (direct antiviral action, stimulates α-, β-, γ-interferon creation; activizes phagocytosis, natural killers cells, cytotoxic T-lymphocytes, strengthens the effect of the antibiotic therapy in case of intestinal infections, supresses autoimmune reactions. It has an anticancerogenic and antimetastatic action (Cicloferon);
- *amizonum* (inhibiting influence on the flu viruses, antioxidant action, increases the level of the endogenic interferon and lysocim. It also stimulates the functional activity of T-cells, macrophages and has antipyretic effect (Amizon);
- *umifenovir* (direct antiviral action on the flu viruses and other respiratory viruses, an inductor of interferon. It strengthens the phagocytic function of macrophages) (Immustat, Arbidol, Arbimax, Arbivir-Zdorovje);
- *heponum* (direct antiviral action, stimulates α-, β-interferons creation, activates macrophages, lessens the HIV concentration in cells and plasma, increases the organism resistance to infections coused by bacteria and fungi (Hepon).

4. Preparations of thymic origin.

1) Natural compounds:

- *timaline* (regulates the number and correlation of T- and B-cells and their subpopulations, stimulates reactions of cellular immunity, strengthens phagocytosis, stimulates processes of regeneration and blood creation in case of their suppresion (Timaline);
- *vilosen* (stimulates proliferation and differentiation of T-cells, suppresses the creation of reagins) (Vilosen).

2) Synthetic compounds:

- *immunofan* (produces formation of specific antiviral and antibacterial antibodies, strengthens the phagocytosis reactions, has detoxicating and hepatoprotecting action (Immunofan);
- *thymosin alpha* (induces markers of differentiation of mature T-cells on lymphocytes of the peripheral blood, strengthens T-cells functions, efficiency of their maturation and capacity of producing cytokines (INF-y, IL-2, IL-3), increases the activity of natural killer cells (zadaxin).

5. Other preparations.

- *glutoxim* (strengthens bone marrow blood creation, activates the phagocytosis system, has cytoprotecting action (Glutoxim).

6. Preparations of bacterial origin.

- *liastenum* (stimulates function of macrophages – phagocytosis and cytotoxic effect on tumor cells, normalizes the number of T-cells, strengthens the synthesis of antiinflammatory cytokines, support leukopoiesis, reduces side effects of chemio- and radiation-therapy (Liasten);
- *IRS 19* (causes protective immune reactions in the tunica mucosa which are identical to those on pathogenic microorganisms, stimulates and reproductes immunocompetent cells, phagocytosis, increases the level of lysocim, interferon and IgA in nasal secretion (IRS 19);
- *imudon* (activates phagocytosis, increases the quantity of immunocompetent cells, raises levels of lysocim and secretory IgA in saliva (Imudon);

- *ribomunyl* (strengthens phagocytosis, stimulates T-cells and production of serumal and secretory IgA (Ribomunile);
- *respibron* (as a vaccine stimulates durable specific immune response, as an immunomodulator increases the level of serumal and secretory antibodies, activates cellular and humoral factors of nonspecific immunity (Respibron);
- *bifiform* (restores normal balance of the intestinal microflora (Bifiform, Bifi-form);
- *lactobacterinum* (an antagonist of the pathogenic and prearranged pathogenic microorganisms – restores normal balance of the intestinal microflora) (Lactobacterin dry);
- *bifidumbacterinum* (restores the normal balance of the intestinal microflora) (Bifidumbacterin).

7. Preparations of fungal origin.

- *ribonucleinic acid* (stimulates leukopoesis in the bone marrow, increases migration and cooperation of T- and B-cells, phagocytic activity of macrophages, production of interferons and endogenic glucocorticoids, suppresses increased aggregation of the thrombocytes, accelerates processes of regeneration and the antiinflammatory action (Nucleinas);
- *baker's yeast* (vitamin B group, aminoacids and mineral elements activates the enzyme system, the processes of metabolism and stimulates nonspecific resistance and regeneration of tissues (Encad, Brewers yeast).

8. Preparations of animal origin.

- *erbisolum* (low molecular biologically active peptides activate the macrophagal system, IL-1-cells and T-killers, inhibits the activity of IL-2 and B-lymphocytes; increases the α-, β-, and γ-interferon's and the tumor necrosis factor synthesis has a hepatoprotective effect (Erbisol);
- *extra erbisolum* (decreases processes of peroxidative oxidation of lipids, provides membrane stabilizing effect of plasmatic membranes, improves microcirculation (Extra erbisol);

- *inflamafertin* (stimulates phagocytic activity of the tunica mucosa and blood cells, strengthens synthesis of the inflammatory cytokines, influences the activity of the regulatory subpopulations of the lymphocytes; act as anti-inflammatory drug; reduces the formation of commissures (Inflamafertin).

