MEDICAL MICROBIOLOGY AND PARASITOLOGY			
GENERAL INFORMATION			
Course coordinator	Associate Professor Domagoj Drenjančević, MD, PhD		
Assistant/Associate	Asst. Prof. Arlen Antolović-Požgain, MD, PhD Asst. Prof. Maja Bogdan, MD, PhD Marijan Orlović, MD Marko Živkov, MD Ivana Roksandić- Križan, MD Dinko Paulić, MD		
Study Programme	Integrated undergraduate and graduate university study of Medicine		
Status of the course	Mandatory		
Year of study, semester	3 <sup>rd</sup> year, 5 <sup>th</sup> semester		
ECTS	8		
Workload (hours)	Lectures (20); Seminars (20); Exercise (40)		
Expected number of students	70		
COURSE DESCRIPTION			
Course objectives			

Get to know, understand and independently interpret the basic biological characteristics of microorganisms (bacteria, viruses, fungi, parasites) that cause infections in humans as well as pathogenic properties of these microorganisms, their prevalence and resistance to environmental conditions, ways of their transmission among humans, their sensitivity to antimicrobial drugs and the basics of persons defense from infection. Students will also get to know and understand the types of vaccines with individual microorganisms, as well as the role of vaccination in protecting the population from infectious diseases. The special goal is for students to get acquainted with the basic groups of antimicrobial drugs from the point of view of spectrum of action, mechanism of action and mechanism of resistance of microorganisms to antimicrobial drugs, understand their action and critically interpret their application. At the end of the class, students will be able to independently determine the type of the most common microorganisms, according to a microscopic specimen or other features, interpret a susceptibility test, and determine the mode of transmission and defense against a specific microorganism. Also, students will be able to independently take a swab of the nose and throat, and plant biological materials on microbiological media. The aim of the course is also to acquire knowledge about diagnostic procedures in all branches of medical microbiology bacteriology, virology, mycology and parasitology, namely: direct diagnosis, cultivation, serological diagnosis, molecular diagnostics and rapid diagnostic procedures.

Enrolment requirements and entry competencies

No additional conditions

Learning outcomes at the Programme level

1.1., 2.1., 3.1., 3.5.

### Learning outcomes (5-10)

After passing the exam, students will acquire basic skills and knowledge in the field of medical microbiology and will be able to:

1. independently describe and analyze the characteristics of pathogenic microorganisms (bacteria, viruses, fungi and parasites), clinical conditions that cause them, epidemiological characteristics of

individual pathogens, samples and diagnostic procedures in the detection of individual pathogens and treatment and prevention procedures.

2. will acquire knowledge of procedures for isolation and cultivation and detection of microorganisms of medical significance and knowledge of serological and molecular methods in microbiology.

3. independently make microscopic preparations from biological samples and determine the presence of microorganisms

4. interpret procedures for testing the sensitivity of an isolated microorganism to antimicrobial chemotherapeutics (dilution and diffusion methods of antibiogram, E test).

5. recognize and distinguish adult parasites

The student will be able to apply all acquired knowledge and skills at the analytical level by synthesizing the adopted theoretical foundations and practical methods and procedures.

### Course content

### Lectures (20 hours)

### L1: Introduction to medical microbiology. General bacteriology

Introduction to medical microbiology. Structure, physiology, genetics and metabolism of bacteria. Bacterial morphology and cell wall synthesis. Preparation and microscopy. Basic staining in bacteriology: simple and complex staining. Immune response to bacterial infections. Pathogenesis of bacterial infections. Bacterial toxins.

### L2: Antimicrobial chemotherapeutics, disinfection and sterilization

Antimicrobial chemotherapeutics: types, division, mechanisms of action, therapeutic spectrum, testing of bacterial susceptibility to antibiotics, bacterial resistance and mechanisms of resistance. Disinfection and sterilization: types of disinfectants and mechanism of action, selection of disinfectant.

Sterilization procedures: types of sterilization, control of sterilization procedure.

### L3: General diagnostic principles in microbiology

Diagnostic procedures in bacteriology. Diagnostic procedures in virology. Diagnostic procedures in parasitology. Diagnostic procedures in mycology. Sampling of materials for microbiological tests. Direct and indirect diagnostics. Cultivation and isolation of microorganisms. Serological methods in microbiological diagnostics. Molecular diagnostics.

### L4: Diagnosis and significance of clostridia

Description and characteristics of bacterial species of the genus Clostridium: C. perfrigens, C. tetani, C botulinum, C. difficile and others. Pathogenesis of diseases caused by species of the genus Clostridium. Cultivation characteristics: anaerobic cultivation. Sporogenesis and germination. Exotoxins. Vaccines. Laboratory diagnosis of clostridial infections. Treatment, prevention and control of infections caused by clostirdia.

