MEDICAL UNIVERSITY – PLOVDIV

FACULTY OF MEDICINE

SYLLABUS

IN

MICROBIOLOGY

Approved by the Department Council - Protocol №6/13.06.2022

Confirmed by the Faculty Council - Protocol №6/15.06.2022

MICROBIOLOGY

Syllabus

Discipline	Final exam/ semester	Academic hours				Extracurricular load/activity	Total credits	Academic hours in years and semesters	
								II/III year	
		Total	Lectures	Practices	ECTS			IV	V
Microbiology	**V**	**135**	**60**	**75**	**4.5**	**3.5**	**8.0**	**2/3**	**2/2**

DISCIPLINE:
Microbiology

TYPE OF DISCIPLINE ACCORDING TO THE UNIFORM STATE REQUIREMENTS: Mandatory

LEVEL OF EDUCATION:
Master's Degree /**M/**

FORMS OF TRAINING: Full-time study

YEAR OF TRAINING: 2nd and 3rd year

DURATION OF TRAINING: 1 year (2 semesters)

ACADEMIC HOURS:
60 hours of lectures, 75 hours of practical classes

TECHNICAL EQUIPMENT APPLIED IN THE TRAINING:
- multimedia
- demonstration materials
- lectures
- handbooks
- electronic platforms (MS Office 365, Zoom, etc.)

FORMS OF EVALUATION: Semester exam

EVALUATION CRITERIA:
The final grade is multicomponent and includes the marks from the written final exam and the following components:
- average mark from ongoing control (colloquiums, tests)
- mark from the practical final exam
- mark from the oral final examination

If one of the components of the final grade is poor 2, then the final grade is necessarily poor 2.

ASPECTS OF EVALUATION CRITERIA:
For each component participating in the final evaluation, a significance coefficient (from 0 to 1) is determined, and the total sum of the coefficients must always be 1. The overall mark is obtained as the sum of the evaluation marks on a six-point scale from the various components multiplied by the respective coefficients of significance.
Q final grade = K1 Q grade from current control + K2 Q grade from written exam + K3 Q grade from the oral exam

K1 = 0.20; K2 = 0.50; **K3** = 0.30

SEMESTER EXAM: Yes

STATE EXAM: No

LECTURER:
Habilitated lecturer from the Department of Medical Microbiology and Immunology
Prof. Mariana Murdjeva, MD, PhD, MHM

DEPARTMENT: Medical microbiology and Immunology "Prof. dr. Elissey Yanev"

ANNOTATION
The main goal of the Microbiology course is to thoroughly introduce medical students to the morphological and biological characteristics of microorganisms, the patterns of the development of the infectious process, the specific and nonspecific immune defence of the body, the diagnosis of infectious diseases, the main point and problems of antimicrobial chemotherapy, specific prevention and control of infection.

The goal is in accordance with:
• the scope and credits of the course (according to the ECTS system), as noted in the curriculum, available on the website of MU – Plovdiv;
• the qualification characteristics of the specialty;
• the educational degree (master's degree).

The goal conforms with the place of the microbiology discipline in the specialty of Medicine in its importance and chronology in the curriculum. As a fundamental discipline, it predominantly serves the next stages of training.

BASIC AIMS OF THE DISCIPLINE

- Introduction to the morphology, physiology, and pathogenicity factors of microorganisms that play a role in human pathology;
- Studying the patterns of occurrence and course of the infectious process, the pathogenesis of infectious diseases and various forms of infection;

- Studying the mechanisms for the protection of macroorganisms - natural resistance and acquired immunity, as well as the principles of immunoprophylaxis and immunotherapy of infectious diseases;

- Antimicrobial chemotherapy - mastering the mechanisms of action of the main groups and types of antimicrobial agents, as well as the mechanisms for the development of bacterial resistance;
- Principles and basic methods for sterilization and disinfection;
- Mastering the microbiological diagnostics of infectious diseases; the structure and role of the microbiological laboratory for the etiological diagnosis of infectious diseases; skills for correct clinical interpretation and analysis of laboratory results;
- Learning the methods for microbiological, immunological, and molecular-biological diagnostics of the infectious diseases, as well as the correct interpretation of the obtained results;
- Studying the composition and role of the normal microflora of the human body;
- Studying the external environment's role in the spread of infectious agents and methods and means for microbiological control of the environment.

➢ EXPECTED RESULTS

After the microbiology course, medical students should be familiar with the morphological and biological characteristics of the most important microorganisms for human pathology, their pathogenic factors, patterns for the development of the infectious process, and the forms of specific and nonspecific immune defense against a given microorganism. They must have mastered the rules for collecting and sending pathological material for microbiological examination, the methods for microbiological examination, the interpretation of the obtained results depending on the clinical syndrome, as well as the diagnosis, prevention, and control of the infection.

LECTURES

LECTURE PROGRAM
II year, IV semester

№	TOPIC	HOURS	DATE
1.	Subject, tasks, historical development, and achievements of microbiology. Introduction to general microbiology.	2	
2.	Morphology and structure of microorganisms.	2	
3.	Physiology of bacteria. Bacterial genetics – I part.	2	
4.	Genetics of microorganisms – 2 part.	2	
5.	Influence of environmental factors on microorganisms. Disinfection and sterilization.	2	
6.	Antimicrobial therapy of infectious diseases. Classes and groups of antibiotics. Antimicrobial resistance.	2	
7.	The study of infection. Characteristics and forms of the infectious process. The role of the microorganism in the infectious process. Pathogenic factors.	2	
8.	The role of the external environment in the occurrence of the infectious process. Epidemic process. Factors and mechanisms for the transmission of infectious agents in the epidemic process.	2	
9.	Immunity. Natural resistance. Protective role of skin, mucous membranes, normal microflora. Cellular and humoral factors of natural resistance. Phagocytosis. Inflammation.	2	
10.	Antigens and antibodies.	2	
11.	Specific humoral immunity. Specific cellular immunity. Immunological tolerance.	2	
12.	Immunopathology. Allergies - definition and forms.	2	

13.	Immunopathology. Immunodeficiency conditions and diseases. Autoimmunity.	2	
14.	Immunoprophylaxis and immunotherapy.	2	
15.	Scheme and course of microbiological research.	2	

HOURS: 30

LECTURE PROGRAM
III year, V semester

№	TOPIC	HOURS	DATE
1.	Cocci - staphylococci, streptococci.	2	
2.	Streptococcus pneumoniae. Mycobacterium *tuberculosis*. The causative agent of leprosy. Causative agents of mycobacteriosis.	2	
3.	Corynebacteria. Pertussis bacteria. Hemophilus spp.. Neisseria spp.	2	
4.	Anaerobes. Spore-forming - tetanus bacillus, gas gangrene bacilli, botulinum bacillus. Non-spore-forming anaerobes.	2	
5.	Causes of particularly dangerous infections. The causative agent of plague. Vibrio cholerae. Bacillus anthracis.	2	
6.	Causes of particularly dangerous infections. Brucella spp. Tularemia bacterium. Legionellosis. Facultative pathogenic intestinal bacteria - Escherichia coli, Klebsiella spp., Proteus spp., and others.	2	
7.	Pathogenic enteric bacteria: Dysenteric bacteria. Salmonella - Salmonella typhi, Salmonella paratyphi A and B, Salmonella spp., causes of food poisoning.	2	
8.	Spirochetes. Cause of syphilis. Cause of typhus. Cause of Lyme disease. Leptospira. Spirilla.		
9.	Mycoplasma spp. and Rickettsia spp. Chlamydia spp.	2	
10.	Pathogenic fungi – Candida spp., actinomycetes, Aspergillus spp., cryptococci.	2	

11.	Viruses - nature and properties. Picornaviruses.	2	
12.	Ortomyxoviruses. Paramyxoviruses. Cause of COVID-19 (SARS-2)	2	
13.	Adenoviruses. Togavirses. Flaviviruses. Rhabdoviruses. Ebola and Zika viruses.	2	
14.	Hepatitis viruses. AIDS viruses.	2	
15.	Herpesviruses. Poxviruses.	2	

HOURS: 30

PRACTICES

PROGRAM FOR PRACTICAL CLASSES
II year, IV semester

№	TOPIC	HOURS	DATE
1.	Structure and equipment of the microbiological laboratory and rules for work in it. Methods for studying the morphology of microorganisms. Types of microscopes. Immersion system microscopy. Study of the morphology of microorganisms in a colored state. Simple staining methods - Löffler and Pfeiffer stain.	3	
2.	Complex methods for staining the microorganisms. Gram and Neisser stain. Ziehl-Neelsen staining (acid-fast bacteria). Möller staining (spores).	3	
3.	Cultivation of microorganisms. Types of nutrient media. Methods for isolation of microorganisms in pure culture. Types of cultures and colonies.	3	
4.	Biochemical activity of bacteria. Pathogenic factors in bacteria.	3	
5.	Determination of the in vitro susceptibility of bacteria to antibiotics (antibiogram). Other methods for detection of antimicrobial resistance.	3	
6.	Resistance of microorganisms. Sterilization and sterilization methods. Disinfection and disinfectants.	3	
7.	Recapitulation of the studied material in practical classes from №1 to №6 included.	3	

