

Ministry of Education and Science of Ukraine

Ukrainian Medical Stomatological Academy
Microbiology, Virology and Immunology Chair

Microbiology, Virology and Immunology

***TRAINING MANUAL
for foreign faculty students
specialty “Medicine”***

Part 3

III year _____group

Student:

Poltava 2020

Training manual on Microbiology, Virology and Immunology for students of faculty of foreign students training specialty “Dentistry” was created by:

1. LOBAN’ Galina Andriivna - the head of the department, MD, professor
2. ZVYAGOLSKA Irina Mykolaivna – Ph.D., associate professor
3. POLYANSKA Valentina Pavlivna – Ph.D., associate professor
4. FEDORCHENKO Vira Ivanivna - Ph.D., associate professor
5. ANANIEVA Maiia Mykolaivna - Ph.D., associate professor
6. HANCHO Olha Valeriivna – Ph.D., associate professor
7. FAUSTOVA Mariia Oleksiivna - Ph.D., lecturer

The training manual is an intellectual property and protected by the Law of Ukraine "On Copyright and Related Rights". It can not be copied and reproduced in full or in parts without the written permission of the authors, except for the handwritten form.

STAFF OF THE DEPARTMENT

1. LOBAN’ Galina Andriivna - the head of the department, MD, professor
2. ZVYAGOLSKA Irina Mykolaivna – Ph.D., associate professor
3. POLYANSKA Valentina Pavlivna – vice principal of the department, Ph.D., associate professor
4. FEDORCHENKO Vira Ivanivna - Ph.D., associate professor
5. ANANIEVA Maiia Mykolaivna - Ph.D., associate professor
6. HANCHO Olha Valeriivna – Ph.D., associate professor
7. HORDIENKO Liudmyla Petrivna - Ph.D., associate professor
8. BOBROVA Nellia Olexandrivna – Ph.D., lecturer
9. ZACHEPYLO Svitlana Viktorivna – Ph.D., lecturer
10. FAUSTOVA Mariia Oleksiivna – Ph.D., lecturer
11. CHUMAK Yuliia Viktorivna – lecturer
12. SOTNYCHENKO Svitlana Mykolaivna – senior laboratory assistant
13. KSONZ Angelina Olahivna - laboratory assistant
14. DUDKA Tetiana Andriivna – support worker
15. HORODCHANINA Lyudmila Volodymyrivna – support worker

Literature for self-directed work:

Basic

1. Medical microbiology, virology and immunology - T. V. Andrianova, V. V. Bobyr, V. V. Danyleichenko, etc. / Ed. by V. P. Shyrobokov/ - Vinnytsia: Nova Knyha, 2019. - 744 p.
2. Cappuccino G. Microbiology: A Laboratory Manual, Global Edition, 11th Edition / G.Cappuccino, Chad T. Welsh. - Pearson Higher Ed USA, 2017. - 560 p.
3. Medical Microbiology 27 E (Lange) / K. C. Carroll, S. Morse, T. Mietzner [et al.]. - McGraw-Hill Education, 2016. - 864 p.
4. Murray P.R. Medical Microbiology 8th Edition / P.R. Murray, K.S. Rosenthal, M.A. Pfaller. - Elsevier, 2016. - 848 p.
5. Murray P.R. Basic Medical Microbiology 1st Edition / P.R. Murray. - Elsevier, 2018. - 240 p.
6. Medical Microbiology, International Edition, 19 Ed / M.R. Barer, W. Irving, A. Swann [et al.]. - Elsevier, 2018. - 760 p.
7. Engelkirk P.G. Burton's Microbiology for the Health Sciences / P.G. Engelkirk, J. Duben-Engelkirk, R. Fader. - Wolters Kluwer Health, 2015. - 488 p.
8. Hawley L. Microbiology and Immunology (Board Review Series) Sixth Edition / L. Hawley, R.J. Ziegler, B. L. Clarke. - Lippincott Williams & Wilkins, 2014. - 320 p.
9. General Medical Microbiology, Virology and Immunology. Part I. Manual for practical lessons / Comp. by Loban G.A., Hanch O.V. - Poltava, 2005. -153 p.
10. General Medical Microbiology, Virology and Immunology. Part II. Manual for practical lessons/ Comp. by Loban G.A., Hanch O.V. - Poltava, 2007.- 104 p.
11. Pathogenic cocci. Gramnegative intestinal pathogens. Manual for practical lessons /Composed by Hanch O.V. - Poltava, UMSA, 2006. - 113 p.
12. Oral cavity flora. Manual of Microbiology, Virology and Immunology for dental faculty students/ Composed by Hanch O.V.- 2010, Poltava. - 88 p.

Additional

1. Ananthanarayan R., Paniker C.K. Textbook of Microbiology. - International edition. - 2003. - 612 p.
2. MIMs' Medical Microbiology and Immunology 6th / Richard Goering, Hazel Dockrell, Mark Zuckerman [et al.]. - Elsevier, 2019. - 568 p.
3. Harriott M. Microbiology in Your Pocket: Quick Pathogen Review 1st Edition / M. Harriott. - Thieme, 2018. - 330 p.
4. Medical Microbiology / Patrick R. Murray [et al.]. - 4th ed. - An Affiliate of Elsevier Science, 2002. - 826 p.
5. Marsh, P.D. and Martin, M.V. Oral Microbiology, 5th edn. Elsevier: Amsterdam, The Netherlands, 2009. - 300 p.
6. Richard J. Lamont, Robert Burne, Marilyn Lantz, Donald Leblanc. Oral Microbiology and Immunology. Wiley-Blackwell, 2006. - 276 p.

Information resources:

1. President of Ukraine, official website <http://www.president.gov.ua/>
2. Verkhovna Rada of Ukraine, official web portal <http://www.rada.gov.ua/>
3. Government Portal, official web portal <http://www.kmu.gov.ua/>
4. Ministry of Education and Science <http://www.mon.gov.ua/>
5. Ministry of Ecology and Natural Resources of Ukraine <http://www.menr.gov.ua/>
6. The State Emergency Service of Ukraine <http://www.dsns.gov.ua/>
7. National Security and Defense Council of Ukraine <http://www.rnbo.gov.ua/>
8. Permanent Mission of Ukraine to the United Nations <http://ukraineun.org/>
9. North Atlantic Treaty Organization (NATO) <http://www.nato.int/>
10. World Health Organization <http://www.who.int/en/>
11. Microbiology and immunology on-line <http://www.microbiologybook.org/>
12. On-line microbiology note <http://www.microbiologyinfo.com/>
13. Centers for diseases control and prevention www.cdc.gov

Date: _____

Practical lesson № 1

Topic: Staphylococci and Streptococci. Microbiological diagnosis of diseases caused by Staphylococci and Streptococci

Self-assessment tasks

a) Questions list: theoretical issues:

1. Evolution of the coccid group of bacteria and their general characteristics.
2. Classification. Biological properties of staphylococci and streptococci.
Factors of pathogenicity of staphylococci and streptococci.
3. Staphylococci and streptococci as causes of human pathology.
Epidemiology and pathogenesis of infections caused by them.
4. The role of streptococcus group A in aetiology and pathogenesis of erysipelas, scarlet fever, and rheumatism. Scarlatinous stomatitis.
5. Immunity and its features in conditions of staphylococcal and streptococcal infection.
6. Prevention and treatment of staphylococcal and streptococcal infections.
Drugs for specific prevention and therapy.
7. Methods of microbiological diagnosis of staphylococcal and streptococcal diseases.
8. Age features of children's susceptibility to staphylococcal infection.
9. Local and generalized forms of staphylococcal infection in children.
10. Streptococcal infections in children. Localized and generalized forms.
11. Pathogenetic role of streptococci as the cause of scarlet fever, peculiarities of immunity in children.
12. The significance of Streptococcus pneumoniae in pediatric pathology.
Localized and generalized forms.
13. The material for investigating pneumococcal infection and its taking in children of different age groups.

b) The list of practical skills and abilities:

1. Compliance with the rules of the anti-epidemic regime and safety in the microbiological laboratory.
2. Preparation of preparations for microscopic examination of pathological material (manure, sputum).
3. Coloring of drugs by complex methods (by Gram, Burri-Gins).
4. Microscopy of preparations in a light microscope using an immersion lens.
5. Differentiation of microorganisms by morphological and tinctorial characteristics.
6. Selection of pure cultures of aerobic bacteria.
7. Sowing the test material to the loop on a solid nutrient medium.
8. Determination of the sensitivity of isolated bacterial cultures to antibiotics.
9. Filling the forms of directions of the investigated material in the laboratory for bacteriological research.

Practical lesson protocol

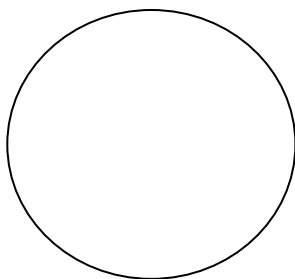
Practical assignments

Task № 1. Make a macroscopic and microscopic examination of the isolated colonies on blood- and yolk salt meat peptone agars (inoculation of the studied material from a patient with suspected furuncle of the left wing of the nose).

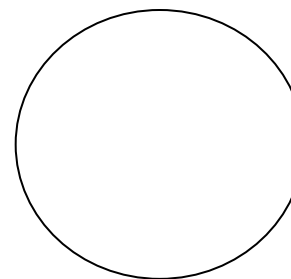
Cultural properties	BloodMPA	Yolk salt MPA
Research in transmitted light		
Size (diameter)		
Form		
Degree of transparency		
Research in reflected light		
Pigmentation		
The pattern of the surface		
Position on a media		
Microscopic research		
The pattern of the margin		
Structure		
Other properties		
Consistency		
Hemolytic activity		
Lecithinase activity		

Task № 2. Make a smear from the isolated colonies, and stain it with the Gram method. Microscopy, draw.

Blood MPA



Yolk-salt MPA

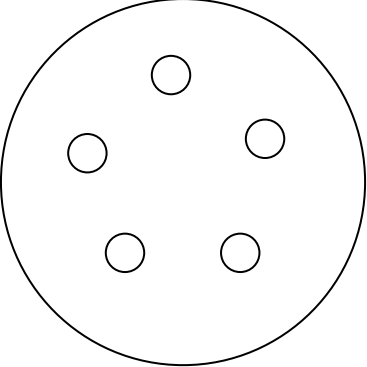


Denote the morphological and tinctorial properties of the detected microorganisms

Task № 3. Observe and evaluate the enzymatic properties of the selected pure culture of bacteria from the pus of a patient with an abscess in the submandibular area. Fill in the result in the table.

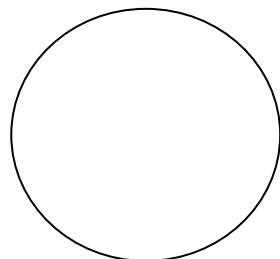
Index Species name	Glucose	Lactose	Maltose	Saccharose	Mannitol	Milk	MPG	H ₂ S	Indole

Task №4. Observe the antibiotic susceptibility test, which has done with a selected strain of staphylococcus. Mark the areas in the picture without growth. Fill in the result in the table. Conclude.

	№ p/p	The names of antibiotics	Diameter of the areas with the absence of the growth (mm)	Susceptibility
	1			
	2			
	3			
	4			
	5			
Conclusion: _____ _____				

Task № 5. Identify the pure culture of bacteria selected from submandibular abscess pus of the patient by morphological, tinctorial, cultural and enzymatic properties. Conclusion:

Task №6. Make a smear from the hemoculture isolated from the patient's blood with suspected sepsis and stain with the Gram method.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №7. Fill in the general microbiology request to a bacteriological laboratory for pus from the nose left wing furuncle of a patient.

GENERAL MICROBIOLOGY REQUEST FORM

PATIENT NAME:			DOB / /			OLS LAB NO.		
LAST	FIRST	MI	MO	DAY	YR		AGE	
PATIENT ADDRESS:					SEX: ☐ M ☐ F	OLS DATE RECEIVED		
COUNTY OF ORIGIN OF PATIENT:								
PROVIDER ID NO.: (optional)		DATE OF COLLECTION:				OLS TIME RECEIVED: ☐ AM ☐ PM		
TO BE FILLED IN BY PROVIDER								
EXAMINATION REQUESTED: ☐ BACTERIOLOGY ☐ ENTERIC BACTERIOLOGY ☐ PERTUSSIS ☐ PARASITOLOGY ☐ OTHER _____								
BACTERIOLOGY					PARASITOLOGY			
SOURCE: ☐ STOOL ☐ BLOODY STOOL ☐ URINE ☐ BLOOD ☐ OTHER _____					☐ FECES IN FORMALIN			
					☐ FECES IN PVA			
SUSPECTED ORGANISM:					☐ CELLULOSE TAPE MOUNT			
☐ PERTUSSIS CULTURE SOURCE: _____					☐ OTHER (SPECIFY) _____			
FOR LAB RESULTS ONLY								
☐ BACTERIOLOGY ID:					☐ PARASITES: ☐ NOT FOUND ☐ FOUND _____ _____			
☐ ENTERIC PATHOGENS: ☐ NOT FOUND								
☐ FOUND _____								
☐ PERTUSSIS ☐ ISOLATED FROM CULTURE ☐ NOT ISOLATED FROM CULTURE								
MAIL REPORT TO:								
ADDRESS:								
CITY:		ZIP	TELEPHONE:		FAX:			

Task № 8. Characterize the immunobiological preparations for specific prophylaxis and treatment of infections caused by staphylococci and streptococci.

