Ministry of Health of Ukraine
Poltava State Medical University

Approved"
at the meeting of the Department of
of Internal Medicine No. 3, Phthisiology
“___”_____20___
p.
Minutes № from
Head of the Department
Associate Professor _____ PhD
Borzykh O.A.

METHODOLOGICAL RECOMMENDATIONS FOR CONDUCTING AND PREPARING FOR PRACTICAL CLASSES

Academic discipline	Clinical immunology and allergology
*Module **4***	Clinical immunology and allergology
Content module	Clinical immunology and allergology
Topic №4	Congenital immunodeficiency diseases. Age-related immunology
Course	5
Hours	2

Poltava 2023

Methodological recommendations for the practical training for independent work of students in preparation for the practical training and during the class were prepared by:
Assistant of the Department of Internal Medicine №3 with phthisiology Bilko V.V.

Methodological recommendations were re-approved at the meeting of the Department of Internal Medicine of Internal Medicine №3 with Phthisiology_____
_____
_____
_____

1. Relevance of the topic.

Resistance to infections is due to the body's defence mechanisms. The first line of defence is represented by mechanical barriers of the skin and mucous membranes. The barrier function of the mucous membranes is complemented by the functioning of the ciliated epithelium, the protective properties of mucus, lysozyme, lactoferrin and interferons. Complement, neutrophils and macrophages, which are the body's second line of defence against foreign matter, are involved in the removal of microbes that have penetrated the skin and mucous membranes. However, the main role in resistance to infection is played by antibodies, T- and B-lymphocytes. Therefore, congenital defects in the structure and function of lymphocytes most often lead to primary immunodeficiencies. Although primary immunodeficiencies are rare, their detection should be intensively pursued, because such children can be foci of spread of a wide variety of pathogens against the background of suppression of the immune system function.

Overall objective: To study the mechanisms of development, clinical signs, peculiarities of immunodiagnostics, approaches to the treatment of congenital T- and B-dependent immunodeficiency diseases caused by impaired phagocytic immunity and complement protein deficiency**.**

Specific objectives:

1. To determine the specifics of primary immunodeficiencies.
2. To make a programme of examination of the patient.
3. Learn the classification of primary immunodeficiencies.
4. Determine the severity of clinical manifestations and learn to differentiate the symptoms of a particular primary immunodeficiency.
5. Establish a diagnosis and identify its complications.
6. Characteristics of the main types of primary immunodeficiencies.
7. Determine the tactics of treatment and prevention of the disease, taking into account clinical manifestations and features of the course.

Theoretical questions for practical training:

1. Primary immunodeficiencies, classification, diagnosis.
2. Study of humoral immunity.
3. Studies of cellular immunity.
4. General principles of treatment of immunodeficiencies.
5. Clinical forms of primary immunodeficiencies.
6. Etiology, pathogenesis of primary immunodeficiencies.
7. Clinical manifestations, immunological status, treatment of congenital immunodeficiencies.

Approximate basis of action

Primary immunodeficiencies are disorders of the immune system with which a person is born. Most often they are detected in the first months of life, in some cases the first manifestations occur in adolescence, or, even less often, in adults.

Patients with severe primary immunodeficiencies usually die in childhood. In case of moderate and mild clinical manifestations of primary immunodeficiencies, patients can reach adulthood. However, in almost all cases of primary immunodeficiency, the prognosis is favourable.

Thus, primary immunodeficiencies are disorders associated with genetic defects in the development of the immune system, which sooner or later lead to certain clinical manifestations.

Classification of primary immunodeficiencies

I. Deficiency of humoral immunity:

1. X-linked agammaglobulinaemia (hypogammaglobulinaemia) (Bruton's syndrome).

2. General variable immunodeficiency (general variable hypogammaglobulinaemia).

3. Transient hypogammaglobulinaemia in children (slow immunological start).

4. Selective immunoglobulin deficiency (dysgammaglobulinaemia).

II. Deficiency of the cellular immune system:

1. DiGiorgi syndrome (hypo-, aplasia of the thymus).

2. Chronic mucocutaneous candidiasis.

III. Combined T- and B-immunodeficiencies:

1. Severe combined immunodeficiency:

a) X-linked;

b) autosomal recessive.

