



**Department of: Biology**

**College of: Education**

**University of: Salahadin**

**Subject: Practical Microbiology**

**Course Book – 3<sup>rd</sup> Year**

**Lecturer's name: Pishtiwan A. Hamad**

**Academic Year: 2022/2023**

# Course Book

<b>1. Course name</b>	<b>Practical Microbiology</b>
<b>2. Lecturer in charge</b>	<b>Pishtiwan Ahmad Hamad</b>
<b>3. Department/ College</b>	<b>Biology/ Education</b>
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<b>5. Time (in hours) per week</b>	<b>Practical: 8 Hours</b>
<b>6. Office hours</b>	<b>Approximately 20 Hours per week</b>
<b>7. Course code</b>	<b>EdB0302</b>
<b>8. Teacher's academic profile</b>	Education is the process of acquiring knowledge, and aims based on objective scientific thinking and confirm rational thinking based on reason and logic and cash Spirit. Teaching is a message written and oral knowledge-based and goal of success in an atmosphere of close communication between education and convincing officials. There is no doubt that the teacher as a main factor of the teaching process, has a very good and important role in performance the teaching program and preparing the students. The teacher is an affected factor among the teaching factors, and has effect on the student's characters and their future, therefor; the teacher must beware in his treatment with the students and the teaching staff, and must involve students in discussions in order to encourage them to express their point of view.as the modern learning process requires educational exchange between the key parties to this process.
<b>9. Keywords</b>	
<b>10. Course overview:</b>	<p>Microbiology involves the study of microscopic organisms - viruses, bacteria, fungi and protozoa. Microorganisms are everywhere - in our bodies, our food, the air, soil, and water. Because they're everywhere, they're involved in almost every aspect of our lives. They are used in producing foods such as cheese, wine, and beer, as well as many pharmaceutical, chemical, and agricultural products. They are important for soil fertility and the decomposition of materials, but can cause major diseases in humans, animals, and plants.</p> <p>This course will introduce the basic principles of Microbiology (especially bacteriology). The course related to basic principles of bacteria, detailed consideration of bacterial structures, and an integrative approach to understand how you can recognize bacteria to prevent from create and spreading of human diseases.</p> <p>The first weeks of the course will address general and special rules, which make the insurance of the health of all students that participate in the work of all labs. We will next focus on tests and address topics in sterilization, instruments which used for this purpose,</p>

bacteria, shapes of it, colonies, media which used for culturing and store the bacterial isolates, bacterial counts to detect the normal flora in the source which take from it. The laboratory will cover the basic techniques and procedures commonly used in clinical bacteria. Student presentations during the last few weeks of the course will address special topics in Microbiology or all procedures, which related and perform in the bacterial labs.

**11. Course objective:**

Upon the completion of the course, students will have

- Become proficient at laboratory skills and safety procedures.
- Learn to follow experimental procedures.
- Develop skills to formulate answerable questions/hypotheses, predict expected results.
- Learn how to make careful observations, collect and analyze data, and draw appropriate conclusions.
- Utilize active learning opportunity in the laboratories.
- Demonstrate good lab citizenry and the ability to work with others.
- Practical skills for the detection and isolation of bacteria from mixed normal flora from various sources and environments. the ability to identify bacteria based on biochemical testing and growth characteristics
- knowledge of factors most affecting microbial growth

**12. Student's obligation**

The role of students and their obligations throughout the academic year include:

- A. Quizzes and daily activities
- B. 1<sup>st</sup> Practical Examination
- C. 2<sup>nd</sup> Practical Examination
- D. 3<sup>rd</sup> Practical Examination
- E. Lab Activities
- F. Practice and Lab. reports
- G. Final Practical Exam

**13. Forms of teaching**

Teaching method used in our lab:

- A. Data show and power point
- B. White board
- C. Paper of lectures

**14. Assessment scheme**

The over all marks are of two part daily quizzes and monthly exams The daily tests (quizzes) will be given 10 marks and finally calculated on 3% in addition to the monthly tests (2-3 tests), all these marks calculated as the yearly attempt mark 13% this is the yearly quest degree and the final exam will be done on 20%.

