



PROBLEM BASED MICROBIOLOGY

**Module Infection:
MICROBIOLOGY
HANDBOOK**

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PROBLEM BASED MICROBIOLOGY

Module Infection: Microbiology



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NOTICE

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PREFACE

During the first couple of years, after adoption of Problem Based Learning (PBL) in Lithuanian University of Health Sciences (LUHS) a mild disorientation of students' attending the Tutorial sessions, Practical Works, Lectures & Seminars have been observed.

This book is aimed to help medical students to obtain the knowledge in Medical Microbiology, Virology, Mycology and Immunology science and navigate them through the PBL – Microbiology studies in the most comprehensive and organised way.

The book is divided into the sections. The “Tutorials” present interesting and challenging clinical cases making the study of medical microbiology and infectious diseases enjoyable. The “Lectures” enhance the body of knowledge and position every bit of information into the practice and self-directed learning. Each “Practical work” includes models of clinical situations which enable students to develop a working knowledge of the variety of microorganisms involved into the infectious maladies. The “Seminars” encourage and direct the students to read more widely about the subjects. A guide to using up-to-date molecular biology and immunological techniques for the undergraduates is presented in “Laboratory diagnosis” section.

An “Abbreviations” and the extensive “Glossary” of medical terms which are frequently used in medical practice is available at the end of the book.

Written by three MD-microbiologists and experienced senior University lecturers at the same time - this book is tailored specifically for medical students and is an invaluable for junior doctors, primary care physicians and all other health care professionals who deal with medical microbiology and infectious diseases on a daily basis.

We hope that we succeeded in this and that our readers find this book useful.

Authors

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TUTORIALS

4.1. First problem. Respiratory Tract Infections

The patient was a 3-year-old male who presented with a 4-day history of fevers. He became acutely ill and vomiting during lunch. Over the next 4 days he developed fevers as high as 40°C that were controlled by Tylenol. He also developed cough, rhinorrhea, and conjunctivitis. He appeared to be fatigued, and his parents reported that he was “very sleepy”. Over the past two days, his eyes began to itch and were painful. His parents noted that his eyes were puffy and he was sensitive to light. He had no rashes. The patient’s lips were dried and cracked, and he had a greatly reduced urinary output.

Other history pertinent to his illness is that he attended preschool twice per week, where he had multiple sick contacts. His 1-year old sibling had otitis, some wheezing, vomiting, and a productive cough.

On physical examination he had a temperature of 38.6°C pulse rate of 126/min, and respiratory rate of 28/min. The significant findings included bilateral conjunctivitis with exudate in the left eye, bleeding, cracked lips, and rhinorrhea. He had shotty lymphadenopathy, but no rash. His feet were slightly edematous. His respiratory examination was normal. Laboratory findings were all normal. A nasopharyngeal swab was sent for rapid antigen testing for RSV and influenza A virus.

Questions:

- *What is the most likely diagnosis of this patient? What symptoms does he have which are consistent with his illness?*
 - *How do you think this child became infected?*
 - *What is the agent causing his infection? What are the key virulence factors of this agent?*
 - *What is the usual outcome of this infection in this patient population?*
 - *The finding of conjunctivitis in this child made the paediatrician examining him attempt to elicit a history of rash. What was the clinical syndrome that this physician was concerned about in this child given his clinical presentation?*
 - *The infection this child has a vaccine preventable. Briefly describe the vaccine that is used to prevent this disease. How this vaccine currently employed in the industrialized world?*
-

The Aim:

- Knowledge of the bacteria and viruses involved into the Respiratory Tract Infections (RTI): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning:

- *Bacteria (H. influenza, M. catarrhalis, M. pneumonia, S. pneumoniae.):* microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Viruses (Influenza viruses, Adenoviruses, Rhinoviruses, RSV, VZV):* virology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

TUTORIALS

4.2. Second problem. Skin and Soft Tissue Infections

The patient was a 45-year-old male who was in his usual state of good health when he awoke at 3.am with pain in the lateral aspect of his left calf. He looked at his calf and thought that the pain was due to an ingrown hair and went back to sleep. At 10am, he expressed a small amount of pus from the ingrown hair. Over the next 8h, the patient developed an area of cellulitis on the lateral aspect of the calf of approximately 5 to 10 cm. At that time, a small amount of pus was again extended from just below the knee to just above the ankle. The patient visited his physician. His vital signs at that visit, including pulse, respirations, blood pressure, and temperature, were all within normal limits.

Physical exam was significant for an area of obvious fluctuante. No lymphadenopathy was observed. The central area of the cellulitis, near the area that the patient described as where the ingrown hair had been, was punctured three times, but no pus was drained. The patient was referred to the surgery service. The surgeons examined the patient and said they would follow him. The patient was given 2g ceftriaxone i/m and begun on oral cephalixin.

The patient returned to the surgical clinic 48h later with an obvious area of fluctuance in the centre of the area of cellulitis. Over the preceding 48h, the patient reported low-grade fevers. Approximately 1ml of pus was aspirated and was sent to the laboratory. Gram stain revealed gram-positive cocci in clusters, and yellowish β -hemolytic colonies on blood agar. When pus was aspirated from the lesion, the surgeon decided to excise and drain the lesion.

Questions:

- *What is the most likely diagnosis of this patient?*
 - *What is the organism most likely causing this patient's infection? What other type of infections does this organism frequently cause?*
 - *How do you think he became infected with this organism?*
 - *Why were incision and drainage necessary to treat this infection? Why would antimicrobial agents alone not be effective in treatment of this infection?*
 - *A new drug resistance problem is emerging with the organism infecting this patient. What is it? Is this patient at risk for becoming infected with a strain of this organism?*
-

The Aim:

- Knowledge of the microorganisms involved into the Skin, Soft tissue, Bone & Joint infections: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning:

- *Bacteria (S. aureus, S. pyogenes, H influenza, E. aerogenes, M. tuberculosis):* microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Fungi (B. dermatitidis, C. neoformans):* mycology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

TUTORIALS

4.3. Third problem. Sexually Transmitted Diseases

The patient was a 20-year-old female who presented to the emergency room with a 4-day history of fever, chills, and myalgia. Two days prior to this she had noted painful genital lesions. On the day of admission she developed headache, photophobia, and a stiff neck. Previously, she had been in good health. She admitted to being sexually active, but had no history of sexually transmitted diseases.

On physical examination, she was alert and oriented. Her vital signs were normal except for a temperature of 38.5°C; pulse rate was 80/min, and blood pressure was 130/80. A general examination was unremarkable except for slight nuchal rigidity. Her throat was clear, and there was no lymphadenopathy. A pelvic examination revealed extensive vesicular and ulcerative lesions on the left labia minora and majora with marked edema. The cervix had outward-growing necrotic ulcerations. Specimens were taken to culture *Neisseria gonorrhoeae*, viruses, and *Chlamydia trachomatis*.

General laboratory tests were unremarkable. The VDRL test was negative. A lumbar puncture was done. The opening pressure was normal. The CSF showed a mild pleocytosis with leucocyte count of 41/μl with 21% polymorphonuclear leucocytes and 79% mononuclear cells, glucose level 2.6mmol/l, and a protein level of 0.06g/l. The CSF VDRL test was negative. A rapid diagnostic test was done on the genital lesion and gave positive results. Cultures from the genital lesions and CSF verified the diagnosis 2 days later. By that time the patient's condition had improved after 2 days of intravenous therapy. She was discharged home on oral medication.

Questions:

- What is the most likely diagnosis of this patient?
 - What is the differential diagnosis of ulcerative genital lesions?
 - Which rapid test was used so that specific therapy could be started?
 - Which complication of her underlying illness she developed?
 - There are two different serotypes of the agent causing her infection. What similarities do they share and what are the differences between these agents?
-

The Aim:

- Knowledge of the microorganisms involved into the Sexually Transmitted Diseases (STD): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning:

- Bacteria (*N. gonorrhoeae*, *C. trachomatis*, *H. ducreyi*, *M. genitalium*, *T. pallidum*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Viruses (*HSV*, *HIV*, *Epstein Barr virus (HHV-4)*): virology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

TUTORIALS

4.4. Fourth problem. Gastrointestinal Tract Infections

The patient was a 4-year-old male who resented to emergency room with a 2-hour history of vomiting, diarrhea, fever, irritability, and lethargy.

In the emergency room, he had two episodes of vomiting. He had a temperature of 38.9°C, pulse rate of 160/min, and respiratory rate of 36/min, and he noted to be dehydrated.

His stool contained bloody streaks; A CSF examination was within normal limits; a peripheral blood white blood cell (WBC) count of 13.2×10^9 /liter with 85% neutrophils; a negative blood culture; and a negative stool examination for parasites. MacConkey agar plate shows the colonial morphology and inability to ferment lactose. The triple sugar iron (TSI) agar slant shows an organism that does not ferment sucrose or lactose and does not produce H₂S. In the urea-motility-indole (UMI) tube, the isolate is non-motile and urea and indole are negative.

Questions:

- *What is the most likely diagnosis of this patient?*
 - *What microorganism is likely causing illness in this patient?*
 - *Describe the pathogenesis of this organism*
 - *Describe the epidemiology of this organism. What special characteristics of this organism lead to its spread?*
 - *What would be the appropriate treatment strategy for this child?*
-

The Aim:

- Knowledge of the microorganisms involved into the Gastrointestinal Tract Infections (GI): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning:

- Bacteria (*E. coli* (EHEC, EPEC, ETEC, EIEC), *Salmonella spp.*, *Shigella spp.*, *C. jejuni*, *Y. enterocolitica*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Viruses (*Rotavirus*, *Norovirus*): virology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

TUTORIALS

4.5. Fifth problem. Central Nervous System Infections

This 19-year-old student was in his usual state of health until the evening prior to admission, when he went to bed with a headache. He told his mother that he felt feverish, and on the following morning his mother found him in bed, moaning and lethargic. He was brought to the emergency room, where he appeared toxic and drowsy oriented.

His temperature was 40°C, his heart rate was 126/min, and his blood pressure was 100/60. His neck was supple. He had an impressive purpuric rash, not blanching, most prominent on the trunk, legs, and wrists. A Gram stain of a skin lesion revealed gram-negative diplococci. His white blood cell (WBC) count was $26 \times 10^9/l$ with 25% band forms. The platelet count was $80 \times 10^9/l$.

Blood cultures were obtained, a lumbar puncture was performed, and the patient was begun on intravenous ceftriaxone. CSF glucose, protein, and white blood cell (WBC) count were normal, and CSF bacterial culture was negative. Blood cultures grew the gram-negative diplococcus.

Questions:

- *What is the most likely diagnosis of this disease?*
 - *What organism was causing this patient's illness? Is the finding of a normal SCF profile, without evidence of meningitis, commonly observed in infection with this organism?*
 - *Is this organism ever part of the normal oropharyngeal flora?*
 - *Which immunologic abnormalities predispose individuals to infection with this organism?*
 - *Which serogroup(s) causes illness?*
 - *Which prophylactic strategies are useful for large populations and for exposed individuals?*
 - *What types of antibiotic resistance have been found in this organism?*
 - *What is a purpuric rash, and which virulence factors play a central role responsible for its appearance?*
-

The Aim:

- Knowledge of the microorganisms involved into the CNS Infections: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning:

- *Bacteria (N. meningitidis, N. gonorrhoeae, S. pneumoniae, H. influenza (Hib), E. coli, L. monocytogenes, P. aeruginosa, K. pneumoniae):* microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Viruses (HSV, Polioviruses, HIV, Coxsackievirus):* virology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Fungi (Aspergillus spp., C. neoformans):* micology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

TUTORIALS

4.6. Sixth problem. Emerging Infectious Disease

The patient was an 8-year-old male with a 2-day history of diarrhea. He presented with worsening diarrhea (14 movements that day) which had become bloody. He also complained of pain on defecation. He had vomited once. He had attended a cookout 6 days previously. He claimed that his mother made him eat a hamburger which was “pink inside” even though “he did not like it”.

His physical examination was benign except for obvious dehydration. The laboratory findings were significant for a white blood cells (WBC) count of $13 \times 10^9/l$ with $9.7 \times 10^9/l$ neutrophils, a methylene blue stain of feces that showed abundant polymorphonuclear cells, and a positive stool guaiac. He was treated with trimethoprim-sulfamethoxazole and i/v fluid therapy for dehydration. He quickly improved and was discharged within 24h. Colonies of his stool specimen on sorbitol-MacConkey (SMAC) agar were clear and colorless.

Questions:

- *What is the most likely diagnosis of this disease?*
 - *What is the most likely etiologic agent of his infection? What are the major virulence factors produced by this organism and what are their roles in the pathogenesis of disease?*
 - *Why are these organisms so difficult to detect in feces?*
 - *Besides cultures, what other methods could be used to detect this organism?*
 - *Was using antibiotic therapy in this patient an appropriate clinical decision?*
-

The Aim:

- Knowledge of the microorganisms involved into the Diarrhea: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning:

- *Bacteria (E. coli (EHEC, EPEC, ETEC, EIEC), Salmonella spp., Shigella spp., C. jejuni, Y. enterocolitica, S. aureus, Vibrio spp.):* microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Viruses (Enteroviruses, Calicivirus, Rotavirus, Norovirus):* virology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Table of Normal Values			
Glucose (serum)	3.3-5.5 mmol/l	Osmolality (serum)	280-300 mmol/kg
Cholesterol (serum)	<5.2 mmol/l	Total serum protein	60-80 g/l
Folic (Folate)	7-36 nmol/l	Albumin	35-50 g/l
Low density lipoprotein (LDL)	<3.37 mmol/l	Blood urea nitrogen (BUN)	7-18 mmol/l
High density lipoprotein (HDL)	>0.9 mmol/l	Lactate dehydrogenase (LDH)	95-195 U/l

	Female	Male	Newborn	Age ≥1
ALT	7-30 U/l	10-52 U/l	1-25 U/l	16-36 U/l
AST	9-26 U/l	11-40 U/l	18-74 U/l	20-60 U/l
Creatinine	53-107 μmol/l	70-130 μmol/l	<90 μmol/l	18-36 μmol/l
Creatinine kinase	30-125 U/l	40-200 U/l	22-267 U/l	0-47 U/l
Alkaline phosphatase	42-98 U/l	53-128 U/l	60-275 U/l	115-460 U/l

Blood gas		Electrolytes	
pH	7.35-7.45	Potassium (K ⁺)	3.5-5.5 mmol/l
p _a CO ₂ (arterial)	35-45 mm Hg	Sodium (Na ⁺)	135-145 mmol/l
p _a O ₂ (arterial)	85-105 mm Hg	Chlorine (Cl ⁻)	96-106 mmol/l
S _a O ₂ (arterial)	96-100%	Calcium (Ca ²⁺)	2.3-2.7 mmol/l
Bicarbonate (HCO ₃ ⁻)	24-30 mmol/l	Magnesium (Mg ²⁺)	0.8-1.2 mmol/l
Anion gap (AG) ³	3-11 mmol/l	Phosphate (PO ₄ ³⁻)	1.0-1.4 mmol/l

Red blood cells	Female	Male	White blood cells	
Red blood cells (RBC)	4.5-5.2 10 ¹² /l	4.5-5.9 10 ¹² /l	White blood cell count (WBC)	4-12 × 10 ⁹ /l
Hemoglobin (Hb) ¹	120-160 g/l	140-180 g/l	Segmented neutrophils	2-7 × 10 ⁹ /l
Hematocrit (Hct) ¹	36-46%	41-53%	Band neutrophils	<0.7 × 10 ⁹ /l
Erthyrocyte sedimentation rate (ESR) ²	< 30 mm/h	<20 mm/h	Basophils	<0.10 × 10 ⁹ /l
			Eosinophils	<0.45 × 10 ⁹ /l
			Lymphocytes	1.5-3.4 × 10 ⁹ /l
			Monocytes	0.14-0.86 × 10 ⁹ /l
			Platelet count	150-350 × 10 ⁹ /l

¹Hb <100g/l or Hct <30% sign of anemia
²usually calculated by age: ESR (female)=0.5×age, ESR (male)=0.5×(age+10)

Vital Signs (VS)	
Body temperature (T)	36.6°C
Pulse (P) [*]	60-100/min
Respiratory rate (R) [*]	9-18/min
Blood pressure [*] (BP)	90-150/50-90

^{*}different for infants and children

Age	P	R
Newborn	100-160/min	30-50/min
0-5 months	90-150/min	25-40/min
6-12 months	80-140/min	20-30/min
1-5 years	80-120/min	20-30/min
6-14 years	60-110/min	12-30/min

LECTURE

5.1 Normal Microbial Flora of Human Body

Objectives:

- Understand the development of Natural Microbial Flora (NMF) during the age and the establishment of NMF in the human body (colonization); also it's role in the ethiopathogenesis of infections
- Know the differences between the permanent (autochthonous) and transient (allochthonous) microflora
- Understand natural mechanisms on mucosal surfaces and skin that safeguard natural microflora
- Know the NMF of the skin, mouth, intestinal, genitourinary and respiratory tracts
- Understand the role of NMF in the pathogenesis of opportunistic infections
- Understand the Dysbiosis
- Know the differences between Probiotics & Prebiotics

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- The Normal Microbial Flora (NMF): Permanent (autochthonous) and Transient (allochthonous) and its role in physiological and pathological processes
- The development of NMF during the age
- The establishment of NMF in the human body (colonization) and it's role in the ethiopathogenesis of infections
- Natural mechanisms on mucosal surfaces and skin that safeguard natural microflora
- NMF of the skin, mouth, intestinal tract, genitourinary tract and respiratory tract
- NMF - secondary and opportunistic pathogens
- Dysbiosis
- Probiotics
- Prebiotics

LECTURE

5.2. Infection & Infectious process & Infectious disease. Bacterial Pathogenicity

Objectives:

- Understand the Host-Microbial Relationship
- Understand the Differences between The Infectious Process & Infectious Disease
- Understand the Development of Infectious Disease
- Know the Properties of Infectious Disease
- Understand the Pathogenesis of Viral Diseases
- Know the Microbial Mechanisms of Pathogenicity
- Understand the Epidemiology & Know the Control of Communicable Diseases

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Host-Microbial Relationship
- Development of Infectious Disease (ID):
 - Incubation Period (ID)
 - Prodrom Period
 - Period of Illness
 - Period of Convalescence
- Koch's Postulates
- Infections: Acute, Asymptomatic, Reinfection, Superinfection, Reccurent, Persistent
- Infections: Local & Systemic
- Infections: Exgoneous & Endogenous
- Microbial Mechanisms of Pathogenicity:
 - Portals of entry
 - Numbers of microorganisms
 - Adherence
 - Penetration & Invasiveness
 - Damage of the host cells
- Toxins: Exotoxins & Endotoxins
- Pathogenesis of Viral Diseases
- Hospital (acquired) Infections & Hospital Strains
- Epidemiology & Control of Communicable Diseases

LECTURE

5.3. Mechanisms of Bacterial Resistance to Antimicrobial Drugs

Objectives:

- Know the General Characteristics of Antimicrobial Drugs
- Know the Mechanism of Action of Antimicrobial Drugs
- Know the Mechanisms of Bacterial Resistance
- Know the Test for Antibiotic Sensitivity