8.	SEMINAR on the topic: Morphology, physiology, and genetics of microorganisms. Test №1.	3	
9.	Cellular and humoral basis of the immune response. Antigen-antibody reactions. Agglutination reaction. Precipitation reaction. Neutralization reaction (ASO).	3	
10.	Antigen-antibody reactions. Bacteriolysis, hemolysis, cytolysis. Complement fixation test. Immune reactions with labeled antibodies or antigens: immunofluorescence assay (IFA), radioimmunoassay (RIA), enzyme-linked immunosorbent assay (ELISA).	3	
11.	Flowcytometry to determine cell subpopulations (immunophenotyping). Examination of the allergic condition. Bioproducts - vaccines and sera.	3	
12.	SEMINAR on the topic: Infection and Immunity. Test №2	3	
13.	Laboratory diagnosis of diseases caused by viruses and rickettsiae. Chlamydia?	3	
14.	Methods for microbiological diagnosis of infectious diseases. General scheme for microbiological research.	3	
15.	Assessment of the practical skills of students, acquired during the semester.	3	

HOURS: 45

PROGRAM FOR PRACTICAL CLASSES
III year, V semester

№	TOPIC	HOURS	DATE
1.	Microbiological diagnosis of staphylococcal and streptococcal infections. Microbiological examination of pus.	2	
2.	Microbiological examination in diseases caused by *Streptococcus pneumoniae*. Microbiological diagnosis of tuberculosis and leprosy. Microbiological examination of sputum.	2	
3.	Microbiological diagnosis of diphtheria and pertussis. Microbiological examination of throat swabs.	2	
4.	Microbiological diagnosis of gas gangrene and tetanus. Microbiological examination of wound secretions.	2	

5.	Microbiological examination of CNS materials. Microbiological diagnosis and differential diagnosis of bacterial meningitis *(Neisseria meningitidis* and *Haemophilus influenzae*).	2	
6.	Microbiological diagnosis of particularly dangerous infections plague, cholera, and anthrax.	2	
7.	SEMINAR on the topic: Microbiological diagnostics of microorganisms, studied in practical classes from №1 to №7. Test №1.	2	
8.	Microbiological examination of materials from the digestive system (feces). Microbiological diagnosis of bacterial dysentery, *E. coli* enteritis, salmonellosis. Food poisoning by *Staphylococcus aureus*, clostridia (*C. botulinum*, *C. perfringens*). Microbiological examination of gastric mucosa biopsy material (*Helicobacter pylori*).	2	
9.	Microbiological examination of urine. Microbiological diagnosis of pathogens causing urinary tract infections: opportunistic pathogens (*E. coli, Klebsiella, Proteus-Providencia-Morganella* group, *Pseudomonas spp*.) and obligatory pathogenic (streptococci, salmonella, *Leptospira spp., Mycobacterium tuberculosis*).	2	
10.	Microbiological examination for sexually transmitted infections caused by Neisseria gonorrhoeae, *Treponema pallidum, Candida albicans, Chlamydia spp.,* and mycoplasmas.	2	
11.	Microbiological examination of blood - blood culture. Causes of septic conditions: obligatory pathogenic (*Salmonella typhi, Brucella spp., Borrelia spp.)* and facultative pathogenic.	2	
12.	Problematic microorganisms causing healthcare-associated infections (HAIs) - *Pseudomonas spp., Enterococcus spp.,* MRSA, *C. difficile*). Systemic mycoses, caused *by Candida, Aspergillus, Actinomyces, Cryptococcus.* Antimitotic therapy.	2	
13.	Microbiological diagnosis of diseases caused by viruses – HIV and hepatitis A, B, C D and E, influenza and coronaviruses – SARS-CoV-2.	2	
14.	Sanitary-microbiological examination of water, air, hospital environment. Sanitary-indicative microorganisms - *E. coli, Enterococcus spp., C. perfringens*, staphylococci, streptococci. Test 2 (includes practicals 8-13)	2	
15.	Recapitulation of the students' practical skills, acquired during the two semesters with an evaluation mark.	2	

HOURS: 30

LECTURES

LECTURE №1 – 2 hours

Subject, tasks, historical development, and achievements of microbiology. Introduction to general microbiology.

Introduction. Subject, tasks, historical development, and achievements of microbiology. The contribution of L. Pasteur and R. Koch. General Microbiology. Taxonomy of microorganisms. Characteristics of major groups of microorganisms: higher protists (Eucaryote) and lower prostists (Procaryotae).

LECTURE №2 – 2 hours

Morphology and structure of microorganisms.

Morphology of microorganisms – size, shape and arrangement. Major types according to the morphology – rod-shaped, round-shaped and curved bacteria; structure of the bacterial cell, essential and non-essential organelles. Methods of studying the morphology of bacteria, fungi, mycoplasmas, and viruses.

LECTURE №3 – 2 hours

Bacterial physiology

Chemical composition of bacteria – water, minerals, proteins, carbohydrates, lipids, and nucleic acids. The significance of bacteria for diagnosis, pathogenesis, and therapy of infectious diseases. Bacterial enzymes. Bacterial metabolism – catabolic (dissimilation). Types of bacteria according to the mechanism of biological oxidation. Anabolic processes (assimilation). Bacterial productivity and its significance in infectious diseases pathogenesis, diagnosis, and therapy. Bacterial growth and reproduction. Principles of in vitro culturing and nutritional requirements of bacteria. Prototrophs and auxotrophs.

LECTURE №4 – 2 hours

Bacterial genetics

Genotype and phenotype in bacteria, viruses and phages. Bacterial genome – chemical composition, structure, and functions. Plasmids (extrachromosomal elements). Types of plasmids and their significance. Bacteriophages and their importance – structure, temperate phages, lytic phages. Inheritance and mutation in microorganisms – definitions. Types of mutations; factors causing mutations. Recombination – significance for biological and medical practice. Gene engineering – significance for the medical theory and practice. Molecular methods (PCR, DNA probes) in the diagnosis of infectious diseases.

LECTURE №5 – 2 hours

Influence of environmental factors on microorganisms. Disinfection and sterilization.

Microbiota of the human body – role in the normal physiological processes, protection, and pathology. Distribution of microorganisms in soil, water, food, hospital environment, instruments, and the objects in the surrounding environment. Influence of physical factors on microorganisms – heat, desiccation, pH, osmotic pressure, light, ultrasound, ionizing

radiation. Sterilization. Methods of sterilization. Influence of chemical factors on microorganisms. Oligodynamia. Disinfections. Types of disinfectants. Mechanism of action. Influence of biological factors on microorganisms – symbiosis, synergy, antagonism, bacteriocins, bacteriophages.

LECTURE №6– 2 hours

Antimicrobial therapy of infectious diseases. Classes and groups of antibiotics. Antimicrobial resistance.

Chemotherapy. Antibiotics. Types of antimicrobial drugs. Classification of antibiotics according to: origin, spectrum of action, chemical composition. Antibiotic drugs by groups according to chemical composition and mechanism of action. Basic principles of chemotherapeutic and antibiotic drug application. Mechanism of resistance in microorganisms. Measures against drug resistance. Side effect of antibiotic treatment. Antimicrobial susceptibility testing to antibiotics. Antibiograms. Other methods for in vitro antimicrobial susceptibility testing in microorganisms.

LECTURE №7 – 2 hours

The study of infection. Characteristics and forms of the infectious process. The role of the microorganism in the infectious process. Pathogenic factors.

Relationships between micro- and macroorganism - mutualism, commensalism, parasitism, saprophytism. Infection, infectious process, infectious disease – definition. Role of the microorganism in infectious process: pathogenicity, virulence, infectious dose, contagiosity, invasiveness, toxigenicity. Virulence factors: adhesion factors, factors of invasion, factors of aggression – endotoxins, exotoxins, etc. Mechanism of action.

LECTURE №8 – 2 hours

The role of the external environment in the occurrence of the infectious process. Epidemic process. Factors and mechanisms for the transmission of infectious agents in the epidemic process.

Pathogenesis of the infectious process – entry, dissemination, localization, and damage to the host. Characteristics of the infectious disease. Types of infectious process: exogenic and endogenic infection. Primary infection, re-infection, secondary infection; superinfection, co-infection; localized and systemic infection; focal infection; septicaemia, bacteraemia, viremia, toxaemia, pyemia, systemic inflammatory response syndrome (SIRS). Role of the host in the infectious process. Role of the environment for the origination and course of the infectious process. Epidemic process – factors and mechanisms. Factors for origination of the epidemic process: source of infection, mechanism of transmission, and susceptible population. Primary and secondary forces of the epidemic process.