### L5: Fundamentals of parasitology

Parasitism as an ecological and medical concept - definitions and basic concepts. Biological associations. Parasites - pathogens and vectors, epidemiological concepts. Classification and nomenclature of medically important parasites. Parasitic pathogenicity. Fundamentals of laboratory diagnosis of parasitosis: samples and sampling. Methods of direct detection of the causative agent. Methods of indirect proof of the cause. Serological methods in the detection of parasitosis.

### L6: Medically significant yeasts and molds

Cultivation and identification of medically significant fungi. Yeasts and molds. Structure, physiology and metabolism of fungi. Pathogenesis of fungal diseases. Skin mycoses. Systemic mycoses.

Opportunistic mycoses. Mycotoxicosis. Laboratory diagnostics of fungal diseases - breeding characteristics, breeding media and identification. Antifungals. Medically important fungi: genus Candida, genus Cryptocccus, genus Aspergillus, genus Penicillium, dermatophytes.

### L7: General virology

Structure and definition of the virus. Cultivation of the virus in cell culture, on a fertilized hen's egg and in an experimental animal. Preparation of cell cultures - primary and continuous cell cultures, diploid cell cultures. Cytopathic effects of the virus in cell culture. Viral inclusions. Viral vaccines types, production and application. Antiviral drugs: chemoprophylaxis and chemotherapy of viral diseases. highly effective antiretroviral therapy (HAART). Laboratory diagnostics of viral diseases: samples, direct virus detection, isolation, molecular diagnostics, serology.

### L8: Hepatitis viruses. Retroviruses. Prions

Structure, replication, pathogenesis, epidemiology, clinical syndromes, laboratory diagnostics, treatment, prevention and control of hepatitis viruses: HAV, HBV, HCV, HDV, HEV, VHG. Structure, replication, pathogenesis, epidemiology, clinical syndromes, laboratory diagnosis, treatment, prevention and control of retroviruses: human immunodeficiency virus 1 and 2 (HIV-1 and HIV-2), human leukemia / lymphoma virus of human T cells type 1 and 2 (HTLV -1, HTLV-2).

Prions - structure and physiology, pathology and pathogenesis, epidemiology, clinical syndromes, laboratory diagnostics, treatment, prevention and control of prion diseases.

### L9: Biosafety Conditions (BSL Level 2-4). Diagnosis of arenavirus and filovirus

Isolation of especially infectious agents in biosafety laboratories. Biosafety working conditions. Filoviruses and arenaviruses - characteristics, epidemiology, pathogenesis, clinical entities, laboratory diagnosis, treatment, prevention and control.

## L10: Serological methods in the diagnosis of viral infections. Molecular diagnostics of the virus. Hepatitis viruses. Hepresviruses.

Serology - double serum, antibody titer and titer dynamics: complement fixation reaction (CFR), neutralization test (NT), Mason and Paul-Bunell reaction. Enzyme-linked immunosorbent assay (EIA), indirect immunofluorescence (IFA), Western Blott (WB) - RIBA and immunoperoxidase test. Polymerase chain reaction - principle and diagnostic application. Diagnosis of herpes virus: EBV and infectious mononucleosis. CMV. XXV. Diagnosis of infections caused by hepatitis viruses: HAV, HBV, HCV, HDV, HEV. Diagnosis of HIV.

### Seminars (20 hours)

### S1: Disinfection and sterilization

Disinfection and sterilization: types of disinfectants and mechanism of action, selection of disinfectant. Sterilization procedures: types of sterilization, control of sterilization procedure.

### S2: Special bacteriology I

Description and characteristics of genera: Staphylococcus. Streptococcus. Neisseria, Haemophilus, Bordetella, Brucella. Breeding traits. The most important representatives and infectious diseases they cause. Samples for microbiological examination and microbiological laboratory diagnostics. Sensitivity to antibiotics.

Treatment, prevention and control of infections caused by the species of the above genera. **S3:Special bacteriology II** 

Description and characteristics of the family Enterobacteriaceae (genera: Escherichia, Salmonella, Shigella, Klebsiella, Proteus, Enterobacter, Yersinia, Serratia, Citrobacter, Providentia, Morganella), genus Vibrio, Campylobacter, Pseudomonas, Acinetobacter, Legionelium, Cory. Breeding traits. The most important representatives and infectious diseases they cause. Samples for microbiological examination and microbiological laboratory diagnostics. Sensitivity to antibiotics. Treatment, prevention and control of infections caused by the species of the above genera.

### S4: Special bacteriology III. Anaerobic bacteria. Genus Bacillus. Mycoplasmas. Chlamydia. Rickettsiae

Description and characteristics of sporogenic and asporogenic anaerobic bacteria. Breeding traits, anaerobic cultivation. Sporogenesis. The most important representatives and infectious diseases they cause. Intracellular bacteria. Arthropod-borne bacteria. Genus Bacillus: B. anthracis. Mycoplasmas: M. pneumoniae, M. hominis, Ureaplasma urealyticum; Chlamydia: C. trachomatis, C. pneumoniae, C. psittaci; rickettsiae: rickettsiae from the group of typhus, rickettsiae from the group of typhus. Samples for microbiological examination and microbiological laboratory diagnostics. Sensitivity to antibiotics. Treatment, prevention and control of infections caused by the species of the above genera

### S5: Mycoplasmas. Chlamydia. Rickettsiae

Mycoplasmas: M. pneumoniae, M. hominis, Ureaplasma urealyticum; Chlamydia: C. trachomatis, C. pneumoniae, C. psittaci; rickettsiae: rickettsiae from the group of typhus, rickettsiae from the group of typhus. Samples for microbiological examination and microbiological laboratory diagnostics. Sensitivity to antibiotics. Treatment, prevention and control of infections caused by the species of the above genera.