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Antimicrobial Agents: Bactericidal & Bacteriostatic
- Antimicrobial Agents: Broad spectrum & Narrow spectrum
- Mechanism of Action of Antimicrobial Drugs:
 - Inhibition of cell wall synthesis
 - Inhibition of protein synthesis
 - Inhibition of nucleic acid synthesis
 - Alteration of cell membrane
 - Inhibition of synthesis of essential metabolites
- Antibiotic Resistance: Natural & Acquired
- Antimicrobial Drug Resistance:
 - Altered target site
 - Altered permeability
 - Inactivation
 - Replacement of a sensitive pathway
- Multiple Drug Resistance
- Antibiotic susceptibility testing (*in Vitro*) : MIC & MBC
- Antibiotic susceptibility testing:
 - Disk Diffusion Test
 - Kirby-Bauer Disk Test
 - E-test

LECTURE

5.6. Cocci: Gram - positive & Gram – negative

Objectives:

- Know the *Staphylococcus spp.* (*S.aureus*, *S.epidermidis*, *S. saprophyticus*, *MRSA*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Streptococcus spp.* (*S.pneumoniae*, *S.pyogenes*, *S.agalactiae*,) : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Enterococcus spp.* (*E. faecalis*, *E.faecium*, *VRE*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Neisseria spp.* (*N. meningitidis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Reading Assignment:

*these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- *Staphylococcus spp.*:
 - Normal flora (NF)
 - Suppurative - pyogenic infections
 - Systemic infections
 - Toxin-mediated infections
 - Methicillin Resistant *S. aureus* (MRSA)
 - Vancomycin Resistant *S. aureus* (VRSA)
- *Streptococcus spp.*:
 - Normal flora (NF)
 - Suppurative - pyogenic infections
 - Nonsuppurative diseases
 - *Viridans streptococci* - associated endocarditis
 - *S.pneumonia & bacterial pneumonia*
- *Enterococcus spp.*:
 - Infections: septicaemia, endocarditis, Urinary tract Infections (UTI)
- *Neisseriae spp.*:
 - *Gonorrhoea*
 - *Bacterial meningitis*

LECTURE

5.7. Enterobacteriaceae

Objectives:

- Know the *Escherichia spp.* (*E. coli*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Salmonella spp.* (*S. typhi*, *S. enteritidis*, *S. schottmulleri*, etc.): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Shigella spp.* (*S. dysenteriae*, *S. flexneri*, *S. boydii*, *S. sonnei*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Yersinia spp.* (*Y. pseudotuberculosis*, *Y. pestis*, *Y. enterocolitica*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Klebsiella spp.* (*K. pneumonia*, *K. oxytoca*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Enterobacter spp.* (*E. aerogenes*, *E. cloacae*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Proteus spp.* (*P. vulgaris*, *P. mirabilis*, *P. penneri*, *P. hauseri*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Serratia spp.* (*S. marcescens*, *S. ficaria*, *S. fonticola*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Reading Assignment:

*these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Primary pathogens
- Secondary (opportunistic) pathogens
- *Enterobacteria* (bacteria of the intestines):
 - Virulence factors
 - Endotoxin-mediated toxicity
 - Suppurative - pyogenic infections
 - Gastrointestinal tract infections
 - Salmonellosis
 - Shigellosis
 - *E. coli* - associated gastroenteritis
- *Yersinia spp.*
 - Zoonotic infections
- Laboratory diagnosis, Treatment & Prevention

LECTURE

5.8 *Vibrios, Aeromonas, Campylobacter & Helicobacter, Spirochetes*

Objectives:

- Know the *Vibrio spp. (V. cholerae)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Campylobacter spp.(C. jejuni)* : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Helicobacter spp. (H. pylori)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Aeromonas spp. (A. hydrophila)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Treponema spp. (T. pallidum)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Borrelia spp. (B. burgdorferi)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Leptospira spp.(L.interrogans, L.noguchii)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- *Vibrionaceae* family
 - *V. cholerae*: biotypes, serotypes, virulence factors
 - Cholera
 - Laboratory diagnosis, treatment, prevention
- *Campylobacter* genus
 - Gastroenteritis
 - Septicemia
 - Antropozoonoses & zoonoses
- *Helicobacter pylori*
 - Gastritis, peptic ulcer, gastric carcinoma
- *Spirochetes*
 - Syphilis
 - Lyme Disease
 - Weil's Disease

LECTURE

5.9 *Pseudomonadaceae* & *Pasteurellaceae*

Objectives:

- Know the *Pseudomonas* spp. (*P. aeruginosa*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Burkholderia* spp. (*B. cepacia*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Stenotrophomonas* spp. (*S. maltophilia*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Haemophilus* spp. (*H. influenzae*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Pasteurella* spp. (*P. multocida*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Actinobacillus* spp. (*A. hominis*, *A. actinomycetemcomitans*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Reading Assignment:

*these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- *Pseudomonadaceae* family
 - *P. aeruginosa*
 - Hospital infections
 - Antibiotic resistance
- *Pasteurellaceae* family:
 - *H. influenzae*: biotypes (I-VIII), serotypes (a-f), biogroups
 - *H. influenzae* Type b
 - Hib vaccine
 - *P. multocida*
 - Pasteurellosis
 - *A. actinomycetemcomitans*
 - Periodontal diseases

LECTURE

5.10. *Mycobacteria, Corynebacteria, Bordetella & Listeria*

Objectives:

- Know the *Mycobacterium spp.* (*M. tuberculosis*, *M. avium*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Corynebacterium spp.* (*C. diphtheriae*) : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Bordetella spp.* (*B. pertussis*, *B. parapertussis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Listeria spp.* (*L. monocytogenes*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- *Mycobacteriaceae* family
 - *M. tuberculosis* Complex (MTBC)
 - Epidemiology
 - Tuberculosis
 - Multi-drug Resistance
 - Laboratory diagnosis
 - Tuberculin skin test (Mantoux test/PPD)
 - BCG (Tuberculosis) vaccine
 - *M. avium* Complex (MAC)
 - *Nontuberculous mycobacteria* (NTM)
- *Corynebacteriaceae* family:
 - *C. diphtheria*
 - Diphtheria
 - DTP (DTwP) vaccine
- *Bordetella pertussis* & Whooping cough
- *Listeria monocytogenes* & Listeriosis (zoonosis)

LECTURE

5.11. *Mycoplasmataceae* & *Chlamydiaceae*; *Anaplasma*, *Ehrlichia* & *Coxiella*

Objectives:

- Know the *Mycoplasma* spp. (*M. pneumoniae*, *M. genitalium*, *M. hominis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Ureaplasma* spp. (*U. urealyticum*) : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Chlamydia* spp. (*C. trachomatis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Chlamydophila* spp. (*C. pneumoniae*, *C. psittacii*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Anaplasma* spp. (*A. phagocytophilum*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Ehrlichia* spp. (*E. ewingii*, *E. chaffeensis*) : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Coxiella* spp. (*C. burnetii*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Reading Assignment:

*these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Intracellular parasitism
- *M. pneumoniae* & atypical pneumonia
- *M. genitalium* & pelvic inflammatory disease
- *U. urealyticum* & non-specific urethritis (NSU), meningitis, infertility
- *C. trachomatis* & trachoma & lymphogranuloma venereum (LGV)
- *C. pneumoniae* & pneumonia (possibly atherosclerosis)
- *C. psittacii* & pneumonia (psittacosis)
- *A. phagocytophilum* & human granulocytic anaplasmosis
- *E. ewingii* & human ewingii ehrlichiosis
- *E. chaffeensis* & human monocytic ehrlichiosis
- *C. burnetii* & Q fever
 - Q-vax vaccine

LECTURE

5.12. *Viruses*

Objectives:

- Know the Virus Classification: ICTV, Baltimore & Holmes
- Know the *DNA viruses* (*Adenovirus, Papillomavirus, HSV-1 & HSV-2, Smallpox virus, HBV, etc.*): virology, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *RNA viruses* (*Rubella virus, Influenzavirus A, B, C, measles virus, Mumps virus, Rabies virus, HEV, Rotavirus, etc.*) : virology, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the *Oncogenic viruses* (*Papillomavirus, Hepatitis B virus, HTLV-1, etc.*) : virology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- Know the Prions & understand the Slow virus infections

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Virus Classification:
 - ICTV classification
 - Baltimore classification
 - DNA viruses
 - RNA viruses
 - Reverse transcribing viruses
 - Holmes classification
- Oncogenic viruses:
 - *Papillomavirus* (some) & uterine (cervical) cancer
 - *Hepatitis B virus* & Liver cancer
 - HTLV-1, HTLV-2 & Adult T-cell leukemia
 - FeLV & feline leukemia
- Conventional viruses & slow viral infections:
 - Progressive multifocal leukoencephalopathy (PML)
- Unconventional viruses (Prions) & slow viral infections
 - Creutzfeldt-Jakob disease (CJD)

LECTURE

5.13. Septicemia & Bacterial Endocarditis

Objectives:

- Know the causative agents of septicaemia: *Staphylococcus spp*, *Enterobacteriaceae*, *Pseudomonas spp.*, *H. influenzae*, *Salmonella spp.*, *N. meningitidis*, *L. monocytogenes*, *Mycobacterium spp.* & viruses: *HIV*, *EBV*, *CMV*, *Hepatitis viruses*
- Understand the Systemic Inflammatory Response Syndrome (SIRS)
- Understand the pathogenesis of Septic Shock
- Know the causative agents of Bacterial Endocarditis: *S. sanguis*, *S. oralis*, *S. mitis*, *S. aureus*, *Gram-negative bacteria* & Microbial diagnosis of Infective endocarditis

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Bacteremia
- Septicemia
- Causative bacteria of septicaemia: *Staphylococcus spp*, *Enterobacteriaceae*, *Pseudomonas spp.*, *H. influenzae*, *Salmonella spp.*, *N. meningitidis*, *L. monocytogenes*, *Mycobacterium spp.* & viruses: *HIV*, *EBV*, *CMV*, *Hepatitis viruses*
- Systemic Inflammatory Response Syndrome (SIRS)
- Septic Shock
- Bacterial Endocarditis
- Causative agents of Bacterial Endocarditis: *S. sanguis*, *S. oralis*, *S. mitis*, *S. aureus*, *Gram-negative bacteria*
- Blood cultures & Microbial diagnosis

LECTURE

5.14. Upper Respiratory Tract Infections (URTI)

Objectives:

- Know the frequency and pathogenesis of URTIs
- Know the causative m/o of URTIs - viruses: *Rhinovirus*, *Coxsackie*, *Influenza*, *Parainfluenza*, *RSV*, *Coronavirus*, *Adenovirus*, *Echo viruses*, etc. & bacteria: *S. pneumoniae*, *S. pyogenes*, *H. influenzae*, *Bacteroides spp.*, *N. gonorrhoeae*, *C. diphtheriae*, *Borrelia vincenti*, *Prevotella spp.*, *Porphyromonas spp.*, *Fusobacterium spp.*, *Peptostreptococcus spp.* & fungi: *Candida spp.*
- Understand the pathogenesis of Pharyngitis, Tonsillitis, Acute Epiglottitis, Otitis & Sinusitis, Gingivitis, Laryngitis & Tracheitis
- Know the Microbiological tests and their practical use
- Understand the Antibioticotherapy

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Local mucosal immunity in the URT and factors for resistance
- Frequency of URTIs
- Causative m/o of URTIs: *Rhinovirus*, *Coxsackie*, *Influenza*, *Parainfluenza*, *RSV*, *Coronavirus*, *Adenovirus*, *Echo viruses*, etc.
- Pharyngitis & Tonsillitis: *S. pneumoniae*, *S. pyogenes*, *H. influenzae*, *Bacteroides spp.*, *N. gonorrhoeae*, *C. diphtheriae*, *Borrelia vincenti* & viruses: *Adenovirus*, *Rhinovirus*, *Coronavirus*, *Parainfluenza*, *Influenza*, *EV*, *HSV*, *CMV*
- Acute Epiglottitis: *H. influenzae (type b)*
- Otitis & Sinusitis: *S. pneumoniae*, *S. aureus*, *S. pyogenes*, *H. influenzae*, *P. aeruginosa*, *proteus spp.*, *Candida spp.* & viruses
- Gingivitis: *Prevotella spp.*, *Porphyromonas spp.*, *Fusobacterium spp.*, *Peptococcus spp.*, *Peptostreptococcus spp.*
- Laryngitis & Tracheitis: *Parainfluenza virus*, *S. pyogenes*
- Microbiological tests
- Antibioticotherapy: ineffective and effective, side effects and treatment recommendations

LECTURE

5.15. Lower Respiratory Tract Infections (LRTI)

Objectives:

- Know the frequency and pathogenesis of LRTIs
- Know the causative m/o of LRTIs
- Know the pathogenesis and causative agents of: Bronchiolitis (*RSV*); Bacterial pneumonia (*S. pneumoniae*, *S. aureus*, *H. influenzae*, *K. pneumoniae*, *Enterobacteriaceae*, *P. aeruginosa*, *Acinetobacter spp.* Etc.); Atypical pneumoniae (*M. pneumoniae*, *L. pneumophila*, *C. pneumoniae*, *C. psittaci*, *M. catarrhalis*); Viral pneumoniae (*RSV*, *Influenza virus*, *Parainfluenza virus*, *Adenovirus*, *VZV*, *Coronaviruses* & *Severe Acute Respiratory Syndrome (SARS)*); Pertussis (*B. pertussis*)
- Understand the importance of specimens & to know the role of Microscopy, Microbiological tests & Immunological tests
- Understand the role of Antibioticotherapy & Vaccinoprophylaxis

Reading Assignment:

*these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Local mucosal immunity in the LRTIs and factors for resistance
- Predisposing factors increasing the risk to develop LRTIs
- Causative m/o of LRTIs: *Rhinovirus*, *Coronavirus*, *Influenza*, *Adenovirus* & *M. pneumoniae*
- Bronchiolitis: *RSV*
- Bacterial pneumonia: *S. pneumoniae*, *S. aureus*, *H. influenzae*, *K. pneumoniae*, *Enterobacteriaceae*, *P. aeruginosa*, *Acinetobacter spp.* Etc.
- Atypical pneumoniae: *M. pneumoniae*, *L. pneumophila*, *C. pneumoniae*, *C. psittaci*, *M. catarrhalis*
- Viral pneumoniae: *RSV*, *Influenza virus*, *Parainfluenza virus*, *Adenovirus*, *VZV*, *Coronaviruses* & *Severe Acute Respiratory Syndrome (SARS)*
- Pertussis (Whooping cough): *B. pertussis*
- Specimen collection, Microscopy, Microbiological tests & Immunological tests
- Antibioticotherapy
- Vaccinoprophylaxis

LECTURE

5.16. Neonatal Infections

Objectives:

- Know the peculiarities of newborns immune system
- Know the causative agents of Intrauterine Infections: *CMV, Rubella virus, Measles virus, HBV, HIV, Toxoplasma, T. pallidum*
- Know the causative agents of Perinatal Infections: *Group B streptococci, L. monocytogenes, E. coli, N. gonorrhoeae, HSV, HBV*
- Know the causative agents of Postnatal Infections: *S. aureus, S. epidermidis, S. pyogenes, E. coli, M. tuberculosis, N.gonorrhoeae, C. trachomatis, CMV, RSV*
- Understand the pathogenesis of Nosocomial (hospital) neonatal infections
- Know the pathogens of Suppurative-inflammatory newborns infections
- Understand the Infections in premature infants & Know the Prophylaxis against infections in newborns & in prematures

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- The peculiarities of newborns immune system predisposing susceptibility to certain infection & the ways of transmission
- Intrauterine Infections: *CMV, Rubella virus, Measles virus, HBV, HIV, Toxoplasma, T. pallidum*
- Perinatal Infections: *Group B streptococci, L. monocytogenes, E. coli, N. gonorrhoeae, HSV, HBV*
- Postnatal Infections: *S. aureus, S. epidermidis, S. pyogenes, E. coli, M. tuberculosis, N.gonorrhoeae, C. trachomatis, CMV, RSV*
- Nosocomial (Hospital) neonatal infections
- Suppurative-inflammatory newborns infections: *N. meningitidis, Chlamydia spp., Pseudomonas spp.*
- Infections in premature infants
- Prophylaxis against infections in newborns & in prematures

LECTURE

5.17. Genital & Urinary Tract Infections (UTI)

Objectives:

- Know the local mucosal immunity in the genito-urinary tract and factors for resistance
- Learn Bacterial Urinary Tract Infections (BUTIs): pyelonephritis, cystitis, urethritis
- Know the causative m/o of UTIs: *E. coli*, *P. mirabilis*, *Klebsiella spp.*, *Enterobacter spp.*, *Serratia spp.*, *Pseudomonas spp.*, *Candida spp.*
- Know how to handle urine specimen, including transportation and bacteriological investigation
- Understand “asymptomatic bacteriuria”
- Learn the causative agents of colpitis & vaginitis: *S. aureus*, *S. epidermidis*, *S. pyogenes*, group *B streptococci*, *C. albicans etc.*
- Know the causative m/o of Sexually Transmitted Diseases (STD): *T. pallidum*, *N. gonorrhoeae*, *C. trachomatis*, *T. vaginalis*, *Papillomavirus*, *HSV*, *HIV*

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Local mucosal immunity in the genito-urinary tract and factors for resistance
- Bacterial Urinary Tract Infections (BUTIs): pyelonephritis, cystitis, urethritis
- Causative m/o of UTIs: *E. coli*, *P. mirabilis*, *Klebsiella spp.*, *Enterobacter spp.*, *Serratia spp.*, *Pseudomonas spp.*, *Candida spp.*
- Nephrogenic strains of m/o
- Urine specimen collection, transportation and bacteriological investigation
- Asymptomatic bacteriuria
- Causative agents of colpitis & vaginitis: *S. aureus*, *S. epidermidis*, *S. pyogenes*, group *B streptococci*, *C. albicans etc.*
- Sexually Transmitted Diseases (STD): *T. pallidum*, *N. gonorrhoeae*, *C. trachomatis*, *T. vaginalis*, *Papillomavirus*, *HSV*, *HIV*

LECTURE

5.18. Bacterial & Viral CNS Infections

Objectives:

- Know the Meningitis: *N. meningitidis*, *H. influenzae*, *S. pneumoniae*, Group B *Streptococci*, *E. coli*, *L. monocytogenes*
- Know the Encephalitis: *HSV*, *VZV*, *CMV*, *Enterovirus*, *Echovirus*, *HIV*, *Measles* & *Parotitis viruses*
- Know the Rabies virus & Rabies disease
- Understand Prion diseases: Creutzfeld-Jakob disease
- Know the *C. botulinum* toxin-induced CNS infections
- Know the Bacteriological tests & Immunoprophylaxis of Infectious Diseases

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Entry & Spread of m/o into the CNS
- Resistance to the pathogenic m/o
- Meningitis: *N. meningitidis*, *H. influenzae*, *S. pneumoniae*, Group B *Streptococci*, *E. coli*, *L. monocytogenes*
- Encephalitis: *HSV*, *VZV*, *CMV*, *Enterovirus*, *Echovirus*, *HIV*, *Measles* & *Parotitis viruses*
- Rabies
- Prion diseases: Creutzfeld-Jakob disease
- *C. botulinum* toxin-induced CNS infections
- Bacteriological Diagnosis (CSF microscopy & culture)
- Treatment (antibioticotherapy)
- Vaccinoprophylaxis