**15. Student learning outcome:**

A microbiology laboratory, or lab, is the primary place that a working microbiologist not in the field can be found. It is in the lab that most of the testing, culturing, and research that they do occurs. This location contains the supplies and equipment needed for these activities, as well as provide an extremely clean and sterile place to work.

In order to understand what occurs in a microbiology lab, what a microbiologist does

should be understood. A microbiologist studies very small life forms, including bacteria, viruses, and fungi. These life forms live everywhere — in the soil, in the air, in the water, and even inside animals. Many of these life forms are so small that they cannot be seen by the unaided eye, and are called microorganisms. Often, a microbiologist will have to separate and grow the microorganisms in order to better see, study, and experiment on them. All of these activities occur in the lab.

Most look like any other biology or chemistry lab. They will probably have long lab benches where scientists can easily set up their equipment and work. Large, partially clear fume hoods will be present in order to keep the scientists safe from any dangerous experiments, though the fume hoods in a microbiology lab may contain special ultraviolet (UV) lights that hinder and kill some microorganisms.

1. Review of historical development of microbiology
2. Recognize types of microorganisms that cause infectious diseases.
3. Interpret diagnostic methods and laboratory findings to make the ultimate diagnosis.
4. Understanding principles and methods of sterilization relative to health care.
5. Using microscope perfectly and demonstrate slide preparation processes.
6. The principles of chemotherapy through the use of appropriate antimicrobial agents and lab techniques.
7. Collecting clinical specimens and disposal of contaminated materials.
8. Introducing to the principles of body defence against infections.

**16. Course Reading List and References:**

- A. Brock Biology of Microorganisms by Michael T. Madigan, John M. Martinko, David A. Stahl and David P. Clark. 2012, 13th Ed.
- B. Color Atlas of Diagnostic Microbiology by Luis M. de la Maza, Marrie T. Pezzlo and Ellen Jo Baron. 2007. 3rd Ed.
- C. Laboratory Manual and Workbook in Microbiology by Josephine A. Morello, Paul A. Granato and Helen Eckel Mizer. 2003. 7th Ed.
- D. Laboratory Exercise in Microbiology by Harley and Prescott. 2002. 1st Ed.
- E. Diagnostic Microbiology by Betty A. Forbes, Daniel F. Sahm and Alice S. Weissfeld. 2007. 12th Ed.
- F. Antimicrobial Susceptibility Testing Protocols by Richard Schwalbe, Lynn Steele-Moore and Avery C. Goodwin. 2007. 1st Ed.
- G. Basic Laboratory Procedure in Clinical Bacteriology by J. Vandepitte, J. Verhaegen, K. Engbaek, P. Rohner, P. Piot and C. C. Heuck. 2003. 2nd Ed.
- H. Clinical Microbiology Procedures Handbook by Lynne S. Garcia and Henry D. Isenberg. 2007. 2nd Ed.
- I. Microbiology Application; Laboratory Manual in General Microbiology by Benson. 2009. 8th Ed.

**17. The Topics:**

**Microbiology Lab Practices and Safety Rules**

**The instruments and tools used in microbiology lab**

**Aims of Culture Media**

To know the safety rules of Lab. and also to know how using the instruments and tools in microbiology lab.