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 2

Topic: Meningococci and gonococci. Microbiological diagnosis of diseases caused by meningococci and gonococci

Self-assessment tasks

a) Questions list: theoretical issues:

1. Biological properties of pathogenic Neisseria. Classification.
2. Biological properties and classification of meningococci.
3. Pathogenicity factors of meningococci.
4. Epidemiology and pathogenesis of meningococcal diseases. Carrier state.
5. Immunity to meningococcal diseases.
6. Methods of microbiological diagnosis of meningococcal diseases and carrier state.
7. Prophylaxis and therapy of meningococcal diseases.
8. Differentiation of meningococci and gram-negative diplococci of the nasopharynx.
9. Biological properties of gonococci, their variability. Pathogenicity for humans. Epidemiology and pathogenesis of gonorrhoea. Acute and chronic gonorrhoea.
10. Immunity in gonorrhoea.
11. Methods of gonorrhoea microbiological diagnosis.
12. Prophylaxis and therapy of gonorrhoea and gonococcal ophthalmia neonatorum.
13. Age characteristics of susceptibility in children to meningococcal infection.
14. Localized and generalized forms of meningococcal infection in children.
15. Gonococcal ophthalmia neonatorum. Epidemiology, pathogenesis, prophylaxis.

b) The list of practical skills and abilities:

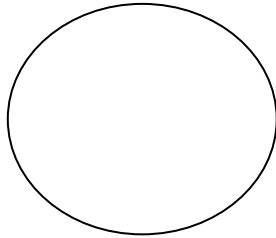
1. Microscopy of preparations in a light microscope using an immiscible lens.
2. Coloring of preparations by simple and complex methods (aqueous solution of methylene blue, for Gram).
3. Differentiation of microorganisms by morphological and tinctorial characteristics.
4. Selection of pure cultures of aerobic bacteria, identification of selected cultures.
5. Ability to record and evaluate the results of serological reactions (complement fixation reaction).

Practical lesson protocol

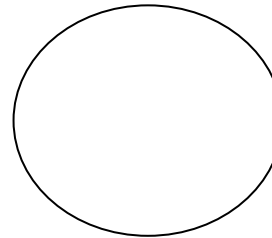
Practical assignments

Task №1. Microscopy and draw the smears from the spinal liquid stained by methylene blue and Gram method.

Staining with methylen blue



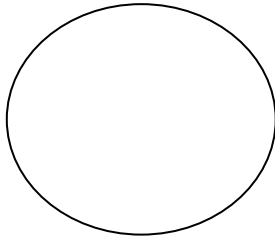
Staining by Gram



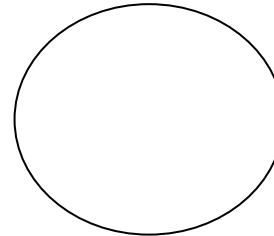
Denote the morphological and tinctorial properties of the detected microorganisms.

Task № 2. Microscopy and draw the smears of urethral pus stained by methylene blue and Gram method.

Staining by methylen blue



Staining by Gram



Denote the morphological and tinctorial properties of the detected microorganisms

Conclusion (denote the microscopic signs of preparations that are the base for the diagnosis: acute gonorrhoea)

Task № 3. Make the complement fixation test (CFT) with the sera of a patient and gonococcal antigen for confirmation of diagnosis: chronic gonorrhoea.

Ingredients (ml) Number of tubes	Investigated serum (dilution 1:10)	Antigen	Complement	Saline solution	37°C – 1 hour	Hemolytic system		37°C – 1 hour	Results	
						Anti-sRBC antibodies	Sheep red blood cells (sRBCs)		Hemolysis	CFT
1 (research)	0,5	0,5	0,5	-		0,5	0,5			
2 (control of serum)	0,5	-	0,5	0,5		0,5	0,5			
3 (control of antigen)	-	0,5	0,5	0,5		0,5	0,5			

Note: "-" negative "+" – positive results

Conclusion: _____

Task № 4. Characterize the immunobiological preparations for specific prophylaxis and treatment of the gonococcal and meningococcal infections.

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 3

Topic: *Escherichia*. Microbiological diagnosis of the diseases caused by colon bacilli.

Self-assessment tasks

a) Questions list: theoretical issues:

1. Classification and general characteristics of the Enterobacteriaceae family.
2. Biological properties of the *Escherichia* genus.
3. Classification of the *Escherichia* genus by antigenic structure and division into categories according to virulence factors, serological markers and clinical and epidemiological features.
4. Epidemiology and pathogenesis of diseases caused by *Escherichia coli*. Immunity. Principles of prevention and therapy.
5. The role of *E.coli* in the aetiology of purulent-inflammatory diseases.
6. Methods of microbiological diagnosis of infections caused by *Escherichia coli*.
7. The physiological role and sanitary-indicative value of *Escherichia coli*. Coli-titer and Coli-index.
8. The *Klebsiella* Genus. Characteristics and biological properties. *Klebsiella pneumoniae*, *ozaenae* and *rhinoscleromatis*. Role in pathology. Microbiological diagnosis.
9. Causative agents of escherichiosis in children.
10. Features of pathogenesis, immunity, and microbiological diagnosis of escherichiosis in children.

b) The list of practical skills and abilities:

1. Making preparations for microscopic research of pathological material.
2. Staining preparations by complex methods (by Gram).
3. Microscope preparations in the light microscope with immersion lens
4. Differentiation of microorganisms by morphological and tinctorial characteristics.
5. Isolation of pure cultures of aerobic microorganisms. Identification of the isolated cultures by morphological, tinctorial, cultural, biochemical, and antigenic properties.
6. Making, estimation and evaluation of reaction on glass agglutination.

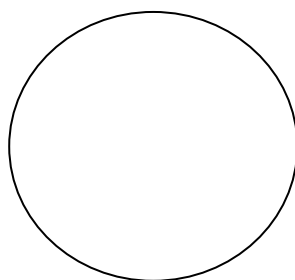
Practical lesson protocol
Practical assignments

Task № 1. Make a macroscopic and microscopic examination of the isolated colonies on differential Endo and Levin media.

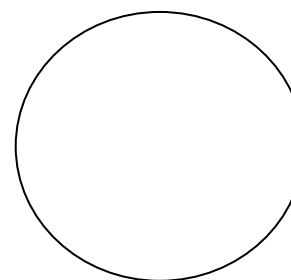
Cultural properties	Endo	Levin
Research in transmitted light		
Size (diameter)		
Form		
Degree of transparency		
Research in reflected light		
Pigmentation		
The pattern of the surface		
Position on a media		
Microscopic research		
The pattern of the margin		
Structure		
Other properties		
Consistency		

Task № 2. Make a smear from the isolated lactose positive colonies from the diagnostic media Endo's and Levin's, stain by Gram's method. Microscopy, draw.

Endo's media



Levin's media



Denote the morphological and tinctorial properties of the detected microorganisms

Task № 3. Make the slide agglutination test of diagnostic escherichia antisera (O26, O55, O111, dilution 1:10) with a one-day bacterial culture. Observe, draw and evaluate the result.

Experiment	Control of antiserum	Control of culture
		

Conclusion: _____

Task № 4. Observe and evaluate the enzymatic properties of the selected pure culture of bacteria from a patient with colitis. Fill in the result in the table.

Index Species name	Glucose	Lactose	Maltose	Saccharose	Mannitol	Milk	MPG	H ₂ S	Indole

Task № 5. Identify an isolated pure bacterial culture by morphological, tinctorial, cultural, enzymatic, and antigenic properties.

Conclusion: _____

Task № 6. Characterize the immunobiological preparations for specific prophylaxis and treatment of infections caused by Escherichia

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 4

Topic: **Salmonella. Microbiological diagnosis of typhoid and paratyphoid fevers.**

Self-assessment tasks

a) Questions list: theoretical issues:

1. The Salmonella genus. General characteristics of the genus.
2. Classification of the genus Salmonella by antigenic structure (Kaufman - White).
3. Pathogenicity of Salmonella for humans and animals.
4. Salmonella - agents of generalized infections (typhoid fever and paratyphoid A and B). Biological properties. Antigenic structure, pathogenicity factors.
5. Epidemiology and pathogenesis of diseases. Phases of pathogenesis. Bacteriocarrierity.
6. Immunogenesis of diseases.
7. Methods of microbiological diagnosis of typhoid, paratyphoid A and B.
8. Specific prevention and treatment of typhoid, paratyphoids A and B.
9. Features of pathogenesis and immunity in conditions of typhoid in children

b) The list of practical skills and abilities:

1. Making preparations for microscopic research of pathological material.
2. Staining preparations by complex methods(by Gram).
3. Microscope preparations in the light microscope with immersion lens
4. Differentiation of microorganisms by morphological and tinctorial characteristics.
5. Isolation of aerobic microorganisms pure cultures, identifying the isolated cultures by morphological, tinctorial, cultural, biochemical, and antigenic properties.
6. Fulfilment, estimation and evaluation of agglutination reaction on glass.

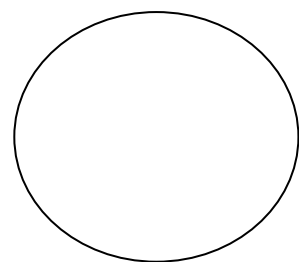
Practical lesson protocol

Practical assignments

Task №1. Make a macroscopic and microscopic examination of the isolated colonies on differential Ploskirev's media.

Cultural properties	Ploskirev
Research in transmitted light	
Size (diameter)	
Form	
Degree of transparency	
Research in reflected light	
Pigmentation	
The pattern of the surface	
Position on a media	
Microscopic research	
The pattern of the margin	
Structure	
Other properties	
Consistency	

Task №2. Make a smear from the isolated colonies on differential Ploskirev's media, stain by Gram’s method. Microscopy, draw.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №3. Make the slide agglutination test a one-day investigated bacterial culture with diagnostic antisera (dilution 1:10). Observe, draw, and evaluate the results.

	Control of culture	Experiment		
		Typhoid antiserum	Paratyphoid A antiserum	Paratyphoid B antiserum
Slide agglutination test:				
Observation:				

Conclusion; _____

Task №4. Re-inoculate the isolated lactose negative colony from Ploskirev's media to MPA in a tube to accumulate pure bacterial culture.

Task №5. Observe and evaluate the results of the Widal test with the patient's serum and the following standard bacterial suspensions: typhoid O-antigen, paratyphoid A O-antigens, paratyphoid B O-antigens, typhoid H-antigen, paratyphoid A H-antigens, paratyphoid B H-antigens. Conclude.

Tube number			1	2	3	4	5	6	Serum control	Antigen control
Patient's serum (1:50) (ml)			1	1	1	1	1	1	1	-
Saline solution (ml)			-	1	1	1	1	1	-	1
Dilution			1:50	1:100	1:200	1:400	1:800	1:1600	1:50	-
Diagnostic bacterial suspensions (ml)			1	1	1	1	1	1	-	1
EVALUATION	O-antigen	Typhoid O-antigen								
		Paratyphoid AO-antigens								
		Paratyphoid BO-antigens								
	H-antigen	Typhoid H-antigen								
		Paratyphoid AH-antigens								
		Paratyphoid BH-antigens								

Conclusion: _____

Teacher's signature _____

Date: _____

Practical lesson № 5

Topic: Salmonella. Microbiological diagnosis of salmonella gastroenteritis

Self-assessment tasks

a) Questions list: theoretical issues: 1. Salmonella - causative agents of acute gastroenteritis. 2. Features of epidemiology and pathogenesis. 3. Salmonella as a microorganism that most often causes nosocomial infection. Features of hospital strains. Nosocomial toxicoseptic salmonellosis. 4. Methods of microbiological diagnosis of salmonellosis. 5. Specific prevention and treatment. 6. Features of pathogenesis and microbiological diagnosis under conditions of salmonellosis in children. Application of serological method.	b) The list of practical skills and abilities: 1. Making preparations for microscopic research of pathological material. 2. Staining preparations by complex methods(by Gram). 3. Microscopy preparations in the light microscope with immersion lens. 4. Differentiation of microorganisms by morphological and tinctorial characteristics. 5. Isolation of aerobic microorganisms pure cultures, identification of isolated cultures by morphological, tinctorial, cultural, biochemical, and antigenic properties. 6. Production, consideration and evaluation of reaction on glass agglutination.
--	--

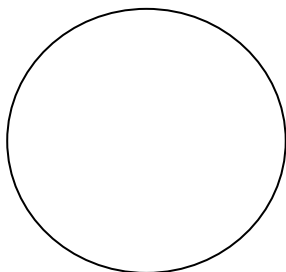
Practical lesson protocol

Practical assignments

Task №1. Make a macroscopic and microscopic examination of the isolated colonies on differential Ploskirev's media.