2. Ataxia - telangiectasia (Louis-Bar syndrome).

3. Wiskott-Aldrich syndrome.

4. Immunodeficiency with increased levels of immunoglobulin M (linked to the X chromosome).

5. Immunodeficiency with dwarfism.

IV. Deficiency of the phagocyte system:

1. Chronic granulomatosis.

2. Cedak-Steinbrink-Higashi syndrome.

3. Hyperimmunoglobulinaemia E syndrome (Job's syndrome).

4. Deficiency in the expression of adhesion molecules.

V. Deficiency of the complement system.

1. Congenital angioedema.

Diagnosis of primary immunodeficiencies

Anamnesis is taken and a physical examination is performed. This allows you to assume which part of the immune system is mainly affected and plan laboratory tests. Physical examination is very important for assessing the effectiveness of treatment of immunodeficiencies. Primary immunodeficiencies are usually congenital and are detected in the first year of life.

Investigation of humoral immunity

Determination of the number of B-lymphocytes. Determination of B-lymphocytes by flow cytometry is based on the detection of immunoglobulins fixed on the cell surface, CD19 and CD20. In older children and adults, B-lymphocytes make up 10-20% of all blood lymphocytes, and in younger children, their number is higher.

Determination of antibody titer. If humoral immunity deficiency is suspected, the titer of antibodies to protein and polysaccharide antigens is assessed. They are usually determined after vaccination or infection. Antibodies to protein antigens. In most cases, IgG to diphtheria and tetanus toxoids are tested before and 2-4 weeks after DPT or DTaP vaccination. Since almost all adults are vaccinated with DPT, the level of antibodies after revaccination is an indicator of the secondary immune response. Antibodies to the PRP antigen can also be detected after administration of Haemophilus influenzae type B vaccine. Although this antigen is a polysaccharide, it acts as a protein antigen in a conjugate vaccine. Antibodies are sometimes tested

after immunisation with inactivated polio vaccine and recombinant hepatitis B vaccine.

Live viral vaccines are contraindicated in case of suspected immunodeficiency. Antibodies to polysaccharide antigens. To assess the humoral immune response to polysaccharide antigens, pneumococcal and meningococcal vaccines that do not contain protein carriers are used. The antibody titer is determined before and 3-4 weeks after vaccination. Some research laboratories use an unconjugated Haemophilus influenzae type B vaccine for this purpose. The results are evaluated taking into account the age of the patient. For example, in children under 2 years of age, the immune response to polysaccharide antigens is weak, and in some children it remains so for up to 5 years. In this regard, the use of polysaccharide vaccines in young children is inappropriate and even contraindicated, as it can lead to immunological tolerance and ineffectiveness of revaccination at an older age.

Assessment of primary and secondary humoral immune response. To determine antigen clearance, IgM (primary immune response) and IgG (secondary immune response) levels, bacteriophage phyci 174, a bacterial virus that is safe for humans, is used as a protein antigen.

Gastropod haemocyanin, recombinant hepatitis B vaccine, monomeric flagellin, and tick-borne encephalitis vaccine are also used to assess the primary humoral immune response. Natural antibodies (isohemagglutinins, antibodies to streptolysin O, heterophilic antibodies, such as antibodies to sheep red blood cells) are normally present in the serum of almost all people. This is because the antigens against which these antibodies are directed are very common and are found in food, inhaled particles, and respiratory tract microflora.

Determination of IgG subclasses. If, in recurrent bacterial respiratory tract infections, the total IgG level is normal or slightly decreased, or an isolated IgA deficiency is detected, IgG subclass determination is indicated. In this case, an IgG2 deficiency can be detected (IgG2 makes up about 20% of IgG), which can be isolated or combined with an IgA or IgG4 deficiency. It should be remembered that the functional assessment of the humoral immune response is a more informative method of investigation than the quantification of IgG subclasses. Thus, with a normal level of IgG2, the level of antibodies to polysaccharide antigens of Streptococcus pneumoniae is often reduced. At the same time, congenital IgG2 deficiency due to impaired synthesis of heavy chains is possible in the absence of any clinical manifestations of immunodeficiency.