LECTURE №9 – 2 hours

Immunity. Natural resistance. Protective role of skin, mucous membranes, normal microflora. Cellular and humoral factors of natural resistance. Phagocytosis. Inflammation.

Types of protection of the host – natural resistance, acquired immunity. Protective role of skin and mucous membranes, their secretions, resident microbiota. Cellular factors of the innate immunity – macrophages, microphages, NK cells. Phagocytosis. Humoral factors of

the innate immunity: complement system, interferons, lysozyme, cytokines, acute phase proteins. Inflammations – protective and pathologic mechanisms.

LECTURE №10 – 2 hours

Antigens and antibodies.

Antigens – characteristics; antigen determinants (epitopes), antigen valence, haptens; types of antigens; microbial antigens. Structure of antibodies. Classes of antibodies and their function.

LECTURE №11 – 2 hours

Specific humoral immunity. Specific cellular immunity. Immunological tolerance.

Immune system: anatomy and structure. Central and peripheral organs of the immune system. Formation of immunocompetent cells – T-cells and B-cells and their subpopulations. CD-molecules, defining lymphocyte subpopulations and the significance of the immune reaction. T-cell receptor and B-cell receptor for antigens. Cell-mediated immunity – types. Humoral immunity. Antigen presentation. Development of the immune response; cell-to-cell cooperation. Primary and secondary immune response. Genetic control and regulation of immune response. HLA system. Immune tolerance – mechanisms.

LECTURE №12 – 2 hours

Immunopathology. Allergies - definition and forms.

Allergic reactions. Type of allergens. Types of hypersensitivity reactions according to the Cooms and Gell classification: Type I anaphylactic and atopic allergic reactions with participation of IgE and release of biological active substances – tissue damage and clinical presentation; type II (cytotoxic); type III (Ag-Ab complexes); type IV (infectious, delayed). Autoimmune reactions. Definition. Types. Mechanisms of origin.

LECTURE №13 – 2 hours

Immunopathology. Immunodeficiency conditions and diseases. Autoimmunity.

Types of pathological conditions and diseases of the immune system – primary and secondary; Causes – defects in the cell-mediated immunity, humoral immunity, phagocytosis, and complement system; combined immunodeficiencies. Clinical presentation. Laboratory diagnosis. Immune status.

LECTURE №14 – 2 hours

Immunoprophylaxis and immunotherapy.

Immunoprophylaxis: vaccine prophylaxis – types of vaccines according to the origin; characteristics, duration of the postvaccinal immunity. Specific immunotherapy with serum and immunoglobulins. Types of immune serums – antitoxic, antibacterial, antiviral. Other methods and approaches for immunotherapy.

LECTURE №15 – 2 hours

Scheme and course of microbiological research.

Type of clinical specimens and tools for collection. Principles of collection and transport of clinical specimens for microbiological examination. Purpose of the microbiological

examination. Course of microbiological examination: direct microscopy (types of smears – wet mount, stained – simple and complex staining methods, immunofluorescent smears); culturing – types of growth media, other requirements for culturing – atmosphere, temperature, humidity, time, etc.; method for obtaining a pure culture; identification of pure cultures – morphological, culture, biochemical, antigenic. Antibiogram. Serological diagnosis. Rapid methods for microbiological diagnosis – ELISA, RIA, IFA, molecular methods – DNA probes, PCR. Automated systems – for blood cultures, identification of microorganisms, etc.

LECTURE №16 – 2 hours

Cocci - staphylococci, streptococci.

Family *Micrococcaceae*. *Staphylococcus spp.: S. aureus*. *Streptococcus* spp.: *S. pyogenes.*

LECTURE №17– 2 hours

***Streptococcus pneumoniae*. *Mycobacterium tuberculosis*. The causative agent of leprosy. Causative agents of mycobacteriosis.**

Streptococcus spp.: *S. pneumoniae*. Family *Mycobacteriaceae*. *Mycobacterium spp*.: *M. tuberculosis, M. leprae. M. kansasii, M. avium complex, etc.*

LECTURE №18 – 2 hours

Corynebacteria. Pertussis bacteria. *Hemophilus spp., Neisseria spp.*

Corynebacterium spp.: C. diphtheriae. Haemophilus spp.: H. influenzae. Neisseria spp.: N. meningitidis, N. gonorrhoeae. Bordetella spp.: B. pertussis, B. parapertussis.

LECTURE №19 – 2 hours

Anaerobes. Spore-forming - tetanus bacillus, gas gangrene bacilli, botulinum bacillus. Non-spore-forming anaerobes.

Family Clostridiaceae. Clostridium spp. C. tetani – causative agent of tetanus. Causative agents of gas gangrene - *C. perfringens, C. novyi, C. septicum, C. histolyticum*. Causative agent of botulism - *C. botulinum*. *Clostridioides spp.* Causative agent of pseudomembraneous collitis - *C. difficile*. Family *Bacteroidaceae*: *Bacteroides spp*. *Fusobacterium spp*. *Leptotrichia spp.*

LECTURE №20 – 2 hours

Causes of particularly dangerous infections. The causative agent of plague. *Vibrio cholerae*. *Bacillus anthracis*.

Family *Yersiniaceae: Yersinia spp.: Y. pestis, Y. enterocolitica.* Family *Vibrionaceae*. *Vibrio spp.: V. cholerae* biotype cholerae, *V. cholerae* biotype El Tor. *Bacillus spp.: B. anthracis.*

LECTURE №21 – 2 hours

Causes of particularly dangerous infections. *Brucella spp*. Tularemia bacterium. Legionellosis. Facultative pathogenic intestinal bacteria - *Escherichia coli, Klebsiella spp., Proteus spp.*, and others.

Brucella spp. Francisella spp.: F. tularensis. Legionella spp.: L. pneumophila. Order *Enterobacterales. Family Enterobacteriaceae. Escherichia spp.: E. coli, Enterobacter spp., Citrobacter spp., Klebsiella spp.* Family. *Yersiniaceae: Serratia spp.* Family *Morganellaceae: Proteus spp. Providencia spp. Morganella spp.*

LECTURE №22 – 2 hours

Pathogenic enteric bacteria: Dysenteric bacteria. *Salmonella - Salmonella typhi, Salmonella paratyphi* A, B and C, *Salmonella spp.*, causes of food poisoning.

Family Enterobacteriaceae. Shigella spp. Salmonella spp.: S. typhi, S. paratyphi A, B, C. Salmonella spp., causing food toxoinfectious disease.

LECTURE №23 – 2 hours

Spirochetes. Cause of syphilis. Cause of typhus. Cause of Lyme disease. *Leptospira spp*. Spirilla.

Order *Spirochaetales*. Familly *Treponemataceae*. *Treponema spp*. T. *pallidum* – the causative agent of syphilis. Family *Borreliaceae*. *Borrelia spp.*: B. r*ecurrentis* – the causative agent of relapsing fever, *B.* b*urgdorferi* – the causative agent of Lyme disease. Family *Leptospiraceae*. *Leptospira spp. – L. interrogans, L. biflexa*.

LECTURE №24 – 2 hours

Mycoplasma and Rickettsia. Chlamydia spp.

Family Mycoplasmataceae. Genus Mycoplasma: M. pneumoniae, M. hominis, M. orale, M. salivarium, M. fermentans. Family Rickettsiaceae. Genus Rickettsia: R. prowazekii – causative agent of epidemic typhus, R. conorii – causative agent of Mediterranean spotted fever, etc. Family Coxiellaceae. Genus Coxiella. C. burnetii. Family. Chlamydiaceae. Genus Chlamydia: C. trachomatis. Genus Chlamydophila. C. pneumoniae. C. psittaci.

LECTURE №25 – 2 hours

Pathogenic fungi – Candida spp., Actinomycetes, Aspergillus spp., Cryptococci.

Genus Candida: C. albicans. Genus Actinomyces: A. bovis, A. israeli. Genus Aspergillus, Genus Cryptococcus.

LECTURE №26 – 2 hours

Viruses – nature and properties. Picornaviruses.

History of virology. General characteristics of viruses. Viral taxonomy. Morphology and structure of viruses: DNA/RNA genome, capsid, supercapsid; biology of virues: viral reproduction, cultivation methods. Epidemiology and pathogenesis of viral diseases; immunity, specific prophylaxis, therapy. Laboratory diagnosis. Family Picornaviridae. Genus Enterovirus: Human polioviruses 1, 2, 3. Human coxsackieviruses A, B. Human echoviruses. Human enteroviruses 68 – 71.

LECTURE №27 – 2 hours

Orthomyxoviruses. Paramyxoviruses. Causative agent of COVID-19 (SARS-CoV-2).