### S6: Protists of the digestive and genitourinary systems. Blood and tissue protists

Life cycle, epidemiology, laboratory diagnostics, treatment and prevention of blood and tissue protists: Trypanosoma spp ,. Leishmania spp. Genus Plasmodium (P. malariae, P. vivax, P. ovale and P. falciparum), filariasis; Toxoplasma gondii - serological diagnosis of toxoplasmosis. Arthropods - vectors for the transmission of blood and tissue protists. Life cycle, epidemiology, laboratory diagnostics of digestive and genitourinary system protists: Entamoeba histolytica, Giardia lamblia, Cryptosporidium parvum, Trichomonas vaginalis.

### **S7: Roundworms and flatworms**

Life cycle, epidemiology, laboratory diagnosis of roundworms: Ascaris lumbricoides, Trichuris trichiura, Ancylostoma duodenale, Strongyloides stercoralis, Enterobius vermicularis, Trichinella spiralis - MIFC for stool eggs finding, perianal imprint, trichinoscopy, coproculture. Serological diagnosis. Life cycle, epidemiology, laboratory diagnostics of flatworms: Taenia spp., Echinococcus granulosus, Hymenolepis nana, Fasciola hepatica, Shistosoma spp. - MIFC from stool, serological methods for the diagnosis of echinococcosis.

### S8: DNA viruses: family Adenoviridae, Papovaviridae, Poxviridae RNA viruses: family Orthomyxoviridae, Paramyxoviridae, Coronaviridae, Rhabdoviridae

Structure, replication, pathogenesis, epidemiology, clinical syndromes, laboratory diagnosis, treatment, prevention and control of adenovirus, papillomavirus, poliomavirus, orthomyxovirus, paramyxovirus, coronavirus, rhabdovirus. Influenza virus - "shift" and "drift". Pandemics and flu epidemics. Respiratory syncytial virus and human metapneumovirus. Measles virus. Mumps virus. Viruses that cause respiratory diseases. SARS virus. Rabies.

### S9: Viruses that cause congenital infections. Family Herpesviridae

Structure, replication, pathogenesis, epidemiology, clinical syndromes, laboratory diagnostics, treatment, prevention and control of viruses that cause congenital infections - families: Togaviridae (Rubivirus), Parvoviridae (parvovirus B-19), Hepresviridae, hepatitis viruses and HIV.

TORCH - serological testing of pregnant women. Structure, replication, pathogenesis,

epidemiology, clinical syndromes, laboratory diagnosis, treatment, prevention and control of Herpesviridae viruses: herpes simplex virus (VHS), varicella zoster virus (VZV), Epstein-Barr virus (EBV), Cytomegalovirus (CMV), human herpes virus 6,7,8 (HHV-6, HHV-7 and HHV-8).

S10: Viruses that cause gastrointestinal infections. Family *Picornaviridae* Arboviruses. *Arenaviridae*. *Filoviridae* 

Structure, replication, pathogenesis, epidemiology, clinical syndromes, laboratory diagnostics, treatment, prevention and control of viruses causing gastrointestinal infections: rotaviruses, adenoviruses, astroviruses, caliciviruses - norovirus. Viruses that enter the body's digestive system: picornaviruses. Structure, replication, pathogenesis, epidemiology, clinical syndromes, laboratory diagnosis, treatment, prevention and control of viruses transmitted by arthropods: family Togaviridae (Alphavirus), family Flaviviridae, family Bunyaviridae (genera: Bunyavirus, Phlebovirus, Nairova, Nairovirus). Structure, replication, pathogenesis, epidemiology, clinical syndromes, laboratory diagnostics, treatment, prevention and control of arenaviruses and filoviruses. Ebola virus and Marburg virus. Biosafety working conditions in the BSL 2-4 laboratory.

### **PRACTICES (40 hours)**

### P1: Microorganisms around us and on us. Microorganisms from the air

Bacterial micromorphology (basics of microscopy, bacterial forms). Native preparation. Staining in microbiology. Gram staining. Methylene blue staining. Normal human flora. Basics of bacterial cultivation in a microbiological laboratory. Pure and mixed bacterial culture. Basics of cultivation (media, incubation) identification of bacteria. Hygienic hand washing. Practical work: Taking fingerprints before and after hygienic hand washing. Installing the air sampling plate. Microscopy of native yeast preparation and staining of yeast preparation with methylene blue. Microscopy of colored preparations from the collection.