Determination of IgA. Isolated deficiency of secretory IgA with normal serum IgA levels is rare. As a rule, there is a simultaneous deficiency of secretory and serum IgA. Isolated IgA deficiency is not clinically detectable or is accompanied by mild upper respiratory tract infections. This is due to the fact that in case of IgA deficiency, the level of IgG in the serum and IgM in the mucosal secretions compensatory increases. The level of IgA is measured in tears, saliva and other biological fluids. There are two subclasses of IgA - IgA1 and IgA2. IgA1 predominates in the blood and respiratory tract secretions, and IgA2 in the gastrointestinal tract secretions.

In vitro synthesis of immunoglobulins. This assay allows to evaluate the production of IgM, IgG and IgA by stimulated B-lymphocytes. By mixing T- and B-lymphocytes from healthy and sick people treated with different stimulants, the function of T-helper and B-lymphocytes can be assessed. In most cases, antibody deficiency is caused by impaired differentiation of B lymphocytes into plasma cells.

Lymph node biopsy is not usually performed in case of suspected primary immunodeficiency. It is indicated only in cases where the diagnosis is unclear and the patient has enlarged lymph nodes, which requires the exclusion of haemoblastosis.

General principles of treatment of immunodeficiencies

Patients with immunodeficiencies require special attention and need not only constant medical care, but also psychological and social support. Diet. In the absence of impaired absorption syndrome, no diet is required. In the presence of gastrointestinal disorders, a dietitian's consultation is necessary. The diet should satisfy the need for proteins, vitamins and microelements and be sufficiently caloric to ensure normal growth and development. Insufficient nutrition in case of immunodeficiency can lead to even greater immune suppression. Prevention of infections is indicated for all patients with immunodeficiencies, especially in severe combined immunodeficiency. Complete isolation of infants with severe combined immunodeficiency and keeping them in sterile boxes eliminates contact with microbes. Incomplete isolation is less effective, as severe infections in immunodeficiencies are caused even by microorganisms that are not pathogenic to healthy people. To reduce the risk of infection at home, it is necessary that the patient sleeps in a separate bed, has his own room, and avoids contact with infectious patients, especially if the infection is caused by herpes simplex viruses or varicella-zoster. Immunoglobulin replacement therapy allows many patients with humoral

immunity deficiency to lead a normal life. Parents of a sick child are explained that he or she does not need excessive care, should not avoid walking outdoors, can play with other children and attend preschools and school.

Clinical forms of primary immunodeficiencies

Hereditary hypogammaglobulinaemia (HHG) Bruton's disease

Specific defect. Absence of B cells, low levels of all Ig. Defect in cytoplasmic tyrosine kinase (Scr family), a signal transducer to the B-cell nucleus for its activation and transformation into a plasma cell. The defect is located on chromosome Xq 21.3 - 22(b+k). X-linked form.

Clinical features. It is characterised by recurrent purulent infectious diseases of the lungs (pneumonia, chronic bronchitis), paranasal sinuses (sinusitis), middle ear (otitis media), central nervous system (meningitis), intestines (enteritis, colitis), eyes (conjunctivitis), skin (pyoderma), lymph nodes (lymphadenitis) caused by Streptococcus, Haemophilus, Staphylococcus, Pseudomonas, etc. Resistance to viral infections is generally preserved, although there are cases of severe enteroviral polyradiculoneuritis and post-vaccination poliomyelitis. Hypoplasia of the palatine tonsils and peripheral lymph nodes, physical developmental delay, arthritis, and agranulocytosis are typical for patients with SGGG. The disease usually begins in the 5-9th month of life, when maternal IgG ceases to protect the child's body. The disease is rare (1:50,000) and has a recessive type of inheritance linked to the X chromosome.

Only boys are affected; when collecting a family history, it is very important to clarify whether the male lineage has had similar diseases. The course of the disease is severe, with frequent relapses.