LECTURE

5.19. Gastrointestinal Tract Infections (GI)

Objectives:

- Know predisposing factors: seasonality, climate change, food quality, status of water supply & sewage, personal hygiene, etc.
- Understand the factors of Immunity & Resistance in gastrointestinal tract
- Know the causative m/o of bacterial diarrhoea & Intoxications: *S. aureus*, *Shigella spp.*, *Salmonella spp.*, *Vibrio spp.*, *E. coli (EIEC & ETEC)* *Streptococcus spp.* & viral diarrhoea: *Rotaviruses*, *Noroviruses*, *Adenoviruses*, *Caliciviruses*, *Enteroviruses*
- Know the Antibiotic-Associated Pseudomembraneous Colitis caused by *C. difficile*
- Know the Viral Hepatitis: A, B, C, D, E
- Know the *Helicobacter pylori* induced GI diseases
- Know the Microbiological diagnosis & Prophylaxis of diarrhoea

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Predisposing factors: seasonality, climate change, food quality, status of water supply & sewage, personal hygiene, etc.
- Factors of Immunity & Resistance in gastrointestinal tract
- Causative m/o of bacterial diarrhoea & Intoxications: *S. aureus*, *Shigella spp.*, *Salmonella spp.*, *Vibrio spp.*, *E. coli (EIEC & ETEC)* *Streptococcus spp.*
- Ecology, Transmission, Mono & Poli Toxigenicity of m/o
- Parasites of GIT
- Antibiotic-Associated Pseudomembraneous Colitis (*C. difficile*)
- Viral Diarrhoeas: *Rotaviruses*, *Noroviruses*, *Adenoviruses*, *Caliciviruses*, *Enteroviruses*
- Viral Hepatitis: A, B, C, D, E
- *Helicobacter pylori* induced GI diseases
- Microbiological diagnosis
- Prophylaxis of diarrhoea

LECTURE

5.20. Nosocomial (Hospital) Infections

Objectives:

- Understand the development of Nosocomial (Hospital) Infections (NI)
- Know the pathogens (bacteria, viruses, fungi) involved into the Nosocomial Infections
- Know the prevalence of NI
- Know the risk factors of NI
- Know the Types of NI
- Know the reservoir/sources and routes of transmission of NI
- Understand the role of Normal Microbial Flora (NMF) in the development of NI
- Know the Hospital strains
- Know and understand the prevention and control of NI

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008

Lecture Outline:

- Definition of Nosocomial (Hospital) Infections (NI)
- Prevalence of NI
- The principal factors influencing NI
- The types of NI: Lower respiratory Tract Infections (LRTIs), Urinary Tract Infections (UTIs), Post- surgical Wounds, Blood Infections, Etc.
- The patients-associated risk factors predisposing NI
- Reservoirs/sources and Routes of transmission of NI
- The role of Normal Microbial Flora (NMF) in the development of NI
- The major bacteria associated with NI: *S. aureus*, *S. epidermidis*, *P. aeruginosa*, *Acinetobacter baumannii*, *proteus spp.*, *Klebsiella spp.*, *Serratia spp.*, *Candida spp.*, *C. difficile*, etc.
- The major viruses associated with NI: *Influenza virus*, *VZV*, *HSV*, *RSV*, *CMV*, *Rotavirus*, *Enterovirus*, *HBV* & *HCV*.
- Hospital strains – specific markers and their identification
- Control of Nosocomial (Hospital) Infections

GENERAL MICROBIOLOGY LABORATORY SAFETY GUIDELINES & CONTRACT

Any microorganism used in this Laboratory is considered a potential pathogen. It is necessary to work with a set of rules that will preclude any possibility of infection by microorganisms used in this class

- No food or drinks are permitted in the laboratory at any time
- Only closed-toe shoes are to be worn in the laboratory. Sandals or flip-flops are not permitted
- Keep hands and other objects away from your face, nose, eyes, ears, and mouth
- The application of cosmetics in the laboratory is prohibited in the laboratory
- Work areas/surfaces must be disinfected before and after use
- Laboratory coats must be worn and buttoned while in the laboratory
- Long hair should be secured behind your head
- Hands must be washed before leaving the laboratory
- All unnecessary books, briefcases or any personal items must be kept off the countertops
- Never pipette anything by mouth (including water). Always use pipette devices
- Dispose of wastes in their proper containers
- Do not pour chemicals or biohazardous fluids down the sink
- Return all chemicals, reagents, cultures, and glassware to their appropriate places
- Flame bacteriological loops before and immediately after use to transfer biological material
- Do not walk about the laboratory with bacteriological loops or pipettes containing infectious material.
- Be careful around Bunsen burners. Flames cannot always be seen
- Bunsen burners will be used frequently. Organize your desk so that you are not reaching over other equipment to use the Bunsen. Do not leave your Bunsen burner on if you are away from your desk
- Turn off Bunsen burners before leaving the laboratory
- Report any injuries, broken equipment, chemical or biological spills to your instructor immediately
- Follow all instructions given by your course instructor cleaning up any spills or broken glass
- Familiarize yourself with safety equipment in the laboratory and emergency escape routes
- Always wipe and clean the lenses of your microscope before putting it away. Use the appropriate tissue paper and cleaning solution for this purpose
- Use appropriate universal precautions with all biological fluids
- Do not remove any materials from the laboratory without the written permission of the course instructor or chief of the Department of Microbiology

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Microbiology Handbook

You must keep track of all your experiments in a “Microbiology Handbook”. It serves as your written memory. It is also a useful place for you to make notes to yourself. It also works as a document – a **written evidence of your work**. As a decent portion of work must be done during limited time, the work **must be** thoroughly planned and firmly substantiated in theoretically sound manner, to finish the assigned work in time and get your work accepted by the instructor. There is a possibility, that your handbook will be collected and valued at any time. Please, make sure, that upon completion of assigned work, your results and corresponding documentation is presented to instructor. Your work is **only valid with the signature of the instructor**

Code of conduct

Starting with the moment of entering laboratory it is your personal obligation to ensure herein written rules are being followed upon by every student in the Laboratory. In case of observing another student violating rules or being in dangerous situation it is your personal duty to inform the instructor

I have read and agree to follow the above safety rules

Print Student's name	Student's Signature	Date

PRACTICAL WORK

6.1. Laboratory Diagnosis of Septicemia

The purpose of laboratory work:

- Knowledge of the aerobic, anaerobic and facultative anaerobic bacteria involved into the septicemia: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Overview of the laboratory work:

- *Enterobacteriaceae* family (*E.coli*, *Klebsiella spp.*, *Proteus spp*, *Enterobacter spp.*, *Serratia spp.*, *Salmonella spp*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Pseudomonas (P.aeruginosa)*, microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Staphylococcus spp.*(*S.aureus*, . *epidermidis*, *S. saprophyticus*, *MRSA*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Streptococcus spp.*(*S.pneumoniae*, *S.pyogenes*, *S.agalactiae*,) : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Enterococcus spp.* (*E. faecalis*, *E.facium*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Neisseria spp.* (*N. meningitidis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Haemophylus spp (H.influenzae)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Bacteroides spp.*(*B.fragilis*) : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Prevotella spp.*(*P.melaninogenica*, *P.intermedia*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Porphyromonas spp.* (*P. gingivalis*) microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Candida spp.* (*C. albicans*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of laboratory work:

*Solving the tasks of Medical Microbiology by Microbiological Method:

- Patient's samples collection & transportation
- Microscopic identification of microorganisms
- Evaluation of bacterial growth in different cultures
- Biochemical identification of microorganisms
- Serological identification of microorganisms
- Evaluation of the Disk Diffusion method to determine a successful treatment
- Evaluation of the results, filling in the tables & writing down the conclusions
- Discussions with medical doctor-microbiologist

SEPTICEMIA

Septicemia is a medical term referring to the presence of pathogenic organisms in the bloodstream, leading to sepsis - a potentially deadly medical condition that is characterized by a whole-body inflammatory state. In the setting of more severe disturbances of temperature, respiration, heart rate or white blood cell count, the response is characterized as septic shock, and may result in multiple organ dysfunction syndrome. Septicemia not necessary has consequences on health of the individual: e.g. microorganism can be introduced into the bloodstream during tooth brushing, but this form of septicemia in healthy individuals normally does not lead to sepsis. However, some patients with prosthetic heart valves need antibiotic prophylaxis for dental surgery because bacteremia might lead to endocarditis.

Bacteremia is the presence of viable bacteria in the bloodstream. Accordingly, the terms **viremia** and **fungemia** refer to viruses and fungi in the bloodstream. Bacteria spread to other parts of the body and causes infections not in the original site of the infection. Bacteremia is the principal means by which local infections are spread to distant organs (referred to as **hematogenous spread**). Hematogenous dissemination of bacteria is frequently a part of the pathophysiology of infectious diseases (e.g. meningitis, endocarditis, osteomyelitis). Bacteremia is most commonly diagnosed by blood culture. Any bacteria that incidentally find their way to the culture medium will also multiply. For this reason, blood cultures must be drawn with great attention to sterile process. Occasionally, blood cultures will reveal the presence of bacteria that represent contamination from the skin through which the culture was obtained. Blood cultures must be repeated at intervals to determine if persistent — rather than transient — bacteremia is present. Bacteria can enter the bloodstream as a severe complication of infections (like pneumonia or meningitis), urinary tract infections, peritonitis, *C. difficile* colitis, colorectal cancer, during surgery (especially when involving mucous membranes such as the gastrointestinal tract), or due to catheters and other foreign bodies entering the arteries or veins (including intravenous drug abuse).

Fungemia is most commonly caused by *Candida* spp., but can be caused by other fungi as well, including *Aspergillus* and *Cryptococcus*. It is most commonly seen in immunosuppressed or immunocompromised patients with severe neutropenia, oncology patients, or in patients with intravenous catheters. The diagnosis is complicated, as routine blood cultures have poor sensitivity.

Viremia can be classified to primary and secondary viremia. **Primary viremia** refers to the initial spread of virus in the blood from the first site of infection. **Secondary viremia** occurs when primary viremia has resulted in infection of additional tissues via bloodstream, in which the virus has replicated and once more entered the circulation. Usually secondary viremia results in higher viral shedding and viral loads within the bloodstream. Usually the virus (e.g. *rabies virus*) will replicate briefly within the first site of infection, within the muscle tissues. Viral replication then leads to viremia and the virus spreads to its secondary site of infection - the CNS. Upon infection of the CNS, secondary viremia results and symptoms emerge. Vaccination at this point is useless, as the spread to the brain is unstoppable. Vaccination must be done before secondary viremia takes place for the individual to avoid brain damage or death.

6.1/1. A patient (an intravenous drug user) diagnosed with pneumonia developed a septic fever. Venous blood has been taken and inoculated into broth culture under aerobic and anaerobic conditions. After 24h of incubation at 37°C the broth culture has been inoculated onto blood agar. The growth then inoculated onto the differential-diagnostic media. Biochemical activity has been established and test for antibiotic susceptibility has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the septic fever (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.1/1 Identification of the microorganism

Gram stain	
Culture (blood agar)	
Culture (egg yolk agar)	
Culture (mannitol salt agar)	
Catalase test	
Coagulase test	
Mannitol fermentation	
Susceptibility to Novobiocin	
Susceptibility to Bacitracin	
Agglutination test	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.1/2. A patient with artificial heart valve developed a septic fever. Venous blood has been taken and inoculated into broth culture under aerobic and anaerobic conditions. After 24h of incubation at 37°C the broth culture has been inoculated onto blood agar. The growth then inoculated onto the differential-diagnostic media. Biochemical activity has been established and test for antibiotic susceptibility has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the septic fever (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.1/2 Identification of the microorganism

Gram stain	
Culture (blood agar)	
Culture (egg yolk agar)	
Culture (mannitol salt agar)	
Catalase test	
Coagulase test	
Mannitol fermentation	
Susceptibility to Novobiocin	
Susceptibility to Bacitracin	
Agglutination test	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.1/3. A woman is diagnosed with bacterial endocarditis (*an infection of the heart's inner lining of the heart (endocardium) or the heart valves*). Venous blood has been taken and inoculated into broth culture under aerobic and anaerobic conditions. After 24h of incubation at 37°C the broth culture has been inoculated onto blood agar. The growth then inoculated onto the differential-diagnostic media. Biochemical activity has been established and test for antibiotic susceptibility has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the bacterial endocarditis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.1/3 Identification of the microorganism

Gram stain				
Culture (blood agar)				
Culture (differential-diagnostic medium)				
Susceptibility to Bacitracin				
Susceptibility to Optochin				
Susceptibility to Bile				
Agglutination test:				
▪ A serum				
▪ B serum				
▪ D serum				
Antibiotic susceptibility testing:				
▪ Disk diffusion test	1.	S	M	R
	2.	S	M	R
	3.	S	M	R
	4.	S	M	R
	5.	S	M	R
	6.	S	M	R
	7.	S	M	R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.1/4. A 3-year old child is suspected of having meningitis. Venous blood has been taken and inoculated into broth culture under aerobic and anaerobic conditions. After 24h of incubation at 37°C the broth culture has been inoculated onto blood agar and chocolate agar. Latex agglutination test has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the meningitis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.1/4 Latex agglutination test

Ingredients	Wells				
	1	2	3	4	5
Ig against <i>N. meningitis</i>	A group	B group	C group	Y group	W-B5 group
Isolated bacteria	+	+	+	+	+
Results:					

Conclusions:

6.1/5. A woman diagnosed with urinary tract infection (UTI) developed septic fever. Venous blood has been taken and inoculated into broth culture under aerobic and anaerobic conditions. After 24h of incubation at 37°C the broth culture has been inoculated onto blood agar. The first (#1) plate has been cultivated in anaerobic conditions, and the second (#2) - in aerobic conditions. The growth then inoculated onto the differential-diagnostic media. Biochemical activity has been established and test for antibiotic susceptibility has been performed.

- Evaluate the results and identify the microorganism responsible for the septic fever
- Which microorganism is likely to be responsible for the septic fever (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.1/5 Identification of the microorganism

Gram stain	
Culture (blood agar) anaerobic conditions	
Culture (blood agar) aerobic conditions	
Culture (differential-diagnostic medium)	
CFU/ml	
Biochemical test:	
▪ Lactose	
▪ Glucose	
▪ H ₂ S	
▪ Mannitol	
▪ Lysine decarboxylase	
▪ Indol	
▪ Ornithine	
▪ Urease	
▪ Oxidase	
▪ Phenylalanine	
▪ Sucrose	
▪ Simmons' citrate	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

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6.1/6. A patient is diagnosed with fever of unknown origin (FUO). Venous blood has been taken and inoculated into broth culture under aerobic and anaerobic conditions. After 24h of incubation at 37°C the broth culture has been inoculated onto blood agar. The first (#1) plate has been cultivated in anaerobic conditions, and the second (#2) –in aerobic conditions. The growth then inoculated onto the differential-diagnostic media. Biochemical activity has been established and test for antibiotic susceptibility has been performed.

- Evaluate the results and identify the microorganism responsible for the FUO
- Which microorganism is likely to be responsible for the FUO (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6..
 - 7.

Table 6.1/6 Identification of the microorganism

Gram stain	
Culture (blood agar) anaerobic conditions	
Culture (blood agar) aerobic conditions	
Culture (differential-diagnostic medium)	
Biochemical test:	
▪ Mannitol	
▪ Lysine decarboxylase	
▪ Indol	
▪ Ornithine	
▪ Urease	
▪ Lactose	
▪ Glucose	
▪ Sucrose	
▪ Simmons' citrate	
▪ Phenylalanine	
Agglutination test	
▪ O antigen	
▪ H antigen	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.1/7. A patient diagnosed with pulmonary abscess developed septic fever. Venous blood has been taken and inoculated into broth culture under aerobic and anaerobic conditions. After 24h of incubation at 37°C the broth culture has been inoculated onto blood agar. The first (#1) plate has been cultivated in anaerobic conditions, and the second (#2) –in aerobic conditions. Biochemical activity has been established and test for antibiotic susceptibility has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
 - Which microorganism is likely to be responsible for the septic fever (options)?
- 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.1/7 Identification of the microorganism

Gram stain	
Culture (blood agar) anaerobic conditions	
Culture (blood agar) aerobic conditions	
Culture (egg yolk agar) Lecithinase/lipase	
Catalase test	
Bile	
Vancomycin	
Kanamycin	
Colistin (polymixin E)	
Biochemical test:	
▪ Indol	
▪ Esculine	
▪ Glucose	
▪ Salicin	
▪ Xylose	
▪ Sucrose	
▪ N-acetylglucosamine	
Agglutination test	
▪ O antigen	
▪ H antigen	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

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PRACTICAL WORK

6.2 Laboratory Diagnosis of GIs

The purpose of laboratory work:

- Knowledge of the bacteria and viruses involved into the Gastrointestinal Tract Infections (GIs): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Overview of the laboratory work:

- *Staphylococcus spp. (S. aureus)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *E. coli (Enteropathogenic (EPEC), Enterotoxigenic (ETEC), Enteroinvasive (EIEC), Enteragggregative (EAEC), Verotoxin-producing (VTEC), E. coli O157:H7)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Salmonella spp. (S. enterica, S. bongori)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Shigella spp. (S. sonnei, S. dysenteriae, S. boydii, S. flexneri)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Clostridium spp. (C. perfringens, C. botulinum, C. difficile)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Vibrio (V. cholerae, V. parahaemolyticus)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Campylobacter spp. (C. jejuni, C. coli, C. fetus)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Helicobacter spp. (H. pylori)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Yersinia spp. (Y. enterocolitica)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Reoviridae (Rotavirus A, dsRNA)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Picornaviridae (ECHO virus & Hepatitis A virus, ssRNA)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Hepeviridae (Hepatitis E virus, ssRNA)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Caliciviridae (Norovirus, ssRNA)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning

Solving the tasks of Medical Microbiology by Microbiological Method (*see the p. 26)

GASTROINTESTINAL TRACT INFECTIONS

Gastrointestinal Tract Infections (GIs) are very common & a wide variety of microorganisms may cause these infections. GI often have specific features that distinguish them, although stool examination and culture are required for a definitive diagnosis. Most infections are self-limited and easily treated once a diagnosis is established.

The **route of infection** of most of these pathogens is *fecal-oral route (person-to-person or animal-to-person)*; also *directly from drinking water*; *seafood-borne bacterial (Vibrio spp.)* & *viral (Hepatitis A virus, Norovirus)* illness becomes popular.

Illnesses caused by gastrointestinal pathogens can generally be divided into four general syndromes: **(1) gastritis; (2) food poisoning; (3) infectious diarrhea; (gastroenteritis); (4) typhoid (enteric) fever.**

(1) Acute infection with *H. pylori*, acquired via fecal-oral mode, leads to a severe **gastritis**; most infected individuals remain asymptomatic their entire lives. However, individuals who are at risk (poor socioeconomic status) may develop peptic ulcer disease (PUD), atrophic gastritis, gastric adenocarcinoma & B-cell lymphoma.

(2) Ingestion of preformed bacterial toxins in food (**food poisoning** by *S. aureus* enterotoxins) can cause rapid onset of symptoms: nausea & vomiting, less frequently diarrhea (1-3h) and rapid resolution of as the toxin is removed (12-36h); although *adult botulism (C. botulinum)* is considered a preformed toxin-associated illness, Unlike other food poisoning, botulism can be life threatening without appropriate support care.