### P2: Bacterial susceptibility tests to antimicrobial chemotherapeutics

Basics of antibiotic susceptibility testing - disc diffusion method, determination of minimum inhibitory concentration (MIC) of antibiotics by microdilution, minimum bactericidal concentration (MBC) of antibiotics, determination of MIC agar by dilution, break point method and correlation with disc diffusion. Interpretation and reading of the prepared antibiogram. Bacterial resistance to antibiotics - examples of MRSA, VRE, ESBL, resistant Pseudomonas aeruginosa and Acinetobacter baumannii. Practical work: Preparation of antibiograms by disc diffusion method. Description of bacterial colonia on blood agar exposed to air in the clasroom and calculating the number of bacteria in 1 m3. Description of grown bacterial colonies on blood agar with fingerprints of the right hand before and after hand washing and disinfection.

### P3: Genus Staphylococcus. Wipe of nose, throat and nasopharynx

Staphylococci: basic clinical samples, description of micromorphology and macromorphology, basic tests for identification (catalase, coagulase, DNA-za), detection of MRSA. Antibiogram for susceptibility testing. Making swabs of the nose, throat and nasopharynx. Clinical indication for nasal swab sampling. Practical work: Fundamentals of microbiological diagnostics of staphylococci and description of staphylococcal colonies (S. aureus, S. epidermidis). Preparation of staphylococcal preparations (S. aureus, S. epidermidis) from the plate and Gram staining, performing and reading the catalase and coagulase test. Reading the DNA test for S. aureus. Taking nasopharyngeal and pharyngeal swabs. Reading antibiograms from the previous exercise (disc diffusion). Reading of a previously prepared test of susceptibility of S. aureus to vancomycin in the dilution method and determination of MIC and MBK.

### P4: Genus Streptococcus and genus Enterococcus

Serological groups of streptococci. Micro and macromorphology of streptococci and enterococci. Microbiological properties and tests for identification: S. pyogenes, S. agalactiae and S. pneumoniae. Cultivation characteristics of enterococci. Normal flora of the nasopharynx and pharynx. Clinical indication for nasal, nasopharyngeal and pharyngeal swabs. Clinically significant bacteria in nasopharyngeal and pharyngeal swabs. Other samples from resp. tract (sputum, tracheal aspirate, BAL). Practical work: Basics of microbiological diagnostics of streptococci: description of streptococcal colonies on blood agar, description of pneumococcal and enterococcal colonies on KA, production of streptococcal preparations from broth and Gram staining, performing and reading catalase test, microscopy of Gram stained streptococcal preparations. Microscopy of Gram-stained S.pneumoniae and methylene blue from a biological sample. Read the bile-esculin test and grow 6.5% NaCl broth for enterococci. Bacitracin test for group A streptococcus. Optochin test for pneumococcus. Read the implanted throat and nasopharyngeal swab.

### P5: Genus Haemophilus, genus Neisseria, genus Brucella

Micromorphology and macromorphology of hemophilus, neisseria and brucella. Microbiological properties and tests for the identification of H. influenzae, N. meningitidis. N. gonorrohoeae, Brucella spp. Nitrocefin test. Serological diagnostics in bacteriology and principles of agglutination and complement fixation reactions (CFR). Indirect diagnosis of brucellosis: Wright agglutination, CFR. Practical work: Description of colonies of Haemophilus spp. and saprophytic neisseria on Blood agar and chocolate agar. Observation of the satellite phenomenon. Making of neisseria preparations and Gram staining. Microscopy of Gram-stained hemophilus preparations. Microscopy of Gram-stained preparations of N. meningitidis from cerebrospinal fluid sediment. Microscopy of N. gonorrhoeae from a methylene blue urethral swab. Development of oxidase test and nitrocefin test. Gram-stained microscopy of Brucella preparations. Readings of serological reactions for the detection of brucellosis: CFR and Wright agglutination.

## P6: Identification of the most common causes of urinary tract infections. Family *Enterobactreiaceae*

Biochemical tests to identify members of the Enterobacteriaceae family. Micromorphology and macromorphology of enterobacteria - description of colonies and preparations. Antibiogram and antibiotic resistance: multidrug-resistant enterobacteria (beta lactamases, ESBL). E.coli, Klebsiella, Serratia, Proteus, Morganella, Enterobacter - clinical samples for isolation, atb. Microbiological treatment of urine. Practical part: Making and microscopy of different types of Gram-stained enterobacteria. Microscopy of Klebsiella indigo stained preparation. Describe colonies of E. coli, Klebsiellae on blood agar and differential media. Description of Proteus sp. on blood agar. Read the biochemical sequence for bacteria E.coli, Klebsiella, Proteus (demonstration). Urine seeding. Reading Uriselect agar sown with urine from routine. Reading antibiogram and double synergistic test for ESBL detection.

