	KULLIYYAH OF MEDICINE & HEALTH SCIENCES
Course/Course Code	Medical Parasitology/ PPB62303
Topic	Introduction to Parasitology (Practical)
Year/Semester/Session	2 (Cohort 5) / 3 / 2018 (3)
Date	30.09.19
Time	1400 – 1600
Student's Name/ ID	
Lecturer's Name	Dr. Lee Ii Li

At the end of the practical session, students should be able to:

TLO No	Topic Learning Outcome (TLO)	Course Learning Outcome (CLO)
1	recognize the various components of a microscope	5
2	demonstrate the use of the following parts in a microscope a) intensity switch b) adjustment of diopter and interpuppilary distance c) engaging of objective lens d) placement of slide and navigating of slides or stage movement e) usage of vernier scale, X-axis and Y-axis f) coarse and fine and focusing g) setting of condenser aperture h) usage of immersion oil	5
3	demonstrate the maintenance procedure (cleaning) for the following parts in a microscope a) eyepieces b) objective lens c) condenser top lens d) field diaphragm lens	5
4	prepare wet mount using saline and iodine	5

References	1. http://www.matrixoptics.com
	2. http://www.olympusmicro.com/primer/anatomy/anatomy.html
	3. whqlibdoc.who.int/publications/9241544104_(part1).pdf

Introduction:

Medical parasitology is the subject which deals with the parasites that infect humans, the diseases caused by them, clinical picture and the response generated by humans against them. It is also concerned with the various methods of their diagnosis, treatment and finally their prevention and control.

Laboratory diagnoses of parasites are an important stage to search and detect the parasites that infect humans. Since most of the diagnostic stages of parasites are microscopic, the proper use of microscope is very important.

Therefore, in the introduction to Parasitology practical class, the introduction to basic microscopy will be dealt with. **It will start with the introduction to the various components of a microscope, followed by the use and maintenance of a microscope.**

In addition to that, students will be exposed to the **preparation of wet mount using saline and iodine**. However, students must realize that there are other laboratory procedures available for stool examination and the preparation of a blood film.

Principle:

The principle of microscopy will be elaborated during practical session and the power point presentation slides will be e-mailed after the session.

Wet mounting is the simplest and easiest technique for the examination of faeces. Therefore, it can be performed in all laboratories at any level. A wet mount can be prepared directly from faecal material of from concentrated specimens. The basic types of wet mount that should be used for each faecal examination are saline, iodine and buffered methylene blue (Table 1 and Table 2).

Table 1: Various wet mount used for faecal examination.

	Wet mount	Description
1.	Saline	• Initial microscopic examination of stools. • Employed primarily to demonstrate worm eggs, larvae, protozoan trophozoites and cysts. • Can reveal the presence of red blood cells and white blood cells.
2.	Iodine	• Mainly to stain glycogen and the nuclei of cysts (if any).
3.	Buffered methylene blue	• Should be employed if amoebic trophozoites are present in a saline wet mount or whenever their presence is suspected. • It stains amoebic trophozoites, not amoebic cysts, flagellate trophozoites or flagellate cysts. • It is appropriate only for fresh unpreserved specimens (live organisms).

Table 2: Appropriate technique according to consistency of stools.

	Consistency	Protozoan stage most likely to be found*	Wet mount technique		
			Saline	Iodine	Buffered methylene blue (if amoebic trophozoites are seen)
1.	Formed	Cysts	+	+	
2.	Soft	Cysts (occasionally trophozoites)	+	+	+
3.	Loose	Trophozoites	+		+
4.	Watery	Trophozoites	+		+

* Eggs, larvae and worms may be found in stools of any consistency

Material/Reagents Preparation:

Materials:

1. Faeces
2. Glass slides
3. Cover glass
4. Applicator stick
5. Microscope
6. Gloves
7. Stool container
8. Lens paper

Reagents:

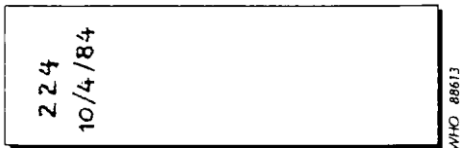
1. Normal saline
2. Lugol's iodine

Procedures:

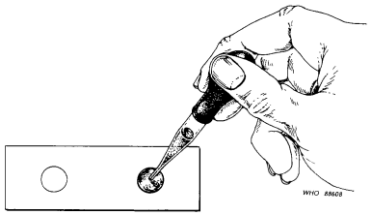
Preparation of wet mount - saline and iodine mounts

(Source: whqlibdoc.who.int/publications/9241544104_(part1).pdf)

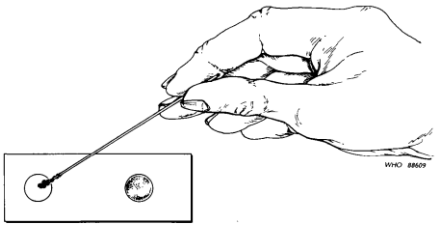
1. Label two patient's identifiers and the date on the left-hand end of the slide.



2. Place a drop of saline in the centre half of the slide and place another drop of iodine solution in the centre of the right half of the slide.
   Note: If the presence of trophozoites is suspected, warm saline (37°C) should be used.



3. Use an applicator stick (match or toothpick) to pick up a small portion of the specimen (size of a match head) and mix with the drop of saline.

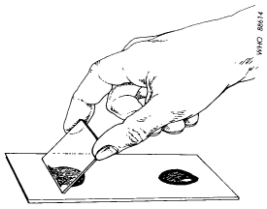


Note:

Stool	Procedure
(a) Formed	Take the portion of stool from an area to include inside and outside parts of the specimen.
(b) With mucus	Whenever mucus is present, label a second slide with the patient's information. Put a drop of saline on the slide, pick up a small portion of mucus and mix with the saline. Trophozoites, if present, are sometimes more readily found in mucus than in the solid part of the stool.
(c) Loose watery stool	If mucus is not present, pick up a small portion of the stool (any part) and mix with the saline.

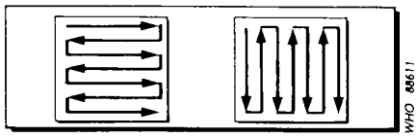
4. Repeat step 3 using iodine.

5. Cover the drop of saline and the drop of iodine with a cover glass, respectively by holding the cover glass at an angle. Touch the edge of the drop and lower gently on to the slide in order to reduce air bubbles in the mount.



6. For examination of glass slide, put the slide with the mounts on the microscope stage and focus on the mount using X10 or low-power objective with appropriate light intensity.

7. Examine the entire cover glass area with X10 objective; focus the objective on the top left-hand corner and move the slide systematically backwards and forwards, or up and down.



When organisms or suspicious materials are seen, switch to higher objective to observe the detailed morphology.

ACTIVITIES

TLO1:	**Recognize the various components of a microscope**

1. With an aid of a diagram, label the parts of a microscope. (20 marks)

TLO 2 – 4:	**Each student will be evaluated through hands-on demonstration.**

1. Demonstrate the use of the following parts in a microscope
   a) Intensity switch (5 marks)
   b) Adjustment of diopter and interpuppilary distance (5 marks)
   c) Engaging of objective lens (5 marks)
   d) Placement of slide and navigating of slides or stage movement (5 marks)
   e) Usage of vernier scale, x-axis and y-axis (5 marks)
   f) Coarse and fine and focusing (5 marks)
   g) Setting of condenser aperture (5 marks)
   h) Usage of immersion oil (5 marks)

2. Demonstrate the maintenance procedure (cleaning) for the following parts in a microscope.
   a) Eyepieces (5 marks)
   b) Objective lens (5 marks)
   c) Condenser top lens (5 marks)
   d) Field diaphragm lens (5 marks)

3. Prepare wet mount using saline. (10 marks)

4. Prepare wet mount and stain with iodine. (10 marks)