Family Orthomyxoviridae. Influenza A, B, C viruses – causative agents of Flu. Causative agents of bird flu and swine flu. Family Paramyxoviridae: Human Parainfluenza

viruses 1-4, Morbillivirus, Mumps virus. Family Pneumoviridae – hRSV, Human Metapneumovirus. Family Coronaviridae. SARS-CoV, MERS-CoV, SARS-CoV-2.

LECTURE №28 – 2 hours

Adenoviruses. Togaviruses. Flaviviruses. Rhabdoviruses. Ebola and Zika viruses.

Family Adenoviridae. Family Togaviridae – Rubella virus and others. Family Flaviviridae – yellow fever virus, Zika virus and others. Family Rhabdoviridae – rabies virus. Family Filoviridae – Ebola virus.

LECTURE №29 – 2 hours

Hepatitis viruses. AIDS viruses.

Hepatitis viruses: Causative agents of viral hepatitis - HAV, HBV, HDV, HCV, HEV. Hepatitis viruses with fecal-oral transmission mechanism - hepatitis A and E viruses. Hepatitis viruses with multiple transmission mechanisms - hepatitis B, D, and C viruses. Characteristics of the virus, the clinical presentation of the disease, laboratory diagnosis (hepatitis markers), specific prevention, and therapy. Fam. Retroviridae. Subfamily Lentivirinae: AIDS viruses (HIV-1 and HIV-2). Acquired Immune Deficiency Syndrome (AIDS): historical data; structure and reproductive cycle of HIV; epidemiology of AIDS; Clinical presentation; laboratory diagnosis; therapeutic approach; prevention

LECTURE №30 – 2 hours

Herpesviruses. Poxviruses.

Family Herpesviridae. Subfamily Alpha herpes virinae: Herpes simplex virus 1, 2; Human herpes virus – 3, Varicella herpetovirus (V. varicellae, V. herpes zoster). Subfamily Beta herpes virinae: Human herpes virus 5 (Human cytomegalovirus). Subfamily Gamma herpes virinae: Human herpes virus 4 (Epstein-Barr herpes virus). Family Poxviridae: Orthopoxvirus variolae.

PRACTICAL CLASSES - THESES

PRACTICAL CLASS №1 – 3 hours

Structure and furnishing of the clinical microbiology laboratory, safety practices and rules. Methods for studying the morphology of microorganisms. Types of microscopes. Immersion oil microscopy. Simple staining methods - Loeffler and Pfeiffer.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the specifics of microbiological practice and basic microbiological materials. Mastering the microscopy slide preparation techniques and staining by the Loeffler and Pfeiffer methods. Working with immersion system on a light microscope.

DEMONSTRATION OF: Structure of a microbiological laboratory and safety rules upon handling infectious materials. Microbiological lab equipment, tools, and glassware. Basic microbiological manipulations and requirements when working with infectious materials. Work rules for immersion oil microscopy on a regular light microscope. Preparation of slides, stained by the Loeffler and Pfeiffer methods.

PRACTICAL TASKS: Preparation and staining microscopy slides by the Loeffler and Pfeiffer Пфайфер from microorganisms cultivated on solid and liquid growth media (S. epidermidis и E. coli). Observation and description of the microscopy field. Immersion oild microscopy of pre-stained slides. Diagnostic application of simple stainings – microscopy of gonococci stained by the Loeffler method on a direct microscopy slide from uretral secretion. Microscopy of Helicobacter pylori in a slide from gastric mucosa biopsy, stained by the Pfeffer method.

PRACTICAL CLASS №2 – 3 hours

Complex staining methods. Gram and Neisser staining. Ziehl-Neelsen staining (for acid-fast bacteria). Moeller staining (spores).

PURPOSE OF THE PRACTICAL CLASS: Mastering the microscopy slide preparation and staining techniques by the Gram, Neisser, Ziehl-Neelsen and Moeller.

PRACTICAL TASKS: Gram staining and observation of a microscopy slide with a mixed culture of Gram / + / and Gram / - / bacteria. Staining by the Neisser method (for metachromatic granules) of a slide with diphtheroid bacteria and observation of a pre-made microscopy slide with Diphtheria bacteria. Preparation of a direct microscopy slide from sputum and staining it by the Ziehl-Neelsen and Kinyon methods. Detection of tuberculosis bacteria in a ready-made and stained microscopy slide from sputum. Preparation of a microscopy slide with spore-bearing bacteria (bacilli) and staining it by the Moeller method. Microscopy of ready-made slides of anthrax bacilli with central non-deforming spores.

PRACTICAL CLASS №3 – 3 hours

Cultivation of microorganisms. Types of growth media. Methods for isolation of microorganisms in pure culture. Types of microbial cultures and colonies.

PURPOSE OF THE PRACTICAL CLASS: To introduce students to the types of growth media and ways of their preparation and use. To master the streaking technique and methods for isolation of microorganisms in pure culture. To be able to characterize the bacterial growth on solid and liquid culture media.

DEMONSTRATION OF: Different types of ready-made solid and liquid sterile growth media. Cultures of various microorganisms on liquid and solid growth media - nutrient broth, glucose broth, nutrient agar, EMB agar, apocholate-citrate agar, blood agar, Lowenstein-Jensen agar, Zeissler agar and others. Different types of colonies. Streaking technique on Petri dishes with nutrient agar. Inoculation on agar slant. Preparation of inoculum in a deep agar.

PRACTICAL TASKS: Streaking *S. epidermidis* on nutrient agar from a pathological material – pus (primary inoculation). Subcultivation of S. epidermidis on agar slant. Inoculating a liquid nutrient medium with pathological material (pus). Description of microbial growth from ready-made cultures on liquid and solid media.

PRACTICAL CLASS №4 – 3 hours

Biochemical activity of bacteria. Pathogenic factors of bacteria.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the methods for biochemical (enzymatic) activity testing – part of the pure culture identification process. Introduction to the pathogenic factors of bacteria and laboratory methods for their determination.

DEMONSTRATION OF: IMVUC tests (conventional – in tubes and semi-automated in microplate systems as API). Tests for saccharolytic activity: decomposition of sugars with and without gas formation; the degree of acidity with methyl-red reagent; Voges-Proskauer test. Tests for proteolytic activity: formation of indole in tryptophan broth; hydrogen sulfide formation; urease activity, etc. Deamination and decarboxylation of the amino acids arginine, lysine, ornithine. Oxidase and catalase activity. Alpha- and beta- hemolysis of blood agar. Plasma coagulase test. Plasma agglutination (clumping-test). Microscopy of encapsulated bacteria (pneumococci and anthrax bacilli) stained by Klett. Identification by MALDI-TOF, Vitek-2 and other automatic methods.

PRACTICAL TASKS: Description of the growth on inoculated plates by the students in the previous practical class. Adding reagents to IMVUC tests and analyzing the results. Testing for indole with Ehrlich reagent. Degree of acidity of bacterial culture with a methyl-red reagent. Inoculating Kligler slant medium. Reporting the results from ready-made biochemical tests on Escherichia coli and Klebsiella pneumoniae. Plasma agglutination.

PRACTICAL CLASS №5 – 3 hours

Determination of the in vitro susceptibility of bacteria to antibiotics (antibiogram). Other methods for antimicrobial resistance detection.

PURPOSE OF THE PRACTICAL CLASS: To master the technique for preparing an antibiogram by the Bauer-Kirby disk-diffusion method and the principles of its reading and interpretation. To get acquainted with the principles of preparation of E-test, microdilution test, D-test, and phenotypic detection of ESBL producing strains and others.

DEMONSTRATION OF: Preparation and interpretation of a Bauer-Kirby antibiogram. Different types of phenotypic tests to determine in vitro antibiotic resistance. Printouts of mPCR samples with resistance gene detection.

PRACTICAL TASKS: Mastering the technique for preparing an antibiogram by the Bauer-Kirby disk-diffusion method. Reporting the results from ready-made antibiograms of different microbes. Reporting the results of pre-made: E-test, microdilution test, D-test, test for ESBL-producing strains.

PRACTICAL CLASS №6 – 3 hours

Resistance of microorganisms. Sterilization and methods of sterilization. Disinfection and disinfectants.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the devices and methods for sterilization and disinfectants.

DEMONSTRATION OF: The sterilization rooms in the department, Koch's apparatus, autoclave, and dry sterilizer. Materials, laboratory vessels and utensils to be sterilized. Method of packaging the laboratory utensils. Disinfectant solutions. Means for sterilization and disinfection control.

PRACTICAL CLASS №7 – 3 hours

Recapitulation of the studied material from practical classes from №1 to №8 included.

PURPOSE OF THE PRACTICAL CLASS: To establish knowledge and practical skills in microbiology.

PRACTICAL CLASS №8 – 3 hours

SEMINAR on the topic: Morphology, physiology, genetics and antimicrobial resistance of microorganisms. Test №1.

PURPOSE OF THE PRACTICAL CLASS: To consolidate the theoretical knowledge of the studied material. To check the knowledge gained from the lectures, practical classes and independent preparation of students on topics 1-8 of the syllabus.