## P7: Identification of the most common causes of intestinal infections. Enterobacteriaceae. Vibrio. Campylobacter. Helicobacter

Microbiological treatment of stool (normal flora and pathogens). Basic selective media and biochemical tests for the identification of Salmonella, Shigella, Yersinia, Campylobacter, Vibrio cholerae. Micromorphology and macromorphology of the causative agent of gastrointestinal tract infections - description of colonies and preparations. Serotyping of salmonella and shigella. Widal agglutination . Helicobacter: lab.diagnosis, Sensitivity of the causative agent of the digestive tract to antibiotics. Practical work: Describe colonies of salmonella, shigella, Escherichia on XLD and SS agar and Yersinia on SS agar. Read the biochemical sequence for salmonella, shigella and yersinia (demonstration). Serotyping of enteropathogenic E. coli. Read Widal's reaction. Microscopy of Gram-stained Campylobacter preparations. Description of campylobacter and vibrio colonies on selective nutrient media. Microscopy of gastric biopsy preparations (Helicobacter pylori). Seeding of stools on selective and differential substrates.

## P8: Identification of pseudomonas, corynebacteria, listeria, legionella and gardnerella. Biological control of sterilization

Pseudomonas aeruginosa, Co. diphteriae, L. monocytogenes. Legionella pneumophila, Gardnerella vaginalis: clinical specimens, description of micromorphology and macromorphology, identification tests. Coloring according to Lubinski. Rapid test to detect legionella antigen in urine. Susceptibility

of these genera to antibiotics. In vitro and in vivo synergism and antagonism of antibiotics. Practical part: Microscopy of Gram-stained preparations of Pseudomonas aeruginosa, description of colonies on blood and plain agar, biochemical identification (demonstration) and disc diffusion reading for Pseudomonas aeruginosa. Preparation of oxidase test. Description of diphtheroid colonies. Microscopy of Gram and Lubin-stained preparations of Corynebacterium diphtheriae and diphtheroids. Microscopy of L. monocytogenes and description of blood agar colonies. Microsporing of G. vaginalis preparation from Gram-stained vaginal swab. Read the antagonism and synergism of the tested antibiotics.

**P9: Identification of anaerobic bacteria. Sporogenic bacteria. Genus** *Clostridium.***Genus** *Bacillus* Cultivation and identification of sporogenic anaerobic bacteria. Clinical samples for isolation of anaerobic bacteria. Mixed infections. Sensitivity testing. Sporogenic bacteria. Coloring by Fulton. Micromorphology and macromorphology of bacterial species: Genus Clostridium - laboratory diagnostics C..tetani, C. botulinum, C. perfringens. Genus Bacillus - B. anthracis. Practical work: B.anthracis: microscopy of a preparation stained with methylene blue peritoneal mouse exudate. Prepare, stain by Gram, microscopy and paint B.subtilis. Clostridium sp. - Gram and Fulton stained microscopy. Description of colonies of Bacillus subtilis and Clostridium spp. on Columbia agar. **P10: Identification of asporogenic anaerobic bacteria** 

### Asporogenic anaerobes: Bacteroides sp, Prevotella, Peptostreptococcus. Genus Actynomices. Micromorphology and macromorphology of asporogenic anaerobes. Laboratory diagnosis and isolation of anaerobes. Susceptibility testing of anaerobic bacteria. Commercial systems for anaerobic identification. Processing of primarily sterile materials (blood culture, cerebrospinal fluid, biopsies). Practical part: Microscopy of Gram-stained preparations of asporogenic anaerobic bacteria (Bacteroides, Prevotella, Peptostreptococcus). Description of colonies of Bacteroides sp. and Actinomyces viscosus on Columbia agar. Reading of a commercial identification system (demonstration). Reading of a commercial system for testing the susceptibility of anaerobic bacteria (demonstration). Planting of blood cultures and abscesses - aerobic and anaerobic. Microscopy of direct preparations from primarily sterile samples.

### P11: Mycobacteria and nocardia

Mycobacteria: Mycobacterium tuberculosis - micromorphology and macromorphology of mycobacteria, laboratory diagnostics (cultivation, susceptibility testing, rapid diagnostic methods, clinical specimens, antituberculosis susceptibility testing. Practical work: Describe the colonies of Mycobacterium tuberculosis on a Lowenstein-Jensen substrate. Stain, microscopy and paint a Ziehl-Neelsen-stained sputum preparation. Microscopy and paint a Kinyon-stained nocardium preparation with color. Read a previously prepared test for M.tuberculosis resistance to antituberculotic drugs.

### P12: Basic principles of serological reactions. Spirochete.

Basic principles of serological reactions. Genus Treponema: T.pallidum: laboratory diagnostics (non-treponemal VDRL, RPR and treponemal ITFA, TPHA tests), atb. genus Borrelia: lab. diagnostics (direct diagnosis of B. reccurentis, ELISA of B. burgdorferi), atb. Leptospire - laboratory diagnostics, atb. Practical part: Read and draw the result of the VDRL test. Determine the titer of acute and convalescent serum in the TPHA test for T. pallidum. Read the ELISA test result on B.burgdorferi Preparation and staining of gingival carbol with fuchsin.