(3) **Diarrhea** is the **most common manifestation of GIs**. Nausea, vomiting & abdominal pain are accompanying symptoms. **Secretory** (watery) **diarrhea** usually is self-limited, caused by viruses (*Rotavirus, Norovirus, Adenovirus, Astroviruses*) & bacteria (*enterotoxigenic E. coli (ETEC), V. cholerae*). **Dysentery** (bloody diarrhea with mucus & fever) is mainly caused by: *Campylobacter spp., Salmonella spp., Shigella spp., Yersinia spp.* **C. difficile colitis** is almost always related to earlier antibiotic use and should always be considered in hospital patients. **Hemorrhagic colitis** (frank bloody diarrhea) is caused by *enterohemorrhagic E. coli (EHEC, O157:H7)*, which is a common cause of sporadic diarrhea.

(4) **Typhoid (enteric) fever** is a systemic febrile illness of prolonged (3-5 weeks) duration marked by fever, persistent bacteremia, and metastatic spread- leading to multiple –organ dysfunction. Most *patients do not have diarrhea (S. typhi* is the main cause and is transmitted via the ingestion of material contaminated with human feces from chronic carriers).

Hepatitis (acute), an inflammation of the liver, can be caused by *Hepatitis A virus (HAV)*, by eating contaminated seafood or imported berries.

Immunosuppressed (AIDS) patients may develop enteric infections with microorganisms that rarely caused disease in healthy individuals (*Cryptosporidium spp., CMV, M. avium complex* & *C. difficile-antibiotic-associated diarrhea*)

The human body has developed several **defense mechanisms** to prevent infection through the GI tract: *stomach acid* destroys most organisms, *gut motility*, which helps prevent adherence of microorganisms to mucosal surfaces, *secretory IgA* which bind to recognized pathogen & *normal microflora*, which is important in preventing overgrowth of pathogens.

6.2/1. A stool sample has been obtained from a patient with diarrhea and plated into two media (one of them is differential Endo/MacConkey's medium). After 24h 37°C incubation period, the growth has been inoculated onto the agar slant. Biochemical test has been performed. Serological (slide agglutination) test should be performed. Antibiotic susceptibility test has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for diarrhoea (options) ?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.2/1 Identification of Enterobacteriaceae

Gram stain	
Culture (blood agar)	
Culture (Endo medium)	
Culture (MacConkey's medium)	
Catalase test	
Oxidase test	
Biochemical test:	
▪ H ₂ S	
▪ Indol	
▪ Urease	
▪ Lysine decarboxylase	
▪ Ornithine	
▪ Phenylalanine	
▪ Nitrates	
▪ Arginine	
▪ Glucose	
▪ Lactose	
▪ Sucrose	
▪ Mannitol	
Agglutination test	
▪ O serum	
▪ H serum	
▪ K serum	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.2/2. A man is noted to have shigellosis. A stool sample has been obtained from a patient and plated into two media (one of them is differential Endo/MacConkey's medium). After 24h 37°C incubation period, the growth has been inoculated onto the agar slant. Biochemical tests have been performed. Serological (slide agglutination) test should be performed. Antibiotic susceptibility test has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for shigellosis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.2/2 Identification of Enterobacteriaceae

Gram stain	
Culture (blood agar)	
Culture (Endo medium)	
Culture (MacConkey's medium)	
Catalase test	
Oxidase test	
Biochemical test:	
▪ H ₂ S	
▪ Indol	
▪ Urease	
▪ Lysine decarboxylase	
▪ Ornithine	
▪ Phenylalanine	
▪ Nitrates	
▪ Arginine	
▪ Glucose	
▪ Lactose	
▪ Sucrose	
▪ Mannitol	
Agglutination test	
▪ polyvalent <i>Shigella</i> serum	
▪ monovalent <i>S. flexneri</i> serum	
▪ monovalent <i>S. sonnei</i> serum	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.2/3. A patient is diagnosed with salmonellosis (enteritis). A stool sample has been obtained from the patient and plated onto the differential Endo/MacConkey's medium and Bismuth sulfite agar (BSA). After 24h 37°C incubation period, the grown inoculated onto the slant meat-peptone agar (MPA). Biochemical tests have been performed. Serological (slide agglutination) test should be performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for salmonellosis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.2/3 Identification of Enterobacteriaceae

Gram stain	
Culture (blood agar)	
Culture (Endo medium/ MacConkey's medium)	
Culture (BSA medium)	
Culture (MPA medium)	
Catalase test	
Oxidase test	
Biochemical test:	
▪ H ₂ S	
▪ Manitol	
▪ Lysine decarboxylase	
▪ Indol	
▪ Ornithine	
▪ Urease	
▪ Lactose	
▪ Glucose	
▪ Sucrose	
▪ Simmons citrate	
▪ Phenylalanine	
Agglutination test	
▪ O serum	
▪ H serum	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
	1. S M R
	2. S M R
	3. S M R
	4. S M R
	5. S M R
	6. S M R
	7. S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

--

6.2/4. A man is diagnosed with campylobacteriosis. A stool sample has been obtained from a patient, plated onto the three selective media and each of them have been grown under microaerophilic conditions, in different temperatures, at 25°C, 37°C and 42°C. After 48h incubation period, the grown inoculated onto the slant meat-peptone agar (MPA). Biochemical tests have been performed. Serological (slide agglutination) test should be performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for campylobacteriosis (options) ?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.2/4 Identification of *Campylobacter* spp.

Gram stain	
Culture (25°C)	
Culture (37°C)	
Culture (42°C)	
Catalase test	
Hippurate hydrolysis	
Nitrate reduction	
H ₂ S	

* Hydrolysis of hippurate: :“+” for red color, “-“ for yellow color;

Conclusions:

6.2/5. A man is diagnosed with a peptic ulcer disease (PUD). A blood serum has been obtained from a patient and EIA Immunoassay has been performed [Table 6.2/5].

- Evaluate the results and identify the microorganism responsible for the PUD

Table 6.2/5 EIA Immunoassay

Ingredients	Wells		
	1	(-) control	(+) control
<i>H. pylori</i> antigen	+	+	+
Patient's serum	+	-	-
Immunoglobulin (Ig) against <i>H. pylori</i>	-	-	+
Peroxidase-marked anti-human IgM	+	+	+
Substrate for peroxidase	+	+	+
Results:			

Note: wells are washed with physiological solution after every infusion of ingredient

Conclusions:

6.2/6. Suspecting an enterovirus infection, a patient’s “A” & patient’s “K” stool specimens have been collected and EIA Immunoassay has been performed [Table 6.2/6].

- Evaluate the results and identify the microorganism responsible for the infection

Table 6.2/6 EIA Immunoassay

Ingredients	Wells			
	1 “A”	2 “K”	3 (control)	4 (control)
Immune serum against <i>Rotavirus</i>	+	+	+	+
Patient’s stool’s filtrate	+	+	-	-
<i>Rotavirus</i> antigen	-	-	-	+
Peroxidase-marked anti- <i>Rotavirus</i> Ig	+	+	+	+
Substrate for peroxidase	+	+	+	+
Results:				

<i>Conclusions:</i>

6.2/7. Stool specimen has been taken from a child with symptoms of acute gastrointestinal tract infection and EIA immunoassay has been performed [Table 6.2/7].

- *Evaluate the results and identify the microorganism responsible for an acute gastrointestinal infection*

Table 6.2/7 *EIA Immunoassay*

Ingredients	Wells		
	1	2	3
Immune serum	Polyvalent serum against <i>Enterovirus</i>	Polyvalent serum against Adenovirus (1-41)	Polyvalent serum against Rotavirus
Patient's stool's filtrate	+	+	+
Peroxidase-marked immune serum	Polyvalent serum against <i>Enterovirus</i>	Polyvalent serum against Adenovirus (1-41)	Polyvalent serum against Rotavirus
Substrate for peroxidase	+	+	+
Results:			

Conclusions:

6.2/8. A stool sample has been obtained from a patient with diarrhea after a long treatment with antibiotics. Latex agglutination test should be performed with *C. difficile* toxin diagnosticum.

- *Evaluate the results and identify the microorganism responsible for diarrhea*

<i>Conclusions:</i>

PRACTICAL WORK

6.3. Laboratory Diagnosis of UTI & STD

The purpose of laboratory work:

- Knowledge of the microorganisms involved into the UTI & STD's: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Overview of the laboratory work:

- *Enterobacteriaceae* family (*E.coli*, *Klebsiella spp.*, *Proteus spp*, *Enterobacter spp.*, *Serratia spp.*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Pseudomonas (P.aeruginosa)*, microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Staphylococcus spp.*(*S.aureus*, *S.epidermidis*, *S. saprophyticus*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Streptococcus spp.*(*S.pyogenes*, *S.agalactiae*,) : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Enterococcus spp.* (*E. faecalis*, *E.faecium*, *VRE*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Neisseria spp.* (*N. gonorrhoeae*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Mycoplasma spp* (*M. genitalium*, *N.hominis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Ureaplasma spp.* (*U. urealyticum*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Chlamydia spp.* (*C.trachomatis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Leptospira spp.* (*L.interrogans*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Treponema spp.* (*T. pallidum*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Gardnerella spp.* (*G. vaginalis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Trichomonas spp.* (*T. vaginalis*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Herpesviridae* (*Herpes simplex virus*, *dsDNA*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Papillomaviridae* (*Human papillomavirus (HPV)*, *DNA*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Retroviridae* (*Human immunodeficiency virus (HIV)*, *ssRNA*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Candida spp.* (*C. albicans*): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning

Solving the tasks of Medical Microbiology by Microbiological Method (*see the p. 26)

UROGENITAL TRACT INFECTIONS & SEXUALLY TRANSMITTED DISEASES

The urogenital tract is a common source of infection in men and women. The pathogens involved are similar for both sexes, but the clinical syndromes vary considerably depending on the anatomic location.

Acute urinary tract infections (aUTI) can be subdivided into two general anatomic categories: lower UTI (cystitis/uncomplicated UTI) & upper UTI (pyelonephritis/complicated UTI)

(1) **Uncomplicated UTI** (community-acquired) typical symptoms: dysuria and/or frequent urination and suprapubic pain; risk factors: recent sexual intercourse & pregnancy; women > men (10:1); monomicrobial: *E. coli*, *S saprophyticus*, *Proteus spp.*, *Klebsiela spp.*, *Enterococcus spp.*; laboratory findings: bacteriuria & pyuria. ***significant bacteriuria is >10⁵cfu/ml of urine (“midstream” specimen); pyuria - the presence of >4 neutrophils per high power field of unspun, voided mid-stream urine.**

(2) **Complicated UTI** (community-acquired) typical symptoms: fever, flank pain, dysuria, nausea, vomiting & malaise; risk factor: obstruction (urinary stasis); can be polymicrobial: *E. coli*, *Proteus spp.*, *Klebsiella spp.*, *S. aureus*, *Candida spp.*, *S. pyogenes*; laboratory findings: leucocytosis, pyuria & bacteriemia (may or may not be present)

***Nosocomial UTI (uncomplicated or complicated);** risk factor: urinary catheterization; pathogens: *E. coli*, *Proteus spp.*, *Klebsiella spp.*, *Serratia spp.*, *Pseudomonas spp.*, *S. epidermidis*, *Candida spp.*

Sexually transmitted diseases (STDs) are usually associated with few clinical syndromes:

(1) **urethritis (in men)** - painful urination or a penile discharge; pathogens: *N. gonorrhoeae* (purulent discharge) & *C. trachomatis* (clear discharge);

(2) **cervicitis (in women)** –significant tenderness on pelvic examination; pathogens: *N. gonorrhoeae*, *C. trachomatis*, *M. hominis* & *M. genitalium* ; *asymptomatic gonorrhea* & *Chlamydia* may result in *pelvic inflammatory disease (PID)* – *infertility-ectopic pregnancy-chronic pelvic pain* & **neonatal ophthalmia.**

(3) **vaginitis** (vaginal discharge) pathogens: *Trichomonas hominis* & *C. albicans*; and **vaginosis** pathogens: *Gardnerella spp.* & *Mobiluncus spp.*

(4) **genital ulcers (nonexudative infections);** penile ulcers in men can be painful (*HSV-1/HSV-2*) or painless (syphilis/*T.pallidum*); also chancroid/ *H.ducreyi* & lymphogranuloma venereum/*C. trachomatis*.

AIDS is sexually transmitted systemic infection, caused by *HIV-1*, *HIV-2*.

Cervical carcinoma is caused by HPV & **Kaposi sarcoma** is caused by HHV-8.

6.3/1. A man is suspected of having acute gonorrhoeae. Gram staining and microscopy on urethral discharge has been performed.

- *Evaluate the results and identify the microorganism responsible for the infection*
- *Which microorganism is likely to be responsible for this infection?*
- *Is microscopy enough to be 100% certain of a right diagnosis?*

<i>Conclusions:</i>

6.3/2. A woman has an external genitalia wound. The pus coming from a wound has been plated onto chocolate media and onto blood media. After 24h 37°C incubation period, the growth has been inoculated into differential-diagnostic media with X, V and XV discs on the agar surface. Antibiotic susceptibility test has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the wound (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.3/2 Identification of the microorganism

Gram stain				
Culture (blood agar)				
Culture (chocolate agar)				
Catalase test				
Oxidase test				
Growth factors:				
▪ X				
▪ V				
▪ XV				
Antibiotic susceptibility testing:				
▪ Disk diffusion test	1.	S	M	R
	2.	S	M	R
	3.	S	M	R
	4.	S	M	R
	5.	S	M	R
	6.	S	M	R
	7.	S	M	R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.3/3. A patient is diagnosed with secondary syphilis. A complement fixation test (CFT) has been performed [Table 6.3/3].

- *Evaluate the results and identify the microorganism responsible for secondary syphilis*

Table 6.3/3 *Complement fixation test (CFT)*

Ingredients	Test-tubes		
	1	2 (+) control	3 (-) control
Patient's blood serum, ml	0,5	-	-
Sick man blood serum, ml	-	0,5	-
Healthy man blood serum, ml	-	-	0,5
Cardiolipin antigen, ml	0,5	0,5	0,5
Complement, ml	0,5	0,5	0,5
Incubation at 37°C/30 min			
Hemolytic system, ml	1,0	1,0	1,0
Incubation at 37°C/30 min			
Results:			

Conclusions:

6.3/4. A woman has endometritis (*inflammation of the endometrium*). An endocervical discharge has been inoculated onto blood agar. After 24h 37°C incubation period gram staining, microscopy, latex agglutination test (slide agglutination test) and a disk diffusion test for antibiotic susceptibility have been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for endometritis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.3/4 Identification of the microorganism

Gram stain				
Culture (blood agar)				
Agglutination test:				
▪ A serum				
▪ B serum				
▪ D serum				
Antibiotic susceptibility testing:				
▪ Disk diffusion test	1.	S	M	R
	2.	S	M	R
	3.	S	M	R
	4.	S	M	R
	5.	S	M	R
	6.	S	M	R
	7.	S	M	R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.3/5. A woman has urinary tract infection (UTI). Urine has been collected and inoculated onto blood agar. After 24h of incubation at 37°C the growth has been re-inoculated into the differential-diagnostic media. Biochemical tests and the disk diffusion test for antibiotic susceptibility have been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for UTI (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.3/5 Identification of the microorganism

Gram stain	
Culture (blood agar)	
Culture (mannitol salts agar)	
Culture (thioglycol medium)	
Catalase test	
Coagulase test	
Mannitol fermentation	
Susceptibility to Bacitracin	
Susceptibility to Novobiocin	
Agglutination test	
CFU/ml	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.3/6. A patient is diagnosed with prostatitis (*an inflammation of the prostate gland*). Urine has been collected and inoculated onto blood agar. After 24 hours of incubation at 37°C the growth has been re-inoculated into the differential-diagnostic media. Biochemical tests and the disk diffusion test for antibiotic susceptibility have been performed.

- Evaluate the results and identify the microorganism responsible for the infection?
- Which microorganism is likely to be responsible for prostatitis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.3/6 Identification of the microorganism

Gram stain				
Culture (blood agar)				
Culture (aesculin agar)				
Culture (thioglycol medium)				
Catalase test				
Susceptibility to Bacitracin				
Susceptibility to Optochin				
Susceptibility to Bile				
CFU/ml				
Agglutination test <ul style="list-style-type: none"> ▪ A serum ▪ B serum ▪ D serum 				
Antibiotic susceptibility testing: <ul style="list-style-type: none"> ▪ Disk diffusion test 				
	1.	S	M	R
	2.	S	M	R
	3.	S	M	R
	4.	S	M	R
	5.	S	M	R
	6.	S	M	R
	7.	S	M	R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.3/7. A man is diagnosed with nephrolithiasis (*urinary stones in the kidney*). Urine has been collected and inoculated onto blood agar. After 24h of incubation at 37°C the growth has been re-inoculated into the differential-diagnostic media. Biochemical tests and the disk diffusion test for antibiotic susceptibility have been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for nephrolithiasis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.3/7 Identification of the microorganism

Gram stain	
Culture (blood agar)	
Culture (thioglycol medium)	
Culture (mannitol salts agar)	
CFU/ml	
Biochemical test:	
▪ Lactose	
▪ Glucose	
▪ H ₂ S	
▪ Mannitol	
▪ Lysine decarboxylase	
▪ Indol	
▪ Ornithine	
▪ Urease	
▪ Oxidase	
▪ Phenylalanine	
▪ Sucrose	
▪ Simmons citrate	
Agglutination test	
▪ O serum	
▪ H serum	
Antibiotic susceptibility testing:	
▪ Disk diffusion test	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R
6.	S M R
7.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

--

6.3/8. A woman is diagnosed with vaginal candidiasis (*vaginal yeast infection*) related to a long-term antibiotic therapy. Vaginal scrapings from the affected area has been collected for a Gram stain and then inoculated onto blood agar and Sabouraud's agar. After 24h of incubation at 37°C the growth has been re-inoculated into the differential-diagnostic (CHROMagar) media. Biochemical tests and the disk diffusion test for antibiotic susceptibility have been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for candidiasis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.3/8 Identification of the microorganism

Gram stain	
Culture (blood agar)	
Culture (Sabouraud's agar)	
Culture (CHROMagar)	
Pseudohyphae	
Blastoconidia	
Chlamyospores	
Biochemical test:	
▪ Glucose	
▪ Maltose	
▪ Lactose	
▪ Sucrose	
▪ Galactose	
▪ Urease	
▪ Antifungal drugs:	
1.	S M R
2.	S M R
3.	S M R
4.	S M R
5.	S M R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

PRACTICAL WORK

6.4. Laboratory Diagnosis of RTI

The purpose of laboratory work:

- Knowledge of the bacteria, viruses & fungi involved into the Respiratory Tract Infections (RTI): microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

Overview of the laboratory work:

- *Streptococcus spp. (S.pneumoniae, S.pyogenes)* : microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Staphylococcus spp.(S.aureus)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Haemophilus spp (H.influenzae)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Klebsiella spp. (K. pneumoniae)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Pseudomonas (P.aeruginosa)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Corynebacterium spp. (C.diphtheriae)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Bordetella spp. (B.pertusis, B. parapertusis)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Mycoplasma spp. (M. pneumoniae)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Mycobacterium spp. (M. tuberculosis)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Legionella spp. (L. pneumophila)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Candida spp. (C. Albicans, C. krusei)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Orthomyxoviridae (Influenza virus, dsRNA)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Paramyxoviridae (Human parainfluenza viruses (HPIVs), ssDNA & Human respiratory syncytial virus (RSV), ssRNA)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Adenoviridae (Human adenovirus type B (HAdV-B) & type C (HadV-C), dsDNA)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control
- *Coronaviridae (SARS coronaviruses (SARS-CoV), ssRNA)*: microbiology, pathogenesis, epidemiology, laboratory diagnosis, treatment, prevention and control

The plan of self-directed learning

Solving the tasks of Medical Microbiology by Microbiological Method (*see the p. 26)

RESPIRATORY TRACT INFECTIONS

The respiratory tract (upper & lower) is the most common site of infection & many infectious etiologies can be responsible for RT infections; **person-to-person** transmission, often by **hand contact**.