9. Preparation of plant origin.

1) of one plant:

- *panavir* (inhibition of viral protein synthesis, increases nonspecific resistance; assists interferon synthesis) (Panavir);
- *maximune* (assists in increase of neutrophils and B-lymphocyts; stimulates regeneration of the liver tissues;) increases diuresis; lowers the level of sugar and urea in the blood, has anti-inflammatory and antipyretic action) (Maximune);
- *echinacea purpurea* (stimulates phagocytosis and hemotaxis of leukocytes; increases the level of the factor of tumor necrosis, properdin, glucocorticoids in the blood; stimulates an alternative way of complement activation) (Echinacea tincture, Echinacin Madaus, Echinal, Echinacea purpurea liquid extract, Echinacea purpurea tincture, Echinacea, Echinacea syrup, Echinacea purpurea root and rhizome tincture, Echinacea dried pressed juice, Immuno-plus, Immunal, Immuno Theiss);
- *imunin-norton* (stimulates stimulates nonspecific organism's resistance, activates phagocytosis, stimulates the production of interferons, has an antioxidant and enterosorbing effect; stimulates hemoglobin synthesis) (Imunin-norton);
- *ginseng* (activates metabolism, stimulates nonspecific organism's resistance, has neurotropic action) (Ginseng, Ginseng tincture, Herbion Ginseng, Gerimax Ginseng, Nguyen Nhan Sam);
- *eleutherococcus* (activates metabolism, stimulates nonspecific resistance of the organism, has an insignificant gonadotropic and hypoglycemic action) (Eleutherococcus, Eleutherococcus Extract);
- *licorice root* (activates metabolism, stimulates nonspecific resistance of the organism, has an expectorant effect (Licorice root extract).

2) Composite preparations:

- *eleutherococcus + echinacea purpurea + tutsan* (Immuno-ton);
- *ginseng + schisandra chinensis + royal jelly* (Ginseng, Royal Jelly);
- *narrow-leaved echinacea + small-leaved linden (lime tree) + rosa canina + clasping-leaved mullein + thyme + ascorbic acid + eucalyptus oil + mint oil* (Grippal with lime flowers and vitamin C);
- *echinacea purpurea + wild rose hips* (Echinasal);
- *echinacea purpurea + black leaf tea* (Golden Root).

10. Synthetic immunomodulators.

1) Low molecular:

- *galavit* (favour the normalization of microphages functional states in case of inflammatory processes. It inhibits an excessive synthesis of the tumor necrosis factor, IL-1 for 6–8 hrs, and suppresses devouring of the active oxygen forms by hyperactive macrophages. It leads to the lowering of the inflammatory reactions intensity and reduces the degree of intoxication. It also stimulates the microbicidal system of the neutrophils and accelerates phagocytosis (Galavit);
- *methyluracil* (stimulates the synthesis of nucleic acids that leads to the acceleration of the cellular regeneration processes. It also has an erythro- and leukopoietic effect. It influences the organism (Methyluracil).

2) High molecular:

- *glatiramer acetate* (blocks myelin specific autoimmune response in case of multiple sclerosis, causes activation and proliferation of the clones of Th_2-lymphocytes, participates in the mechanisms of the activated cells apoptosis (Copaxone);
- *polyoxidonium* (direct stimulating influence on the phagocytizing cells and natural killers; stimulation of detoxicating action; increases the resistance of the cell membranes to the cytotoxic substances (Polyoxidonium).

11. Vitamins, minerals.

Composite preparations:

- *ascorbic acid + rutin* (Ascorutin, Imunovit);
- *beta-carotene + vitamin C + vitamin E + selenium* (Vitrum);
- *vitamin E + beta-carotene + vitamin C + selenium* (Trivit+SE-KB);
- *vitamin A + vitamin C + zinc + vitamin E + selenium + copper* (Visital, Tri-V plus);
- *lutein + zeaxanthin + bilberry + vitamin C + vitamin E + beta-carotine + zinc + vitamin B_2 + selenium + rutin* (Vitrum Foreyes Forte);
- *folic acid + cyanocobalamin + iron + selenium + zinc* (Globingen);
- *iron + zinc+ manganese + copper + cobalt + chromium + selenium + molybdenum + vanadium + mefenamic acid* (Esmin);
- *yohimbine + ginseng + ascorbic acid + selenium + zinc* (Yohimbex Garmonia).

12. Homeopathic preparations.

- *anaferon* (antibodies for human INF-γ) (Anaferon);
- *aflubin* (Aflubin);
- *immunokind* (Immunokind);
- *mercuride* (Mercuride).