PRACTICAL CLASS №9 – 3 hours

Cellular and humoral basis of the immune response. Antigen-antibody reactions. Agglutination reaction. Precipitation reaction. Neutralization reaction (ASLO).

PURPOSE OF THE PRACTICAL CLASS: Introduction to the morphology of cells involved in the immune response and the structure of antibodies and their functional areas. Mastering the technique for performing immune diagnostic reactions - agglutination and precipitation, their diagnostic significance, and interpretation.

DEMONSTRATION OF: Microscopy slides of blood smears with macrophages, leukocytes, lymphocytes, blast, and plasma cells. Types of agglutination reactions - Gruber and Widal. Types of precipitation reactions - Ascoli ring test, Mancini test, immunodiffusion in agar. ASLO (AST), turbidimetry and nephelometry.

PRACTICAL TASKS: Observation and drawing cells involved in the immune response. Performing Gruber agglutination test, Ascoli ring test. Interpretation of AST titers.

PRACTICAL CLASS №10 – 3 hours

Antigen-antibody reactions. Bacteriolysis, hemolysis, cytolysis. Complement fixation test (CFT). Immune reactions with labeled antibodies or antigens: immunofluorescence method (IFA), radioimmunoassay (RIA), enzyme-linked immunosorbent assay (ELISA).

PURPOSE OF THE PRACTICAL CLASS: Introduction to the technique of immune reactions CFT and the principles of marked immune reactions, their interpretation, and diagnostic value.

DEMONSTRATION OF: Hemolysis. Wasserman's complement-fixation test. ELISA equipment and ready-made ELISA plate. Immunofluorescence microscope and immunofluorescence test for *Chlamydia trachomatis* in cervical secretions.

PRACTICAL TASKS: Reading a Wasserman sample. Reporting positive and negative ELISA results from sera for anti-HCV, HBsAg or other suitable samples. Reading and reporting immunofluorescence test for Chlamydia and others.

PRACTICAL CLASS №11 – 3 hours

Flow cytometry for the determination of cellular subpopulations (immunophenotyping). Allergy testing. Biological products – vaccines and sera.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the operation principles of the flow cytometer and diagnostic capabilities of the equipment for immune status testing. Mastering the techniques of allergic tests for the diagnosis of fast and delayed type of hypersensitivity. Biological products used for specific therapy and prophylaxis of infectious diseases - vaccines and sera.

DEMONSTRATION OF: Flow cytometry test of a patient whole blood. Different types of vaccines – mandatory, according to the immunization calendar of the Republic of Bulgaria, recommended ones for different groups of people and for travelers. Antitoxic, antiviral and antibacterial sera. Guinea pig Mantoux allergy test.

PRACTICAL TASKS: Interpretation of flow cytometry protocol results Mastering the technique of the Mantoux test - intradermal injection of allergens.

PRACTICAL CLASS №12– 3 hours

SEMINAR on: Infection and immunity. Test №2.

PURPOSE OF THE PRACTICAL CLASS: To consolidate the theoretical knowledge on the studied material. To control the knowledge gained from lectures, exercises and independent preparation of students on topics №11-22 of the syllabus.

PRACTICAL CLASS №13– 3 hours

Laboratory diagnosis of diseases caused by viruses and rickettsia.

PURPOSE OF THE PRACTICAL CLASS: To introduce students to the particularities of the diagnosis of diseases caused by viruses and rickettsia.

DEMONSTRATION OF: Tissue cultures - normal and with cytopathic effect. Chicken embryos Hearst phenomenon and Hearst reaction (HIA). ELISA plate with positive and negative samples for HBsAg and anti-HCV antibodies. Immunofluorescence test.

PRACTICAL TASKS: Observation and drawing tissue cultures - normal and with cytopathic effect. Reporting the results from HIA in influenza, ELISA tests for hepatitis markers and HIV, immunofluorescence test for Chlamydia.

PRACTICAL CLASS №14 - 3 hours

Methods for microbiological diagnosis of infectious diseases. General scheme for microbiological examination.

PURPOSE OF THE PRACTICAL CLASS: Applying the acquired knowledge in a consecutive scheme for microbiological diagnosis of infectious diseases.

DEMONSTRATIONS AND PRACTICAL TASKS: Introduction to the general rules for collecting and sending pathological materials to the microbiological laboratory. Transport culture media. Preparation of a microscope slide from pathological material. Gram staining. Microscopy with immersion oil. Microscopy of ready-made slides. Recognition of pure microbial cultures on various growth media. Identification tests - cultural; biochemical; determination of pathogenic factors; serotyping by Gruber agglutination test. Antibiogram interpretation.

PRACTICAL CLASS №15 - 3 hours
Evaluation of the practical skills acquired during the semester.

PRACTICAL CLASS №16 - 2 hours

Microbiological diagnosis of staphylococcal and streptococcal infections. Microbiological examination of pus.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the most common causative agents of purulent infections in the human body. Methods for microbiological diagnostics.

DEMONSTRATION OF: Prepared microscope slides with Gram-stained staphylococci and streptococci. Demonstration of the cultural features of staphylococci and streptococci on different growth media and tests for their identification. Methods for determination of methicillin-resistant staphylococci.

PRACTICAL TASKS: Identification of staphylococcal and streptococcal cultures on blood agar. Reporting alpha- and beta-hemolysis. Reading bacitracin and optochin test results. Performing a coagulase slide test. Coagulase tube test reading. Result interpretation of antibiograms of staphylococci, beta-hemolytic, and viridans streptococci. Reading and interpretation of ASLO test. Differential diagnosis between alpha-hemolytic streptococci and pneumococci. MRSA screen agar – result interpretation.

PRACTICAL CLASS №17 - 2 hours

Microbiological examination in diseases caused by *Streptococcus pneumoniae.* Microbiological diagnosis of tuberculosis and leprosy. Microbiological examination of sputum.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the methods of collection and microbiological examination of sputum, features of the specific inflammatory process. Introduction to the microbiological diagnosis of tuberculosis, leprosy, and pneumococcal infections. Bioproducts for specific prevention and therapy.

DEMONSTRATION OF: Microscopic slides: a) Pneumococci stained by Klett method; b) Tuberculosis bacteria in sputum smear, stained by Ziehl-Neelsen method. Culture features of pneumococci on blood agar and glucose broth. Optochin and inulin test. Culture of tuberculosis bacteria on Lowenstein-Jensen agar. Gamma-interferon-based tests

PRACTICAL TASKS: Preparation of a microscopic smear of sputum and stain it by Ziehl-Neelsen staining for the detection of tuberculosis bacteria. Reporting the positive and negative optochin tests of alpha-hemolytic microorganisms. Reading an antibiogram of pneumococci on blood agar. Introduction to the principles of gamma-interferon-based tests. Introduction to bioproducts for specific prophylaxis (pneumococcal vaccine, BCG - vaccine) and for allergic diagnosis (PPD) of tuberculosis.

PRACTICAL CLASS №18 - 2 hours

Microbiological diagnosis of diphtheria and pertussis. Microbiological examination of throat swabs.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the microbiological diagnosis of diphtheria and pertussis and bioproducts for specific prophylaxis and treatment.

DEMONSTRATION OF: Ready-made microscope slides with diphtheria bacteria stained by the Neisser method. Cultural features of diphtheria bacteria in the Löffler and Klauberg media. Growth of *Bordetella pertussis* on the Bordet-Gengou culture media.

PRACTICAL TASKS: Microscopic observation of a ready-made microscope slide with *Bordetella pertussis,* Gram-stained. Preparation of a slide with pseudodiphtheria bacteria and Neisser staining. Introduction to bioproducts for specific prophylaxis (DTP and DT vaccines) and specific therapy for diphtheria (diphtheria antitoxin).

PRACTICAL CLASS №19 - 2 hours

Microbiological diagnosis of gas gangrene and tetanus. Microbiological examination of wound secretions.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the microbiological diagnosis of gas gangrene and tetanus, the particularities of collecting and sending pathological materials for anaerobic bacteria, and bioproducts for specific prophylaxis and therapy.

DEMONSTRATION OF: Ready-made microscope slides of *C. perfringens* and *C. tetani*, stained by the Gram method. Culture media for anaerobic bacteria - Kitt-Tarozzi, thioglycolate broth, Zeissler, and Wilson-Blair agars and the cultural features of gas gangrene agents and the tetanus bacillus. Tetanic seizure of a mouse injected with blood from a sick patient.

PRACTICAL TASKS: Preparation of a microscope slide with Gram-stained wound swab containing clostridia. Introduction to bioproducts for specific prophylaxis and therapy of gas gangrene and tetanus.

PRACTICAL CLASS №20 - 2 hours

Microbiological examination of CNS materials. Microbiological diagnosis and differential diagnosis of bacterial meningitis (*Neisseria meningitidis and Haemophilus influenzae*).