### P13: Mycoplasmas. Chlamydia. Rickettsiae

Mycoplasmas: laboratory diagnosis of M. pneumoniae, clinical samples. laboratory diagnostic M. hominis and Ureaplasma urealyticum, clinical specimens, antibiogram. Laboratory diagnosis of chlamydia and rickettsia. Microbiological diagnosis of bacterial sexually transmitted diseases. Practical work: microscopy of C. trachomatis preparations and rickettsiae from the collection of premade preparations. Microscopy of mycoplasmas and ureaplasmas on PPLO agar. Readings of

CFR and cold agglutination for M. pneumoniae. Weil-Felix agglutination reading for rickettsia diagnosis.

### P14: Medically significant fungi I: Yeast cultivation and identification

Yeast cultivation and identification. Yeast structure. Fermentation and assimilation test. Genus Candida: C. albicans and Candida sp., clinical samples, sensitivity to antifungals. Genus Cryptococcos - C. neoformans: laboratory diagnostics and clinical samples. Practical work: Description of colonies of Candida albicans, Candida krusei. Geotrichum spp. Grown on Saburaud agar. Microscopy of native yeast preparations. Microscopy of a microscopic preparation of Gramstained yeasts. Microscopy of the C. albicans germination test. Description of Cryptococcus colonies on solid medium, microscopy of cryptococcal preparations, cryptococcal shower preparation.

P15: Medically significant fungi II: Cultivation and identification of molds. *Pneumocystis jirovecii* Mold breeding and identification. Mold structure. Penicillium, Aspergillus, Mucor, Rhisopus clinical specimens, interpretation of mold isolates depending on the specimen, susceptibility testing for antifungals. Serodiagnosis of systemic mycoses. Identification of P. jirovecii. Pneumocystis jirovecii - laboratory diagnosis (preparation, DFA, PCR), treatment. Practical work: Description of the colonies of Aspergillus, Mucor and Penicillium on Sabouraud agar. Microscopy of preparations with lactophenol Aspergillus, Mucor and Penicillium. microscopy of P. jirovecii preparations.

### P16: Blood and tissue protists I

Trypanosoma spp. - life cycle, epidemiology, laboratory diagnostics: blood smear and thick drop stained by Giemsa-Romanowski (GR) method. Toxoplasma gondii - epidemiology, life cycle, prevention and treatment, lab. diagnostic. Practical part: Microscopy of Trypanosoma spp. From GR-stained blood smear. Microscopy of T. gondii stained by GR. Serological diagnosis of toxoplasmosis. Leishmania spp. - diagnosis of leishmaniasis- direct bone marrow preparation stained with GR. Cultivation (NNN media), preparation and staining of preparations according to Giemsa-Romanowski.

### P17: Blood and tissue protists II. Genus Plasmodium. Microfiralia

Genus Plasmodium: life cycle, epidemiology, clinical disease, laboratory diagnosis, treatment, prevention and control of malaria. P. malariae, P. vivax, P. ovale and P. falciparum. Filariasis - life cycle, epidemiology, clinical picture, laboratory diagnostics. Practical work: microscopy of blood smears and thick drops stained according to Giemsa-Romanowski and recognition of developmental and diagnostic forms of species of the genus Plasmodium. Microscopy of microfilariae from a blood smear stained according to Giemsa-Romanowski.

### P18: Protists of the digestive and genitourinary systems

Life cycle, epidemiology, laboratory diagnostics of digestive and genitourinary system protists: Entamoeba histolytica, Giardia lamblia, Cryptosporidium parvum, Trichomonas vaginalis. Practical work: Microscopy of native and stained preparation of stool and secretions of the genitourinary system, MIFC method for finding cysts: Entamoeba histolytica, Giardia lamblia, Cryptosporidium parvum, Trichomonas vaginalis. Cultivation of protists. Entamoeba moshkowskii - a display of amoeboid movement. Other protists in the digestive system - microscopy preparations (MIFC): Entamoeba coli, Blastocystis hominis, Iodamoeba butschlii.

### P19: Identification of eggs, larvae and adult roundworms

Life cycle, epidemiology, laboratory diagnosis of roundworms: Ascaris lumbricoides, Trichuris trichiura, Ancylostoma duodenale, Strongyloides stercoralis, Enterobius vermicularis, Trichinella spiralis - MIFC for stool egg finding, perianal imprint, coproculture, trichinoscopy . Serological diagnosis. Practical part: microscopy of stool preparations (MIFC): Ascaris lumbricoides, Trichuris trichiura, Ancylostoma duodenale, Strongyloides stercoralis; Enterobius vermicularis - microscopy

of the Graham perianal imprint; Trichinella spiralis - microscopy of preparations of infected meat with Trichinella larvae.

Identification of parasite adults from the collection of durable preparations.