13. Others.

- *mumiyo* (induces the production of endogenic interferons and natural killers, assists the acceleration of reparative processes, positively influences the liver function (Mumiyo, Tien Shan);
- *apilac* (stimulates metabolism and nonspecific resistance of the organism) (Apilac);
- *pantocrine* (activates enzyme systems, stimulates metabolism and nonspecific resistance of the organism) (Pantocrine)
- *pidotimod* (in case of T-lymphocytes deficiency it induces maturation and differentiation of T-lymphocytes, stimulates macrophages (Imunorix).

Immunodepressants

1. Glucocorticosteroids:

1) systemic;
2) topical.

2. Natural compounds:

- *tacrolimus* (suppresses the formation of cytotoxic lymphocytes, reduces the T-lymphocytes activation, B-lymphocytes proliferation, production of lymphokines, expression of IL-2 receptor) (Advagraf, Vingraf, Prograf, Protopic, Tagraf, Tacrol, Tacrolimus, Strides);
- *cyclosporine* (syppresses development of the reactions of the cellular type and T-depending formation of antibodies, formation of lymphokines including IL-2 (Ivax, Lifemun, Sandimmun, Bioral, Equoral);
- *anti-thymocyte immunoglobulin* (selective immunodepressant of T-lymphocytes) (Atgam).

3. Synthetic compounds:

- *leflunomide* (has an antiproliferative action regarding the activated lymphocytes, plays an important role in the pathogenesis of autoimmune T-cellular mediated diseases (Arava, Lefno);
- *mycophenolic acid* (inhibits T- and B-lymphocytes proliferation (Mycophenolate mofetil, Mofilet, Myfortic, Imufet, Cellcept);
- *azathioprine* (suspension of the proliferation of cells participating in the immune response) (Imuran);
- *everolimus* (inhibitor of T-cells activation and proliferation, suppresses the proliferation of blood creating cells that is vascular cells of muscles stimulated by the growth factor) (Sertican);
- *pimecrolimus* (inhibits the formation and release of anti-inflammatory cytokines by T-cells and mast cells (Elidel).

4. Immunosuppressors based on monoclonal antibodies.

- *infliximab* (with a high affinity ties together the early and transmembrane forms of tumor necrosis factor α) (Remicade);
- *retuximab* (initiates immune responses which cause B-lymphocytes lysis (Mabthera);
- *bevacizumab* (neutralizes the growth factor of the vessels of endothelium which leads to the lessening of vascularization and suppression of the tumor growth (Avastin);

- *cetuximab* (neutralizes receptors of the epidermic growth factor that reduces the activity of the tumor neovascularization and metastasis. This preparation induces the apoptosis of the tumor cells which express the receptor of the epidemic growth factor) (Erbitux);
- *alemtuzumab* (by means of binding with CD52 it causes the lysis of the T- and B-lymphocytes, monocytes, thymocytes and macrophages) (Mabcampath);
- *basiliximab* (monoclonal antibodies against A-chain of the IL-2 receptor which express on the surface of T-cells as a result to the antigenic stimulation; it leads to the persistent blocking of the IL-2 receptor and to the inhibition of the T-cells proliferation) (Simulect).

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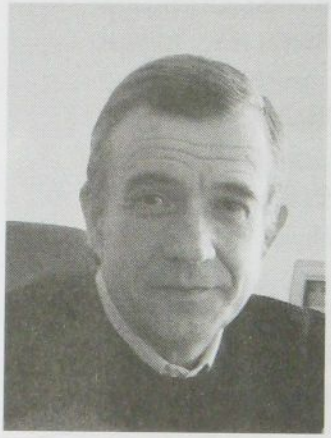

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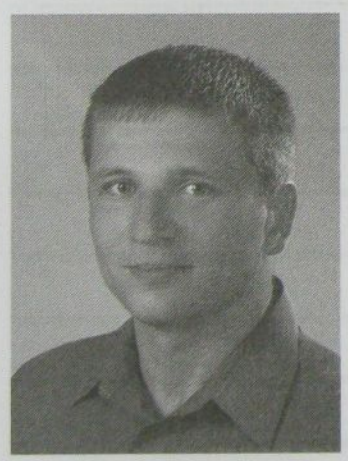

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У підручнику викладені основні теми з клінічної імунології та алергології відповідно до типової програми, затвердженої МОЗ України в 2013 р. Враховуючи клінічну спрямованість програми, в кожній темі викладаються, в основному, питання діагностики, особливості патогенезу і перебігу захворювань та ін. Для полегшення засвоєння матеріалу книга містить багато ілюстрацій, тестові завдання і контрольні питання. Для англомовних студентів і викладачів медичних вузів III–IV рівнів акредитації.

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