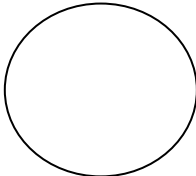
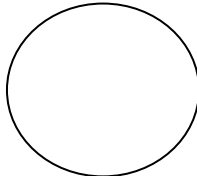
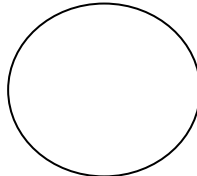
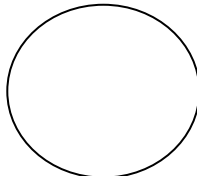
Cultural properties	Ploskirev
Research in transmitted light	
Size (diameter)	
Form	
Degree of transparency	
Research in reflected light	
Pigmentation	
The pattern of the surface	
Position on a media	
Microscopic research	
The pattern of the margin	
Structure	
Other properties	
Consistency	

Task №2. Make a smear from the selected culture of microorganisms (haemoculture), and stain by Gram’s method. Microscopy, draw.



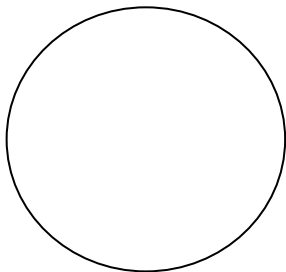
Denote the morphological and tinctorial properties of the detected microorganisms

Task №3. Make the slide agglutination test with an investigated one-day culture of bacteria and diagnostic anti-salmonellosis adsorbed antiserums to Salmonella groups B, C, and D (dilution 1:10). Observe, draw and evaluate the results.

	Control of culture	Experiment		
		Anti-salmonellosis B-antiserum	Anti-salmonellosis C-antiserum	Anti-salmonellosis D-antiserum
Slide agglutination test:				
Observation:				

Conclusion;_____

Task №4. Make a smear from the *S. typhimurium* culture, and stain by Gram’s method. Microscopy, draw.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №5. Characterize the immunobiological preparations for specific prophylaxis and treatment of infections caused by Salmonella.

Preparations	Type	Purpose of application	Orientation of the immunity, that is created
For active immunization			
For passive immunization			

Teacher’s signature _____

Date: _____

Practical lesson № 6

Topic: Shigella. Microbiological diagnosis of shigellosis

Self-assessment tasks

a) Questions list: theoretical issues:

1. Biological properties of the Shigella genus. Classification.
2. Shigella virulence factors.
3. Epidemiology, pathogenesis, the main clinical manifestations of shigellosis.
4. Immunity under shigellosis.
5. Methods of microbiological diagnosis of shigellosis.
6. Prevention and treatment of shigellosis. The problem of specific prevention.
7. Features of epidemiology, pathogenesis and immunity in the case of shigellosis in children.
8. Complications of shigellosis in children.
9. Use of bacterial drugs and the value of breastfeeding in treating intestinal infections in young children age.

b) The list of practical skills and abilities:

1. Making preparations for microscopic research of pathological material.
2. Staining preparations by complex methods(by Gram).
3. Microscopy preparations in the light microscope with immersion lens.
4. Differentiation of microorganisms by morphological and tinctorial characteristics.
5. Isolation of pure cultures of aerobic microorganisms, identification of isolated cultures on morphological, tinctorial, cultural, biochemical, antigenic properties.
6. Production, consideration and evaluation of reaction on glass agglutination.

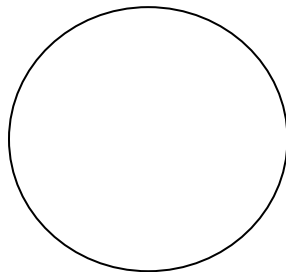
Practical lesson protocol

Practical assignments

Task №1. Make a macro- and microscopic examination of the isolated colonies on differential Ploskirev's media.

Cultural properties	Ploskirev
Research in transmitted light	
Size (diameter)	
Form	
Degree of transparency	
Research in reflected light	
Pigmentation	
The pattern of the surface	
Position on a media	
Microscopic research	
The pattern of the margin	
Structure	
Other properties	
Consistency	

Task №2. Make a smear from the isolated colonies on differential Ploskirev's medium, stain by Gram’s method. Microscopy, draw.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №3. Make the slide agglutination test with an investigated one-day culture of Lac negative bacteria and diagnostic anti-shigellosis adsorbed antiserums to *S. dysenteriae*; *S. sonnei*; *S. flexneri*; *S. boydii* (dilution 1:10). Observe, draw and evaluate the results.

Slide agglutination test:	Control of culture	Experiment			
		Anti- <i>S. dysenteriae</i> antiserum	Anti- <i>S. sonnei</i> antiserum	Anti- <i>S. flexneri</i> antiserum	Anti- <i>S. boydii</i> antiserum
Observation:					

Conclusion; _____

Task №4. Observe and evaluate the enzymatic properties of the selected pure cultures of Shigella. Then, fill in the result in the table.

Index Species names	Glucose	Lactose	Maltose	Saccharose	Mannitol	Milk	MPG	H ₂ S	Indole
S.dysenteriae									
S.sonnei									
S.flexneri									
S.boydii									

Task №5. Characterize the immunobiological preparations for specific prophylaxis and treatment of infections caused by Shigella.

Preparations	Type	Purpose of application	Orientation of the immunity, that is created
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 7

Topic: Vibriones. Microbiological diagnosis of cholera

Self-assessment tasks

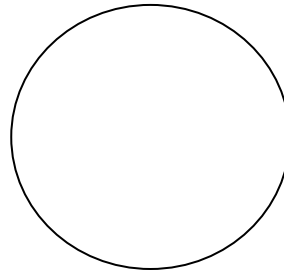
a) Questions list: theoretical issues: 1. General characteristics of vibrios. Classification, mechanism of action. 2. Vibrio cholerae. Biovars (classical and El Tor), their differentiation. 3. Classification of Vibrio by Heiberg. Antigenic structure of biovars. 4. Virulence factors of cholera vibrios. Vibrio cholerae enterotoxin (cholerogen), mechanism of action. 5. The spread of cholera. Epidemiology, pathogenesis, and main clinical manifestations of cholera. Immunity. 6. Methods of microbiological diagnosis of cholera. Accelerated diagnosis of the disease and indication of cholera vibrios in the environment. 7. Specific prevention and treatment of cholera. 8. Vibrio parahaemolyticus. 9. Features of pathogenesis and immunity against cholera in children. 10. Specific prevention of cholera in children.	b) The list of practical skills and abilities: 1. Making preparations for microscopic research of pathological material. 2. Staining preparations by complex methods(by Gram). 3. Microscope preparations with a light microscope with immersion lens. 4. Differentiation of microorganisms by morphological and tinctorial characteristics. 5. Isolation of pure cultures of aerobic microorganisms, identification of isolated cultures on morphological, tinctorial, cultural, biochemical, antigenic properties. 6. Production, consideration and evaluation of glass agglutination reaction.
---	---

Practical lesson protocol

Practical assignments

Task № 1. Make a description of the *V. cholerae* cultural properties in 1% alkaline peptone water.

Task № 2. Make a smear from the culture of *V. cholerae*, stain by Gram's method. Microscopy, draw.



Denote the morphological and tinctorial properties of the detected microorganisms

Task № 3. Make an observation of the agglutination test for rapid detection of cholera vibrios in drinking water.

№ test tubes	1	2	3	4	5	6	Control of serum	Control of water
Dilution of O-cholerae serum	1:100	1:200	1:400	1:800	1:1600	1:3200	1:100	-
Observation:								

Conclusion: _____

Task № 4. Characterize the immunobiological preparations for specific prophylaxis and treatment of infections caused by *Vibrio*.

Preparations	Type	Purpose of application	Orientation of the immunity, that is created
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 8

Topic: **Corynebacteria. Microbiological diagnosis of diphtheria.**

Self-assessment tasks

a) Questions list: theoretical issues:

1. Biological properties of the diphtheria pathogen. Classification. Biovars. Resistance.
2. Pathogenicity factors. Diphtheria toxin, mechanism of action. Toxigenicity due to phage conversion, the molecular mechanism of the diphtheria toxin action.
3. Epidemiology and pathogenesis of diphtheria. Antitoxic immunity. Bacteriocarrierity.
4. Methods of microbiological diagnosis of diphtheria. Immunological and genetic methods for determining the toxigenicity of a diphtheria pathogen.
5. Differentiation of diphtheria pathogen from other pathogenic and non-pathogenic corynebacteria, toxigenicity control.
6. Specific prevention and treatment of diphtheria.
7. Specimens for diphtheria diagnosis and it's obtaining from children of different age groups.
8. Planned specific prevention of diphtheria in children.

b) The list of practical skills and abilities:

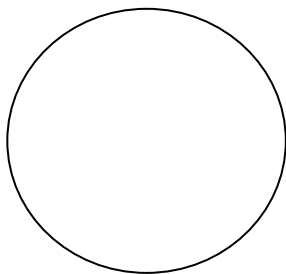
1. Microscope preparations in the light microscope with immersion lens.
2. Differentiation of microorganisms by morphological and tinctorial characteristics.
3. Ability to conduct consideration and evaluate the results of serological reactions (precipitation reaction in agar).

Practical lesson protocol

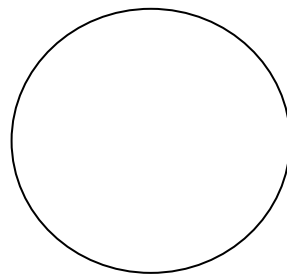
Practical assignments

Task №1. Microscopy and draw the smears of *Corynebacterium diphtheriae*.

Stained by Loeffler



Stained by Neisser



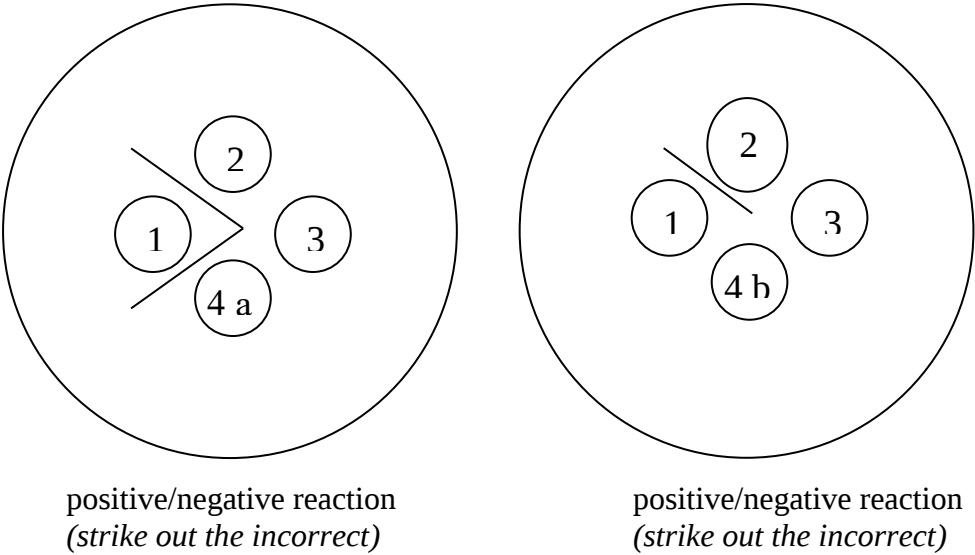
Denote the morphological and tinctorial properties of the detected microorganisms

Task №2. Observe and evaluate the enzymatic properties of the pure cultures of corynebacteria, fill in the results in the table, and conclude about species affiliation.

Indexes Species of corynebacteria	glucose	saccharose	starch	Cystinase test	Ureaza test	renewal of nitrites to nitrites	Conclusion
<i>Corynebacterium diphtheriae</i>							The explored culture №____
<i>Corynebacterium pseudodiphtheriticum</i>							The explored culture №____

Task №3. Observe and evaluate the results of gel precipitation tests to reveal of culture's toxigenicity (demonstration). Conclude.