The most common upper RTI is „**Common cold**“; pathogens: in adults (*Rhinovirus*) & in children (*Parainfluenza virus*, *Coronavirus*); it is more likely due to indoor crowding and contact than it is to colder weather

Pharyngitis is also very common upper RTI. Pathogens: in children <3y.o & adults (*Rhinovirus*, *Coronavirus*, *Adenovirus*), but in children 5-15 y.o. (*Enteroviruses*, *S. pyogenes*, *M. pneumoniae*); in all age groups (*Influenzavirus A & B*); in unimmunized children (*C. diphtheriae*)

Laryngitis – is self limited upper RTI; pathogens: *M. catarrhalis*, *M. tuberculosis*, *viruses*.

Sinusitis – typical viral upper RTI, but often caused by a secondary bacterial infection, *Aspergillus spp.* cause allergic fungal sinusitis

Otitis media may be a primary infection, but more commonly caused by a secondary bacterial infection following an initial viral upper RTI; pathogens: *S. pneumoniae*, *H. influenzae*.

Tracheobronchitis – *viral* in patients with normal lung functions (*Adenovirus*, *Parainfluenzavirus*, *Influenzavirus*) & **bacterial** (*H. influenzae*, *M. catarrhalis*) in patients with chronic obstructive pulmonary disease (COPD)

Croup (laryngotracheitis)- unique clinical syndrome seen in children <3y.o. (*Parainfluenza virus*)

Pneumonia - an inflammatory disease of the lungs; **hospital-acquired pneumonia (HAP)** pathogens: *K. pneumoniae*, *P. aeruginosa*, *M. S. aureus*.

Community-acquired pneumonia (CAP): typical (*S. pneumoniae* & *S. aureus*, *K. pneumoniae*, *H. influenzae*, *M. catarrhalis*) & **atypical** (*M. pneumoniae*, *C. pneumoniae*, *L. pneumophila*).

Viral causes of pneumoniae are uncommon in adults but can be seen in infants and children (*RSV*).

Immunocompromised hosts' pneumonia pathogens: *S. pneumoniae*, *M. tuberculosis*, *Nocardia spp.*, *CMV*, *P. jiroveci*; *A. fumigatus*

6.4/1. A woman has bacterial tonsilitis (*inflammation fo tonsils*). A throat swab has been taken for Gram stain and then inoculated onto blood agar and into thioglycol medium. Susceptibility to bacitracin, optochin and bile has been established. Disk diffusion test for antibiotic susceptibility has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the tonsilitis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.4/1 Identification of the microorganism

Gram stain				
Culture (blood agar)				
Culture (thioglycol medium)				
Catalase test				
Susceptibility to Bacitracin				
Susceptibility to Optochin				
Susceptibility to Bile				
Agglutination test				
▪ A serum				
▪ C serum				
▪ G serum				
Antibiotic susceptibility testing:				
▪ Disk diffusion test	1.	S	M	R
	2.	S	M	R
	3.	S	M	R
	4.	S	M	R
	5.	S	M	R
	6.	S	M	R
	7.	S	M	R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.4/2. A patient has sinusitis (*an inflammation of paranasal sinuses*). The pus coming out from the sinuses has been plated onto blood agar and chocolate agar. After 24h 37°C incubation period, the growth has been inoculated into differential-diagnostic media with X, V and XV discs on the agar surface. Antibiotic susceptibility test has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the sinusitis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.4/2 Identification of the microorganism

Gram stain				
Culture (blood agar)				
Culture (chocolate agar)				
Catalase test				
Oxidase test				
Growth factors:				
▪ X				
▪ V				
▪ XV				
Antibiotic susceptibility testing:				
▪ Disk diffusion test	1.	S	M	R
	2.	S	M	R
	3.	S	M	R
	4.	S	M	R
	5.	S	M	R
	6.	S	M	R
	7.	S	M	R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.4/3. A child is diagnosed with diphtheria. A throat swab has been obtained and cultured on a Tellurite plate. After 24h 37°C incubation period, the growth has been inoculated onto the blood agar and Loeffler’s media. Biochemical tests and the disk diffusion test for antibiotic susceptibility have been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the diphtheria (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.4/3 Identification of the microorganism

Gram stain				
Loefflers’ stain				
Neisser’s stain				
Culture (blood agar)				
Culture (Loefflers’ medium)				
Biochemical test:				
▪ Urease				
▪ Glucose				
▪ Sucrose				
▪ Maltose				
▪ Starch				
Antibiotic susceptibility testing:				
▪ Disk diffusion test	1.	S	M	R
	2.	S	M	R
	3.	S	M	R
	4.	S	M	R
	5.	S	M	R
	6.	S	M	R
	7.	S	M	R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.4/4. Ten children were diagnosed with pertussis (*whooping cough*). Nasal swabs have been obtained and an EIA Immunoassay has been performed [Table 6.4/4].

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the pertussis (options)?
 - 1.
 - 2.
 - 3.

Table 6.4/4 EIA Immunoassay

Ingredients	Wells	
	1-10	11-20
Ig against <i>B. pertussis</i>	+	-
Ig against <i>B. parapertussis</i>	-	+
Nasal swabs	+	+
Peroxidase-marked Ig against <i>B. pertussis</i>	+	-
Peroxidase-marked Ig against <i>B. parapertussis</i>	-	+
Substrates for peroxidase	+	+
Results:		

<i>Conclusions:</i>

6.4/5. A patient is diagnosed with bacterial pneumonia. Sputum has been collected for a Gram stain and then inoculated onto chocolate agar. Susceptibility to optochin, bacitracin and bile has been established and a disk diffusion test for antibiotic susceptibility has been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the pneumonia (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.4/5 Identification of the microorganism

Gram stain				
Culture (blood agar)				
Culture (chocolate agar)				
Susceptibility to Bacitracin				
Susceptibility to Optochin				
Susceptibility to Bile				
CAMP test				
Agglutination test				
Antibiotic susceptibility testing:				
▪ Disk diffusion test				
1.	S	M	R	
2.	S	M	R	
3.	S	M	R	
4.	S	M	R	
5.	S	M	R	
6.	S	M	R	
7.	S	M	R	

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.4/6. A patient is diagnosed with tuberculosis. The sputum has been obtained from the patient and stained by Ziehl-Neelsen (ZN) staining method and plated onto the selective Lowenstein-Jensen (LJ) medium and incubated for 2 weeks at the 37°C. The biochemical tests and drug susceptibility testing of *M. tuberculosis* have been performed.

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the tuberculosis (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.

Table 6.4/6 Identification of the microorganism

ZN stain				
Culture (LJ medium)				
Niacin test				
Nitrate reduction				
Urease				
Catalase (68°C)				
Antibiotic susceptibility testing:				
▪ Disk diffusion test	1.	S	M	R
	2.	S	M	R
	3.	S	M	R
	4.	S	M	R
	5.	S	M	R
	6.	S	M	R
	7.	S	M	R

*(S) sensitive; (M) medium sensitive; (R) resistant

Conclusions:

6.4/7. Throat secret has been taken from a patient with symptoms of acute respiratory infection and inoculated into embryonated egg culture. Hemagglutination inhibition test (HIT) has been performed [Table 6.4/7].

- Evaluate the results and identify the microorganism responsible for the infection
- Which microorganism is likely to be responsible for the infection (options)?
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.4/7 Hemagglutination inhibition tests (HIT)

Ingredients	Test-tubes			
	1	2	3	4 (control)
Allantoic liquid	0,5	0,5	0,5	0,5
Antiviral <i>influenza</i> serum A I (H1N1)	0,5	-	-	-
Antiviral <i>influenza</i> serum A II (H2N2)	-	0,5	-	-
Antiviral <i>influenza</i> serum B	-	-	0,5	-
Physiological solution	-	-	-	0,5
Chicken's erythrocytes suspension (2%)	0,5	0,5	0,5	0,5
Results:				

Conclusions:

6.4/8. Throat secret has been taken from a child with symptoms of acute respiratory infection and EIA immunoassay has been performed [Table 6.4/8].

- *Evaluate the results and identify the microorganism responsible for the infection*
- *Which microorganism is likely to be responsible for the infection (options)?*
 - 1.
 - 2.
 - 3.
 - 4.
 - 5.
 - 6.
 - 7.

Table 6.4/8 *EIA Immunoassay*

Ingredients	Wells		
	1	2	3
Immune serum	Polyvalent serum against <i>Influenza virus</i>	Polyvalent serum against <i>Adenovirus</i> (1-41)	Polyvalent serum against <i>Rhinovirus</i> (1-115)
Throat secret	+	+	+
Peroxidase-marked immune serum	Polyvalent serum against <i>Influenza virus</i>	Polyvalent serum against <i>Adenovirus</i> (1-41)	Polyvalent serum against <i>Rhinovirus</i> (1-115)
Substrate for peroxidase	+	+	+
Results:			

<i>Conclusions:</i>

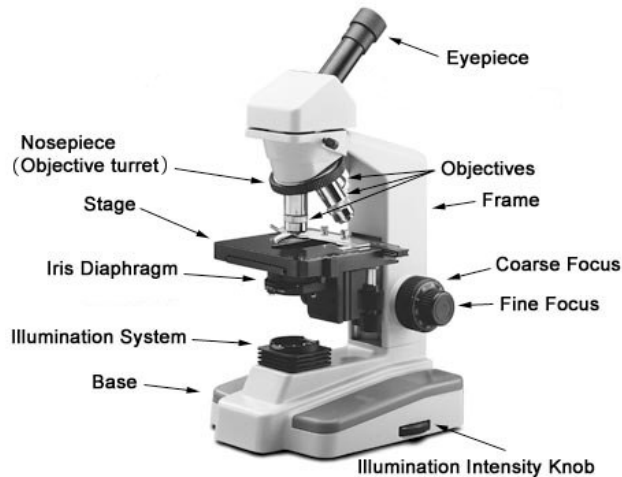
LABORATORY TECHNIQUES

STAINING

- **GRAM STAINING (Gram's method)** was first described by Danish scientist Hans Christian Gram (1884); It is an empirical method of differentiating bacterial species into two large groups: Gram-positive (dark blue or violet) & Gram-negative (red or pink)
- The method consists of four following steps:
 - (1) applying a primary stain (*gentian violet*) to a heat-fixed smear of a bacterial culture;
 - (2) the addition of a trapping agent (*Lugol's iodine*);
 - (3) rapid decolorization with *alcohol (95%)*;
 - (4) counterstaining with *fuchsine* & washing the preparation with water.
- **ZIEHL –NEELSEN (ZN) STAINING (acid-fast stain)** was first described by two German doctors: Franz Ziehl & Friedrich Neelsen; It is a special method used to identify acid-fast microorganisms, mainly: *Mycobacterium spp.* & *Nocardia spp.*(bright red)
- **GIEMSA STAINING** was first described by Gustav Giemsa; It differentially stains human and bacterial cells purple and pink respectively; It can be used to study the adherence of pathogenic bacteria to human cells, for histopathological diagnosis of malaria, some other *spirochete* & *protozoan* blood parasites; also stains the *Histoplasma capsulatum* (fungus) & *Chlamydia spp.*
- **SCHAEFFER-FULTON STAINING** was first designed by Alice B. Schaeffer & MacDonald Fulton (1930); It is a technique designed to isolate endospores by staining any present endospores green (green stain is malachite green) & any other bacteria are red (stain is safranin).

LABORATORY TECHNIQUES

MICROSCOPY



- **Light microscopy:** the common light microscope used in the laboratory & called a compound microscope.

- **Dark-field microscopy:** is used to observe live spirochetes (*T. pallidum*); the dark-field microscope contains a special condenser that scatters light and causes it to reflect off the specimen at an angle. A light object is seen on a dark background.

- **Phase-contrast microscopy:** is used to observe live, unstained organisms. In effect, the phase contrast technique employs an optical mechanism to translate

minute variations in phase into corresponding changes in amplitude, which can be visualized as differences in image contrast.

- **Fluorescent microscopy (fluorescent-antibody technique):** is used to identify unknown bacteria; the fluorescent microscope uses ultraviolet light as its light source.

- **Electron microscopy (EM):** was first used in 1950s; is used to identify *viruses*, mainly *viral gastroenteritis viruses (Rotavirus, Norovirus, Calicivirus, Adenovirus 40/41)*.. *Disadvantages:* viruses belonging to the same family can not be distinguished from each other as they will have the same *morphology (size, shape, surface characteristics)*; *It is expensive, technically demanding, requires specialist training and relatively insensitive (requires a minimum of one million viral particles/ml of 1g of specimen) & has been limited to research institutions.*

“A weak mind is like a microscope, which magnifies trifling things, but cannot receive great ones”

Lord Chesterfield

LABORATORY TECHNIQUES

BACTERIOLOGICAL CULTURE (bacteria)

This method is used to determine the type of organism, its abundance in the sample being tested, or both.

A *growth medium (culture medium)* is a liquid (*broth*) or gel (*agar*) designed to support the growth of microorganisms; there are five types of growth media:

- (1) **NUTRIENT MEDIA:** contain all the elements that most bacteria need for growth & used for the general cultivation and maintenance of bacteria kept in laboratory culture collections
- (2) **ENRICHED MEDIA:** contain the nutrients required to support the growth of a wide variety of organisms, including some of the more fastidious ones; it is commonly used to harvest as many different types of microbes as are present in the specimen
 - **Blood agar**
 - **Chocolate agar** - heat-treated blood (40-45°C), which turns brown
- (3) **SELECTIVE MEDIA:** used for the growth of only selected microorganisms;
 - **Eosin methylene blue (EMB) agar** designed to grow *only Gram-negative bacteria*
 - **MacConkey agar** designed to grow *Gram-negative bacteria* & stain them for *Lactose fermentation (Lac+/Lac-)*
 - **Mannitol Salt agar (MSA)** designed to grow *Gram-positive bacteria* & differential for *mannitol*; *Coagulase-positive Staphylococci* produce yellow colonies with yellow zones, whereas *Coagulase-negative Staphylococci* produce small pink/red colonies with no color change to the medium
- (4) **DIFFERENTIAL MEDIA:** used to distinguish one microorganism type from another growing on the same media
 - **Eosin methylene blue (EMB) agar** distinguishing between Lac+ organisms (*E. coli* colonies purple black with distinctive metallic green sheen) & those Lac- (*Salmonella spp.* & *Shigella spp.* colonies colorless)
 - **MacConkey agar:** Lac+ bacteria such as *E. coli*, *Enterobacter spp.*, *Klebsiella spp.* will produce acid, which lowers the pH of the agar below 6.8 and results in the appearance of *red/pink colonies*; Lac- bacteria such *Salmonella spp.*, *Proteus spp.*, *P. aeruginosa* & *Shigella spp.* (except *S. sonnei*) raises the pH of the agar, and leads to the formation of *colorless colonies*;
- (5) **TRANSPORT MEDIA:** used for temporary storage of specimens being transported to the laboratory for cultivation
 - **Thioglycolate broth** - for strict anaerobes
 - **Stuart Transport Medium (STM)** - for *Neisseria spp.*, *Haemophilus spp.*, & etc.
 - **Venkat-Ramakrishnan (VR) medium** -for *V. cholerae*

LABORATORY TECHNIQUES

CELL CULTURE (viruses)

- Viruses are fastidious *intracellular organisms* and therefore *living cells are required* to grow viruses in laboratory; cell lines can be *from human or non-human sources*
- *Continuous cell lines*- they can be maintained in indefinite growth cycles; they are immortalized cell lines derived mostly from tumour cells (*HeLA*- human cervical cancer cell line) or cells that have been transformed in the laboratory (*hep2* & *Graham293* – transformed human epithelial cell line);
- *Primary or semi-continuous cell lines* –they can be maintained for only one or a limited number of growth cycles; they are more sensitive to infection by viruses and *VZV, CMV, Influenza virus will only grow in them*; *MRC5* (human lung fibroblast cell line) & *PMK* (primary monkey kidney cell line)
- The *advantages*: very sensitive & specific technique;
- The *disadvantages*: labour intensive and required considerable technical expertise, results may take up to 2 weeks and the specimens need to be transported quickly and under correct conditions to the laboratory to maintain the viability of the infecting virus; Many viruses (*Hepatitis viruses, Papilloma viruses, Parvovirus B19, Rotavirus & Norovirus*) *cannot be grown* in cell culture; The *EBV & HIV need special cells*; therefore cell lines are not suitable in a routine diagnostic laboratory
- A suspension of cells in *growth medium* (consists of a buffer +calf serum to provide protein and amino acids + antibiotics to prevent bacterial overgrowth) is put in glass or plastic tubes; the cells attach to the sides of the container and grow until they become confluent. The *patient's specimen is then added* and *cells incubated at 37C* to allow the virus to grow. The tubes are examined daily to look for evidence of *virus growth*, which may take *from a day to weeks*. If virus is present then it kills off the cells and is referred to as *cytopathic effects (CPE)*; *diagnosis is made by EM, IF, etc.*

LABORATORY TECHNIQUES

BIOCHEMICAL TESTS

CATALASE TEST

The catalase test involves adding hydrogen peroxide to a culture sample. If the bacteria produce catalase, they will convert the hydrogen peroxide and oxygen gas will be evolved. The evolution of gas causes bubbles to form and is indicative of a positive test.



Positive (+) on the left, negative (-) right

COAGULASE TEST

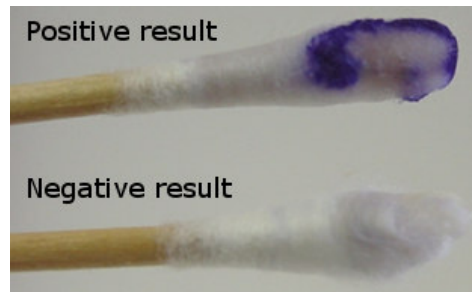
The sample in question is usually inoculated onto 0.5 ml of rabbit plasma and incubated at 37°C for 1-2 h. A positive test is denoted by a clot formation in the test tube



Positive (+) result on the left, negative (-) right

OXIDASE TEST

Cytochrome oxidase is an enzyme found in some bacteria that transfers electrons to oxygen, the final electron acceptor in some electron transport chains. Thus, the enzyme oxidizes reduced cytochrome c to make this transfer of energy. Presence of cytochrome oxidase can be detected through the use of an Oxidase reagent which acts as an electron donor to cytochrome oxidase. If the bacteria oxidize the disk (remove electrons) the disk will turn purple, indicating a positive test. No color change indicates a negative test.