An important diagnostic symptom is that the lymph nodes, spleen, and liver do not respond to the inflammatory process with an increase in size. It is possible to develop sluggish arthritis, 210 allergic reactions to antibiotics, slowly progressive neurological diseases, malignant lymphoma. Immunological examination (at least twice) reveals: very low levels of all Ig classes (G, M, A, D and E), serum concentration of IgG < 200 mg/dl, IgA, IgM < 20 mg/dl; absence of circulating B-lymphocytes.

Generalised variable immune deficiency (GVHD, generalised variable hypogammaglobulinaemia)

GVHD is a primary immunodeficiency condition manifested in persons of either sex by repeated bacterial infections, the laboratory diagnosis of which is based on the detection (at least twice) of a total serum concentration of IgG, IgA, IgM < 300 mg/dl. The disease can occur at any age, although in children it most often manifests itself at an early age.

In fact, ZVIN is a variant of hereditary hypogammaglobulinaemia (D80.0) and requires the same set of treatment and diagnostic measures as SGGG. Decreased levels of IgM, IgA, IgG. The number of B-lymphocytes is normal or slightly reduced. Deficiency of antibody formation. T-lymphocyte function defects are often detected. The defect is located in chromosome 6p21.3.

Clinical features. The clinical picture is very similar to Bruton's hypogammaglobulinaemia (recurrent pyogenic lung infection), but the main difference is that the disease does not begin in childhood, but usually at 15-35 years of age. There are stomach and intestinal diseases. Resistance to viral infections is generally preserved, although there are cases of severe enteroviral polyradiculoneuritis and post-vaccination poliomyelitis. Arthritis and agranulocytosis are typical for patients with ZVN.

Both sexes are affected.

Immuno-laboratory examination reveals: 1) a normal or slightly decreased content of circulating B-lymphocytes; 2) a decrease in the synthesis and/or secretion of immunoglobulins, which is manifested by a decrease in the level of serum Ig; 3) the T-cell component is usually preserved, but in some cases there is a decrease in the level of T-helper cells and an increase in the level of T-suppressors. In 25-30% of cases, the following additional symptoms are noted: 1) malabsorption with frequent impaired absorption of cyanobalamin (vitamin B12); 2) presence of giardiasis; 3) lactose intolerance; 4) abnormalities of small intestinal villi. Patients with general variable hypoimmunoglobulinaemia often develop autoimmune pathology.

Treatment. Patients with VVHD require lifelong intravenous immunoglobulin (IVIG) replacement therapy. If it is not available, native plasma can be used in the treatment.

Deficiency of cellular (T-link) immunity Di Giorgi syndrome

DiGiorgi syndrome (DGS) is an isolated T-cell immunodeficiency. It is characterised by a triad of clinical manifestations: hypoplasia of the thymus and/or parathyroid glands and congenital heart disease. It accounts for 5-10% of the total number of primary immunodeficiencies.

A specific defect. Dysembryogenesis: a defect in the development of the third or fourth pharyngeal pockets that occurs between the sixth and tenth weeks of gestation, which leads to impaired development of the thymus, thyroid and parathyroid glands, malformation of facial structures, congenital heart disease with aortic arch lesions. The defect is located on chromosome 22qII.

Clinical features. Most patients have dysplastic facial features. The most characteristic are dysplastic auricles, hypertelorism, a wide bridge of the nose, "fish mouth", anti-Mongoloid cut of the eyes. Some children have grosser abnormalities, such as micrognathia and non-union of the hard and soft palate. Hypocalcemia of various degrees of severity and the absence of a shadow of the thymus gland on chest X-rays are frequent manifestations. Hypoparathyroidism is manifested by hypocalcemic seizures that occur from the first days of life. All patients have a delay in mental development. Congenital defects of the heart and main vessels are also among the most characteristic and severe signs of the disease. T-cell immunodeficiency leads to recurrent viral, parasitic, bacterial infections and mycoses.

However, the level of serum immunoglobulins in such patients is not disturbed. Children may experience unusual reactions, up to death, when vaccinated with live, attenuated vaccines of the measles virus, poliomyelitis, when vaccinated with BCG. Clinical features. Most patients have dysplastic facial features. The most characteristic are dysplastic auricles, hypertelorism, a wide bridge of the nose, "fish mouth", anti-Mongoloid cut of the eyes. Some children have grosser abnormalities, such as micrognathia and non-union of the hard and soft palate.