1. Specific immune precipitated serum (antidiphtherial);
2. Known antigen
(toxigenic culture of diphtheria pathogen – *Corynebacterium diphtheriae*);
3. Normal serum;
4. Unknown antigen
(investigated cultures *Corynebacterium diphtheriae* 4a and 4b).



Conclusion:

Task №4. Characterize the immunobiological preparations for specific prophylaxis and treatment of diphtheria.

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 9

Topic: The causative agent of whooping cough. Microbiological diagnosis of pertussis. Test computer control on topics 1 - 9.

Self-assessment tasks

a) Questions list: theoretical issues:

1. The causative agent of pertussis (*Bordetella pertussis*). Morphological, cultural, antigenic properties.
2. Pathogenesis and immunity of the disease.
3. Microbiological diagnosis of pertussis.
4. Differentiation of pathogens of pertussis, parapertussis and bronchosepticosis.
5. Specific prevention of pertussis.
6. Etiotropic therapy of pertussis.
7. Hemophilic bacteria. *Legionella*.
8. Peculiarity of the epidemiology of pertussis in children.
9. Rules for taking a specimen from children for bacteriological diagnosis of pertussis.

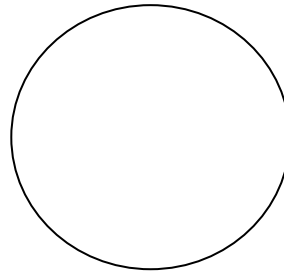
b) The list of practical skills and abilities:

1. Microscope preparations in the light microscope with immersion lens.
2. Differentiation of microorganisms by morphological and tinctorial characteristics.
3. Ability to conduct consideration and evaluate the results of serological reactions (agglutination tests).
4. Ability to record and evaluate the results of serological reactions (indirect hemagglutination reaction).

Practical lesson's protocol

Practical assignments

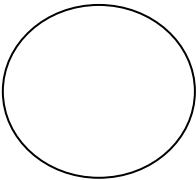
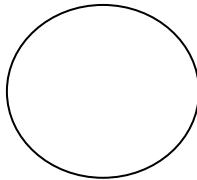
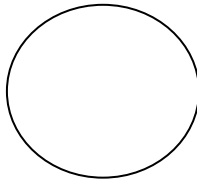
Task №1. Microscopy and draw the smears of pertussis pathogen stained by Gram.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №2. Observe the slide agglutination test with an investigated culture of bacteria and diagnostic antiserums to *B. pertussis* and *B. parapertussis* (dilution 1:10).

Draw and evaluate the results.

	Control of culture	Experiment	
		Anti- <i>B. pertussis</i> antiserum	Anti- <i>B. parapertussis</i> antiserum
Slide agglutination test:			
Observation:			

Conclusion; _____

Task №3. Observe and evaluate the results of the indirect hemagglutination test (IHAT), performed as a double-antibody technique, with the paired serums of a sick child and erythrocyte whooping-cough standard antigen. Conclude.

№ of the plastic wells in the plastic plate		1	2	3	4	5	Serums control	Antigen control
Dilution of serum		1:4	1:8	1:16	1:32	1:64	1:4	-
Observation	Serum №1							
	Serum №2							

Conclusion: _____

Task №4. Characterize the immunobiological preparations for specific prophylaxis and treatment of pertussis.

Preparations	Type	Purpose of application	Orientation of the immunity, that is created
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 10

Topic: Mycobacteria. Microbiological diagnosis of tuberculosis

Self-assessment tasks

a) Questions list: theoretical issues:

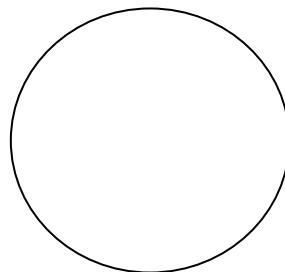
1. Pathogenic, conditionally pathogenic and saprophytic mycobacteria.
2. Biological properties of tuberculosis pathogens.
3. Variability of tuberculous bacteria, pathogenicity factors. Tuberculin.
4. Epidemiology and pathogenesis of tuberculosis.
5. The peculiarity of immunity, the role of cellular mechanisms in the context of tuberculosis.
6. Methods of microbiological diagnosis of tuberculosis.
7. Specific prevention of tuberculosis.
8. Pathogens of mycobacteriosis. Classification, properties. Role in human pathology. Mycobacteriosis as a manifestation of HIV infection.
9. Mycobacterium of leprosy. Actinomycetes. Nocardia.
10. Peculiarity of microbiological diagnosis of tuberculosis in children.
11. Planned specific prevention of tuberculosis in children. Evaluation of tuberculin test in children.

b) The list of practical skills and abilities:

1. Making preparations for microscopic examination of pathological material (mucus).
2. Staining preparations by complex methods (by Ziehl – Neelsen)
3. Microscope preparations in the light microscope with immersion lens.
4. Differentiation of microorganisms by morphological and tinctorial characteristics.

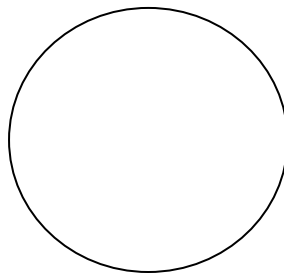
Practical lesson protocol
Practical assignments

Task №1. Make a smear from the mucus of a patient with tuberculosis, stain by Ziehl- Neelsen. Microscopy, draw.



Mark acid-fast bacteria

Task №2. Microscopy and draw the smear of *M. tuberculosis*, and reveal cord-factor as a virulence factor.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №3. Observe and evaluate the results of polymerase chain reaction (PCR), performed with pathological material from a patient with a previous diagnosis of "infiltrative form of right lung tuberculosis" to determine the presence of *Mycobacterium tuberculosis* DNA. Conclude.

Results of amplified products electrophoresis:

№№ of wells	1	2	3	4	5	6	7	8
	Clinical samples.					Controls		
						Control “+” № 1	Control “+” № 2 (Master Mix)	Control “-“
Line of the start of DNA electrophoretic separation								
Internal standard								
Target								
Observation								

Notes:

- 1 – 5 – clinical samples;
- 6 – 7 – positive controls;
- 8 – negative control;
- Internal standard - strip in a gel, that answers the size of amplicon internal standard;

5. Target - a strip in a gel, that answers the area of *Mycobacterium tuberculosis* DNA.

Task №4. Characterize of the immunobiological preparations for specific prophylaxis and treatment of tuberculosis.

Preparations	Type	Purpose of application	Orientation of the immunity, that is created
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 11

Topic: Pathogenic agents of anaerobic infections. Microbiological diagnosis of gas gangrene (clostridial mionecrosis).

Self-assessment tasks

a) Questions list: theoretical issues:

1. Classification of clostridia. Ecology, properties. Resistance to environmental factors.
2. Toxigenicity of clostridia. Genetic control of toxin formation.
3. Clostridia as causative agents of clostridial mionecrosis. Types.
4. Biological properties of clostridial mionecrosis pathogens. Pathogenicity factors, toxins.
5. Epidemiology, pathogenesis, and leading clinical manifestations of clostridial mionecrosis. Antitoxic immunity.
4. Methods of microbiological diagnosis of clostridial mionecrosis.
5. Prevention and treatment of clostridial mionecrosis.
6. Group of anaerobic gram-negative rods (bacteroids, fusobacteria).
7. Anaerobic cocci of the genus Peptococcus, Peptostreptococcus and Veillonella.
8. The role of clostridial mionecrosis pathogens in the occurrence of complications in newborns.

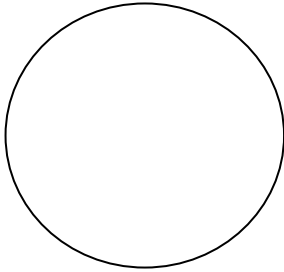
b) The list of practical skills and abilities:

1. Make preparations for microscopic research.
2. Stain preparations by sophisticated methods.
3. Microscope preparations in the light microscope with immersion lens.
4. Differentiation of microorganisms by morphological and tinctorial characteristics.
5. Sowing the test material by a pipette into a semi-fluid nutrient medium.

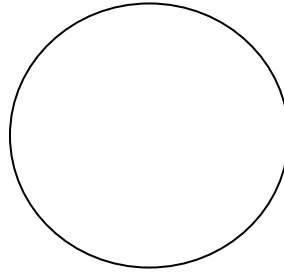
Practical lesson protocol
Practical assignments

Task №1. Microscopy and draw the smears of gas gangrene pathogens from the wounds (stained by Ojeshco, Peshcov, and Gram).

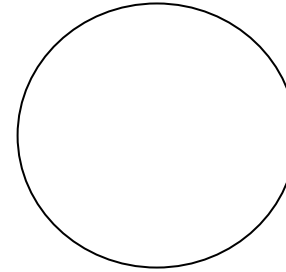
Stained by Ozheshko



Stained by Peshcov



Stained by Gram



Denote of the morphological and tinctorial properties of the detected microorganisms

Task №2. Acquaint with the features of *Clostridium perfringens* growth on the special medias:

a) Media of Vilson–Bler _____

b) Media of Kitt-Tarozzi _____

c) Sterile fat free litmus milk _____

Task № 3. Characterize the immunobiological preparations for specific prophylaxis and treatment of gas gangrene.

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 12

Topic: Pathogens of anaerobic infections. Microbiological diagnosis of tetanus and botulism.

Self-assessment tasks

a) Questions list: theoretical issues:

1. The biological properties of causative agents of tetanus and botulism.
Pathogenicity factors, toxins.
2. Epidemiology, pathogenesis, the main clinical manifestations of tetanus and botulism. Immunity.
3. Methods of microbiological diagnosis of tetanus and botulism.
4. Specific treatment and prevention of tetanus and botulism.
5. Tetanus neonatorum (umbilical tetanus).

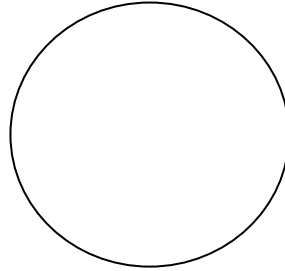
b) The list of practical skills and abilities:

1. Make preparations for microscopic research.
2. Stain preparations by sophisticated methods (by Gram)
3. Microscope preparations in the light microscope with immersion lens.
4. Differentiation of microorganisms by morphological and tinctorial characteristics.
5. Isolation of pure cultures of anaerobic bacteria, identification of isolated cultures.

Practical lesson protocol

Practical assignments

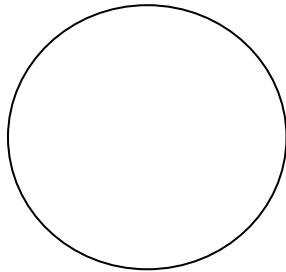
Task №1. Make a smear from the culture of anaerobic bacteria in Kitt-Tarozzi media, and stain by Gram's method. Microscopy, draw.



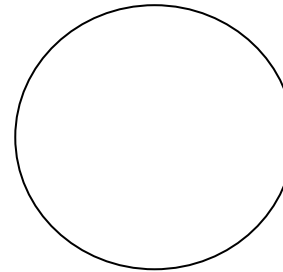
Denote of the morphological and tinctorial properties of the detected microorganisms

Task № 2. Microscopy and draw the smears of *C. tetani* and *C. botulinum* stained by Gram.

