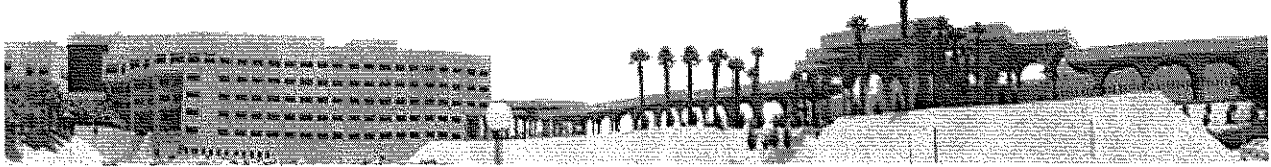




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Basic Microbiology	
Course No.	Mic.1202
Credit hour	٣ (٢ Theoretical + 3 Practical)
Prerequisite	General Biology

Aims & Objectives:

This Course is designed to build on work carried out in general biology code No.1151 and to introduce the student to important topics which are preparatory to other courses in the Medical laboratory Technology Clinical Curriculum. The Course involves a study of major topics in microbiology, and immunology. But covering the basic elements only in each case. The Course incorporates a practical Study Unit, which introduces students to some of the fundamental principles and techniques, met in lectures. It is an essential feature of this Course, that it provides the basis, not only, for further studies in Mic. 1203 and Imm 1202, but also, complements other clinical and professional courses in the MLT Clinical Curriculum.

اسم المقرر : أساسيات علم الأحياء الدقيقة رقمه : ١٢٠٢ رمزه : حيد
عدد الساعات المعتمدة : ٣ (٢ نظري + ٣ عملي) المستوى الثانى

يحتوي هذا المقرر على أساسيات علم الأحياء الدقيقة من حيث تصنيف مجموعات الميكروبات المختلفة مع تقديم وصفاً لكل مجموعة من حيث الشكل والوظيفة ، كما يشمل المقرر الطرق المختلفة لزراعة الميكروبات في المختبر والعوامل المختلفة التي تؤثر على النمو، طرق الفصل والتنقية والصباغة والتعرف عليها بالإضافة إلى الطرق المختلفة للتعقيم والتطهير وأساسيات كل منها.

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Laboratory Rules

1

- 1. Always wear a laboratory coat when working in the laboratories (to prevent your clothes from the contaminated material and from the stains)**
- 2. Should cover any injury or cuts by bandage to protect yourself from any micro-organisms**
- 3. Clean your working area with disinfectant before and after laboratory experiment**
- 4. Don't eat , smoke , and drink in the laboratories**
- 5. Wear gloves through working in the lab**
- 6. Never mouth-pipette any culture of micro-organisms**
- 7. Sterile your laboratory material before and after the work to prevent contamination by suitable sterilization method**
- 8. Always wash and dry your hands thoroughly before leaving the laboratory**

Some instruments and materials may used in this course and their functions :

- 1- Microscope : used to see micro-organisms
- 2- Incubator : used to incubate the organisms , its temperature is 37C to culture organisms
- 3- Water bath : a pool of heat water used to increase the temperature of some organisms and the liquid agar
- 4- Hot plate or slide warmed L used to fix the organisms on the slide , and also to kill the organisms
- 5- Autoclave : used to sterile the plates m media m broth , and other solution used in the laboratory
- 6- Hot air oven : used for drying and sterilizing the plates
- 7- Petri-dishes : used to culture the organisms after addition of the media
- 8- Wire loop L made of nichrome wire , used for inoculating bacteria into agar media
- 9- Straight wire : made of nichrome wire , used to pick up bacterial colonies for inoculation into broth media
- 10- Bunsen burner or flame : used for sterilization of loops , mouth of tubes , and air near the flame
- 11- Forsted end slide : used to examine the organism under the microscope after staining the slide
- 12- Cover slide : a very thin glass slide used solution (should be sterile by autoclave and dispose in the disinfectant jar)
- 14- Serological pipette : it's a measured pipette used to take an exact amount of the solution
- 15- Sterile physiological saline : its .08% of NaCl = .085gm of NaCl+100ml of water
* it sterile by autoclave and used to emulsify the bacteria on the slide
- 16- Oil immersion : use with the X100 objective of the microscope

Disposal unwanted cultures and other contaminated materials :

- 1\ Plastic Petri-dishes : should be disposed in an autoclave-bag which then autoclaved**
- 2\ Contaminated glass wares : should be put in the basket , autoclaved then washed for re-use**
- 3\ Contaminated slide and pipettes : should be disposed on disinfectant jar , then autoclaved and disposed it after that .**