Oxydase test

LECITHINASE activity results in the production of an opaque zone of precipitation around the area of growth

EVALUATION OF BIOCHEMICAL TEST

Ingredients	Positive	Negative
Indole	<i>red</i>	<i>yellow</i>
Urea	<i>red</i>	<i>yellow</i>
Esculin	<i>black</i>	<i>colorless</i>
Glucose	<i>yellow</i>	<i>blue</i>
Lactose	<i>yellow</i>	<i>blue</i>
Cucrose	<i>yellow</i>	<i>blue</i>
Maltose	<i>yellow</i>	<i>blue</i>
Mannitol	<i>yellow</i>	<i>blue</i>
H ₂ S	<i>black</i>	<i>colorless</i>
Lizine	<i>red</i>	<i>yellow</i>
Ornithine	<i>red</i>	<i>yellow</i>
Arginine	<i>red</i>	<i>yellow</i>
Simmons citrate	<i>green</i>	<i>yellow</i>
Trehalose	<i>yellow</i>	<i>blue</i>
Arabinose	<i>yellow</i>	<i>blue</i>
Rhamnose	<i>yellow</i>	<i>blue</i>
Xylose	<i>yellow</i>	<i>blue</i>
N-acethylglucosamine	<i>colorless</i>	<i>yellow</i>
Foges-proskauer test	<i>red</i>	<i>yellow</i>
Salicin	<i>yellow</i>	<i>blue</i>
Inozit	<i>yellow</i>	<i>blue/green</i>
Starch	<i>blue</i>	<i>colorless</i>
Phenylalanin	<i>blue</i>	<i>colorless</i>
Oxidase	<i>blue</i>	<i>colorless</i>
Twin 80	<i>blue</i>	<i>colorless</i>

Table 1 *Differential characteristics of Staphylococcus spp.*

<i>Species</i>	β Hemolysis	Coagulase	Mannitol	Novobiocin	Protein A
<i>S. aureus</i>	+	+	+	S	+
<i>S. epidermidis</i>	\pm	–	–	S	–
<i>S. saprophyticus</i>	–	–	+	R	–

S-sensitive; *R*-resistant

Table 2 *Differential characteristics of Streptococcus spp. and Enterococci*

<i>Species</i>	Hemolysis	Bacitracin	Optochin	Bile	6.5% NaCl	Aesculin
<i>S. pyogenes</i>	β	S	R	–	–	–
<i>S. agalactiae</i>	β	R (S)	R	–	\pm	–
<i>Enterococci</i>	α, β, γ	R	R	+	+	+
<i>S. pneumoniae</i>	α	R	S	–	–	–
<i>S. viridans group</i>	α	R (S)	R	–	–	–

Table 3 *Biochemical activity of Gram-negative microorganisms*

	<i>Pseudomonas</i>	<i>E. coli</i>	<i>P. vulgaris</i>	<i>P. mirabilis</i>	<i>K. pneumoniae</i>
Lactose	–	+	–	–	+
Glucose	+	+	+	+	+
H ₂ S	+	–	+	+	–
Mannitol	–	+	–	–	+
Lizin	–	+	–	–	+
Indole	–	+	+	–	–
Ornithin	–	\pm	–	+	–
Urea	+	–	+	+	+
Oxidase	–	–	–	–	–
Phenylalanine	–	–	+	+	–
Sucrose	–	+	+	\pm	+
Simmons citrate	+	–	–	\pm	+

Table 4 *Differential characteristics of common members of the Family Neisseriaceae*

	<i>Neisseria</i> <i>spp.</i>	<i>Kingella</i> <i>spp.</i>	<i>Moraxella</i> <i>spp.</i>	<i>Acinetobacter</i> <i>spp.</i>
Hemolysis (blood agar)	–	±	+	±
Oxidase test	+	+	+	–
Glucose	±	+	–	±
Lactose	±	–	–	–
Sucrose	±	–	–	–
Mannitol	±	–	–	–

Table 5 *Differential characteristics of Anaerobes*

Genus	<i>Bacteroides</i> <i>spp.</i>	<i>Prevotella</i> <i>spp.</i>	<i>Porphyromonas</i> <i>spp.</i>	<i>Fusobacterium</i> <i>spp.</i>
Catalase test	V	–	–	–
Bile	R	S	S	V
Vancomycin	R	R	S	R
Kanamycin	R	R	R	S
Colistin (polymixin E)	R	V	V	S
Pigment (black colonies)	–	V	+	–

R-resistant S-sensitive V-variable

Table 6 *Biochemical activity of Bacteroides spp.*

	<i>B.</i> <i>fragilis</i>	<i>B.</i> <i>vulgatus</i>	<i>B.</i> <i>distasonis</i>	<i>B.</i> <i>ovatus</i>	<i>B.</i> <i>uniformis</i>	<i>B.</i> <i>splanchnicus</i>
Indole	–	–	–	+	–	–
Trehalose	–	–	+	+	–	–
Sucrose	+	+	+	+	+	–
Arabinose	–	+	–	+	+	+
Rhamnose	–	+	±	+	±	–
Catalase test	+	±	+	+	–	–

Table 7 *Biochemical activity of Prevotella spp.*

	<i>P. oralis</i>	<i>P. buccalis</i>	<i>P. intermedia</i>	<i>P. melaninogenica</i>	<i>P. denticola</i>
Indole	–	–	+	–	–
Esculin	+	+	–	–	±
Glucose	+	+	+	+	+
Salicin	+	–	–	–	–
Xylose	–	–	–	–	–
Sucrose	+	+	+	+	–
N-acetylglucosamine	–	–	–	+	+

Table 8 *Biochemical activity of Porphyromonas spp.*

	<i>P. asacharolytica</i>	<i>P. gingivalis</i>	<i>P. endodontalis</i>
Indole	±	+	+
Esculin	–	–	–
Glucose	–	–	–
Salicin	–	–	–
Xylose	–	–	–
Sucrose	–	–	–
N-acetylglucosamine	–	+	–

Table 9 *Characteristics of Candida spp.*

Species	Blastoconidia	Pseudohyphae	Chlamydoconidia	Glucose	Maltose	Lactose	Sucrose	Galactos	Urease
<i>C. albicans</i>	+	+	+	+	+	–	+	+	–
<i>C. krusei</i>	+	+	–	+	–	+	–	–	+
<i>C. parapsilosis</i>	+	+	–	+	+	–	+	+	–
<i>C. tropicalis</i>	+	+	–	+	+	–	+	+	–
<i>C. guilliermondii</i>	+	+	–	+	+	–	+	+	–

LABORATORY TECHNIQUES

ANTIBIOTIC SUCCEPTIBILITY TESTING (AST)

Is carried out to determine which antibiotic will be most successful in treating a bacterial infection *in vivo*

KIRBY-BAUER ANTIBIOTIC TESTING (KB)/ disk diffusion antibiotic sensitivity testing uses antibiotic impregnated disks to test whether particular bacteria are susceptible to specific antibiotics

E-TEST (Epsilometer test) is used to determine whether or not a specific strain of bacterium or fungus is susceptible to the action of a specific antibiotic

Minimum inhibitory concentration (MIC) is the lowest concentration of an antimicrobial agent that will inhibit the visible growth of a microorganism after 24h incubation; a lower MIC is an indication of a better antimicrobial agent

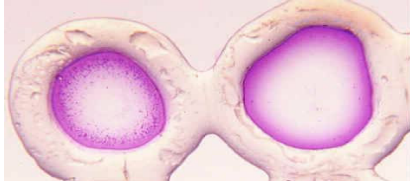
LABORATORY TECHNIQUES

SEROLOGICAL TECHNIQUES

- “*Serology*” means the study of serum and can be used to detect both Antibody (Ab) and Antigen (Ag). Several techniques have been developed, but the fundamental principles are similar for all.
- *Acute or recent infection can therefore be diagnosed by:* demonstrating the presence of (virus) specific IgM (IgG may or may not be present); *past infection or immunity is diagnosed:* demonstrating of (virus) specific IgG alone (and absence of IgM)
- IgA is produced at the local site of infection and provides *local immunity* (gut, respiratory tract); the *first* to appear is of the IgM class, which can be detected as early as a couple of days after an acute infection; **IgG appears** from 7-15 days after onset of infection. Both IgM & IgG continue to rise in response to the infection, peaking at about 6 weeks post infection; (viral) specific IgM then declines and is normally undetectable by about 3 months after infection; *IgG persists for life* and is responsible for *providing lifelong immunity* to the particular (virus)
- Serological assays can be *qualitative* (give only a yes or no answer) or to be *quantitative* (measures the Ab level); Assays that utilize the presence of IgM/IgG to make diagnosis are qualitative; If diagnosis relies on detection of a rising Ab “titer”, then the assay needs to measure the level of Ab response (quantitative). Ab “titer” is expressed as the inverse of the highest serum dilution at which the Ab is detected
- All serological techniques are based on the principle of adding specific (viral) Ag to patient serum; if virus-specific Ab is present in the serum then it will bind the Ag to form an Ag-Ab complex; an indicator system (depending on the technique) is then used to detect whether such a complex has been formed
- The latest generation of *automated machines* are called “*random access*”, as the specimens do not have to be batched and an urgent specimen can be put on the machine at any time without disrupting the other assays already on it; results can be obtained <1h; most laboratories linking their machines to a laboratory computer system/to hospital-based IT system – that allows the clinician to access results as soon as possible

SLIDE AGGLUTINATION TEST:

The aggregation of a suspension of bacteria (Ag) in the presence of specific Ab on glass slides



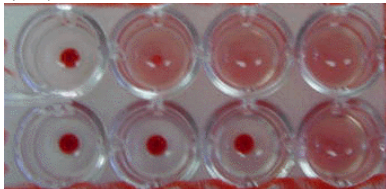
Positive (+) on the left, negative (-) right

LATEX AGGLUTINATION TEST:

In this reaction, the Antigen (Ag) or Antibody (Ab) is adsorbed on an inanimate particle (latex) and a positive reaction is indicated by agglutination of the particles.

COMPLEMENT FIXATION TEST (CFT):

The test is based on the principle that when an Ag-Ab complex is formed it will bind Complement (C'), so free C' is not available to lyse sensitized red cells that are added as indicator.



Positive (+) on the left, negative (-) right

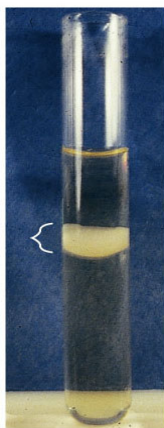
The reaction consists of two following steps: (1) A & Ab - one known and another is unknown- are mixed and C' -usually from guinea pig- is added; (2) An indicator system - "*sensitized*" red blood cells (red blood cells plus red-blood cell antibody) is added.

- If the Ab matched the Ag in the first step, C' was fixed and less available to the sensitized red blood cell; the red blood cells remain *unhemolyzed*. *the test is positive*
- If the Ab did not match the Ag in the first step, C' is free to attach to the sensitized red blood cell and they are lysed, red cells are *hemolyzed*, *the test is negative*

HAEMAGGLUTINATION (HA) & HAEMAGGLUTINATION INHIBITION TEST (HIT)

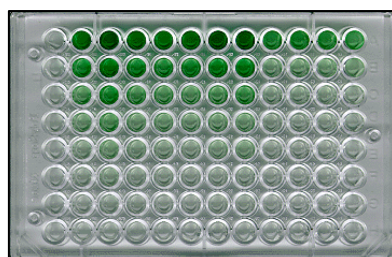
These tests detect Ab to *viruses (rubella, influenza)* that possess a *haemagglutinin A*; they are relatively insensitive and have been replaced by more sensitive and specific techniques

PRECIPITATION (TUBE) TEST:



In this reaction, the Ag is solution; in the *zone of equivalence*, optimal proportion of Ag and Ab combine; if the "precipitation ring" appears - the test is positive.

"Precipitation ring" – zone of equivalence shown.



ENZYME LINKED IMMUNOSORBENT ASSAY (ELISA/EIA):

- These are the most widely used serological assays in routine diagnostic laboratories;
- The test is performed in a 8x12 cm plastic *microtitre plate* which contains an 8x12 matrix of 96 wells, each of which are about 1 cm high and 0.7 cm in diameter
- There are four steps:
 - (1) Ag is attached to the base of a plastic microtitre well (solid phase);
 - (2) *patient's serum* is added to this microtitre well. If specific Ab is present in the serum it will attach to the Ag on the solid phase. Excess serum is washed off;
 - (3) *Anti-human Ig "labeled" with enzyme* is added to bind to this Ag-Ab complex. Excess enzyme is washed off;
 - (4) A *substrate* (chromogen) for the enzyme is added – a colour change indicates a *positive reaction*; *Positive & Negative controls* are added to the assay- to ensure the quality of the assay system
- The colour change in the EIA can be detected *by eye or measured in a spectrophotometer*; the intensity of the colour can indicate how much Ab is present in the serum. If the well is coloured, result is positive, and contrary if colorless - negative.
- The assay can also be *done in reverse to detect (viral) Ag* simply by coating the solid phase by Ab (monoclonal/polyclonal)

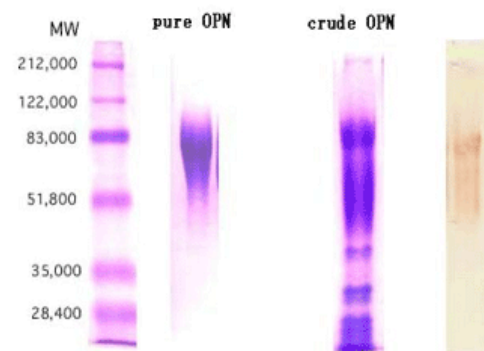
EIA is advantageous because of *rapidity (2-3h), objectivity and very high sensitivity & specificity (>95%)*

Newer ELISA-like techniques utilize fluorogenic, electrochemiluminescent, and real-time PCR reporters to create quantifiable signals.

WESTERN BLOT

Specific (viral) proteins are transferred on blotting paper from a gel. Further steps are similar of those of EIA.

The (viral) Ag band on the blotting paper develops colour if specific Ab to that particular Ag is present in the serum. The *advantage* of this technique is that the assay is able to distinguish Ab directed against specific (virus) protein and therefore is *very specific*.

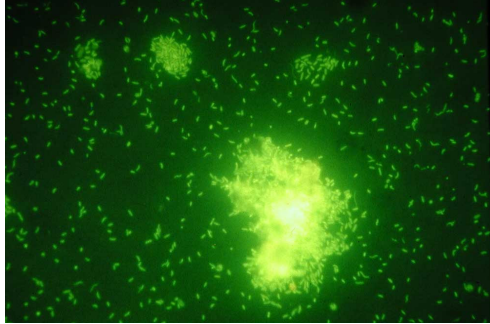


Western blot

RADIOIMMUNOASSAY (RAI):

This method is used for “*quantitation*” of *antigens (Ag)* or *haptens (Hp)* that can be *radioactively labeled* ; RIA is *highly sensitive method* - used to *assay hormones and drugs in serum*

IMMUNOFLUORESCENCE (IF)



These assays use the same principal as EIA; instead of the enzyme/substrate detector system of EIA, *fluorescein-labelled anti-human Ig* is used to detect a *positive reaction*, which appears as *apple-green fluorescence under a light (UV) microscope*

The IF *advantage: - is rapid; disadvantage – require subjective interpretation and is labour intensive to carry out and are dependent upon operator expertise*

(1) DIRECT IMMUNOFLUORESCENCE (dIF):

When *known “labeled” A)* interacts directly with *unknown Ag*

(2) INDIRECT IMMUNOFLUORESCENCE (inIF):

There are two steps: (1) *known Ag* is attached to the slide (2) *unknown Serum (Ab)* is added; the preparation is washed.

- If the unknown serum (Ab) matches the Ag, the *Ag-Ab complex will remain fixed on the slide*; it can be detected on addition of a *fluorescent “labeled” anti- human immune serum globulin* and by UV microscope

LABORATORY TECHNIQUES

NUCLEIC ACID AMPLIFICATION TECHNIQUES (NAATs)

POLYMERASE CHAIN REACTION (PCR)

The first NAAT to be described; it is a technique by which a single copy of DNA or RNA can be amplified more than million times. To detect RNA viruses, RNA has to be first transcribed to complementary DNA by means of an enzyme called *reverse transcriptase*; this type of PCR is referred to as *reverse transcription PCR (RT-PCR)*.

PCR used enzyme, *taq polymerase*, to initiate DNA amplification. The first steps in the process though are extraction and denaturation of the DNA or RNA followed by amplification. These steps involve complex chemical reactions, and heating and cooling of the sample mix in thermocycler. Each heating and cooling cycle takes only a few minutes to complete, and doubles the numbers of DNA copies; the amplified DNA product (*amplicons*) can be detected by use of specific probes labelled with a chemiluminescent or fluorescent dye. *Advantage: is exquisitely sensitive and used in routine diagnostic laboratories; Disadvantage: cross-contamination* (automated equipment is completely closed, cross-contamination is less of an issue)

NUCLEIC ACID SEQUENCE BASED AMPLIFICATION (NASBA)

Is used to detect DNA viruses (*HIV, HCV*)

STRAND DISPLACEMENT ASSAY (SDA):

Is used to detect *Chlamydia spp.*

“Seminar is a formal presentation by experts - in which each attendee is encouraged to present and discuss the subject matter”

SEMINAR

7.1 Pathogenicity & Virulence of Microorganisms

Topics for seminar:

1. Methods by which pathogens cause disease
 - Adhesion
 - Colonization
 - Invasion
 - Immune response inhibitors
2. Methods by which pathogens cause disease
 - Exotoxins
 - Endotoxins
3. Sources of infection & Modes of infection
 - Ingestion
 - Respiratory route
 - Mucous membranes
 - Inoculation
 - Animal bites
4. Transmission of infection
 - Person-to-person spread
 - Food-borne infections
 - Water-borne infections
 - Air-borne infections
 - Insect-borne infections
5. Anti-microbial agents
 - Antibiotics vs. Chemotherapeutic agents
 - Mechanism of action of Antimicrobials
 - Inhibitors of cell wall synthesis
 - Inhibitors of cell membrane synthesis
 - Inhibitors of protein synthesis
 - Inhibitors of DNA and cell division
 - Microorganisms that produce antibiotics:
 - *Penicillium and Cephalosporium molds*
 - *Actinomycetes (Streptomyces spp.)*
 - *Bacillus spp.*

- Antivirals
- Antifungals
- Antiparasitics
- Complications of non-rational antibiotic therapy

6. Antibiotic resistance:

- Causes
- Mechanisms
- Resistant pathogens
 - *S. aureus*
 - *S. pyogenes*
 - *S. pneumoniae*
 - *E. faecium*
 - *P.aeruginosa*
 - *C. difficile*
 - *E. coli*
 - *A. baumannii*
- Prevention
- Phage therapy