C. tetani



C. botulinum

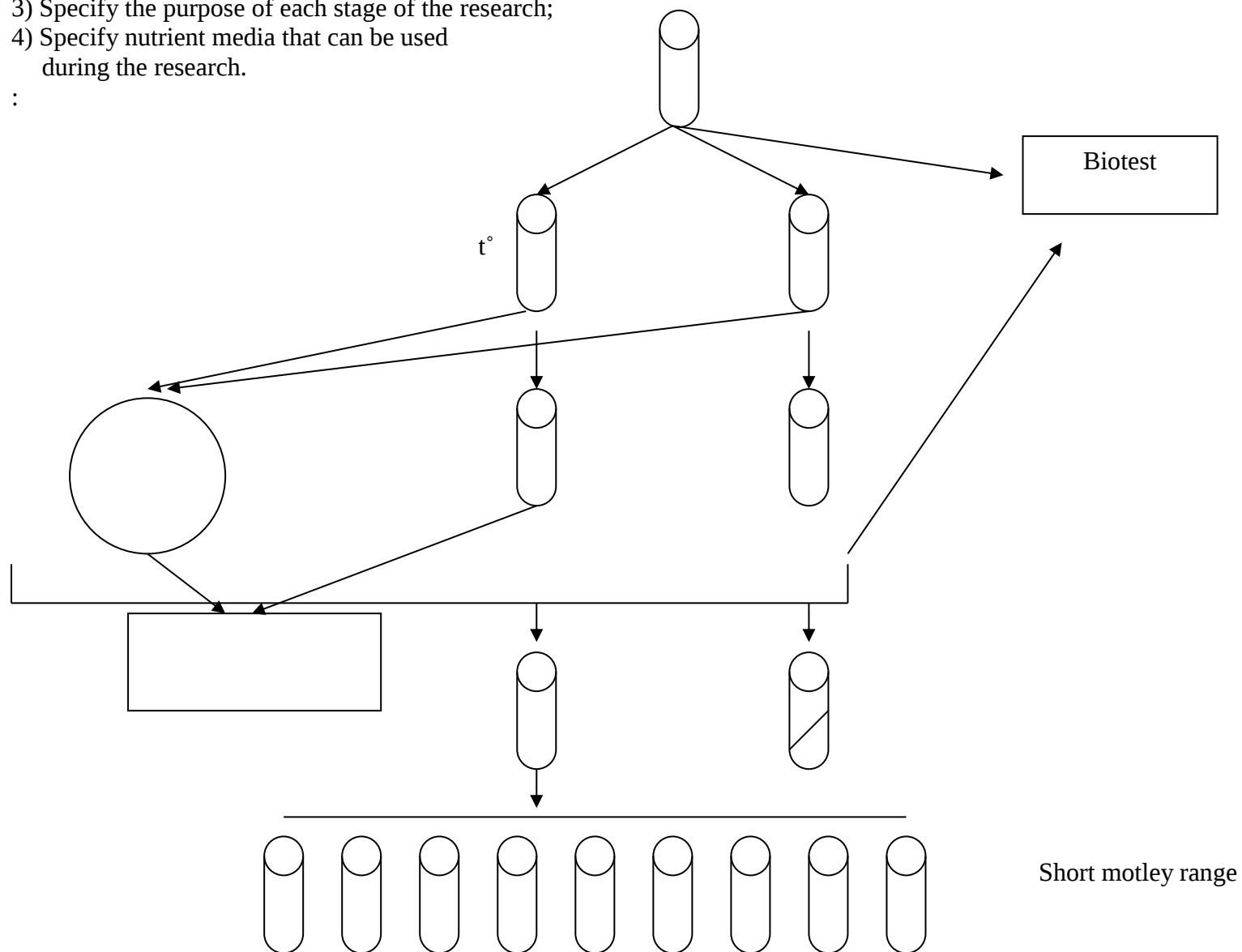


Denote morphological and tinctorial properties of microorganisms

Task № 3. Characterize the bacteriological research under conditions of botulism:

- 1) Specify the purpose of research;
- 2) Specify the material to be researched;
- 3) Specify the purpose of each stage of the research;
- 4) Specify nutrient media that can be used during the research.

:



1. _____
2. _____
3. _____
3. 1 _____
3. 2 _____
3. 3 _____
3. 4 _____
3. 5 _____
4. _____

Task № 4. Characterize of the immunobiological preparations for specific prophylaxis and treatment of tetanus and botulism.

Preparations	Type	Purpose of application	Orientation of the immunity, that is created
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 13

Topic: Pathogenic agents of zoonous infections. Microbiological diagnosis of anthrax and brucellosis

Self-assessment tasks

a) Questions list: theoretical issues:

1. Ecology of agents of anthrax and brucellosis.
2. Biological properties of pathogens of anthrax and brucellosis.
Classification. Resistance. Pathogenicity factors. Pathogenicity to human and animals.
3. Epidemiology and pathogenesis. The main clinical manifestations of anthrax and brucellosis in human.
4. Immunity under conditions of anthrax and brucellosis.
5. Methods of microbiological diagnosis of anthrax and brucellosis.
6. Principles of prevention and treatment of anthrax and brucellosis. Specific prevention and treatment.

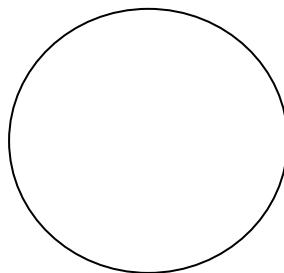
b) The list of practical skills and abilities:

1. Compliance with the rules of anti-epidemic regiment and safety in the microbiology laboratory working with agents of especially dangerous infections.
2. Microscope preparations in the light microscope with immersion lens.
3. Differentiation of microorganisms by morphological and tinctorial characteristics.
4. Ability to perform consideration and evaluate the results of serological reactions (reactions of precipitation, agglutination).

Practical lesson protocol

Practical assignments

Task №1. Microscopy and draw the smear of B.anthraxis stained by Gram.



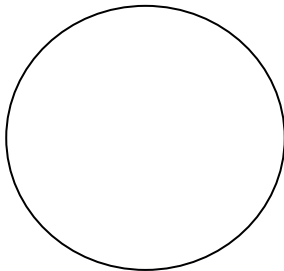
Denote the morphological and tinctorial properties of the detected microorganisms

Task № 2: Make the Ascoli ring precipitation test with Anthrax antiserum and extract dead animal tissues. Observe and evaluate the result.

Tube numbers	Research	Control	Control	Control
Ingredients (ml)	1	2	3	4
Anthrax antiserum	0,5		0,5	0,5
Investigated extract	0,5	0,5		
Normal serum		0,5		
Anthrax positive extract				0,5
Extract without anthrax antigens			0,5	
Observation				

Conclusion: _____

Task №3. Make a smear from the standard Brucella’s antigen, stain by Gram’s method. Microscopy, draw.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №4. Observe and evaluate the agglutination test results (Wrayt's test) with the serums of the patient and standard Brucella’s antigen. Conclude.

№ test tubes	1	2	3	4	5	Serum control	Antigen control
Dilution of serum	1: 50	1: 100	1: 200	1: 400	1: 800	-	-
Observation							

Conclusion:_____

Task №5. Characterize of the immunobiological preparations for specific prophylaxis and treatment of anthrax and brucellosis.

Preparations	Type	Purpose of application	Orientation of the immunity, that is created
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 14

Topic: Pathogens of zoonotic infections. Microbiological diagnosis of plague and tularemia.

Self-assessment tasks

a) Questions list: theoretical issues:

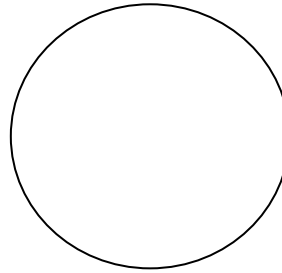
1. Biological properties of the causative agents of plague and tularemia. Virulence factors. Classification.
2. Epidemiology, pathogenesis and clinical forms of plague and tularemia.
3. Immunity in plague and tularemia.
4. Methods of microbiological diagnosis of plague and tularemia. Identification criteria for the plague pathogen.
5. Specific prevention and treatment of plague and tularemia.
6. Enteropathogenic *Yersinia* (pathogens of pseudotuberculosis and intestinal yersiniosis).

b) The list of practical skills and abilities:

1. Compliance with the rules of the anti-epidemic regime and safety in the microbiology laboratory when working with agents of especially dangerous infections.
2. Microscope preparations in the light microscope with immersion lens.
3. Differentiation of microorganisms by morphological and tinctorial characteristics.
4. Ability to perform consideration and evaluate the results of serological reactions (reactions of agglutination).

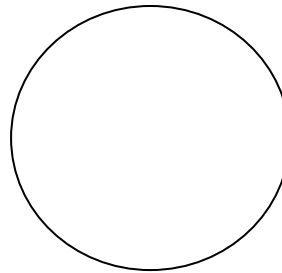
Practical lesson protocol
Practical assignments

Task №1. Microscopy and draw the smear of the *Y. pestis*, stained by methylene blue.



Denote the morphological and tinctorial properties of the detected microorganisms

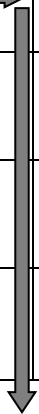
Task №2. Microscopy and draw the smears of the *F. tularensis* stained by Gram.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №3. Observe and evaluate the indirect hemagglutination test (IHAT) results to identify the pure bacterial culture isolated from a patient with suspicion of plague. Conclude.

Reactions:	Investigation					Control of freeze-drying erythrocyte diagnosticum
№ of the plastic wells in the plastic plate	1	2	3	4	5	6
Reaction stabilizer (ml)	0,4	0,4	0,4	0,4	0,4	0,4
Investigated pure culture of bacteria (in the concentration of 4×10^9) (ml)	0,4 →	0,4 →	0,4 →	0,4 →	0,4 →	0,05
Freeze drying erythrocyte diagnosticum (ml)	0,05	0,05	0,05	0,05	0,05	-
Dilution of culture	1:2	1:4	1:8	1:16	1:32	-
Observation						



Conclusion: _____

Task №4. Observe and evaluate the agglutination test results, performed with the patient's serum and tularemia standard antigen. Conclude.

№ test tubes	1	2	3	4	5	Serum control	Antigen control
Dilution of serum	1: 50	1: 100	1: 200	1: 400	1: 800	1: 50	-
Observation							

Conclusion: _____

Task №5. Characterize the immunobiological preparations for specific prophylaxis and treatment of plague and tularemia.

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 15

Topic: Rickettsia, Chlamydia, Mycoplasma. Microbiological diagnosis of rickettsiosis, chlamydiosis and mycoplasmosis

Self-assessment tasks

a) Questions list: theoretical issues:

1. General characteristics of rickettsia, chlamydia, and mycoplasma. Classification.
2. Rickettsia - causative agents of epidemic typhus and Brill-Zinsser disease, and endemic typhus. Biological properties. Ecology of pathogens. Antigenic structure. Toxin formation.
3. Epidemiology, pathogenesis, immunity in the case of typhus.
4. The causative agent of Q-fever. Ecology. Resistance. Antigenic structure. Toxin formation.
5. Epidemiology, pathogenesis, immunity under conditions of Q-fever.
6. Rickettsia of spotted fever group.
7. Biological properties of chlamydia and mycoplasma. Intracellular parasitism of chlamydia. Antigenic structure and pathogenicity factors.
8. The role of chlamydia and mycoplasma in human pathology. Epidemiology, pathogenesis, immunity, infections.
9. Methods for microbiological diagnosis of diseases caused by rickettsiae, chlamydia and mycoplasma.
10. Prevention and treatment of diseases caused by rickettsiae, chlamydia and mycoplasma.
11. Rickettsiosis in children. Specific prevention of rickettsioses.
12. The role of chlamydia in the pathology of pregnancy and fetal lesions.
13. The role of mycoplasma in the pathology of pregnancy and diseases in children.

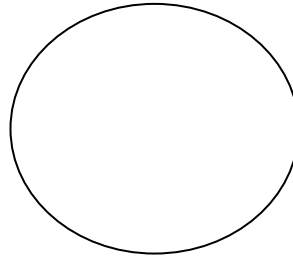
b) The list of practical skills and abilities:

1. Microscope preparations in the light microscope with immersion lens.
2. Differentiation of microorganisms by morphological and tinctorial characteristics.
3. Ability to conduct consideration and evaluate the results of serological reactions (complement fixation, ELISA, PCR).

Practical lesson protocol

Practical assignments

Task №1. Microscopy and draw the smear of rickettsia, stained by Zdrodovsky.



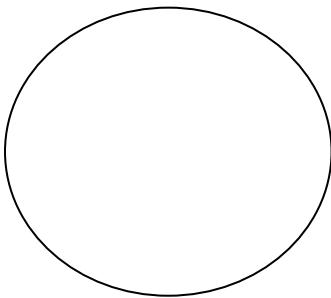
Denote the morphological and tinctorial properties of the detected microorganisms

Task №2. Observe and evaluate the results of the indirect hemagglutination test (IHAT), performed as a double-antibody technique, with the paired serums of a sick child and erythrocyte *Coxiella burnetii* standard antigen. Conclude.

№ of the plastic wells in the plastic plate		1	2	3	4	5	Serums control	Antigen control
Dilution of serum		1:4	1:8	1:16	1:32	1:64	1:4	-
Observation	Serum №1							
	Serum №2							

Conclusion: _____

Task №3. Microscopy and draw the material from the urethra of a patient with chlamydiosis, stained by Romanovsky-Gimza.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №4. Observe and evaluate the results of the complement fixation test (CFT), performed as a double-antibody technique, with the paired serums of a patient and standard antigens of the *Chlamydia psittaci* and *Mycoplasma pneumonia*. Conclude

.