Immunological studies: 1) lymphocytopenia; 2) decrease in the number and proliferative activity of T-lymphocytes; 3) dissociation between reduced levels of T- and NK-cells and increased content of B-lymphocytes; 4) normal or elevated levels of antibodies; 5) reduction of delayed-type hypersensitivity skin reactions. The ability to produce antibodies against certain antigens is reduced due to the lack of T-helpers.

Treatment. Thymus transplantation is used; introduction of thymus hormones with a substitute purpose; symptomatic therapy. In the presence of severe defects, which mainly determine the prognosis for life, thymus transplantation is considered insufficiently justified. If the patient survives 6 months of age, there is a gradual spontaneous recovery of T-cell immunity.

Lymphocytic dysgenesis (Nezelof's syndrome, French type

Lymphocyte dysgenesis (Nezelof's syndrome) is a quantitative and qualitative insufficiency of the T-system as a result of atrophy of the thymus and lymph nodes. Violated maturation of T-lymphocyte precursors, which leads to their functional deficiency; autosomal recessive type of inheritance.

It is characterized by the absence of cellular reactions of immunological protection with a normal content of immunoglobulins in the blood plasma. Appears in the first weeks and months of life. Vestigial thymus; low number of thymocytes; Hassell bodies are absent.

Clinical manifestations: there is a delay in the child's growth and development, a protracted septic process with purulent-inflammatory foci in the internal organs and skin; recurrent pneumonia, diarrhea, eczema, lymphadenitis, hyperplasia of lymphoid tissue. Pronounced susceptibility to infectious agents: bacteria (tuberculosis, listeria, Escherichia coli, salmonella), viruses: (herpes, Epstein-Barr, adenoviruses, enteroviruses), protozoa (pneumocystis, toxoplasma, cryptosporidium) and fungi (candida, cryptococci, nocardia).

Immunological research: lymphocytopenia, an extremely low level of T-lymphocytes is observed in the peripheral blood; the norm of B-lymphocytes; sharply suppressed reaction of lymphocyte blast transformation; weakly expressed hypersensitivity reaction of the delayed type. The content of immunoglobulins of all classes in the blood is within the normal range or increased. The formation of specific immunoglobulins is reduced. Children often die in the first months of life from sepsis. Treatment: stem cell transplantation.

T Severe combined immunodeficiency, X-linked type Specific defect.

Violation of stem cell differentiation into B- and T-lymphocytes. Defect of the IL-2 receptor gamma chain on T-lymphocytes. The gamma chain is a signal transducer when the receptor binds to IL-2. Autosomal recessive type. Specific defect. Mutation of the ZAP-70 tyrosine kinase gene, a signal transducer in T-

lymphocytes necessary for their proliferation. Absence of CD8+ cells in peripheral blood is characteristic. Localization of the defect in chromosome Xq 13-21.1.

Clinical features. Recurrent infectious diseases, weight loss, developmental delay. Lymphopenia and hypoplasia of the thymus are characteristic. The number and function of T-lymphocytes is reduced. Hypogammaglobulinemia, decrease in the level of B-lymphocytes. Decreased skin tests and production of antibodies. Patients die in the first 1-2 years of life from viral, bacterial, protozoal infections or mycosis.

Treatment. Bone marrow transplantation, antibiotic therapy, intravenous immunoglobulin therapy, embryonic liver and thymus cell transplantation.

Louis-Bar syndrome, ataxia - telangiectasia, autosomal recessive type of inheritance Specific defect.

Violation of the function of T- and B-lymphocytes. Decreased level of IgA, IgE and IgG. Hypoplasia of the thymus, spleen, lymph nodes, tonsils. A very high level of α-fetoprotein is detected in the serum. Localization of the defect in chromosome IIq 22.3 (atm).

Clinical features. Telangiectasia of skin and eyes; progressive cerebellar ataxia; recurrent infection of the paranasal sinuses and lungs of a viral and bacterial nature; bronchiectatic disease; increased level of alpha-fetoprotein. In the future - damage to the nervous, endocrine, vascular systems, malignant tumors. The disease is most often diagnosed at the age of 5-7, equally often in boys and girls. Half of the patients have a lag in mental development, adynamia, and limited interests. Some patients live up to 20 and even 40 years.