7. Intracellular parasitism

- Bacteria
- Viruses
- Fungi

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- Brooks GF, Carroll KC, Butel JS, Morse SA. Jawetz, Melnick, and Adelberg's medical microbiology. McGraw Hill Medical; 2010.
- *Levinson W. Review of Medical Microbiology and Immunology. 11th ed. New York [etc.]: McGraw-Hill Medical; 2010.
- *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008.
- *Engelkirk PG. Laboratory Diagnosis of Infectious Diseases: Essentials of Diagnostic Microbiology. Baltimore (Md.): Wolters Kluwer/Lippincott Williams & Wilkins; 2008.
- *Engleberg NC. Schaechter's Mechanisms of Microbial Disease. 4th ed. Philadelphia [Pa.]: Lippincott Williams & Wilkins; 2007.
- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005.
- Greenwood D, Slack RCB, Peutherer JF. Medical microbiology: a guide to microbial infections □: pathogenesis, immunity, laboratory diagnosis, and control. Churchill Livingstone; 2002.
- Levinson W, Jawetz E. Medical microbiology & immunology: examination & board review. Lange Medical Books/McGraw-Hill; 2002.
- Truant AL. Manual of commercial methods in clinical microbiology. ASM Press; 2002

SEMINAR

7.3 Microbiological Diagnosis of Fungal Infections

Topics for seminar:

- Microscopy
 - Yeast
 - *Candida albicans*
 - *Saccharomyces cerevisiae*
 - *Cryptococcus neoformans*
 - Molds (*Aspergillus spp.*)
 - Dimorphic (*Blastomyces dermatitidis*)
- Cultivation of fungi
 - Sabouraud's agar
 - CHROMagar Candida (*C. albicans*)
- Immunological methods
 - ELISA (*C. albicans* IgG)
- Molecular diagnosis
 - PCR

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- Kavanagh K. Fungi: Biology and Applications. John Wiley and Sons; 2011.
- Brooks GF, Carroll KC, Butel JS, Morse SA. Jawetz, Melnick, and Adelberg's medical microbiology. McGraw Hill Medical; 2010.
- *Levinson W. Review of Medical Microbiology and Immunology. 11th ed. New York [etc.]: McGraw-Hill Medical; 2010.
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- *Engleberg NC. Schaechter's Mechanisms of Microbial Disease. 4th ed. Philadelphia [Pa.]: Lippincott Williams & Wilkins; 2007.
- *Murray PR. Medical Microbiology. 5th ed. Philadelphia (Pa.): Elsevier Mosby; 2005.
- Levinson W, Jawetz E. Medical microbiology & immunology: examination & board review. Lange Medical Books/McGraw-Hill; 2002.
- Greenwood D, Slack RCB, Peutherer JF. Medical microbiology: a guide to microbial infections □: pathogenesis, immunity, laboratory diagnosis, and control. Churchill Livingstone; 2002.
- Truant AL. Manual of commercial methods in clinical microbiology. ASM Press; 2002

SEMINAR

7.4 Laboratory diagnosis of viral infections

Topics for seminar:

- Microscopy
 - *cytopathic effects (CPE)* of viral infection in tissue culture
 - *inclusion bodies* in *biopsy tissue specimens*
- *Virus isolation and growth*
 - *Cell culture*
 - *Growth of viruses in embryonated eggs*
- Viral detection
 - Hemagglutination assay
 - Serological methods
 - direct & indirect methods for antigen detection
 - visualization of proteins by Immunoprecipitation
 - visualization of proteins by Immunoblotting
 - detection of viral antigen or antibodies by ELISA
 - the use of green fluorescent protein
 - Nucleic acid detection
 - Southern blot hybridization
 - DNA microarray technology

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- Cann AJ. Principles of Molecular Virology. Academic Press; 2011
- Mahy BWJ, Regenmortel MHV van. Desk Encyclopedia of Human and Medical Virology. Academic Press; 2009.
- *Engelkirk PG. Laboratory Diagnosis of Infectious Diseases: Essentials of Diagnostic Microbiology. Baltimore (Md.): Wolters Kluwer/Lippincott Williams & Wilkins; 2008.
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SEMINAR

7.6 Oncogenic viruses & Slow virus infections. Prions

Topics for seminar:

Oncogenic viruses

- DNA
 - *Papovaviridae* (*Human papillomavirus (HPV)*)
 - HPV-positive- oropharyngeal cancer
 - HPV-positive cervical cancer
 - HPV vaccine
 - *Herpesviridae* *Lymphocryptovirus (Epstein-Barr virus)*
 - Nasopharyngeal carcinoma
 - Burkitt's lymphoma
 - *Hepadnaviridae* (Hepatitis B virus (HBV))
 - Hepatocellular carcinoma
 - Pancreatic cancer
 - Adenoviridae
 - Poxviridae
- RNA
 - Human T-cell leukemia virus (HTLV-1, HTLV-2)
 - Feline Leukemia Virus (FeLV)
- Slow virus infections
 - Slow virus diseases of the nervous system
- Prions
 - structure
 - properties
 - transmission
 - pathogenesis
 - prion diseases
 - Creutzfeldt-Jakob disease (CJD)
 - Gerstmann-Sträussler-Scheinker syndrome (GSS)
 - Fatal insomnia (FI)

Reading Assignment:

**these books are available at the Library of Health Sciences, Eiveniu str. 2, Kaunas*

- Cann AJ. Principles of Molecular Virology. Academic Press; 2011
- Brooks GF, Carroll KC, Butel JS, Morse SA. Jawetz, Melnick, and Adelberg's medical microbiology. McGraw Hill Medical; 2010.
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STUDY PROGRAMME

1. The normal microflora and its role in physiological and pathological processes. Normal microflora - secondary and opportunistic pathogens. Dysbiosis and major reasons for its development. Probiotics and prebiotics.
2. Factors influencing development of infectious process. Primary, secondary and opportunistic pathogens. Peculiarities of the infectious diseases. Infectious disease dynamics and its stages. Primary and secondary infections, superinfections, reinfections and recurrent infections.
3. The peculiarities of viral infections and their different appearance. The stages of viral infections and factors influencing them.
4. Hospital-acquired (nosocomial) infections, the sources and routes of spread of them. Hospital strains of microorganisms: the factors influencing their spreading, general markers of determination. Multiresistance – one of the hospital strain-specific characteristics.
5. Pathogenicity and virulence of microorganisms, virulence factors. Adhesion, colonization, invasion factors, toxigenicity and intracellular parasitism of bacteria.
6. Antimicrobial agents: classification, properties and mechanisms of action. Complications of non-rational antibiotic therapy.
7. Resistance to antibacterial drugs: innate (natural) and acquired (adapted). Antimicrobial drug acquired resistance: principles and mechanisms. Hospital and community acquired resistance. Antibiotic susceptibility testing: disk diffusion tests, minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC).
8. The genus *Staphylococcus*. Species: *S. aureus*, *S. epidermidis*, *S. saprophyticus*, their biological properties, ecology, epidemiology, infections and diseases caused by them. Virulence factors of *S.aureus*. The strains of methicillin-resistant *S. aureus* (MRSA)
9. Classification of *Streptococci*, the most important groups and species: their biological properties, ecology, epidemiology. Virulence factors of *S.pyogenes*, infections and diseases caused by them, immunity and prophylaxis.
10. *S. pneumoniae*: biological properties, ecology, epidemiology, virulence factors. Infections: pneumonia, otitis media, septicemia, meningitis, sinusitis. Immunity and immunoprophylaxis.
11. The genus *Enterococcus*: the most important species, their biological properties and virulence factors, ecology, epidemiology, infections caused by them.
12. *N. gonorrhoeae*: biological properties, ecology, epidemiology, virulence factors, infections caused by them. Pathogenesis and immunity.
13. *N. meningitidis*: biological properties, ecology, epidemiology, and virulence factors, infections caused by them. Pathogenesis and immunity.
14. The family *Enterobacteriaceae*. *E. coli*: biological properties, ecology, epidemiology, virulence factors, and infections caused by them. *E. coli* serological groups and their role in infectious pathology.
15. The family *Enterobacteriaceae*. *Salmonella spp*: biological properties, ecology, epidemiology, virulence factors, and infections caused by them. Immunity and prophylaxis.

16. The family *Enterobacteriaceae*. *Shigella spp*: biological properties, ecology, epidemiology, virulence factors, and infections caused by them. Immunity and prophylaxis.
17. The family *Enterobacteriaceae*. *Klebsiella spp*: biological properties, ecology, epidemiology, virulence factors, and infections caused by them. Immunity and prophylaxis.
18. The family *Enterobacteriaceae*. *Proteus spp*: biological properties, ecology, epidemiology, virulence factors, and infections caused by them. Immunity and prophylaxis.
19. *V. cholerae* – cause of cholera: ecology, biological properties, virulence factors, and epidemiology. Immunity and prophylaxis.
20. *H. pylori* – cause of gastritis, peptic ulcer disease, gastric cancer: its prevalence, epidemiology and virulence factors.
21. The family *Spirochetaceae*. *Treponema, Borrelia, Leptospirae*: biological properties, ecology, epidemiology, virulence factors, and infections caused by them. Prophylaxis.
22. The family *Pseudomonadaceae*. *P. aeruginosa* biological properties, ecology, epidemiology, virulence factors, and infections caused by them. Hospital –acquired infections.
23. The family *Pasteurellaceae*. *H. influenzae* biological properties, ecology, epidemiology, virulence factors, and infections caused by them. Specific prophylaxis.
24. *Mycobacteria*-their structure and chemical composition. *M. tuberculosis* – causative agent of tuberculosis (TB): virulence factors, intracellular parasitism and delayed-type hypersensitivity reaction. Prevalence, modes of transmission and identification of the infection (Mantu test). Specific prophylaxis.
25. The genus *Chlamydia*: species, biological properties, ecology, modes of transmission, intracellular parasitism. Infections caused by *C. trachomatis* and *C.pneumoniae*, their virulence factors.
26. The family *Mycoplasmataceae* - prokaryotes that lack their cell wall. The species, virulence factors and infections caused by them. Ecology and epidemiology of *Mycoplasma* and *Ureaplasma*.
27. Definition of hospital (nosocomial) infections, their frequency in different hospital departments. Endogenous and Exogenous hospital infections, their epidemiology. The source of infection, modes of transmission and prevalence.
28. The role of normal microflora in the etiology of hospital infections. Microorganisms related to hospital infections: pathogenic and opportunistic microorganisms. Ecological variants of hospital strains, their risk factors, virulence, resistance to environmental and chemical factors, multiresistance to antibiotics.
29. The fever of unknown origin (FUO). The most common Gram-positive and Gram-negative infection agents, their pathogenesis. Laboratory diagnosis and prevention of FUO.
30. Bacterial and viral upper respiratory tract infections, the most common causative agents, ways of infections. Laboratory diagnosis and prevention of the upper respiratory tract infections (URTI).

- 31.** Bacterial and viral lower respiratory tract infections (LRTI), the most common causative agents, ways of infections. Hospital-acquired LRTI, the sources and routes of transmission. Laboratory diagnosis and prevention of the LRTI.
- 32.** Congenital and acquired neonatal infections, the most common bacterial and viral causative agents, the routes of transmission. Laboratory diagnosis of the neonatal infections. Prophylaxis of fetus and newborns infections.
- 33.** Bacterial and viral urinary tract and genital tract infections, the most common causative agents, routes of transmission. Hospital-acquired urinary tract infections (UTI), the sources and the routes of transmission. Laboratory diagnosis and prevention of UTI.
- 34.** Bacterial and viral central nervous system (CNS) infections, the most common causative agents and their way of transmission. Laboratory diagnosis and prevention of the CNS infections.
- 35.** Bacterial and viral gastrointestinal tract infections (GIT), the most common causative agents and their routes of transmission. Laboratory diagnosis and prevention of GIT.
- 36.** Infective endocarditis, the most common causative agents their way of transmission. Laboratory diagnosis and prevention of infective endocarditis.

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2. Kavanagh K. Fungi: Biology and Applications. John Wiley and Sons; 2011.
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4. *Levinson W. Review of Medical Microbiology and Immunology. 11th ed. New York [etc.]: McGraw-Hill Medical; 2010.
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9. *Murphy K. Janeway's Immunobiology. 7th ed. New York: Garland Science; 2008.
10. *Mims' Medical Microbiology. 4th ed. Philadelphia (Pa.): Mosby; 2008.
11. *Engelkirk PG. Laboratory Diagnosis of Infectious Diseases: Essentials of Diagnostic Microbiology. Baltimore (Md.): Wolters Kluwer/Lippincott Williams & Wilkins; 2008.
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14. *Nairn R. Immunology for Medical Students. 2nd ed. Philadelphia (Pa.): Mosby Elsevier; 2007.
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29. *Case Studies in Immunology: Companion to Immunology Fifth Ed. Jonathan Brostoff, Alexander Gray, David Male, Ivan Roitt. London [etc.]: Mosby; 1998.

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31. <http://wwwnc.cdc.gov/eid/>
32. <http://www.cdc.gov/DiseasesConditions/>
33. <http://labtestsonline.org/>
34. <http://www.microbelibrary.org/>
35. <http://www.who.int/en/>

ABBREVIATIONS

AIDS	acquired immune deficiency syndrome	Mab	monoclonal antibody
BCG	Bacille Clamette-Guerin	MALTOMA	mucosa-associated lymphoid tissue lymphomas
CBC	complete blood count	MBC	minimal bactericidal concentration
CD	cluster of differentiation	MHC	major histocompatibility complex
CFT	complement fixation test	MBC	minimal bactericidal concentration
CFU	colony forming unit	MIC	minimal inhibitory concentration
CLL	chronic lymphocytic leukemia	MLD	minimal lethal dose
CML	chronic myelogenous leukemia	MMR	measles, mumps, rubella (vaccine)
CMV	cytomegalovirus	MRSA	methicilin (multi-) resistant <i>S. aureus</i>
CNS	central nervous system	MS	multiple sclerosis
CPE	cytopathic effect	MSU	mid-stream urine
CSF	cerebrospinal fluid	NGU	non-gonococcal urethritis
CT	cholera toxin	NLF	non-lactose-fermenter
DFA-TP	direct fluorescent antibody test for <i>T. pallidum</i>	NK	natural killer (cell)
DIC	disseminated intravascular coagulation	NSU	non-specific urethritis
DPT	diphtheria, pertussis, tetanus combined vaccine	NSV	non-specific vaginitis (anaerobic vaginosis)
EBV	Epstein-Barr virus	OPV	oral polio vaccine
ECHO	enteric cytopathic human orphan (viruses)	PAGE	polyacrylamide gel electrophoresis
EIA	enzyme immunoassay	PCR	polymerase chain reaction
ELISA	enzyme-linked immunosorbent assay	PBMC	peripheral blood mononuclear cells
EM	erythema migrans	PGU	post-gonococcal urethritis
EPEC	enteropathogenic <i>Escherichia coli</i>	PMC	pseudomembranous colitis
ETEC	enterotoxigenic <i>Escherichia coli</i>	PMN	polymorphonuclear leukocyte
FIA	fluorescence immunoassay	PNS	peripheral nervous system
GI	gastrointestinal	PPD	purified protein derivative
H	flagellum antigen	PUO	pyrexia of unknown origin
HI	haemagglutination inhibition (test)	RA	rheumatoid arthritis
HAV	hepatitis A virus	RF	rheumatoid factor
Hb	hemoglobin	RIA	radioimmunoassay
Hib	<i>Haemophilus influenzae type b</i>	RIBA	recombinant immunoblot assay
HIV	human immunodeficiency virus	RNA	ribonucleic acid
HPV	human papilloma virus	RPR	rapid plasma regain (test)
HSV	herpes simplex virus	RSV	respiratory syncytial virus
HTLV	human T cell lymphotropic virus	RT-PCR	reverse transcriptase PCR
HTLV-1	human T cell leukemia virus type 1	SMAC	sorbitol-MacConkey agar
IB	immunoblot or immunoblotting	spp.	species, pl.
ID	infective dose	STD	sexually transmitted disease
IFN	interferon	TB	tubercle bacilli (used to denote tuberculosis)
Ig	immunoglobulin	TNF	tumour necrosis factor
IIF	indirect immunofluorescence (test)	TORCH	toxoplasma, <i>Rubella virus</i> , <i>CMV</i> , <i>HSV</i> (acronym for four infectious agents that must be considered in congenital infections)
IL	interleukin	TPHA	<i>T. pallidum</i> haemagglutination (test)
IM	intramuscular	TPI	<i>T. pallidum</i> immobilisation (test)
IPV	inactivated polio vaccine	TSS	toxic shock syndrome
IVIG	intravenous immunoglobulin G	TT	tetanus toxoid
K	capsular or envelope (antigens, antibodies)	UHT	ultra-heat treated (milk)
kb	kilobase	URTI	upper respiratory tract infection
kbp	kilobase pair	UTI	urinary tract infection
LA	latex agglutination	VDRL	Venereal Diseases Research Laboratory (test)
LCR	ligase chain reaction	Vi	virulence factor (antigen of <i>S. typhi</i> , etc.)
LD ₅₀	dose lethal to 50% of experimental animals	VRE	vancomycin-resistant enterococci
LGV	lymphogranuloma venereum	VTEC	verotoxigenic <i>E. coli</i>
LPS	lypopolysaccharide	VZV	<i>varicella-zoster virus</i>
LRTI	Lower respiratory tract infection	WBC	white blood cells
LT	heat-labile toxin	ZN	Ziehl-Neelsen (detect <i>M. tuberculosis</i>)