PURPOSE OF THE PRACTICAL CLASS: Mastering the differential microscopic diagnosis of bacterial meningitis.

DEMONSTRATION OF: Ready-made microscope slides from cerebrospinal fluid with *Neisseria meningitidis*, stained with methylene blue and Gram-stain, *H. influenzae*, stained by the Gram method and other microorganisms, causing bacterial meningitis. Cultural features of *H. influenzae* on chocolate agar, Levinthal agar, and blood agar – satellite phenomenon. Methods for microanaerophilic cultivation and cultures of *Neisseria meningitidis* on Levinthal agar and blood agar. Other causative agents of bacterial meningitis - pneumococci, staphylococci, streptococci and tuberculosis bacteria. Latex agglutination tests to detect microbial antigens in the cerebrospinal fluid of patients

PRACTICAL TASKS: Preparation of a microscope slide with cerebrospinal fluid and stained by the Gram method. Observation of ready-made microscope slides with *Neisseria meningitidis and H. influenzae*. Reporting the phenomenon of satellite growth in *H. influenzae.*

PRACTICAL CLASS №21 - 2 hours

Microbiological diagnosis of particularly dangerous infections plague, cholera, and anthrax.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the microbiological diagnosis of plague, cholera, and anthrax and the relevant bioproducts for specific prophylaxis and therapy.

DEMONSTRATION OF: Ready-made microscopic slides - blood smear with *Bacillus anthracis*, stained by the Klett method for capsules and Moeller - for spores, *Vibrio cholerae* - stained by the Gram method and *Yersinia pestis* - stained by Giemsa method. Culture media, bacterial cultures, and biochemical tests for identification of *Vibrio cholerae*. Ascoli precipitation test for anthrax antigen.

PRACTICAL TASKS: Microscopy and drawing of a slide with B. anthracis, V. cholerae and Y. pestis. Performing the Ascoli precipitation test. Immunofluorescence microscopy of B. anthracis.

PRACTICAL CLASS №22 - 2 hours

SEMINAR on the topic: Microbiological diagnostics of microorganisms, studied in practical classes from №1 to №7. Test №1.

Staphylococcus spp. Streptococcus spp. Streptococcus pneumoniae. Tuberculosis bacteria and other mycobacteria. Mycobacterium leprae. Neisseria meningitidis. Corynebacterium diphtheriae. Bordetella pertussis. Haemophilus influenzae. Causative agents of gas gangrene. Clostridium tetani. Bacillus anthracis. Vibrio cholerae. Yersinia pestis

PRACTICAL CLASS №23 - 2 hours

Microbiological examination of materials from the digestive system (feces). Microbiological diagnosis of bacterial dysentery, E.coli enteritis, salmonellosis. Food poisoning by Staphylococcus aureus, salmonella, and clostridia (C. botulinum, C. perfringens). Microbiological examination of gastric mucosa biopsy material (Helicobacter pylori).

PURPOSE OF THE PRACTICAL CLASS: Introduction to the scheme for bacteriological examination of feces in diseases caused by members of the family Enterobacteriaceae (pathogenic E. coli, Shigella, Salmonella). Introduction to the microbiological diagnosis of food poisoning by bacterial agents and methods for bacterial diagnosis of Helicobacter pylori from biopsy material.

DEMONSTRATION OF: Microscope slide with Gram-negative microorganisms. Selective and differentiating media for enteric bacteria - Levine, deoxycholate-citrate agar with cultures of lactose-positive and lactose-negative bacteria; blood agar with S. aureus; culture media for clostridia. Biochemical tests for identification. Serotyping by agglutinating sera. Microscopic preparation of H. pylori.

PRACTICAL TASKS: Preparation of a microscope slide from a culture of E. coli and Gram staining. Performing biochemical tests - indole formation, acidity (MR), Voges-Proskauer test, etc. Performing type Gruber agglutination test. Inoculation of feces on a differentiating medium.

PRACTICAL CLASS №24 - 2 hours

Microbiological examination of urine. Microbiological diagnosis of pathogens causing urinary tract infections: opportunistic pathogens (E. coli, Klebsiella, Proteus-Providentia-Morganella, Pseudomonas) and obligatory pathogenic (streptococci, salmonella, leptospira, M. tuberculosis).

PURPOSE OF THE PRACTICAL CLASS: To introduce the rules for collection and sending urine for bacteriological examination and the methods for detection and isolation of the most common bacterial agents.

DEMONSTRATIONS OF: Cultural and biochemical features of E. coli, Klebsiella, Proteus, Pseudomonas. Inoculation of urine culture by the quantitative method with a calibrated loop. Demonstration of leptospira in a wet mount slide on dark-field microscopy. Ready-made microscopic preparations of Gram-negative rods, streptococci, tuberculosis bacteria.

PRACTICAL TASKS: Inoculation of urine on Levine agar with calibrated loop. Reading and interpretation of urine cultures with different degrees of bacteriuria - determination of microbial count. Observation and drawing of leptospira. Interpretation of an antibiogram of microorganisms isolated from urine.

PRACTICAL CLASS №25 - 2 hours

Microbiological examination for sexually transmitted infections caused by Neisseria gonorrhoeae, Treponema pallidum, C. albicans, chlamydia, and mycoplasmas.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the morphology and biology of the most common causative agents of sexually transmitted infections.

DEMONSTRATIONS OF: Ready-made microscope slide of urethral smear with N. gonorrhoeae; ready-made microscope slide with Candida albicans. Immunofluorescence slide with elementary bodies of chlamydia in cervix cells. Bacterial cultures of Candida albicans on Sabouraud agar; chlamydospores of Candida albicans on rice agar; filamentation test. ELISA. Incolulation of fungi on ChromagarCandida. sowing of ChromagarCandida fungi. Positive and negative CFT (Wasserman) and VDRL tests - as screening tests for syphilis. Multiplex PCR - results from a patient.

PRACTICAL TASKS: Microscopy and recognition of N. gonorrhoeae in Gram and Loffler-stained urethral secretions. Reporting positive and negative samples of Wasserman and VDRL test. Observation and detection of chlamydospores of Candida albicans on rice agar. Working with mPCR protocols for sexually transmitted infections.

PRACTICAL CLASS №26 - 2 hours

Microbiological examination of blood - blood culture. Causative agents of septic conditions: obligatory pathogenic (Salmonella typhi, Brucella, Borrelia) and facultative pathogenic bacteria.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the microorganism cause bacteremia and sepsis and methods for the microbiological examination of bloodstream infections - conventional and automatic methods.

DEMONSTRATION OF: Blood culture media - soy-casein broth, thioglycollate broth, etc. Salmonella spp. and Proteus spp. cultures on Levine agar, deoxycholate-citrate agar and selenite broth. IMVUC tests of *Salmonella typhi* and *Proteus mirabilis*. Brucella culture media. Microscopic preparations of Salmonella spp. and Brucella spp. according to Gram stain. Cultures of streptococci and staphylococci on blood agar and tests for their identification. Analytical Vidal for typhoid fever and Right's agglutination for brucellosis.

PRACTICAL TASKS: Observation of ready microscopic slides. Reporting of positive and negative blood cultures cultured in the BactAlert apparatus. Characterization of salmonella and proteus growth on differentiating media. Reporting conventional tests for identification and the API system for Salmonella typhi and Proteus mirabilis. Determination of sample titers of Vidal and Right.

PRACTICAL CLASS №27 - 2 hours

Problematic microorganisms causing nosocomial and iatrogenic infections - Pseudomonas, Acinetobacter, Enterococcus, MRSA, C. difficile. Systemic mycoses caused by Candida, Aspergillus, Actinomyces, and Cryptococcus. Antifungal therapy.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the most common microorganisms that caused nosocomial and iatrogenic infections, methods for microbiological diagnosis, and interpretation of results. Introduction to the causative agents systemic mycoses - Candida, Aspergillus, Actinomyces, Cryptococcus and modern antifungal therapy

DEMONSTRATION OF: Cultures, identification tests, and AST of Pseudomonas, Acinetobacter, Enterococcus, MRSA. ELISA test for C. difficile toxins. Cultures, tests for the identification of Candida, Aspergillus, Actinomyces, Cryptococcus, and E-tests for antifungal drugs.

PRACTICAL TASKS: Reporting and identification of cultures, AST of multidrug-resistant Pseudomonas and Acinetobacter. Determination of ESBL (+) E. coli. Determination of MRSA and MSSA strains on MRSA screen agar. Reading AST of vancomycin-resistant enterococci - VRE. Resistance gene detection by mPCR. Characterization of cultures of Candida, Aspergillus, Actinomyces, Cryptococcus and reporting E-tests for antifungal drugs - voriconazole, anidulafungin, caspofungin, and others.

PRACTICAL CLASS №28 - 2 hours

Microbiological diagnosis of diseases caused by viruses - HIV, hepatitis A, B, C, D, and E, flu, and coronaviruses - SARS-CoV-2.