Treatment. Symptomatic means. Bone marrow transplant. Thymus hormones. Intravenous immunoglobulin therapy.

Tasks for checking the initial level of knowledge

1. Specify the main types of congenital immune deficiency:

A. Defects of phagocytic cells.

B. Deficiency of the complement system.

C. T-cell deficiency.

D. B-cell deficiency.

E. Insufficiency of stem cells.

F. None of the listed types of immunodeficiency can be congenital.

2. What diseases can be caused by a decrease in the efficiency of phagocytosis?

A. Chediak-Higashi disease.

B. Pustular infections.

C. Systemic candidiasis.

D. None of the listed diseases.

3. Insufficiency of the complement system can be clinically manifested:

A. Autoimmune reactions.

B. Allergic diseases or conditions.

C. Infectious diseases.

D. None of the specified clinical manifestations.

4. Define the most frequent defect in the synthesis of complement components, which causes the development of angioedema:

A. Components C1, C2, C4.

B. Component C3 and factor B.

C. Factor I.

D. C1 inhibitor.

5. Boys with Bruton's congenital agammaglobulinemia are most prone to re-infections?

A. Caused by Staphylococcus aureus.

B. Caused by pyogenic streptococcus.

C. Caused by pneumococcus.

D. Caused by meningeal neisseria.

E. None of the listed pathogens cause infection in boys with Bruton's congenital agammaglobulinemia.

6. Viral infections (measles, smallpox) in patients with Bruton's agammaglobulinemia usually pass:

A. With severe complications.

B. Without features.

7. With transient hypogammaglobulinemia of childhood, a low level is noted:

A. Immunoglobulins of class A.

B. Immunoglobulins of class E.

C. Immunoglobulins of class M.

D. Immunoglobulins of class G.

8. What are the most common clinical signs of transient hypogammaglobulinemia in childhood?

A. Repeated infections of the respiratory tract.

B. Severe course of childhood infectious diseases.

C. Autoimmune diseases and conditions.

9. At what age does the level of class G immunoglobulins reach the level characteristic of adults with transient hypogammaglobulinemia of childhood?

A. 1-2 years old.

B. 4 years old.

C. 7 years old.

D. 11 years old.

E. 14 years old.

F. 18 years old.

10. What changes in the immune system are noted with underdevelopment of the thymus?

A. Significantly reduced cell content in thymus-dependent zones of lymphoid tissue.

B. Lymphoid follicles are underdeveloped.

C. Reduction in the effectiveness of cellular immunity reactions.

D. Weakened synthesis of antibodies.

E. Only some changes.

Correct answers to questions: 1 – ABCDE; 2 – ABC; 3 – ABC; 4 – D; 5 – ABCD; 6 – B; 7 – ACD; 8 – A; 9 – B; 10 - ABCD.

Sources of educational information:

1. Clinical immunology and allergology: Textbook / [H.M. Dranik, O.S. Prylutskyi, Yu.I. Bazhora and others]; under the editorship Prof. AHEM. Dranika - K.: Zdrovya, 2006. - 888 p.

2. Kazmirchuk V.E. Clinical immunology and allergology / V.E. Kazmirchuk, L.V. Kovalchuk. – Vinnytsia: Nova kniga, 2006, 504 p.

3. Andreychyn M.A. Clinical immunology and allergology: Textbook / Andreychyn M.A., Chop'yak V.V., Gospodarskyi I.Ya. – Ternopil: Ukrmedknyga, 2005. – 372 p.

4. Clinical Immunology and Allergology: Textbook / Ed. A.V. Karaulova - M.: Medical Information Agency, 2002. - 651 p.

5. Nikulin B.A. Evaluation and correction of immune status / Nikulin B.A. - M.: GEOTAR-Media, 2007. - 376 p.

6. Burmester G.R. Supervised immunology, Persian with English. / Burmester G.R. - M.: BINOM. Laboratory of Knowledge, 2007. – 329 p.