**Lecture Date**

Lecturer's name  
**Pishtiwan A. Hamad**  
Time: (2 hrs.)  
**Date: 19/9/2022**

<p><b>Sterilization</b>                  Definition of sterilization, Types of sterilization                  Methods and instruments that used in Sterilization  <b>Aims of Sterilization</b>                  Knowing the types and methods used for killing of bacteria and which one is the best for this purpose.</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>                  Time: (2 hrs.)                  Date: 26/9/2022</p>
<p><b>Bacterial Culture Media</b>                  Definition, sources and classification of Cultural Media                  Preparation of culture media.  <b>Aims of Culture Media</b>                  To know the ingredient and composition of culture media, using for what purpose and classification of culture media and also using the culture media for classification of bacteria.</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>                  Time: (2 hrs.)                  Date: 3/10/2022</p>
<p><b>Preparation of bacterial culture media</b></p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>                  Time: (2 hrs.)                  Date: 10/10/2022</p>
<p><b>Microbial Cultivation and Establishing Pure Culture</b>  <b>Aims of this lab</b>                  For establishing pure culture of bacteria and Preparation of smears to staining of bacteria.</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>                  Time: (2 hrs.)                  Date: 17/10/2022</p>
<p><b>Bacteria</b>                  Definition, shapes, and colonies of Bacteria  <b>Aims of this lab</b>                  Using the shape (morphological properties) and colonies (cultural characteristics) for classification of bacteria.</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>                  Time: (2 hrs.)                  Date: 24/10/2022</p>
<p><b>Bacterial Staining Methods (Staining Methodology)</b>                  Definition of Stain                  The Aims of Staining                  Types of Stains in Microbiology  <b>Simple Stains</b>                  a. Basic, Direct or Positive Stains                  b. Acidic, In – Direct or Negative Stains  <b>Aims of this lab</b>                  To see the shape, and arrangements of bacterial cells under microscope. In the other hand, compare between positive and negative stain to staining the bacterial cells and how can different between them.</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>                  Time: (2 hrs.)                  Date: 31/10/2022</p>
<p><b>Gram Stain</b>                  Components of Gram Stains                  Different between Bacteria according to Gram Stain  <b>Aims of Gram Staining</b>                  Discrimination and differentiation of bacteria in to large groups ( Gram positive and negative bacteria) in response to take the color of each stain which use in the method.</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>                  Time: (2 hrs.)                  Date: 7/11/2022</p>
<p><b>Capsule Stain</b>                  Definition, types, functions and methods of capsule staining.</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b></p>

<p><b>Aims of Capsule Staining</b> Our principal concern is simply to demonstrate the presence or absence of a pronounced capsule. Staining the bacterial capsule cannot be accomplished by ordinary simple staining procedures. This can be easily achieved by combining negative and simple staining techniques. To learn about this technique prepare a capsule “stained” slide of <i>K. pneumoniae</i></p>	<p>Time: <b>(2 hrs.)</b> Date: <b>14/11/2022</b></p>
<p><b>Endospore Stain</b> Definition, types, functions and methods of bacterial endospore staining <b>Aims of Endospore Staining</b> To differentiate between organisms that can produce endospores and those that cannot.</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date: <b>21/11/2022</b></p>
<p><b>Movement of Bacteria (Flagella Stain)</b> What is mean by Bacterial Movement? Types of Bacterial Movement, Structure and Classification of Flagella. Methods for Detection of Bacterial Movement <b>Aims of Bacterial Flagella staining</b> To determine bacterial motility</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date: <b>28/11/2022</b></p>
<p><b>Bacterial Counting methods</b> What is (are) the Purpose (s) of the Bacterial Counting <b>Types of Bacterial Counting</b> Direct Method (Slide Method) <b>Aims of Bacterial counting methods</b> 1. To learn the different techniques used to count the number of microorganisms in a sample. 2. To be able to differentiate between different enumeration techniques and learn when each should be used. 3. To have more practice in serial dilutions and calculations.</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date: <b>5/12/2022</b></p>
<p><b>1. In-Direct Methods</b> <b>A. Standard Plate Count</b></p>	
<p><b>2. In-Direct Methods</b> <b>B. Optical Density method (Spectrophotometer method)</b></p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date: <b>12/12/2022</b></p>
<p><b>3. In-Direct Methods</b> <b>C. Most Probable Number (MPN)</b></p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date: <b>19/12/2022</b></p>
<p><b>Antibiotics</b> Differences between Drugs and Antibiotics Sources of Antibiotics Mechanical Action of Antibiotics Mechanisms of Bacterial Resistances against Antibiotics <b>Aims of antibiotics study</b> 1. To utilize specific monitoring techniques to evaluate the susceptibility of a microbe to different antibiotics.</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>