PURPOSE OF THE PRACTICAL CLASS: Introduction to the specificities of the viral diagnosis, viruses important for human pathology.

DEMONSTRATION OF: Hearst phenomenon and Hearst reaction (HFA) for Influenza viruses. ELISA - conventional and automated and CLIA equipment; real-rime PCR and mPCR. ELISA samples for the diagnosis of viral markers (HBsAg, anti-HCV antibodies, anti-HIV-1,2 antibodies, etc.). mPCR for detection of influenza viruses, SARS-CoV-2, etc. respiratory pathogens.

PRACTICAL TASKS: Performing an ELISA test for anti-HCV antibodies. Reporting and interpretation of the result. Reporting of results from the mPCR respiratory panel - with SARS-CoV-2.

PRACTICAL CLASS №29 - 2 hours

Sanitary-microbiological examination of water, air, hospital environment. Sanitary-indicative microorganisms - E. coli, Enterococcus, C. perfringens, staphylococci, streptococci. TEST (practical classes №8-13)

PURPOSE OF THE PRACTICAL CLASS: Introduction to the methods of collection, transport, and basic scheme for sanitary-microbiological examination. Criteria for the identification of microorganisms as nosocomial agents.

DEMONSTRATION OF: Nutrient media and materials needed to determine the microbial count and *E.coli* titer in the sanitary-microbiological examination of water. Biochemical tests for differentiation of *E. coli* and *Klebsiella* - IMVUC. Sedimentation plates method for microbial air monitoring. Enterococcal nutrient media and cultures. Sherman's test.

PRACTICAL TASKS: Taking a hand swab for sanitary-microbiological examination and broth inoculation. Identification of *E. coli*, *Klebsiella*, *Enterococcus, C. perfringens*. Determination of microbial biotype, serotype, resistotype, etc. Identifying intrahospital infections. Characterization of the growth of *Staphylococcus* and *Streptococcus* strains on blood agar and identification according to specific tests.

EXERCISE №30 - 2 hours

Recapitulation of the students' practical skills, acquired during the two semesters with an evaluation mark.

- **Bibliography:**

Textbooks for medical students in Microbiology - English training

Obligatory:

1. Review of Medical Microbiology and Immunology, Warren Levinson, 13 e. McGraw Hill Education, 2014, ISBN 978-0-07-181811-7, 789 pp or
2. Medical Microbiology. Patrick R. Murray, Ken Rosenthal, Michael Pfaller. 8 e. Elsevier, 2016, ISBN 978-0-323-29956- 5, 848 pp.

Recommended:

1. Medical Microbiology. S. Baron, 4 edt, 2000, ISBN-10: 0-9631172-1-1, http://www.ncbi.nlm.nih.gov/books/NBK7627/ or
2. Todar's online textbook of bacteriology. K. Todar, 2009, http://www.textbookofbacteriology.net/kt_toc.html
3. Medical microbiology / Patrick R. Murray et al . - 8th ed. - Philadelphia : Elsevier, 2016 . - 836 p. - (Student consult)
4. Cases in medical microbiology and infectious diseases / Peter H. Gilligan et al . - 4th ed. . - Washington : ASM Press, 2014 . - 589 p.
5. Laboratory exercises in microbiology : For students of medicine / Galina Zhelezova et al. - Sofia : St. Kliment Ohridski University Press, 2014 . - 256 p.
6. Mims` medical microbiology / Richard V. Goering et al. - 5th ed. . - Philadelphia: Elsevier, Saunders, 2013 . - 565 p. - (Student consult)
7. Manual for practical exercises in microbiology / Mariya Petrova Sredkova et al. - Pleven: Publ. center, Medical University - Pleven, 2012 . - 155 p.
8. Medical microbiology / Ed. Michael Ford . - 2nd ed. . - Oxford : Oxford University Press, 2014. - 484 p.

- **SYLLABUS FOR THE SEMESTER EXAM**

GENERAL MICROBIOLOGY

1. Subject and tasks of microbiology. Pasteur and Koch's contributions to the development of microbiology. Taxonomy of microorganisms - nomenclature and classification. General characteristics of the separate groups of microorganisms.
2. Morphology of bacteria - basic shapes, size. Methods for studying the morphology of bacteria. Bacterial structure - capsule, bacterial wall, cytoplasmic membrane, cytoplasm, and cytoplasmic inclusions. Flagella, pili, spores.
3. Bacterial genetics. Bacterial genotype and phenotype. Genetic apparatus in bacteria. The bacterial chromosome as a genetic system. Extrachromosomal genetic elements. Bacteriophages - main types, structure. Forms of the interaction of bacteriophages with bacteria - lytic cycle, moderate phage, phage conversion. Phage typing. Practical applications.
4. Microbial variability. Mutation. Mutagenic factors - chemical and physical, mechanism of action, practical significance, and application. Genetic exchange between bacteria: transformation, transduction, conjugation - mechanisms. Significance of bacterial and phage genetics. Genetic engineering. Modern genetic methods in clinical microbiology. DNA probes, PCR - polymerase chain reaction..
5. Bacterial physiology. Chemical composition of bacteria. Types of bacterial enzymes and their practical significance. Metabolism in bacteria - catabolic and anabolic processes. Bacterial respiration. Bacterial nutrition. Nutrient transfer.

6. Growth and multiplication of bacteria. Growth phases and growth curves. Bacterial cultivation - basic principles, types of nutrient media. Growth factors in bacteria.
7. Influence of physical factors on microorganisms: heat, drying, lyophilization, light, atmospheric pressure, osmotic pressure, pH, radiation, sound energy. Sterilization. Sterilization methods. Influence of chemical factors on microorganisms; Mechanism of action. Oligodynamic effect. Disinfection. Types of disinfectants. Influence of biological factors on microorganisms: symbiosis, antagonism, antibiosis
8. Antimicrobial agents. Antibacterial drugs - main groups and mechanisms of action. Mechanisms of resistance. Determination of bacterial susceptibility to antibiotics.
9. Viruses. Nature and properties. Cultivation methods. Classification. *Rickettsia*. Nature and properties. Cultivation methods. Classification.
10. The external environment as a factor in the spread of infectious diseases. Microflora of water, soil, and air. Microorganisms in food products, hospital rooms, etc. Sanitary-indicative microorganisms in the environment.

INFECTION AND IMMUNITY

11. Infection and infectious process. The role of microorganisms in the infectious process. Pathogenicity, virulence, contagiousness, invasiveness, toxigenicity. Pathogenicity factors. Pathogenesis of the infectious process. Characteristics of infectious disease. Forms of the infectious process. The role of the macroorganism in the infectious process. The role of the external environment for the occurrence and course of the infectious process. Epidemic process. Factors and mechanisms of transmission of infectious agents in the epidemic process.
12. Natural resistance. Protective role of the skin, mucous membranes, organs, and normal microflora. Humoral factors of natural resistance. Lysozyme. Complement. Interferon. Cellular factors of natural resistance. Phagocytosis. Inflammation.
13. Immunity. Definition. Types of immunity. Anatomy and structure of the immune system. Central and peripheral immune organs. Cells of the immune system.
14. Antigens. Types of antigens. Antigenic characteristics of microorganisms.
15. Humoral immunity. Characteristics of antibodies (immunoglobulins). Structure and functions of different classes of immunoglobulins. Mechanism of action of antibodies. Local immunity.
16. Cellular immunity. Cells and mechanism of action. Forms of cellular immunity. Cellular cooperation in the immune response.
17. Development of the immune response. Dynamics of the immune response – primary and secondary immune response. Humoral regulation of the immune response. Genetics and genetic control of the immune response. APC. The role of MHC - antigen recognition molecules
18. Allergy - definition and forms. Fast type of allergy - anaphylaxis, atopy, clinical significance. Cytotoxic allergic reactions. Allergic phenomena of immune complexes – Arthus phenomenon, serum sickness, clinical significance. Slow type of allergy - cell-mediated hypersensitivity. Contact dermatitis. Clinical significance.
19. Immunopathology. Immunopathological reactions and diseases. Immunological tolerance. Autoimmune diseases. Immunodeficiency conditions and diseases. Infectious diseases of the immune system.
20. Antigen-antibody reaction. Types of immune diagnostic reactions - agglutination, precipitation, neutralization - toxin-antitoxin, AST, virus-neutralizing reaction. Complement dependent - bacteriolysis, cytolysis, hemolysis, complement fixation test (CFT). Mechanism of reactions and application in microbiological diagnostics.

21. Labeled immune reactions - immunofluorescence (IFA), radioimmune (RIA) and enzyme-linked immunosorbent assay (ELISA) tests. Hybridoma biotechnology. Monoclonal antibodies.
22. Immunoprophylaxis and immunotherapy. Vaccines and serums. Immunomodulation.