Dilution of serums							Serums control	Antigens control
Standard antigens		1:8	1:16	1:32	1:64	1:128	1:8	-
Observation	<i>Chlamydia psittaci</i>							
	7th day of disease							
	20th day of disease							
	<i>Mycoplasma pneumoniae</i>							
	7th day of disease							
	20th day of disease							

Conclusion:_____

Task №5. Observe and evaluate the results of polymerase chain reaction (PCR), performed with pathological material from a patient with a previous suspicion of chlamydiosis to determine the presence of *Chlamydia trachomatis* DNA. Conclude.

Results of amplified products electrophoresis:

№№ of wells	1	2	3	4	5	6	7	8
	Clinical samples.					Controls		
						Control “+” № 1	Control “+” № 2 (Master Mix)	Control “-“
Line of the start of DNA electrophoretic separation								
Internal standard								
Target								
Observation								

Notes:

- 1 – 5 – clinical samples;
- 6 – 7 – positive controls;
- 8 – negative control;
- Internal standard - strip in a gel that answers the size of internal amplicon standard;
- Target - a strip in a gel that answers the area of *Mycobacterium tuberculosis* DNA.

Task №6. Characterize the immunobiological preparations for specific prophylaxis and treatment of the diseases caused by Rickettsia, Chlamydia and Mycoplasma.

Preparations	Type	Purpose of application	Orientation of the immunity, that is created
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 16

Topic: Spirochaetes. Microbiological diagnosis of syphilis

Self-assessment tasks

a) Questions list: theoretical issues:

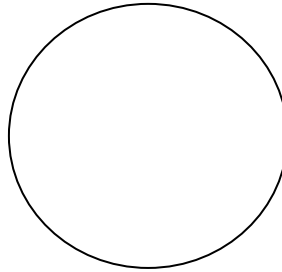
1. General characteristics of spirochetes. Classification.
2. The causative agent of syphilis. Biological properties of Treponema.
3. Epidemiology, pathogenesis and immunogenesis of syphilis.
4. Methods of microbiological diagnosis of syphilis.
5. Prevention and treatment of syphilis.
6. Causes of frambezia, pinta. Epidemiology, pathogenesis, microbiological diagnosis.
7. Features of epidemiology and pathogenesis of acquired syphilis in children.
8. Congenital syphilis.
9. Peculiarities of treatment and microbiological diagnosis of syphilis in children.

b) The list of practical skills and abilities:

1. Microscope preparations in the light microscope with immersion lens.
2. Differentiation of microorganisms by morphological and tinctorial characteristics
3. Ability to conduct consideration and evaluate the results of serological reactions (complement fixation, ELISA).

Practical lesson protocol
Practical assignments

Task №1. Microscopy and draw the smear of dental plaque, made by Burri.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №2. Observe and evaluate the results of the microprecipitation reaction (MPR) performed with the serum of a patient with a previous suspicion of syphilis and standard Cardiolipin antigen. Conclude.

Conclusion: _____

Task №3. Observe and evaluate the results of the Wassermann test performed with a patient's serum with the previous suspicion of syphilis and standard antigens. Conclude.

Tubes numbers	1	2	3
Ingredients, ml			(Serums control.)
Inactivated patient's serum 1:5	0,25	0,25	0,25
Treponemal antigen in the working dose	0,25	-	-
Cardiolipin antigen in the working dose	-	0,25	-
Isotonic sodium chloride solution	-	-	0,25
Compliment in the working title	0,25	0,25	0,25
Shake, put in the thermostat at 37°C for 45 min			
Hemolytic system	0,5	0,5	0,5
Shake, put in the thermostat at 37°C for 45-60 min			
Observation			

Conclusion: _____

Task №4. To estimate ELISA results with serums of donors with antibodies exposure to the antigens of syphilis pathogen.

TEST NO : 50 W/L MODE : DUAL DATE : 17/10/03 **SANOF 1 DIAGNOSIS PASTEUM PR 21.00**
 TEST NAME : SYPHO . 10 TEST FILTER : 490 nm TIME : 12:05 *** INDICATES VALUE OUT OF RANGE
 PLATE : 0038 REF. FILTER : 620 nm OPERATOR : POS INDICATES A POSITIVE REACTION
 QUALITY CONTROL Neg INDICATES A NEGATIVE REACTION
 NCi<0,2

Valid (NC)>=3
 NCi<NCi2

0,018<0,2
 0,022<0,2
 0,022<0,2
 0,021<0,2
 4>=3
 0,018<0,0415
 0,022<0,0415
 0,022<0,0415
 0,021<0,0415
 4>=3
 2,25>0,6

Valid (NC)>=3
 PC>0,6

+ EON = (NC + 0,10) - EON = (NC + 0,10) * 0,9
 = 0,121 = 0,109

1	2	3	4	5	6	7	8	9	10	11	12	
0,018	0,21 neg	0,012 neg	0,018 neg	***	***	***	***	***	***	***	***	A
0,022 NC2	0,22 neg	0,020 neg	0,020 neg	***	***	***	***	***	***	***	***	B
0,022 NC3	0,143 neg	0,022 neg	0,025 neg	***	***	***	***	***	***	***	***	C
0,021 NC4	0,019 neg	0,020 neg	0,038 POS	***	***	***	***	***	***	***	***	D
2,261 PC1	0,016 neg	0,019 neg	0,407 POS	***	***	***	***	***	***	***	***	E
2,243 PC1	0,027 neg	0,021 neg	0,380 POS	***	***	***	***	***	***	***	***	F
0,018 neg	0,015 neg	0,020 neg	2,808 POS	***	***	***	***	***	***	***	***	G
0,018 neg	0,013 neg	0,018 neg	2,872 POS	***	***	***	***	***	***	***	***	H

Conclusion: _____

Task №5. Characterize the immunobiological preparations for specific prophylaxis and treatment of syphilis.

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 17

Topic: Spirochaetes. Microbiological diagnosis of relapsin fever and leptospirosis

Self-assessment tasks

a) Questions list: theoretical issues:

1. Borrelia, Leptospira. Classification. Biological properties of Borrelia and Leptospira.
2. Epidemiology and pathogenesis of relapsing fever and leptospirosis. Immunity.
3. Methods of microbiological diagnosis of relapsing fever and leptospirosis.
4. Specific prevention of relapsing fever and leptospirosis
5. Lyme disease, pathogen, microbiological diagnosis, prevention.
6. Campylobacter. Helicobacter.
7. Features of epidemiology and pathogenesis of relapsing fever in children.

b) The list of practical skills and abilities:

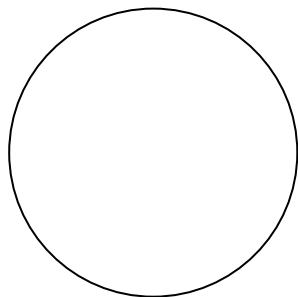
1. Microscope preparations in the light microscope with immersion lens.
2. Differentiation of microorganisms by morphological and tinctorial characteristics.
3. Ability to conduct consideration and evaluate the results of serological reactions (complement fixation).

Practical lesson protocol

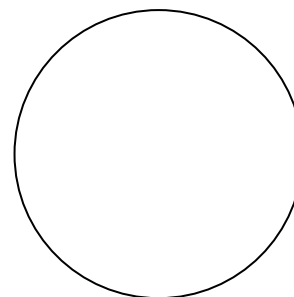
Practical assignments

Task №1. Microscopy and draw the smear of *Borrelia*, stained by Romanovsky-Giemza and *Leptospira* by Burri.

Borrelia



Leptospira



Denote the morphological and tinctorial properties of the detected microorganisms

Task №2. Observe and evaluate the results of the complement fixation test (CFT), performed with the serums of a patient and standard antigens of the *Leptospira*. Conclude

№ test tubes	1	2	3	4	Control of serum	Control of the antigen
Dilution of the serum	1:10	1:100	1:1000	1:10000		
Observation of the hemolysis						
Observation of the reaction						

Conclusion: _____

Task №3. Characterize the immunobiological preparations for specific prophylaxis and treatment of relapsing fever and leptospirosis.

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 18

Topic: Pathogenic fungi. Microbiological diagnosis of mycoses.

Self-assessment tasks

a) Questions list: theoretical issues:

1. Pathogenic fungi. Classification. Biological properties. Resistance. Factors of pathogenicity, toxins. Sensitivity to antibiotics.
2. Dermatophytes - pathogens of dermatomycosis (epidermophytia, trichophytia, microsporia, favus). Properties. Pathogenicity to humans. Microbiological diagnosis.
3. Pathogens of systemic mycoses: blastomycosis, histoplasmosis, cryptococcosis. Properties. Pathogenicity to humans. Microbiological diagnosis.
4. Fungi of the *Candida* genus. Properties. Pathogenicity to humans. Factors that promote candidiasis (dysbiosis, etc.). Microbiological diagnosis. Antimicrobial drugs.
5. Pathogens of aspergillosis and penicilliosis. Properties. Pathogenicity to humans.
6. *Pneumocystis carinii*. Pneumocystis pneumonia in AIDS patients.
7. Candidal lesions in children of different age groups. Epidemiology, pathogenesis.
8. Candidiasis in newborns (particularly thrush or candidiasis stomatitis, etc.).

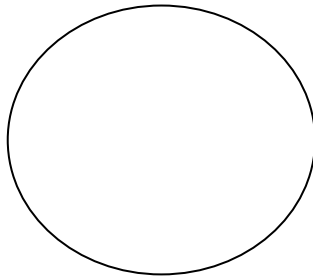
b) The list of practical skills and abilities:

1. Make preparations for microscopic research.
2. Stain preparations by sophisticated methods (by Gram)
3. Microscope preparations in the light microscope with immersion lens.
4. Differentiation of microorganisms by morphological and tinctorial characteristics.
5. Transfer the test material to a loop on solid and liquid nutrient media.

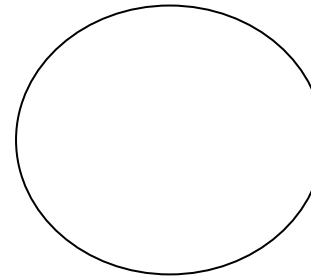
Practical lesson protocol
Practical assignments

Task №1. Microscopy and draw the smears of *Aspergillus* and *Penicillium*.

Aspergillus

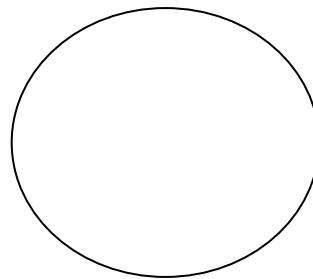


Penicillium



Denote the morphological and tinctorial properties of the detected microorganisms

Task №2. Make a smear from a pathological material of a patient with candidosis stain by Gram's method. Microscopy, draw.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №3. Make a macroscopic and microscopic examination of the isolated colonies on differential Sabouraud media.

Cultural properties	Sabouraud media
Research in transmitted light	
Size (diameter)	
Form	
Degree of transparency	
Research in reflected light	
Pigmentation	
The pattern of the surface	
Position on a media	
Microscopic research	
The pattern of the margin	
Structure	
Other properties	
Consistency	

Task №4. Characterize the immunobiological preparations for specific prophylaxis and treatment of mycoses.

Preparations	Type	Purpose of application	Orientation of the created immunity
For active immunization			
For passive immunization			

Teacher's signature _____

Date: _____

Practical lesson № 19

Topic: Normal microflora of the human body

Self-assessment tasks

a) Questions list: theoretical issues:

1. Normal microflora (normal microbiota) of the human body (eumicrobiocenosis). The autochthonous (indigenous) and allochthonous microflora of the human body. Microbiome.
2. Microflora of skin, respiratory tract, digestive, and genitourinary systems, its anti-infective, detoxifying, immunizing, and metabolic role.
3. Research methods on the normal microflora role in the human body. Gnotobiology, the value of gnotobiological principles in the clinic. Ethology of microbes.
4. Factors that affect the quantitative and qualitative composition of the microflora of the human body. The concept of colonization resistance and its role in infectious pathology.
5. Dysbacteriosis. Methods of determination.
6. Probiotics, prebiotics - preparations for restoring normal microflora of the human body (bifidumbacterin, lactobacterin, colibacterin, bificol, aerococobacterin, biosporin, bactisubtil, multi probiotics of the Symbiter group, etc.). Mechanism of action.
7. Dynamics of normal microflora in human ontogenesis.
8. Pathogenic role of normal microflora and mechanisms of their acquisition of pathogenic properties.
9. Age peculiarities of normal microflora of the human body.
10. Dynamics of intestinal microflora in newborn children. Value of bifidobacteria and lactobacilli.
11. Influence of natural and artificial feeding on the intestinal microflora of a child.
12. The phenomenon of dysbiosis and its importance in childhood pathology.
13. The use of bacterial preparations (bifidumbacterin, bificol, lactobacterin, colibacterin, etc.) to prevent dysbiosis and treat intestinal diseases in children.