Typed of Microscopes

2

* Micro-organisms can not be seen by the naked eyes , so we use microscope to identify them

Functions of microscopes :

1\ to magnify the object being examined

2\ to resolve the image seen

Type of microscopes :

1\ Bright-field microscopes

2\ phase contract microscopes

3\ Dark-field microscopes

4\ Fluorescent microscopes

5\ Inverted microscopes

6\ Dissecting or stereo microscopes

7\ Electron microscopes

Brightfieled Microscopes

Components of bright-field Microscopes :

1.Stage

2.Objective lens

3.Ocular of eye- pieces

4.Nose-pices

5.Condenser

6.Light source

7.Iris diaphragm

8.Adjustment of focusing knobs

Function of the Bright-field components :

1.Stage: the slide is placed on it

2.Objective lens : above the stage produces magnified image of the object into the eye-piece

There are many objective lenses , with different magnifications (X4 , X10 , X40 , X100)

3.Ocular of eye-piece : also provides (fives) a further magnification of the image

4.Condenser : focuses the light from the light source onto the object

5.Iris diaphragm L controls the ring of the light that goes into the condenser

6.Adjustment of focusing knobs are used to get the image into focus by moving the stage to adjust the distance between the object and the objectives lens

* it's used to examine stained smears

Resolution :

.02mm to be able to see a short distance between two points

Magnification : ocular lens X objective lens

Example : Ocular lens = 4

Objective lens = 10

Magnification = $4 \times 10 = X40$

Darkfield Microscope

1.is is used to examine the motility of bacteria and investigate the presence of spirochaetes in clinical specimens

2.reflection of the light into the objective , and it seen as bright shapes

Phase contrast Microscope

1.it has a special condenser and phase-plate

2.the phase-plate retrads the waves of light that go through the specimen

3.the retarded and non-retarded waves produce a contrast between the cells and the background

4.it is suitable for the examination of wet-preparation

Fluorescent microscope

1. we use this microscope when we dealing with organisms that stained by fluorescent stain (fluorochromes)
2. the source of light is ultra-violet light
3. cells appear bright objects against dark background
4. important in the immunology and virology laboratories for the detection of Antigen-Antibody reactions

Inverted Microscope

1. the part of this microscope is similar to the light microscope but the condenser is located above the stage while the objectives are below the stage . The purpose of this set up to allow a bigger working distance
2. it is used in virology for the examination of cell culture flasks

Dissecting of Stereo Microscope

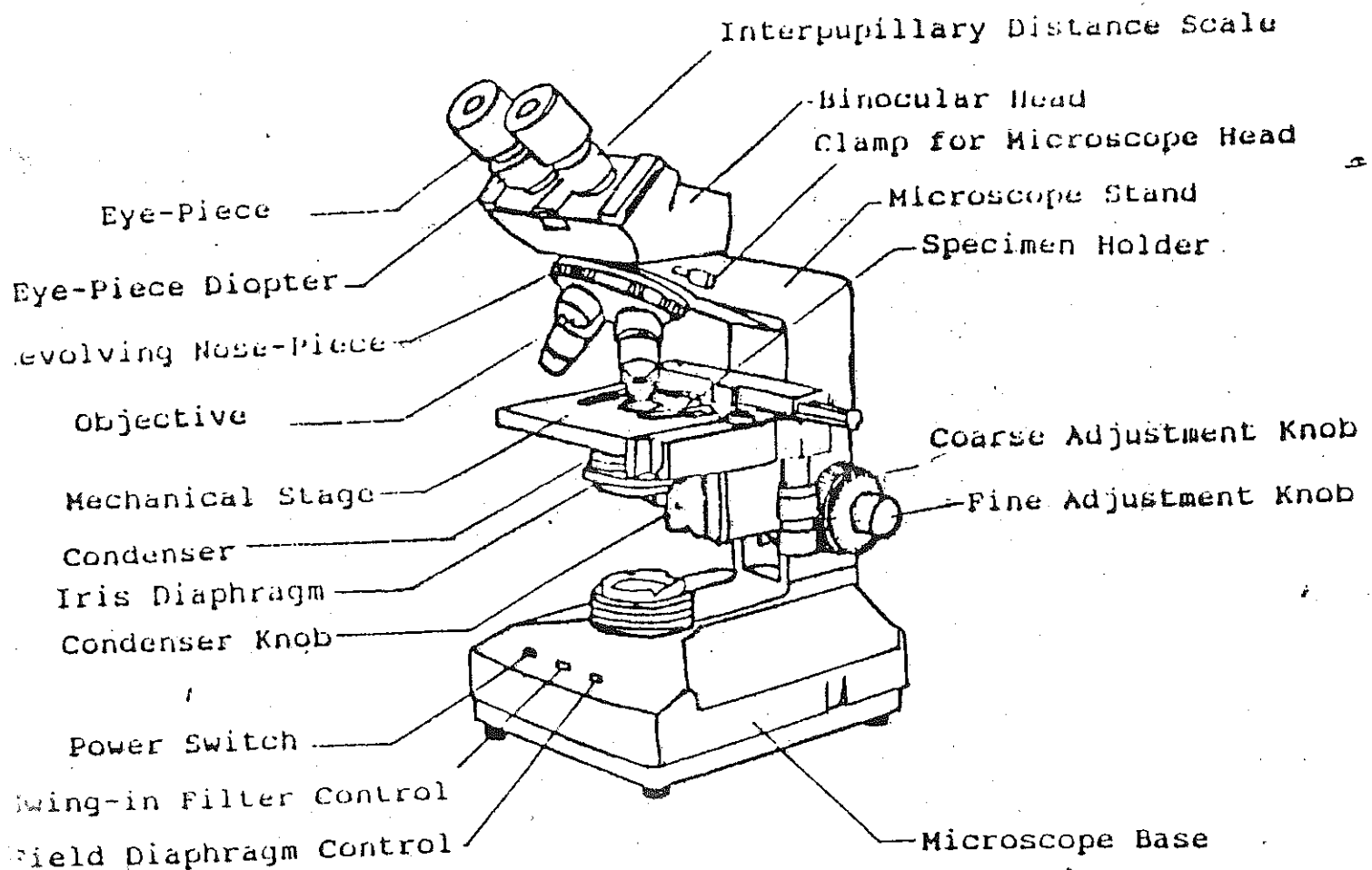
1. gives three dimensional image
2. use for examination of insects or colonies of bacteria and fungi

