

USE, CARE AND MAINTENANCE OF MICROSCOPES

MALARIA MICROSCOPY STANDARD OPERATING PROCEDURE – MM-SOP-12

1. PURPOSE AND SCOPE

To describe the general protocol for the use, care and maintenance of microscopes in laboratories in which malaria microscopy is performed

This procedure is to be modified only with the approval of the national coordinator for quality assurance of malaria microscopy. All procedures specified herein are mandatory for all malaria microscopists working in national reference laboratories, in hospital laboratories or in basic health laboratories in health facilities performing malaria microscopy.

2. BACKGROUND

Compound binocular light microscopes are recommended for malaria microscopy, with 10x and 40x objectives, an oil immersion 100x objective and 10x oculars. The maximum depth of field can be achieved only if the microscope light source has sufficient luminosity to allow the iris diaphragm to be closed optimally without loss of brightness. Preferably, it should have a built-in lamp, an adjustable condenser (a fixed condenser may be acceptable) and a daylight blue filter.

The accuracy of malaria microscopy depends on the correct functioning and use of the microscope. Microscopes should be set up for optimal performance, protected from damage, used ergonomically, regularly maintained and, if required, repaired by qualified personnel. Care of microscopes is particularly important in facilities in areas that are dusty, where microscopes must be covered when not in use and before cleaning the floor to avoid damage to the microscope.

3. SUPPLIES, MATERIALS AND EQUIPMENT

- a compound microscope fitted with paired 10x oculars (eyepieces), 10x, 40x and 100x (oil immersion) objectives and a mechanical stage;
- a dust cover;
- lens paper;
- commercial microscope cleaning solution and
- a soft cloth.

4. PROCEDURE

4.1. Transporting and moving microscopes

- Transport microscopes in the original container with the internal packing materials to prevent it from moving within the container.
- If the original packaging is not available, use a specifically designed microscope transport box supplied by the manufacturer or a fabricated container containing foam or similar packing materials to prevent the microscope from moving within the container during transport.
- During transport, protect the microscope from excessive movement or vibration: pack it so that it does not move around in the vehicle and cannot fall from the top of other items being transported; protect the microscope from exposure to water (such as rain, flooding, falling into water), excessive heat, direct sunlight and rodents.
- Carry a microscope with two hands, one supporting the base and the other holding the arm.

4.2. Placement

- Place the microscope on a flat, level, firm bench, free from vibration. At high magnification, very small movements of the bench will cause large movements in the image being viewed by the microscopist.
- Place the microscope in a position where there is space for the microscopist to place his or her legs fully under the bench. Preferably use height-adjustable chairs.
- Do not place the microscope in front of a brightly lit window. Place it in front of a wall or a darkened window.

4.3. Microscope set-up

- Follow the manufacturer's directions for optimal set-up of the optical system and general use.
- Adjust the iris aperture to the setting recommended by the manufacturer to achieve maximum depth of field.
- If the manufacturer provides an iris diaphragm setting on the condenser, set this to the 100x objective lens setting.
- Use the following method if the eyepiece of the microscope can be removed:
 - raise the condenser;
 - set the lamp to "low";
 - select the 40x objective;
 - close the iris diaphragm;
 - remove one eyepiece;
 - look down the tube, then adjust the condenser until the edge of the diaphragm is in sharp focus without showing an outer fringe ring in green or in red;
 - open the iris diaphragm until the objective lens completely fills with light;
 - the shape of the light is usually octagonal; when the points of the octagon touch the outside of the objective lens, open the iris diaphragm until the light appears circular;
 - replace the eyepiece.
- An alternative method, and one that does not require removal of the eyepieces, is to set the microscope into Kohler illumination.
 - close the iris diaphragm on the condenser;
 - close the light source at the base of the microscope, where the light is reflected up to the condenser;
 - turn the light up as bright as possible;
 - put a stained blood smear on the stage, add immersion oil, and use the 100x oil immersion objective lens;
 - look through the microscope oculars, and focus the condenser up and down until the edge of the field is sharp;
 - use the adjustment screws on the condenser to move the circle of light to the centre of the field if necessary.
 - turn the light down, and open the iris diaphragm and the light source before use.
- Adjust the interpupillary distance.
- Adjust the dioptre.

4.4. Use of the microscope

- Be conversant with the parts of the microscope and their functioning for efficient use.
- Turn on the light source, and adjust the optimum light setting to ensure the correct level of brightness by turning or sliding the brightness adjustment knob at the base.
- Rotate the low power objective into position. Remove the eyepiece, look down the body tube and adjust the mirror and diaphragm setting so that light is reflected up the tube and a circle of evenly illuminated light is visible in the field of view.
- View the specimen with the 10x objective, then with the 40x and then with the 100x oil immersion objective.