### P20: Identification of eggs, larvae and adult flatworms

Life cycle, epidemiology, laboratory diagnostics of flatworms: Taenia spp., Echinococcus granulosus, Hymenolepis nana, Fasciola hepatica, Shistosoma spp. Practical part: microscopy of stool preparations (MIFC): Taenia spp, Fasciola hepatica, Shistostoma spp .. Microscopy of the native preparation from the contents of hydatid cyst (Echinoocccus granulosus). Identification of adult parasites from the collection of durable preparations.

# P21: Taking clinical material and methods of virological diagnosis. Hemagglutination and inhibition of hemagglutination. Diagnosis of orthomyxovirus,

### paramyxovirus and coronavirus

Taking clinical material for virological tests. Cultivation of viruses in cell culture. Cytopathic effects of the virus in cell culture - microscopy of the preparation. Breeding of the virus in a fertilized hen's egg and an experimental animal. Rapid tests for the detection of virus antigens in a clinical sample - latex and immunochromatogenic tests: Adenolex, Directigen RSV, Rotalex. Diagnosis of adenovirus, papovavirus and rhabdovirus. Diagnosis of picornavirus, reovirus and togavirus. Practical work: performing rapid latex tests and immunochromatographic tests for detection of viral agents from the respiratory and digestive systems. Microscopy of virus-infected cell culture preparations and identification of CPE. Haemagglutinins and haemagglutination (HA), haemadsorption (HAD), haemagglutination inhibition (IH) and haemagorption inhibition for virus detection and identification. Determination (titer) of hemagglutinin antigen and serological reaction (IH) for antibody detection. Laboratory diagnosis of infection: influenza virus, parainfluenza virus, respiratory syncytial virus, human metapneumo virus, measles virus, mumps virus, coronaviruses. Practical work: Reading tests: hemagglutination test (HA) for mumps virus, hemagglutination inhibition test (IH) for typing isolated virus, reading hemagglutination reaction -HA (titration of isolated virus), reading IH reaction (titer of specific haemagglutination pairs in patients with from influenza) and complement fixation (CFR) reactions.

### P22: Arthropods. Diagnosis of flavivirus and bunyavirus

Arbo (arthropod borne) viruses and their arthropod vectors. Arthropods as vectors of other infectious diseases. Flaviviruses and bunyaviruses - characteristics, epidemiology, pathogenesis, clinical entities, laboratory diagnostics, treatment, prevention and control. Practical part: Microscopy of preparations and recognition of arthropods. Repetition of preparations from bacteriology, mycology and parasitology.

### Mode of teaching

Lectures; Seminars; Exercises

### **Student obligations**

Attendance at all forms of classes is mandatory, and the student must access all knowledge tests. A student may justifiably miss 30% of each form of instruction. Unfinished exercise must be colloquial. **Monitoring student work (alignment of learning outcomes, teaching methods and grading)** 

Teaching activity	ГСТС	Learning	Student estivity	Assessment	Grade	points
Teaching activity	ECTS	outcome	Student activity	methods	Min.	Max.
Class attendance	0,4	1-5	Attendance in class	Records	3	5

Total	8					100
Oral exam	1,6	1-5	Continuous studying during classes	Oral exam	1	20
Written exam	0,8	1-5	Continuous studying during classes	Written exam	6	10
Partial exams	3,2	1-5	Continuous studying during classes	Partial written exam	10	20
Seminar paper	0,4	1-5	Preparation of seminar paper	Presentation of seminar paper	1	5
Practical work (exercises)	1,6	1-5	Studying for the practical exam and class attendance	Practical examination	10+3	15+5

Student work is evaluated during classes and at the final exam. Students are evaluated numerically and descriptively: insufficient (1), sufficient (2), good (3), very good (4), excellent (5). During the tour, the student will be able to collect a maximum of 100 points. Students can gain a maximum of 70 points during classes through various forms of activities (see Table 1) and continuous testing, and a maximum of 30 points in the final exam.

Continuous testing consists of 2 partial tests by which the student can achieve a maximum of 20 points in each depending on the success of the test (Table 2): I. partial test: bacteriology and mycology and II. partial test: virology and parasitology. Students are entitled to one correction (rewriting) of the partial exam before the exam period for which it is applied.

The final part of the exam consists of a written and an oral part. The student must meet more than 60% on the written part of the exam to earn grades. The final grade is the sum of grade points achieved during classes and at the final exam.

Table 1. Evaluation of student teaching obligations

	EVALUATION	MAX. NUMBER OF POINTS
	Partial test 1	20
Partial tests	Partial test II	20
	Total	40
Practical work/laboratory	Final colloquium - practical part of the exam	15
excercises	Exercise completed	5
excercises	Total	20
	Active participation	5
Seminar	Total	5
	Attendance	5
Lectures	Total	5
	TOTAL	70
Final exam	Written part	10
	Oral part	20
	Total	30
TOTAL		100

Table 2. Evaluation of partial exams

Percentage of correctly solved tasks (%)	Grade points	
50,00-54,99	10	
55,00-59,99	11	
60,00-64,99	12	
65,00-69,99	13	
70,00-74,99	14	
75,00-79,99	15	
80,00-82,49	16	
82,50-84,99	17	
85,00-87,49	18	
87,50-89,99	19	
90,00-100	20	

### **Class attendance**

The student must attend a minimum of 70% of all forms of teaching: exercises, seminars and lectures and access all forms of knowledge testing. A student who misses seminars and / or exercises more than 30% of classes up to a total of 50% of all forms of classes must make up for missed material by colloquium.