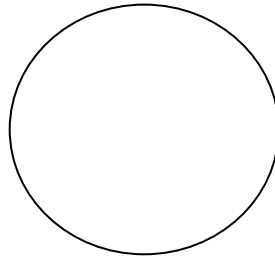
b) The list of practical skills and abilities:

1. Microscope preparations in the light microscope with immersion lens.
2. Differentiation of microorganisms by morphological and tinctorial characteristics.
3. Test material inoculation by loop and pipette to solid, semi-solid and liquid culture media.

Practical lesson protocol

Practical assignments

Task №1. Microscopy and draw the smear of healthy human faeces and stain it by Gram.



Denote the morphological and tinctorial properties of the detected microorganisms

Task №2. Observe and evaluate the results of the patient's faeces bacteriological research. Conclude. (For analysis of the results obtained, see the appendix "Classification of intestinal dysbiosis").

Result of faeces bacteriological research

From « ____ » _____ 20__ year
 Analysis № _____
 The last name, name _____
 Age of patient _____
 Analysis primary _____
 Repeated _____
 Establishment _____

	Microflora	Norm	Patient's results
1.	The common quantity of E.coli	$10^6 - 4 \times 10^8$	
2.	E.coli with the changed enzyme properties	$<10^6$	
3.	Lactosenegative E.coli	$<10^6$	
4.	Hemolytic microorganisms	$<10^6$	
5.	Lactobacterias	$>10^6$	
6.	Bifidobacteria	$>10^7$	
7.	Conditionally pathogenic (CP) bacteria (rod and cocci forms)	$10^3 - 10^6$	
8.	Staphylococci (hemolytic, plasmocoagulative)	$<10^4$	
9.	Staphylococci (non hemolytic, epidermal)	$<10^4 - 10^5$	
10.	Candida	$<10^4$	
11.	Streptococci	$<10^5 - 10^7$	
Date _____ Doctor _____			

Conclusion: _____

Task №3. Inoculate the nose mucus on yolk-salt agar (YSA).

Appendix for the task №2

Classification of the intestinal dysbiosis

1st degree: latent phase of dysbiosis. Anaerobic flora predominates over aerobic. Bifidobacteria and lactobacilli are excreted in dilutions 10^8 - 10^7 or one of these forms in dilution 10^{10} - 10^9 . High-grade *Escherichia coli* constitute up to 80% of the total. In this phase, the vegetation of individual representatives of the conditionally pathogenic flora is possible (no more than two species in dilutions 10^4 - 10^2). The initial phase of dysbiosis occurs as a reaction of an organically healthy baby's body to the influence of some adverse factors, such as impaired diet or quality of nutrition. There is no bowel dysfunction.

2nd degree: start-up phase of dysbiosis. There is an inhibition of anaerobic bacteria, and their sum is approximately equal to the number of aerobes. It reduces the number of high-grade *Escherichia coli*. Conditional pathogens (Plasma-coagulative *Staphylococcus aureus*, *Proteus* or fungi of the genus *Candida*) are excreted in dilutions 10^6 - 10^7 . Normal *Escherichia coli* are replaced by their atypical variants (lactose-negative, hemolytic).

3rd degree: Aggression phase of aerobic flora. Aerobic flora prevails up to the complete absence of bifidobacteria and lactobacilli. The number of opportunistic microbes with aggression properties increases sharply; hemolysis of erythrocytes and coagulation of blood plasma. Plasma-coagulating and haemolytic staphylococcus, haemolytic *Escherichia coli*, *Proteus*, *Klebsiella*, *Pseudomonas aeruginosa*, clostridia, and fungi of the genus *Candida* are particularly common. A common feature of all these bacteria is multiple drug resistance.

4th degree: Associated dysbiosis phase. There is an almost complete absence of bifidobacteria against the background of a sharp decrease in the number of lactic acid bacteria and significant aggressiveness of conditionally pathogenic microorganisms.

Depending on the predominant selected conditionally pathogenic microbes, they divide staphylococcal, proteic, candidiasis, clostridiosis, and associated dysbiosis.

Teacher's signature _____

Date: _____

Practical lesson № 20

Topic: Clinical microbiology. Microbiological research of respiratory system, blood and CNS (central nervous system)

Self-assessment tasks

a) Questions list: theoretical issues:

1. The value of clinical microbiology in the professional work of a doctor.
2. Objects of research. Pathogenic and opportunistic microorganisms.
Pathogenicity. Heterogeneity and variability of microbial populations.
3. Opportunistic infections. Conditions of occurrence, multiorgan tropism, polyetiology, the low specificity of clinical manifestations, and tendency to generalization.
4. The spread of opportunistic infections. Exogenous opportunistic infections (legionellosis, pseudotuberculosis, listeriosis, serraciosis).
5. Endogenous opportunistic infections, the role of representatives of resident microflora in their occurrence. Anaerobic non-clostridial bacteria: bacteroids, fusobacteria, anaerobic cocci.
6. Microbiological diagnosis of opportunistic infections. Criteria for the etiological role of conditionally pathogenic microbes isolated from the pathological focus.
7. Microbiological examination of the respiratory system.
8. Microbiological examination of blood.
9. Microbiological examination of the CNS.

b) The list of practical skills and abilities:

1. Compliance with the rules of the anti-epidemic regime and safety in the bacteriological laboratory.
2. Disinfection of infected material, antiseptic hand treatment contaminated with the test material or culture of microbes
3. Preparation of preparations for microscopic examination of pathological material.
4. Coloring of drugs by complex methods (by Gram).
5. Microscopy of preparations in a light microscope using an immersion lens.
6. Differentiation of microorganisms by morphological and tinctorial characteristics.
7. Filling the forms of directions of the investigated material in the laboratory for bacteriological research.

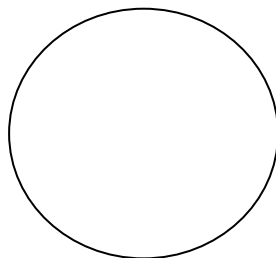
Practical lesson protocol

Practical assignments

Task №1. Make a macroscopic and microscopic examination of the isolated colonies on differential yolk salt agar (YSA).

Cultural properties	YSA
Research in transmitted light	
Size (diameter)	
Form	
Degree of transparency	
Research in reflected light	
Pigmentation	
The pattern of the surface	
Position on a media	
Microscopic research	
The pattern of the margin	
Structure	
Other properties	
Consistency	

Task №2. Make a smear from the isolated colony, and stain by Gram's method. Microscopy, draw.



Denote the morphological and tinctorial properties of the detected microorganisms

Conclude tasks №1 and №2.

Conclusion: _____

Task №3. Microscopy and draw the smears, determine their morphological and tinctorial properties. picturei, characteristics of the studied microorganisms and the names of the nutrient media for their cultivation fill in in Appendix 1: "Pathogens of opportunistic and hospital infections" (columns 6a, 6b, 6c).

Task №4. Fill in the form to the bacteriological laboratory of the test material (blood) from a patient with suspected sepsis.

Task №5. Fill in the form with the results of a blood test from a patient with suspected sepsis.

Teacher's signature _____

Date: _____

Practical lesson № 21

Topic: Clinical microbiology. Microbiological study of the digestive and urogenital systems.

Self-assessment tasks

a) Questions list: theoretical issues:

1. Microbiocenoses of healthy and pathologically altered human body biotopes.
2. Microbiocenoses of pathologically altered human biotopes (under conditions of affection of digestive and urogenital systems).
3. Microbiological examination of digestive and urogenital systems.
4. Dysbiosis. Conditions of occurrence. Consequences of development.
5. Classification of dysbiosis by pathogen and by localization.
6. Principles of diagnosis and treatment of dysbiosis.
7. Opportunistic iatrogenic infections. Etiological structure.
8. Hospital strains and ecovars of opportunistic microbes. Opportunistic infections associated with medical intervention. The peculiarity of immunity.
9. Microbiological bases of prevention and treatment of opportunistic infections.
10. Scientific substantiation of anti-epidemic measures.

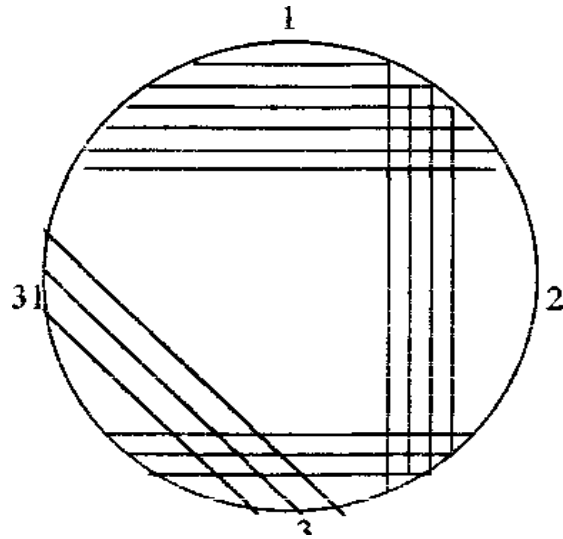
b) The list of practical skills and abilities:

1. Compliance with rules of epidemiological regime and safety in the bacteriological laboratory.
2. Disinfection of infected material, antiseptic of hands contaminated by material or microbes culture studied.
3. Making of preparations for microscopic examination of pathological material.
4. Staining of agents by complex methods (Gram).
5. Microscopy with the light microscope with immersion lens.
6. Differentiation of microorganisms by morphological and tinctorial characteristics.
7. Production, recording and evaluation of slide agglutination.

Practical lesson protocol

Practical assignments

Task №1. Observe and evaluate the results of the patient's urine culture, which have been done by the sectoral method (by Gould). Then, according to the calculation table, denote the degree of its microbial contamination (bacteriuria). Conclude.



Scheme of inoculation by Gould

Calculation table for determination of bacteria quantity in 1 ml of liquid:

Quantity of colonies that have grown in a sector				Quantity of bacteria in 1 ml of liquid
1st	2nd	3d	4Th	
1 - 6	No growth	No growth	No growth	<1 000
8-20	--	--	--	1000
21-30	--	--	--	5000
31-60	--	--	--	10000
70-80	--	--	--	50000
100-150	5-10	--	--	100 000
Very large amount	20-30	--	--	500 000
The same	40-60	--	--	1 000 000
--	100 - 140	10-20	--	5 000 000
--	Very large amount	30-40	--	10 000 000
--	The same	60 - 80	Single colonies	50 000 000
--	--	80 - 140	From single to 25	100 000 000

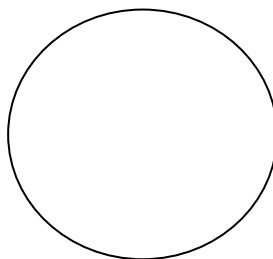
Conclusion:_____

Task №2. Make the slide agglutination test with bacteria from the lactose-positive colony (from Endo medium) and the mixture of standard sera (O1, O8, O62, O75 + K1, K5, K13). Draw the reactions. Observe and evaluate the results. Conclude

Experiment	Control of untisera	Control of culture
		

Conclusion: _____

Task №3. Microscopy and draw the smear of vagina discharge stained by Gram. Observe and evaluate the results. See the appendix for the analysis of the obtained results. Conclude.



Denote the morphological and tinctorial properties of the detected microorganisms

Conclusion: _____

Appendix:

Determination of the degree of purity of the vagina

4 degrees of purity of the vagina are:

1st degree of purity of the vagina is characterized by the presence in the female reproductive organ of sticks of Doderlein lactobacilli. These microorganisms form the basis of a healthy vagina. The medium is acidic. Any pathogenic microbes, blood cells, particularly leukocytes, are absent.

2nd degree of purity of the female vagina occurs in most women of reproductive age because the first degree is very rarely due to sexual activity, violations of the rules of hygiene, and other factors contributing to the emergence of opportunistic flora. This purity is characterized by the presence of the same sticks of Doderlein, lactobacilli. However, in a single amount of cocci present. Additionally, it may be up to 10 cells and no more than 5 epithelial cells.

3rd degree of purity of the vagina is characterized by the presence in the reproductive system and the inflammatory process. In this case, the environment is changed to alkaline, and the number of sticks of Doderlein is drastically reduced. Thus there is an increase in pathogens such as Streptococcus, Staphylococcus, fungi, E. coli. The number of cells increases, and in the microscope's field of view, it can be counted up to 30 such cells, plenty of leucocytes. Typically, this degree of purity of the vagina is accompanied by symptoms like discharge and pruritus.

4 degree is observed in bacterial vaginosis or infectious process. Alkalinity and sticks Doderlein are entirely absent. In this case, all the flora is represented by pathogenic microorganisms, which leads to an increase in the number of cells — they found over 50, many leucocytes. At 3 and 4, the degree of purity of the vagina, the woman needs treatment.