Prevent damage to the 40x objectives

- Immediately remove any immersion oil that accidentally comes into contact with the 40x objective. Because the 40x lens is located next to the 100x lens and because it is also a long lens, it is easy for it to come into contact with immersion oil accidentally. The 40x objective is not sealed against oil penetration, and any immersion oil left in contact with this objective will penetrate the lens and be deposited on the inside of the lower lens. Only the manufacturer can repair, after dismantling the objective lens.

Prevent damage to the 100x objective

- Lower the stage before removing a slide.
- After finishing a microscopic examination, immediately clean the immersion oil from the objective with lens paper and commercially prepared lens cleaner. If this is not done, the oil may thicken and harden over time and affect the optical performance of the lens.
- To protect the 100x objective lens, the objective should be rotated up when the microscope is not in use.

4.5. Daily maintenance

- Inspect the microscope for damage or malfunction.
- Record any damage or malfunction in the daily microscopy maintenance sheet. Clean the parts of the microscope with a clean cloth.
- Clean the objectives after each day's work.
- Ensure that immersion oil residues are removed. Do not clean any part of the microscope with xylene, which will damage the microscope and is toxic.
- Clean the objective lens with lens cleaning tissue only. Never clean lenses with alcohol, ordinary tissues, cleaning paper, toilet paper, cotton wool or hand towels, which will scratch the lens surface.
- Cover the microscope with a dust cover when not in use.
- Turn off the power at the end of the day, and unplug the microscope to protect it from a power surge.
- Do not leave lens ports uncovered; use the port cover or sealing tape.

4.6. Storage

- In warm humid climates, fungal growth is easily established on lenses and prisms. The microscope must be stored in dry conditions when not in use to prevent fungal growth on glass surfaces. For example, the microscope could be kept in a "warm cupboard" with a constant temperature and low humidity. Lenses and prism heads could be stored in airtight boxes containing desiccant (not containing cobalt chloride).

4.7. Repairs

- Site staff can replace a broken bulb or replace a blown fuse by strictly following the manufacturer's instructions. Other repairs must be performed by a qualified service engineer or technician.
- Do not exchange lenses and ports with those of other microscopes.
- Routine servicing, such as realignment of optics, replacement of lens and maintenance and lubrication of the stage, should be performed by a qualified service technician.
- Record all repairs on the microscope maintenance sheet.

5. PROCEDURE NOTES

Precautions and safety

- The power source and connections to the microscope should be secure and stabilized and not expose staff to a risk for electrocution.
- The microscope and electrical connections should not be exposed to water.
- Care should be taken to prevent damage to the eyes from exposure to the high light-intensity halogen lamp.
- The microscope should be used ergonomically to prevent back and neck strain.

Quality assurance

- The set-up and maintenance of the microscope should be inspected during routine supervisory visits. A register for maintaining laboratory equipment should also be inspected.
- A microscope maintenance chart similar to the table at the end of this SOP may be used.

Causes of error

- unskilled repairs attempted on malfunctioning microscopes;
- removal of eyepieces, unless they are designed to slide in and out of the tube; and
- removal of eyepieces from sealed microscopes, exposing the internal optics to dust and fungi.

6. REFERENCES

WHO. Universal access to malaria diagnostic testing. An operational manual. Geneva; 2011.

WHO. Basic malaria microscopy. Part 1: Learner's guide. Geneva; 2010.

7. DOCUMENT HISTORY

Date (mmm/yyyy)	Version	Comments	Responsible person (First name, last name)
Jan 2016	1	Reviewed and finalized by experts, edited and formatted	Glenda Gonzales, Technical Officer, WPRO

EXAMPLE OF MICROSCOPE MAINTENANCE CHART

MICROSCOPE ID: # _____ **Month/Year:** _____
Microscope type: _____ **Serial number:** _____ **Manufacturer:** _____
Date first used: _____

Please tick [✓] the activities. This chart should be filed away at the end of each month.

		Day of month																															
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	
Daily maintenance																																	
Check all optics for damage.																																	
Check coarse and fine adjustments.																																	
Clean surfaces of components.																																	
Clean the external surface.																																	
Remove oil.																																	
Replace bulbs as needed.																																	
Cover microscope after use.																																	
Technician's initials																																	
Comments																																	

QA check _____ **QA review** _____
 Date: _____ **Audit** _____