<p>2. To distinguish the range of activity of an antibiotic. 3. To recognize and define advantages and limitations of two different susceptibility testing procedures. <b>Methods of Antibiotic Susceptibilities</b> 1. Disc Diffusion Methods (Kirby – Bauer Method)</p>	
<p><b>Methods of Antibiotic Susceptibilities</b> 2. Dilution Method (MIC and MBC) Minimum Inhibitory Concentration and Minimum Bactericidal Concentration</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>
<p><b>Methods of Antibiotic Susceptibilities</b> 3. Agar Dilution Method</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>
<p><b>Environmental Factors Directly Affecting on Microbial Growth</b> Temperature, Salinity and Osmosis, and Oxygen Requirement Effects of Antiseptics and Disinfectants on Bacterial Growth <b>1. Effect of temperature on the growth of bacteria</b> <b>Aims of this lab</b> To detect the effect of some environmental factors on bacterial growth to choose the best one for inhibit and stop the bacterial growth around the our environment and prevent the spread the diseases caused by bacteria.</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>
<p><b>Environmental Factors Directly affecting on Microbial Growth</b> 2. Effects of osmosis and water activity on bacterial growth</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>
<p><b>Environmental Factors Directly affecting on Microbial Growth</b> 3. Effects of Oxygen on Bacterial Growth</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>
<p><b>Environmental Factors Directly affecting on Microbial Growth</b> 4. Effects of Antiseptics and Disinfectant on Bacteria Growth</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>
<p><b>Environmental Factors Directly affecting on Microbial Growth</b> 5. The effect of pH on the growth of bacteria</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>
<p><b>API System</b> Types of API Analytical Profile Index 20 Enterobacteriaceae How We do the API 20E?  <b>Aims of API System</b> Learn how to perform and interpret the miniaturized, multi-</p>	<p>Lecturer's name <b>Pishtiwan A. Hamad</b> Time: <b>(2 hrs.)</b> Date:</p>

test technique for bacterial identification.	
<p><b>Bacteriological Test for Water</b>  <b>Detect the bacterial number in the tested water</b></p> <ol style="list-style-type: none"> <li>1. Presumptive test</li> <li>2. Confirmed test</li> <li>3. Complement test</li> </ol> <p><b>Aims of Bacteriological Test for Water</b>  Evaluate the purity of water and determine the number and types of bacteria (if present) in the drinking water.</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>  Time: <b>(2 hrs.)</b>  Date:</p>
<p><b>Bacteriological Test for Water</b>  Detect the bacterial type (fecal bacteria) in the tested water</p>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>  Time: <b>(2 hrs.)</b>  Date:</p>
<p><b>Identification of Unknown Bacteria</b>  <b>Principle of identification and differentiation of Bacteria</b></p> <ol style="list-style-type: none"> <li>A. Direct Examination and Techniques</li> <li>B. Cultural Identification and</li> <li>C. Biochemical Identification</li> </ol>	<p>Lecturer's name  <b>Pishtiwan A. Hamad</b>  Time: <b>(2 hrs.)</b>  Date:</p>
<p><b>18. Examinations:</b></p> <p><b>1. Compositional: Explain the following briefly</b></p> <ol style="list-style-type: none"> <li>1. Insertion of Durham tube in MPN test. <ul style="list-style-type: none"> <li>• Used for the detection of gas production</li> </ul> </li> </ol> <p><b>2. True or false type of exams: Put (T) in front of true sentences and (F) in front of false sentences</b></p> <ol style="list-style-type: none"> <li>1. The first step in the identification procedure is to accumulate information that relates to the organisms' morphological, cultural, and physiological characteristics. True</li> </ol> <p><b>3. Multiple choices: From the options, chose the correct one to complete the meaning of sentences</b></p> <ol style="list-style-type: none"> <li>1. Remove of microorganisms can be performed by.....:</li> </ol> <p>A. Pasteurization. B. Thermal sterilization. C. Cooling. D. <u>Centrifuge.</u></p> <p><b>4. Count only</b></p> <ol style="list-style-type: none"> <li>1. Classification of culture media Based on consistency. <ol style="list-style-type: none"> <li>a. Liquid media</li> <li>b. Solid media</li> <li>c. Semi-solid agar</li> </ol> </li> </ol> <p><b>5. Find the bacterial cells number in a 1 ml of distilled water, when you add 0.1 ml from the tubes into plates and the number of colonies in tubes number 1, 2, 3 and 4 from serial dilution were 350, 80, 28 and 13 respectively.</b></p> <p><b>Bacterial cell/0.1ml = 80 CFU x10<sup>2</sup>= 8x10<sup>3</sup> CFU/0.1ml</b></p> <p><b>Bacterial cell/ml = 8x10<sup>3</sup> /0.1 = 8x10<sup>4</sup> CFU/ml</b></p>	
<p><b>19. Peer review</b></p>	