The microscope has two main functions :

1. To magnify the object being examined.
2. To resolve the image seen.

The object is placed on the stage under the objective lens which produces a magnified image of the object into the ocular or eye-piece. There are many objective lenses, each with a different magnification, attached to the revolving nose-piece. The viewer looks at the image of the object through the ocular, which also provides a further magnification of the image. The condenser 'focuses' the light from the light source on to the object for proper illumination. Under the condenser is the Iris diaphragm which controls the ring of light that goes into the condenser. The adjustment or focussing knobs are used to get the image into focus by moving the stage to adjust the distance between the object and the objective lens.

The microscope should be properly 'set-up' before use to get the maximum resolution and clarity of the image. The light intensity, the position of the condenser, the closing or opening of the iris diaphragm should all be controlled to get a proper magnified and resolved image of the object. The resolution of the brightfield microscope is 0.2 μm . The brightfield microscope is mainly used to examine stained preparations.



STERILIZATION

1- Heat:-

a\ Dry Heat:

like (flame, Bunsen burner) used to sterilize wire loops and straight wire loops.

(oven) used to sterilize glass wares, glass pipettes, oils, and sands.

Its temperature is $160-180^{\circ}\text{C}$ for 1-2 hours.

b\ Moist Heat:

like (autoclaves) used to sterilize metals, water, saline, culture media, plastics, and contaminated materials.

Its temperature is 121°C for 15-20 min at atmosphere pressure 15

Autoclave bags: it made of a specific materials that can not dissolve at high temperature.

Indicators to check oven and autoclave:-

1\ Chemical Indicators:-

- autoclave tape
- brown's tube

2\ Biological Indicators:-

- spore forming bacteria
(*Bacillus steriothermophilus*)

2- Radiation:-

Used to sterile plastic things like Petri-dishes, serological pipettes, operation rooms. *غرف العمليات*

3- Filtration:- *الترشيح*

Used to sterile liquid solutions that components are sensitive to high temperature. *سائل*

Like (serum, antibiotics, sugars) according to the size of the pores of the membrane or filter, if the bacterial size more than the pore size they can not penetrate through the filter.

But if the bacterial size less than the pore size they will penetrate the filter. So the pore size for bacteria is (0.45mm) and for viruses (0.22mm).

4- Chemicals (disinfectants):-

They are sterilize *الطعام* surfaces and items. *الأسطح*

Using chemical agents to inhibit growth of the bacteria.

Types of Disinfectants:-

1\ *Bactericidal:* *مقتل*

It will kill bacteria, like Detol and Clorox.

2\ *Bacteriostatic:* *موقف*

It will inhibit bacteria, like Lux.

It used in the medication and health benches. *الطعام* *الأدوية*

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