GLOSSARY

acid-fast bacteria retaining initial stain and difficult to decolorize with acid fast

aerobe any oxygen-requiring organism

aetiology the study of the cause of a disease

abscess an accumulation of pus in a cavity hollowed out by tissue damage

acute disease a disease that develops rapidly and runs its course quickly

adherence the attachment of a microorganism to a host's surface

aflatoxin fungal toxin that is a potent carcinogen

agar plate a plate of nutrient medium solidified with agar

amantadine an antiviral agent that prevent penetration by influenza A viruses

anaerobe an organism that grows in the absence of molecular oxygen

antagonism the decreased effect when two antibiotics are administered together

anthrax a zoonosis caused by *Bacillus anthracis*

antibiotic a substance of microbial origin that has antimicrobial activity

antibody a protein produced in response to an antigen (immunoglobulin)

antibody titer the quantity of a specific antibody in an individual's blood

antigen a substance that the body identifies as foreign

antimicrobial agent a chemotherapeutic agent used to treat diseases caused by microbes

antiseptic a disinfectant that can be applied to the skin to prevent or stop growth of microorganisms

antiserum, pl. antisera serum that contains antibodies

antitoxin an antibody against a specific toxin

aseptic technique a set of procedures used to minimize chances that cultures will be contaminated by organisms from the environment

aspergillosis skin infection caused by various species of *Aspergillus*, which can cause severe pneumonia in immunosuppressed patients

asthma respiratory anaphylaxis caused by inhaled or ingested allergens or by hypersensitivity to endogenous microorganisms

attenuation weakening; reduction in virulence

atrichous a bacterial cell without flagella

autoantibody an antibody against one's own tissues

autoclave an apparatus using steam under pressure for sterilization

bacillus any rod-shaped bacterium

bacteraemia a condition in which bacteria are present in the bloodstream

bacterium, pl. bacteria diverse and ubiquitous prokaryotic single-celled microorganism

bacterial conjunctivitis a highly contagious inflammation of the conjunctiva caused by various bacterial species (also called pinkeye)

bacterial endocarditis a life-threatening infection of the lining and valves of the heart (also called infective endocarditis)

bacterial enteritis an intestinal infection caused by bacterial invasion of intestinal mucosa or deeper tissues

bactericide an agent that kills bacteria

bacteriophage a virus that infects bacteria and causes lysis of bacterial cells

bacteriostatic an agent that inhibits the growth of bacteria

beta (β) haemolysis complete lysis of red blood cells by bacterial enzymes; a colourless, defined zone of haemolysis surrounding certain bacterial colonies growing on blood agar

binomial nomenclature the system of taxonomy developed by Linnaeus in which each organism is assigned a genus and specific epithet

biological vector an organism that actively transmits pathogens that complete part of their life cycle within the organism

blastomycosis fungal skin disease caused by *Blastomyces dermatitidis*

botulism disease caused by *Clostridium botulinum*

brain abscess a pus-filled cavity caused by microorganisms reaching the brain from head wounds or via blood from another site

bronchial pneumonia type of pneumonia that begins in the bronchi and can spread through surrounding tissue toward the alveoli

bronchitis an infection of the bronchi

cancer an uncontrolled, invasive growth of abnormal cells

candidiasis a yeast infection caused by *Candida albicans* that appear as thrush (in mouth) or vaginitis (also called **moniliasis**)

capside the protective protein coat of a virus particle

capsule an enveloped or slime layer surrounding the cell wall of certain microorganisms

carbuncle a massive pus-filled lesion resulting from an infection, particularly of the neck and upper back

carrier a person in apparently good health who harbours a pathogenic microorganism

cell-mediated immunity the immune response involving the direct action of T cells to activate B cells or to destroy microbe-infected cells, tumor cells, or transplanted cells (organ transplants)

central nervous system the brain and spinal cord

cephalosporin an antibacterial agent that inhibits cell wall synthesis

chancre a hard, painless, nondischarging lesion; a symptom of primary stage syphilis

chemotherapeutic agent any chemical substance used to treat disease (also called **drug**)

chemotherapeutic index the maximum tolerable dose of a particular drug per kilogram body weight divided by the minimum dose per kilogram body weight that will cure the disease

chocolate agar type of medium made with heated blood, so named because it turns a chocolate brown color

chronic disease a disease that develops more slowly than an acute disease, is usually less severe, and persists for a long, indeterminate period

cluster of differentiation marker an antigen found on the cell surface of B and T cells that can be used to distinguish the cells from one another

coagulase an enzyme, produced by pathogenic staphylococci, that causes coagulation of blood plasma

coccus, pl. cocci a spherical bacterium

colicin a protein released by some strains of *Escherichia coli* that inhibits growth of other strains of the same microorganism

colonization growth of microorganisms on epithelial surfaces such as skin or mucous membranes

colony a macroscopically visible growth of microorganisms on a solid culture medium

commensalism a symbiotic relationship between members of different species living in proximity in which one organism benefits and the other one neither benefits nor is harmed by the relationship

complement a set of more than 20 large regulatory proteins that circulate in plasma and when activated form a nonspecific defense mechanism against many different microorganisms

compromised host a person already weakened with debilitating disease

contamination entry of undesirable organisms into some material or object

convalescent stage the stage of an infectious disease during which tissues are repaired, healing takes place, and the body regains strength and recovers

cross-reaction immune reaction of a single antibody with different antigens that are similar in structure

cross-resistance resistance against two or more similar antimicrobial agents through a common mechanism

cystitis inflammation of the bladder

cytopathic effect cellular injury caused by virus infection; the effects of virus infection on cultured cells, visible by microscopic or direct visual examination

cytotoxic T cell (Tc) lymphocyte that destroys virus-infected cells

culture a population of microorganisms cultivated in a medium

dental caries the erosion of enamel and deeper parts of teeth (also called **tooth decay**)

dental plaque a continuously formed coating of microorganisms and organic matter on the tooth enamel

dimorphism the ability of an organism to alter its structure when it changes habitats

diphtheria a severe upper respiratory disease by *Corynebacterium diphtheriae*

dipicolinic acid acid found in the core of an endospore that contributes to its heat resistance

direct fecal-oral transmission direct contact transmission of disease in which pathogens from fecal matter are spread by unwashed hands to the mouth

disease a state of impaired body function occurring as a response to infection, stress, or other conditions

disinfectant an agent that kills most, but not all, microorganisms

disk diffusion method a method used to determine microbial sensitivity to antimicrobial agents in which antibiotic disks are placed on an inoculated petri dish, incubated, and observed for inhibition of growth (also called **Kirby-Bauer method**)

DNA polymerase an enzyme that adds nucleotides and synthesizes DNA in one direction

dysentery a severe diarrhea that often contains mucus and sometimes blood or pus

dysuria pain and burning on urination

ecology the study of relationship among organisms and their environment

edema an accumulation of fluid in tissues that causes swelling

endemic a disease that is constantly present in a specific population

endogenous infection an infection caused by opportunistic microorganisms already present in the body

endospore a resistant, dormant structure, formed inside some bacteria such as *Bacillus* and *Clostridium*, that can survive adverse conditions

endotoxin a toxin incorporated in Gram-negative bacterial cell wall and released when the bacterium dies (also called **lipopolysaccharide**)

enteritis an inflammation of the intestine

enterocolitis disorder caused by *Salmonella typhimurium* and *S. paratyphi* that invade intestinal tissue and produce bacteremia

enteropathogenic an organism that causes intestinal disease

enterotoxin a toxin specific for cells of the intestine. It gives rise to symptoms of food poisoning

epidemic a sudden increase in the incidence of a disease, affecting large numbers of people over a wide area

epidemiologic study a study conducted in order to learn more about the spread of a disease in a population

etiology the assignment of causes and origins of a disease

eukaryote a cell that possesses a definitive or true nucleus

exogenous produced or originating from without

exotoxin a toxin excreted by a microorganism into the surrounding medium

fimbriae surface appendages of certain Gram-negative bacteria composed of protein subunits.

Also called **pili**

fission an asexual process by which some microorganisms reproduce

flagellum, pl. **flagella** whip-like appendage on cells used for locomotion

flora in microbiology, the microorganisms present in a given situation, e.g. intestinal flora

food poisoning a gastrointestinal disease caused by ingestion of foods contaminated with preformed toxins and other toxic substances (also called **enterotoxigenesis**)

fungicide an agent that kills or destroys fungi

gas gangrene a deep wound infection, destructive of tissue, often caused by a combination of two or more species of *Clostridium*

gastroenteritis inflammation of the mucosa of the stomach and intestine

gonorrhoea a sexually transmitted disease caused by *Neisseria gonorrhoeae*

genotype the particular set of genes present in an organism and its cells; an organism's genetic constitution

genus, pl. **genera** a group of very closely related species

germicide an agent capable of killing germs, usually pathogenic microorganisms

Gram-negative bacteria bacteria that appear red after being subjected to the Gram stain

Gram-positive bacteria bacteria that appear blue or violet after being subjected to the Gram stain

H antigen a type of antigen found in the flagella or certain bacteria

haemolysis the process of dissolving red blood cells

helper T cells (T_H) lymphocytes that stimulate other immune cells, such as B cells and macrophages

horizontal transmission direct contact of disease in which pathogens are usually passed by handshaking, kissing, contact with sores, or sexual contact

host an organism that harbors another organism

ID infective dose; the number of microorganisms required to infect a host

ID₅₀ the dose (number of microorganisms) that will infect 50% of the experimental animals in a test series

immortalized cell a cell capable of indefinite growth in culture

immunity the ability of an organism to recognize and defend itself against infectious agents

immunocompromised an individual whose immune defenses are weakened due to fighting another infectious disease, or because of an immunodeficiency disease or an immunosuppressive agent

incubation period in the stages of an infectious disease, the time between infection and the appearance of signs and symptoms

infectious disease disease caused by infectious agents

inoculation the artificial introduction of microorganisms into the body or into a culture medium

intoxication the ingestion of a microbial toxin that leads to a disease

isolation situation in which a patient with a communicable disease is prevented from contact with the general population

in vitro literally, "in glass" and pertaining to biological experiments performed in test tubes or other laboratory vessels

in vivo within the living organism; pertaining to laboratory testing of agents within living organisms

kb a unit of measurement of single-stranded nucleic acid molecules

kbp a unit of measurement of double-stranded nucleic acid molecules

Koch's postulates four postulates formulated by Robert Koch in the 19th century; used to prove that a particular organism causes a particular disease

lactobacilli type of regular, nonsporing, Gram-positive rods found in many foods; used in production of cheeses, yogurt and other fermented foods

LD₅₀ the dose (number of microorganisms) that will kill 50% of the animals in a test series

Legionnaires' disease disease caused by legionella pneumophila, transmitted by airborne bacteria

leptospirosis a zoonosis caused by the spirochete *Leptospira interrogans*

L forms irregularly shaped naturally occurring bacteria with defective cell walls

listeriosis a type of meningitis caused by *Listeria monocytogenes*

Lyme disease disease caused by *Borrelia burgdorferi*, carried by the deer tick

lymphogranuloma venereum a sexually transmitted disease, caused by *Chlamydia trachomatis*, that attacks the lymphatic system

major histocompatibility complex a group of cell surface proteins that are essential to immune recognition reactions

memory cells long-lived B or T lymphocyte that can carry out an anamnestic or secondary response

minimal bactericidal concentration the lowest concentration of an antimicrobial agent that kills microorganisms, as indicated by absence of growth following subculturing in the dilution method

minimal inhibitory concentration the lowest concentration of an antimicrobial agent that prevents growth in the dilution method of determining antibiotic sensitivity

mixed infection an infection caused by several species of organism present at the same time

monoclonal antibody a single, pure antibody, produced in the laboratory by a clone of cultured hybridoma cells

monotrichous a bacterial cell with a single flagellum

morbidity rate the number of persons contracting a specific disease in relation to the total population (cases per 100,000)

mortality rate the number of death from a specific disease in relation to the total population

morphology the branch of biological science that deals with the study of the structure and form of living organisms

mumps disease caused by a *Paramyxovirus* that is transmitted by saliva and invade cells of the oropharynx

mutualism a symbiotic relationship in which both organisms benefit from the relationship

mycelium, pl. mycelia in fungi, a mass of long, threadlike structures (hyphae) that branch and intertwine

mycology the study of fungi

mycoplasmas very small bacteria with cell membranes, RNA and DNA, but no cell wall

mycosis, pl. mycoses a disease caused by fungi

mycotoxin any toxic substance produced by fungi

multiplicity of infection the (average) number of virus particles that infect each cell in an experiment

narrow spectrum the range of activity of an antimicrobial agent that attacks only a few kinds of microorganisms

natural killer (NK) cells a lymphocyte that can destroy virus-infected cells, malignant tumor cells, and cells of transplant tissue

nongonococcal urethritis a gonorrhea-like sexually transmitted disease most often caused by *Chlamydia trachomatis* and *mycoplasmas*

normal flora microorganisms that live on or in the body, but do not usually cause disease (also called **normal microflora**)

nosocomial disease an infection acquired in a hospital or other medical facility

obligate intracellular parasite an organism or virus that can live or multiply only inside a living host cell

obligate psychrophile an organism that cannot grow at temperatures above 20°C

obligate thermophile an organism that can grow only at temperatures above 37°C

oncogene a cancer-causing gene

oedema excessive accumulation of fluid in body tissue

opportunistic microorganism a microorganisms that exists as part of the normal flora but becomes pathogenic when transferred from the normal habitat into other areas of the host or when host resistance is lowered

pandemic an epidemic that has become worldwide

parasite an organism that lives in or on, and at the expense of, another organism, the host

parenteral by some route other than via the intestinal tract

passive immunization the process of inducing immunity by introducing ready-made antibodies into a host

pasteurization the process of heating liquid food or beverages at a controlled temperature to enhance the keeping quality and destroy harmful microorganisms

pathogen an organism capable of producing disease

penetration the entry of the virus (or its nucleic acid) into the host cell in the replication process

peritrichous having flagella around the entire surface of the cell

persistent viral infection the continued production of viruses within the host over many months or years

phage-typing identifying a pathogenic bacterium by the pattern of lysis caused by different phage-types

plasmid an extrachromosomal genetic element capable of autonomous replication

phenotype the observable characteristics of an organism

prion a proteinaceous infectious particle, believed to be responsible for transmissible spongiform encephalopathies such as Creutzfeldt-Jacob disease (CJD) or bovine spongiform encephalopathy (BSE)

poliomyelitis disease caused by any of several strains of *Polioviruses* that attack motor neurons and the spinal cord and brain

polyacrylamide gel electrophoresis a technique for separating proteins from a cell based on their molecular size

portal of entry a site at which microorganisms can gain access to body tissues

portal of exit a site at which microorganisms can leave the body

potable water water that is fit for human consumption

preserved culture a culture in which microorganisms are maintained in a dormant state

prevalence rate the number of people infected with a particular disease at any one time

primary atypical pneumonia a mild form of pneumonia with insidious onset (also called **mycoplasma pneumonia** or **walking pneumonia**)

primary infection an initial infection in a previously healthy person

prodromal phase in an infectious disease, the short period during which nonspecific symptoms such as malaise and headache sometimes appear

prokaryote a type of cell in which the nuclear substance is not enclosed within a membrane: e.g. a bacterium

prophylaxis preventive treatment for protection against disease

protoplast a Gram-positive bacterium from which the cell wall has been removed

pseudomembrane a combination of bacilli, damaged epithelial cells, fibrin, and blood cells resulting in infection with diphtheria that can block the airway, causing suffocation

pure culture a culture that contains only a single species of organism

pyoderma a pus-producing skin infection caused by *staphylococci*, *streptococci*, and *corynebacteria*, singly or in combination

rabies a viral disease that affects the brain and nervous system with symptoms including hydrophobia and aerophobia; transmitted by animal bite

relapsing fever disease caused by various species of *Borrelia*, most commonly by *B. recurrentis*; transmitted by lice

reservoir of infection site where microorganisms can persist and maintain their ability to infect

resident microflora species of microorganisms that are always present on or in an organism

resistance the ability of a microorganism to remain unharmed by an antimicrobial agent

reverse transcriptase an enzyme found in *Retroviruses* that copies RNA into DNA

rheumatic fever a multisystem disorder following infection by β -hemolytic *Streptococcus pyogenes* that can cause heart damage

rubella viral disease characterized by a skin rash; can cause severe congenital damage (also called **german measles**)

salmonellosis a common enteritis characterized by abdominal pain, fever, and diarrhea with blood and mucus; caused by *Salmonella* species

sanitiser an agent that reduces the microbial flora in materials or on such articles as eating utensils to levels judged safe by public health authorities

secondary infection infection that follows a primary infection, especially in patients weakened by the primary infection

selective toxicity the ability of an antimicrobial agent to harm microbes without causing significant damage to the host

septicemia a systemic disease caused by the invasion and multiplication of pathogenic microorganisms in the blood stream

sexually transmitted disease an infectious disease spread by sexual activities

shigellosis gastrointestinal disease caused by several strains of *Shigella* that invade intestinal lining cells (also called **bacillary dysentery**)

shingles sporadic disease caused by reactivation of *Varicella-zoster herpesvirus* that appears most frequently in older and immunocompromised individuals

smallpox a formerly worldwide and serious viral disease that has now been eradicated

species a single kind of microorganism; a subdivision of a genus

spirochete a spiral form of bacterium; most are parasitic

spore a resistant body formed by certain microorganisms; a resistant resting cell; a primitive unicellular dormant body

sporicide an agent that kills spores

sterile free of living organisms

sterilization the process of making sterile by killing all forms of life

strain a pure culture of microorganisms composed of the descendants of a single isolation

streak plate method method used to prepare pure cultures in which bacteria are lightly spread over the surface of agar plates, resulting in isolated colonies

streptomycetes Gram-positive, filamentous, sporing, soil-dwelling bacteria, produces many antibiotics

subacute disease a disease that is intermediate between an acute and a chronic disease

superinfection a secondary infection resulting from the removal of normal microbiota, allowing colonization by pathogenic, and often antibiotic-resistant, microorganisms

suppressor T cell (Ts) a type of cytotoxic or helper T cell that inhibits immune system

susceptibility the state of being open to disease; specifically, capability of being infected; lack of immunity

symbiosis the living together of two or more organisms; microbial association

syndrome a group of signs and symptoms that occur together

synergism an inhibitory effect produced by two antibiotics working together that is greater than either can achieve alone

syphilis a sexually transmitted disease, caused by *Treponema pallidum*, characterized by a chancre at the site of entry and often eventual neurological damage

systemic infection an infection that affects the entire body (also called **generalized infection**)

taxonomy the science of classification of organisms, usually based on natural relationship

teratogen an agent that induces defects during embryonic development

tetanus disease caused by *Clostridium tetani* in which muscle stiffness progresses to eventual paralysis and death (also called **lockjaw**)

thermophile a heat-loving microorganism that grows best at temperature from 50°C

thermolabile destroyed by heat at temperature below 100°C

thermostable resistant to temperature of 100°C

thrush milky patches of inflammation on oral mucous membranes; a symptom of candidiasis, caused by *Candida albicans*

tissue culture a growth of tissue cells **in vitro** in a laboratory medium

toxic shock syndrome (TSS) condition caused by infection with certain toxigenic strains of *Staphylococcus aureus*; often associated with the use of superabsorbent but abrasive tampons

toxin a poisonous substance elaborated by an organism, such as a bacterial toxin

toxoid an exotoxin inactivated by chemical treatment but which retains its antigenicity and therefore can be used to immunize against the toxin

trachoma eye disease caused by *Chlamydia trachomatis* that can result in blindness

transient microflora microorganisms that may be present in or on an organism under certain conditions and for certain lengths of time at site where resident microbiota are found

transplantation the moving of tissue from one site to another

traveler's diarrhea gastrointestinal disorder generally caused by pathogenic strains of *Escherichia coli*

tuberculin skin test an immunological test for tuberculosis in which a purified protein derivative (PPD) from the *Mycobacterium tuberculosis* is injected subcutaneously, resulting in an induration if there was previous exposure to the bacterium

tuberculosis disease caused by *Mycobacterium tuberculosis*

urethritis inflammation of the urethra

urinalysis the laboratory analysis of urine specimen

urinary tract infection (UTI) a bacterial urogenital infection that causes urethritis or cystitis

vaccination inoculation with a biologic preparation (a vaccine) to produce immunity

vaccine a substance that contains an antigen to which the immune system responds

vaginitis vaginal infection, often caused by opportunistic microorganisms that multiply when the normal vaginal flora are disturbed by antibiotics or other factors

vibriosis an enteritis caused by *Vibrio parahaemolyticus*, acquired from eating contaminated fish and shellfish that have not been thoroughly cooked

viral enteritis gastrointestinal disease caused by *Rotaviruses*, characterized by diarrhea

viremia the presence of virus in the blood

viricide an agent that kills viruses

virion morphologically complete (mature) infectious virus particle

virulence the capacity of a microorganism to produce disease; pathogenicity

virus an obligate intracellular submicroscopic parasite composed of a nucleic acid (DNA or RNA) core inside a protein coat

vitamin a substance required for growth that the organism cannot make

wart a growth on the skin and mucous membranes caused by infection with *Human papilloma viruses (HPV)* (also called **papilloma**)

Western blotting a technique used to transfer and identify proteins

whooping cough a highly contagious respiratory disease caused primarily by *Bordetella pertussis* (also called **pertussis**)

yeast a kind of fungus that is unicellular and not characterized by typical mycelia

yersiniosis severe enteritis caused by *Yersinia enterocolitica*

zoonosis a disease that can be transmitted from animals

zygomycosis disease in which certain fungi of the genera *Mucor* and *Rhizopus* invade lungs, the central nervous system, and tissues of the eye orbit

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