

Al-Mustansiriyah University

College of Medicine

Department of Microbiology

**Laboratory Manual
for Bacteriology**

Dr. Riyadh A. Al-Ani

Dr. Orouba K. Abbas

Dr. Shama M.J.Saadaldin

Title of practicals for the first term:

- 1- General safety and precautions**
- 2- Culture media and laboratory equipments**
- 3- Handling of bacterial cultures**
- 4- Hanging drop technique**
- 5- A) Bacteriological stains (simple & differential) B) Cellular morphology**
- 6- Special staining technique**
- 7- Sterilization and disinfection**
- 8- Staphylococcus species**
- 9- Streptococcus species**
- 10- Neisseria**
- 11- Corynebacterium**
- 12- Mycobacterium**

Introduction

This laboratory manual maintains emphasis on the basic principles of diagnostic microbiology for students who will be directly involved in patients care and who wish to learn how microbiological principles should be applied in the practice of their profession.

For students of medicine the clinical and epidemiological applications of microbiology often seem more relevant than its technical details.

For this reason a laboratory manual should direct microbiology toward the clinical setting, and to relate its principles to patients care.

The principles learned in each exercise are then applied practically in diagnostic microbiology.

The contents of this manual are planned to be used in conjunction with the text book.

Practical No. (1)

Orientation to the Laboratory:

General Safety and Precautions

Many of the microorganisms used in this course may be pathogenic for humans and animals. As a result, certain rules are necessary to avoid the possibility of infecting yourself or other people. Anyone who chooses to disregard these rules or exhibits carelessness that endangers others may be subject to immediate dismissal from the laboratory. If doubt arises as to the procedure involved in handling infectious material, consult your instructor. Each student is responsible for the observance of the following rules:

- 1- Place all extra clothing, unnecessary books, purses, backpacks, in an appropriate place. Racks are provided for these materials.
The laboratory working area must be kept free of articles not actually in use.
- 2- Eating, drinking, and smoking are forbidden at all times in the laboratory.
- 3- Keep your locker or laboratory drawer clean. Do not allow your locker or drawer become filled with cultures that have no value in your current work.
- 4- Return all reagents, cultures, and glassware to their proper places.
- 5- Wear a laboratory coat when working in the laboratory. This will protect clothing from contamination or accidental discoloration by staining solutions.
- 6- Do not place anything in your mouth while in the laboratory. This includes pencils, food, and fingers. Learn to keep your hands away from your mouth and eyes.
- 7- Avoid contamination of the benches, floor, and wastebaskets.

- 8- Clean your work area (Laboratory bench) with a phenolic disinfectant such as 50% phenol (or any other

disinfectant supplied) before and after each laboratory period. This standard procedure lessens the chance for accidental infection as well as contamination of cultures.

- 9- Special receptacles will be provided for infectious materials and used glass slides. Place all discarded cultures and contaminated glassware into these receptacles. Do not let unwanted and unneeded materials accumulate. Tall jars filled with a solution such as 5% Lysol or special receptacles will be provided for pipettes.
- 10- When an infectious material is spilled, cover it immediately with a disinfectant such as 5% Lysol or 5% Phenol and notify your instructor at once.
- 11- Flame wire loops and needles before and immediately after transfer of cultures. Do not move through the laboratory with a loop or pipette containing infectious material.
- 12- Wash your hands thoroughly before and after each experiment, using disinfectant soap if possible.
- 13- Label all experimental material with your; a. Name b. Date c. Exercise number d. Lab section e. Specimen/Organism
- 14- Long hair should be tied back to minimize fire hazard and contamination of experiments and cultures.
- 15- Some of the chemicals employed in the various exercises can be hazardous if not handled properly. We have selected experiments to minimize the use of such chemicals; however; where they are necessary, be certain to observe the precautions noted in the exercise and by your instructor.
- 16- Do not stack Petri plates more than three high on incubator shelves. Tall stacks are a potential hazard if they topple when the incubator is opened. If available, special Petri plate holders (cans) may be used to hold large stacks of plates.
- 17- To avoid burns, beware of Bunsen burners that will be used in almost every exercise. Immediately report to your instructor about any cut or injury occurs.
- 18- The Workplace Hazardous Material Information System (WHMIS) requires that all hazardous substances, including microorganisms, be labeled in a specific manner. In addition, there must be a Material Safety Data Sheet (MSDS) available to accompany each hazardous substance. MSDS are now supplied with every chemical sold by supply houses. The person in charge of the microbiology laboratory should ensure that adherence to this law is enforced.
- 19- Finally, write notes about all the specific safety feature that are located in the laboratory. These would include the fire extinguishers, the safety shower, the fire blanket, the eye wash station, the first aid kit, and emergency exit.

If the instructor does not supply this information, please ask him or her to do so.

All laboratory work can be done more effectively and efficiently if the subject matter is understood before coming to the laboratory. To accomplish this, read the experiment several times before the laboratory begins. Know how each exercise is to be done and what principles it is intended to convey.

Also read the appropriate sections in your handout manual concerning the experiment being performed.

This will save you much time and effort during the actual laboratory period.

All laboratory experiments will begin with a brief discussion by your instructor of what is to be done, the location of materials, and other important information. Feel free to ask questions if you do not understand the instructor or the principles involved.

Much of the work in the laboratory is designed to be carried out in groups or with a partner. This is to aid in coverage of subject matter, to save on time and expense, and to encourage discussion of data and results.