Task №4. Microscopy and draw the smears, determine their morphological and tinctorial properties. picturei, characteristics of the studied microorganisms and the names of the nutrient media for their cultivation fill in in Appendix 1, "Pathogens of opportunistic and hospital infections" (columns 6d, 6e).

Teacher's signature _____

Date: _____

Practical lesson № 22

Topic: Nosocomial infections.

Self-assessment tasks

a) Questions list: theoretical issues:

1. Nosocomial (hospital) infections. Definition. Classification. Conditions that promote their occurrence and widespread distribution in hospitals.
2. Microorganisms that most commonly cause nosocomial infection (Staphylococci, Streptococci, Proteus, Serratia, Salmonella, Pseudomonads, Escherichia, Vibrios, Cytophaga, Bronchamella, Moraxella, Listeria, Mycobacteria, Bacteroids, Fusobacteria, Peptostreptococci, Clostridia, Mycoplasma, fungi of the genus Candida, etc.).
3. The most common pathology - wound infections, purulent-inflammatory processes of the skin, fatty tissue, respiratory system, central nervous system, gastrointestinal tract, urinary system, eyes, ears, sepsis, septicemia.
4. Etiology, pathogenesis, clinical forms of hospital infection caused by obligate-pathogenic microbes (nosocomial toxic septic salmonellosis, hospital colienteritis, hepatitis B, adenoviral conjunctivitis, local and generalized cytomegaloviral infections and herpes infections).
5. Opportunistic iatrogenic infections. Etiological structure.
6. Hospital strains and ecovars of opportunistic microbes.
7. Opportunistic infections associated with medical intervention. Features of immunity.
8. Conditions for a successful diagnosis of nosocomial infections.
9. Criteria for the etiological role of microorganisms isolated in the bacteriological diagnosis of nosocomial infections.
10. Prevention of nosocomial infections.
11. The problem of nosocomial staphylococcal infection in pediatric practice.
12. Detection of pathogenic staphylococcus carriers in persons working in children's institutions.
13. Detection of hemolytic streptococcus carriers in a staff of child care institutions.
14. Nosocomial outbreaks of salmonellosis typhimurium in children.

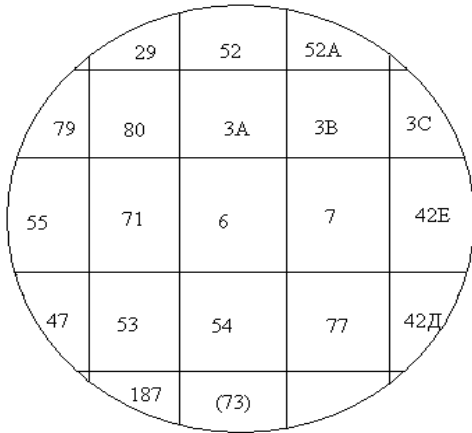
b) The list of practical skills and abilities:

1. Be able to identify bacteria phage-type.
2. Be able to determine the sensitivity of microorganisms to antibiotics.
3. Compliance with rules epidemiological regime and safety in bacteriological laboratories.
4. inoculation loop pathological material on solid culture medium.
5. Decontamination of infected material, antiseptic hand, the investigated material or contaminated culture microbes.
6. Microscopic preparations in the light microscope with immersion lens.
7. Differentiation of organisms based on morphological characteristics.

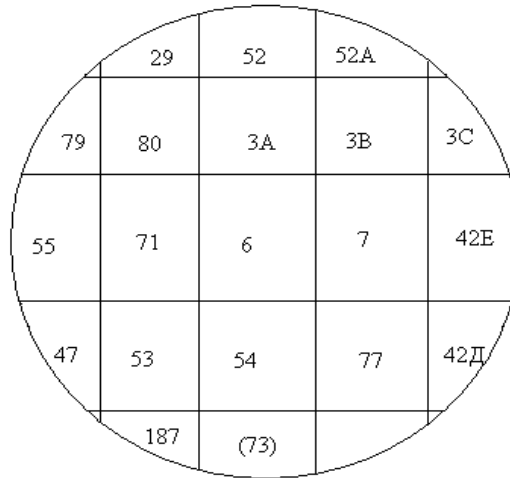
Practical lesson protocol

Practical assignments

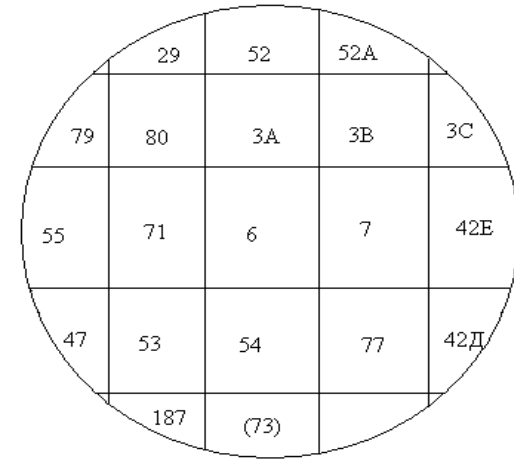
Task №1. Make phagotyping of the Staphylococci cultures: 1)- from a patient; 2), 3) -from medical workers of the surgical department. Denote phagogroups (look at the note) and conclude.



1



2



3

Note:

The first group is lysed by phages: 29, 52, 52A, 79, 80;

The second group is lysed by phages 3A, 3B, 3C, 55, 71;

The third group is lysed by phages 6, 7, 42E, 47, 53, 54, 75, 77;

Fourth group is lysed by phages 42 D;

The mixed group is lysed by phages 187 (73).

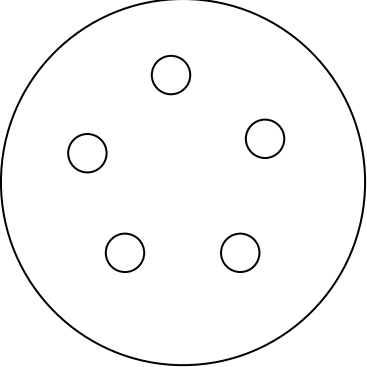
The first group includes pathogenic Staphylococci (are isolated in the conditions of furunculosis, osteomyelitis, and phlegmon).

The second group includes conditionally-pathogenic Staphylococci (isolated from superficial skin lesions, in the conditions of subacute and chronic processes, in the case of quinsy, cystitis).

The third group includes saprophytic Staphylococci.

Conclusion: _____

Task №2. Observe the antibiotic susceptibility test, which has done with a selected strain of staphylococcus. Mark on the picture the areas with an absence of growth. Fill in the results in the table. Conclude.

				
	№ p/p	The names of antibiotics	Diameter of the areas with the absence of the growth (mm)	Susceptibility
	1			
	2			
	3			
	4			
	5			
Conclusion: _____ _____				

Task №3. Microscopy and draw the smears, determine their morphological and tinctorial properties. picture, characteristics of the studied microorganisms and the names of the nutrient media for their cultivation fill in in Appendix 1 "Pathogens of opportunistic and hospital infections" (column 6f).

Teacher's signature _____

Appendix 1. Pathogens of opportunistic and hospital infections

1 №	2 Agent	3 Media	4 Picture	5 Morphological and tinctorial properties	6 (organs and tissues that are affected)					
					a Respiratory system	b Central nervous system	c Blood	d Digestive system	e Genital and urine system	f Postoperative wounds
1	S. aureus									
2.	S. eridermidis									
3.	S. pyogenes									
4.	S. pneumoniae									
5.	N. meningitidis									
6.	Pseudomonas aeruginosa									
7.	E. coli									
8.	Salmonella									
9.	Shigella									
10.	Proteus									
11.	Prevotella									
12.	Enterobacter									
13.	Serratia									
14.	Klebsiella pneumoniae									
15.	Actinomyces									

1	2	3	4	5	6					
					a	b	c	d	e	f
16.	M. tuberculosis									
17.	S. septicum									
18.	S. ramosum									
19.	Bacteroides fragilis									
20.	Moraxella catarrhalis									
21.	Haemophilus influenzae									
22.	Chlamydia psittaci									
23.	Legionella pneumophila									
24.	Mycoplasma pneumoniae									
25.	Pneumocystis carinii									
26.	Pasteurella multocida									
27.	Acinetobacter calcoaceticus									
28.	Listeria monocytogenes									
29.	Cryptococcus neoformans									
30.	Nocardia asteroides									

Date: _____

Practical lesson № 23

Topic: Sanitary microbiology

Self-assessment tasks

a) Questions list: theoretical issues:

1. The value of sanitary microbiology in the professional activity of a doctor. Tasks and methods of microbiological research.
2. Direct methods for detecting pathogens in environmental objects and indirect methods of sanitary-microbiological research. Microbial number.
3. Characteristics of the sanitary-indicative microorganisms. Sanitary-indicative microorganisms (SIM) for soil, water and air. Terms and conditions for the survival of pathogenic microbes in the environment.
4. Sanitary microbiology of water. Methods of sanitary-bacteriological research of water. Determination of microbial number. Determination of bacteria - indicators of faecal contamination: coli-index and coli-titer (by membrane filter method and fermentation method).
5. Varieties of *Escherichia coli* and the question of their sanitary significance. Faecal coliform bacteria (FCFB) of the *Escherichia coli* group as the indicators of fresh faecal contamination.
6. The role of water in the transmission of infectious agents.
7. Sanitary microbiology of the soil. Soil microbiology to infection prevention. Pathogenic microorganisms are detected in the soil. Microbes for which soil is a natural biotope. Microbes that get into the soil with human and animal faeces.
8. Methods of sanitary-microbiological research of soil. Factors that affect the qualitative and quantitative composition of soil microbes. Microbial number, coli-titer, soil perfringens-titer.
9. Sanitary microbiology of air. The role of air in the transmission of infectious diseases causative agents. Methods for determining the microbial number of air. Factors that affect microbial composition.
10. Methods of sanitary-bacteriological examination of air (sedimentation and aspiration). Assessment of the sanitary condition of the indoors by the general microbial contamination and the presence of sanitary indicator bacteria - SIB (staphylococci, α and β - hemolytic streptococci) are indicators of air contamination with the microflora of the human nasopharynx.
11. The role of the air environment in the spread of pathogens of respiratory viral infections. Methods for air sampling and an indication of respiratory viruses.
12. Sanitary-virological criteria for the assessment of water, soil and indoors.
13. Principles of sanitary-microbiological research of food.

b) The list of practical skills and abilities:

1. Sanitary and bacteriological examination of children's institutions and child care items.
2. The value of the air microflora for maternity wards and newborns' chambers.
3. The sanitary value of staphylococci and hemolytic streptococci in the environment of children's institutions.
4. Sanitary and bacteriological research of children's food products: milk, milk products and dairy products

Practical lesson protocol

Practical assignments

Task №1. Determine the drinking-water microbe number.

Conclusion: _____

Task №2. Observe and evaluate the results the coli-index and the coli-titer of the drinking-water revealed by fermentation method. Conclude.

The number of positive results from the analysis of water			ECGB- index (Coli-index)	Coli-titer
three bottles of 100 cm ²	three tubes of cm ²	three tubes of 1 cm ²		
0	0	0	< 3	< 333
0	0	1	3	333
0	1	0	3	333
1	0	0	4	250
1	0	1	7	143
1	1	0	7	143
1	1	1	11	91
1	2	0	11	91
2	0	0	9	111
2	0	1	14	72
2	1	0	15	67
2	1	1	20	50
2	2	0	21	48
2	2	1	28	36
3	0	0	23	43
3	0	1	39	26
3	0	2	64	16

Conclusion: _____

Task №3. Determine the drinking-water coli-index and coli-titer revealed by the membrane filter technique. Conclude.

Conclusion: _____

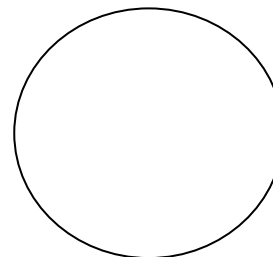
Task №4. Determine the soil microbe number.

Conclusion: _____

Task №5. Determine the common microbe number of classroom air by sedimentation technique.

Conclusion: _____

Task №6. Make a smear from yoghurt, and stain by Gram's method. Microscopy, draw.



Denote the morphological and tinctorial properties of the detected microorganisms

Teacher's signature _____

Date: _____

Practical lesson № 24

Topic: Final computer test control of the students' knowledge

You will pass the exam in the computer class using the test tasks of the licensing exam "Krok 1" to the corresponding module and discipline in general.

Date: _____

Practical lesson № 25

Topic: Examination of practical skills

Look on the University website for the list of the questions.

The final semester attestation of discipline

Look on the University website for the list of the questions.