MICROBIOLOGY

23. Staphylococci. (*Staphylococcus*) Species, morphology, biology, biochemical productivity, pathogenicity factors. Signs of pathogenicity in staphylococci. Diseases, immunity. Microbiological diagnosis. Antibiotic therapy. MRSA - clinical significance and diagnosis.
24. Streptococci (*Streptococcus*). Classifications. Morphology, biology, antigenic structure, pathogenicity factors. Diseases. Immunity. *Streptococcus* as a cause of scarlet fever. Microbiological diagnosis. Antibiotic therapy. Pneumococci (*Streptococcus pneumoniae*). Morphology, biology, biochemical productivity. Antigenic structure. Pathogenicity factors. Diseases. Immunity. Microbiological diagnosis. Therapy and specific prevention.
25. Meningococci (*Neisseria meningitidis*). Morphology, biology, biochemical productivity. Antigenic structure - serogroups. Pathogenicity factors. Pathogenesis and clinical forms of meningococcal infection. Immunity. Microbiological diagnosis. Specific prevention and therapy. Gonococci (*Neisseria gonorrhoeae*). Morphology, biology, biochemical productivity. Pathogenicity factors. Pathogenesis and clinical forms of gonococcal infection. Immunity. Microbiological diagnosis. Prevention and therapy.
26. Family Enterobacteriaceae. Groups of intestinal bacteria according to pathogenicity. General characteristics: morphology, biology, biochemical productivity. Antigenic structure. Pathogenicity factors. Properties of endotoxin. Coli bacteria (*Escherichia coli*). Morphology, biology, biochemical productivity. Antigenic structure. Pathogenicity factors. Diseases. Pathogenic *Escherichia coli* in the intestinal tract. Immunity. Microbiological diagnosis.
27. *Proteus*. *Providencia*. *Morganella*. Species. General characteristics: morphology, biology, biochemical productivity. Diseases. Therapy. Microbiological diagnosis. Tribus *Klebsiellae*. Species. Morphology, biology, biochemical productivity. Pathogenicity factors. Diseases. Immunity. Microbiological diagnosis. Therapy. *Pseudomonas*. Morphology, biology, biochemical productivity. Pathogenicity factors. Diseases. Microbiological diagnosis. Therapeutic problems.
28. *Salmonella*. General characteristics: morphology, biology, biochemical productivity. Kaufman antigenic characterization and classification. Antigenic formulas. Pathogenicity factors. Pathogenesis, immunity, and specific prophylaxis in typhoid and paratyphoid fever. *Salmonella* – as causative agents of food poisoning. Characteristic. Microbiological diagnosis.
29. Dysenteric bacteria (*Shigella*). Classification. Morphology, biology, biochemical productivity. Antigenic structure. Pathogenicity factors. Pathogenesis and immunity. Microbiological diagnosis. *Helicobacter pylori*. Morphology, biology, biochemical productivity. Diseases. Microbiological diagnosis. Therapy. *Clostridium difficile*. Morphology, biology, biochemical productivity. Pathogenicity factors. Pathogenesis and immunity. Microbiological diagnosis. Therapy.
30. The causative agent of plague (*Yersinia pestis*). Morphology, biology, biochemical productivity. Pathogenicity factors. Pathogenesis and immunity. Microbiological diagnosis. Specific prevention and therapy. *Yersinia enterocolitica* - morphology,

biology, biochemical productivity. Pathogenicity factors. Pathogenesis. Microbiological diagnosis.

31. *Vibrio cholerae*. Morphology, biology, biochemical productivity. Antigenic structure. Serological types. Pathogenicity factors. Pathogenesis and immunity. Microbiological diagnosis. Specific prevention and therapy.
32. Pertussis and pertussis bacteria (*Bordetella pertussis, B. parapertussis*). Morphology, biology. Pathogenicity factors. Pathogenesis and immunity. Microbiological diagnosis. Specific prevention and therapy. Genus *Haemophilus*. Morphology, biology. Antigenic structure. Pathogenicity factors. Diseases. Immunity. Microbiological diagnosis. Specific prevention and therapy. *Listeria monocytogenes*. General characteristics.
33. *Brucella*. Species. Morphology, biology, biochemical productivity. Pathogenicity factors. Pathogenesis and immunity. Microbiological diagnosis. Specific prevention. Causative agent of tularemia (*Francisella tularensis*) - general characteristics. *Legionella pneumophila* - general characteristics.
34. Causative agent of diphtheria (*Corynebacterium diphtheriae*). Morphology, biology, biochemical productivity. Pathogenicity factors. Pathogenesis and immunity. Microbiological diagnosis. Specific prevention and therapy. Diphtheroid bacteria (*C. jeikeium, C. urealyticum, C. amiculatum, C. pseudo-diphtheriticum*). Clinical significance.
35. *Mycobacterium*. Causative agent of tuberculosis (*Mycobacterium tuberculosis*). Morphology, biology, pathogenesis, clinical forms, immunity, allergy. Specific prevention of tuberculosis. Therapy. Microbiological diagnosis. The causative agent of leprosy (*Mycobacterium leprae*). Morphology, biology. Pathogenesis. Clinical forms. Prevention. Microbiological diagnosis.
36. Causative agent of anthrax (*Bacillus anthracis*). Morphology, biology. Pathogenesis, clinical forms. Immunity. Specific prevention and therapy. Microbiological diagnosis. The causative agent of typhoid fever (*Borrelia recurrentis*). Morphology, biology. Pathogenesis, immunity. Microbiological diagnosis. The causative agent of Lyme disease (*Borrelia burgdorferi*). Pathogenesis. Immunity. Microbiological diagnosis
37. Anaerobic spore-forming bacteria - genus *Clostridium*. General characteristics - morphology, biology. Tetanus bacillus (*Clostridium tetani*). Pathogenicity factor. Pathogenesis and immunity. Specific prevention and therapy. Microbiological diagnosis. Causative agents of gas gangrene (*C. perfringens, C. novyi, C. septicum C. histolyticum*). Pathogenicity factors. Pathogenesis, immunity, prevention, and therapy. Microbiological diagnosis. The causative agent of botulism (*C. botulinum).* Pathogenicity factor. Pathogenesis and immunity. Prevention and specific therapy. Microbiological diagnosis.
38. Spirochetes (family Spirochaetaceae) - general characteristics. The causative agent of syphilis (*Treponema pallidum*). Morphology, biology. Pathogenesis and immunity. Microbiological diagnosis. *Leptospira*. Species. Morphology, biology. Antigenic structure. Pathogenesis and immunity. Microbiological diagnosis
39. Genus *Mycoplasma*. Classification. Morphology, biology. Diseases. Microbiological diagnosis. L-forms of bacteria. Genus *Chlamydia*. General characteristics. Species. Causative agents of ornithosis and trachoma. Morphology, biology. Pathogenesis. Diseases. Microbiological diagnosis
40. The causative agent of typhus (*Rickettsia prowazekii*). Morphology, biology. Pathogenesis and immunity. Specific prevention. Microbiological diagnosis. The causative agent of Marseille fever (*Rickettsia conorii*). Morphology, biology. Pathogenesis and immunity. Microbiological diagnosis. The causative agent of Q fever (*Coxiella burnetii*). Morphology, biology. Microbiological diagnosis.

41. Pathogenic fungi (*Fungi*). Candida (genus *Candida*). Morphology, biology. Pathogenesis, clinical forms. Microbiological diagnosis. Therapy. *Aspergillus*, *Cryptococcus*, Actinomycetaceae. Morphology, biology, diseases, and microbiological diagnosis.

VIROLOGY

42. Family Picornaviridae. Genus *Enterovirus* - *poliovirus*, *Coxsackie viruses*, *ECHO viruses*. Genus *Rhinovirus*. Genus *Aphtovirus* - the causative agent of foot-and-mouth disease.
43. Family Orthomyxoviridae. *Influenza viruses*
44. Family Paramyxoviridae - *parainfluenza viruses*; the causative agent of mumps; the causative agent of measles. Respiratory syncytial virus.
45. Arbovirus infections and rubella. The family Togaviridae - genus *Alphavirus* and genus *Rubivirus*. Family Flaviviridae - causative agents of yellow fever, dengue, pappataci fever, tick-borne encephalitis. Family Bunyaviridae – causative agents of Crimean hemorrhagic fever and hemorrhagic fever with renal syndrome.
46. Family Poxviridae - the causative agent of smallpox. Family Adenoviridae.
47. Family Retroviridae - the causative agent of AIDS. Family Rhabdoviridae – the causative agent of rabies.
48. Family Herpesviridae - Herpes simplex virus type 1 and 2, Varicella-Zoster virus, *Cytomegalovirus*, Epstein-Barr virus, other herpes viruses.
49. Causative agents of viral hepatitis (HAV, HBV, HCV, HDV, HEV).
50. Family Coronaviridae. SARS-CoV-2, the causative agent of COVID-19.