### Practical work (exercises)

The final colloquium - practical part of the exam consists of five practical tasks which include microscopy of preparations (six pcs.) In the field of bacteriology (3 pcs.), Parasitology (2 pcs.), Mycology (1 pc.), Two cultures on nutrient agar (one bacterial and one fungal), antibiogram readings,

serological reaction readings and identification of parasite adults from the collection of durable preparations. The student must meet at least 60% on the practical part of the exam in order to be able to take the final exam. Students can earn a maximum of 15 marks on the practical exam (see Table 3) depending on the percentage of correctly solved tasks. The final colloquium is organized after the end of classes and within each exam period as a practical part of the exam.

Table 3. Evaluation of the final colloquium - practical exam

Percentage of correctly solved tasks (%)	Grade points
60,00-69,99	10
70,00-79,99	11
80-89,99	12
90,00-94,99	13
95-100	15

### Seminars

During the course, the student can collect a maximum of 5 grade points through active preparation and presentation of seminars, which is mandatory according to the following criteria:

- 1 grade point: the seminar meets the minimum criteria
- 2 grade points: average answer with noticeable errors
- 3 grade points: average answer with minor errors
- 4 grade points: very good answer with slight errors
- 5 grade points: exceptional answer

### Lectures

By attending lectures, a student can achieve 3-5 grade points according to the following scheme: participation in 60-79.99% of lectures is evaluated with 3 grade points, 80-89.99% 4 grade points, 90-100% 5 grade points.

### **Final exam**

A student who has duly completed all forms of teaching has acquired the right to sign and take the final exam. The final exam is mandatory and consists of a written and an oral part. During the final exam, the student can receive a maximum of 30 points, of which 10 points in the written part and 20 in the oral part.

The written part of the final exam consists of questions with five answers offered, of which only one is correct. The minimum criterion for gaining grade points is 60% of correctly resolved questions. Points earned in the written part of the final exam are converted into grade points according to the criteria listed in Table 4. Points earned in the final exam are counted as points earned during classes. In case the student does not meet the minimum criteria in the first exam term, he / she takes the

final exam again in the next exam term, as well as in case he / she does not meet the oral part of the exam.

Table 4. Evaluation of the written part of the final exam

Percentage of correctly solved tasks (%)	Grade points
50,00-99,99	6
60,00-69,99	7
70,00-79,99	8
80-89,99	9
90,00-94,99	10

The oral part of the exam consists of five questions divided into areas: general microbiology, special bacteriology, special virology, special parasitology, special mycology that the student draws.

Evaluation of the oral part of the final exam:

1-5 grade points: the answer meets the minimum criteria6-10 grade points: average answer with noticeable errors11-15 grade points: very good answer with slight errors16-20 grade points: exceptional answer

Forming the final grade:

Grades earned in the final exam are added to the points earned during the course. Assessment in the ECTS system is performed by absolute distribution, ie on the basis of final achievement and is compared with the numerical system as follows:

- A excellent (5): 90-100 grade
- B very good (4): 80-89.99 grade points
- C good (3): 70-79.99 grade points
- D sufficient (2): 60-69.99 grade points
- E sufficient (2): 50-59.99 grade points

Required reading (available in the library and through other media)				
	Number of	Availability		
	copies in the	through other media		
	library			
1. Kalenić i sur. Medicinska mikrobiologija. Drugo,	5			
izmijenjeno i obnovljeno izdanje. Medicinska naklada,				
Zagreb, 2019. (udžbenik)				

### Additional reading

Jawetz, Melnick, & Adelberg's Medical Microbiology, 25nd edition. Brooks GF, Carroll KC, Butel JS, Morse SA, Mietzner TA editors. Lange Medical Books/McGraw-Hill: New York, Chicago, San Francisco, Lisboa, London, Madrid, Mexico City, Milan, New Delhi, San Juan, Seoul, Singapore, Sydney, Toronto, 2019

### **Course evaluation procedures**

The quality and success of the course is monitored through an anonymous student survey that will include assessment of students on the quality of various forms of teaching in the course and on teachers conducted by the Department of Microbiology and Parasitology, Faculty of Medicine Osijek. Also, the output knowledge will be monitored through the success of the students at the end of the course. During the classes, records of student attendance at lectures, seminars and exercises will be used.

In addition to the above, the quality of the teaching process is also monitored by conducting a unique university survey among students for teacher evaluation determined by the Senate of the J. J. Strossmayer University in Osijek.

### Note /Other

E-learning is not included in the norm of subject hours, but it is used in teaching and contains links to various pages, video and audio materials available on the website.