**THE MICROSCOPE**

**Parts of the binocular compound microscope**

* Identify parts, know their functions:
	+ Ocular, diopter adjustment, ocular micrometer, pointer, body tube or head, revolving nose piece, objectives (scanning, low-power, high-power, oil immersion), arm, coarse adjustment knob, fine adjustment knob, base, light source, iris diaphragm, iris diaphragm lever, condenser, condenser height control knob, pull-out phase adapter, stage, graduated mechanical stage controls, power switch, electrical cord, plug.

**Computation of total magnification of specimen being viewed:**

* **Magnification = ocular x objective (ocular on this scope is 10X)**
* Scanning (red band) 4X x 10X = 40X
* Low-power (yellow band) 10X x 10X = 100X
* High-power (blue band) 40X x 10X = 400X
* Oil immersion (white band) 100X x 10X = 1000X

**Care and handling of microscope**:

Report any problems with your microscope to your instructor IMMEDIATELY.

**1.** Locate your microscope by number in the cabinet. Carry with two hands, one under base and other around the arm. Keep the microscope near your body.

**2.** Place scope gently on the lab table on top of plastic sheet.

**3.** Do **not** disassemble your microscope or reorient the oculars.

**4.** Keep the scope and lens systems clean.

* Clean lens with lens paper (in drawer)
* Clean stage and slides with Kim wipes (on lab bench)

**Storage of your microscope**: This is the way microscopes are to be stored in the cabinet.

* Scanning objective (4X with red band) in position!
* Power switch off.
* No slide left on the stage.
* All lenses and the stage **must be clean**.
* Graduated mechanical stage centered.
* Stage in the full down position. (Do not lower the condenser.)
* Cord wrapped around the cord holder.
* Plastic dust cover is on.
* The microscope sets above its number on the cabinet shelf.

**Focusing the Microscope:**

* Clean slide to be viewed. Wipe gently with a Kimwipe. If oily, place a few drops of 70% ethyl alcohol onto the Kimwipe and gently wipe the cover slip and bottomof slide. **Never** place alcohol directly onto a prepared slide.
* Place the slide down on the front of the stage with the label facing up and in position to be read, use stage clip to hold slide in position
* Turn on the light to sufficient intensity to produce a WHITE background (not yellowish). Note that the ***condenser is in the full up position***. Locate the condenser height control knob forward from course / fine adjustment knobs.
* Use the course adjustment knob to bring the slide into focus.
* Use the iris diaphragm lever to adjust the amount of light striking your specimen. More light will be needed for preserved and stained slides and at higher magnifications. Less light is required for thin preparations and unstained slides. Remember the condenser remains in its full uppermost position.

**Moving to next higher magnification power**

* Center the specimen to be examined further in the ***CENTER*** of the field of view. These microscopes are ***parfocal***. This means that the specimen is focus at the center of the field of view will be in partial focus as the next power.
* Grasp the revolving nosepiece and rotate it to the next power lens (low power – yellow band).
* Only minor adjustment with the coarse adjustment (for scanning and low-power objectives ONLY), then fine adjustment (no more that 2 revolutions) if needed.

Again center the specimen being viewed and rotate nosepiece to next power (high power – blue band). Focus using ONLY fine adjustment.

* To use the 100X objective, you need to add a drop of **immersion oil** on the slide.

**After observations have been completed**:

* Move the revolving nosepiece to low and then to scanning. Do not drag the oil immersion objective (longest objective) across the cover slip – scratching it !
* Open the spring stage clip and slide the microscope slide out to the forward edge of the stage.
* Return the clean slide to where you obtained it (your slide box or the side counter).
* Do not lower the stage or turn off the light if you have another slide to view.
* View other slides that are assigned.
* After last slide of the day is finished, prepare the microscope for storage as outlined

**Oil immersion techniques**: (used to visualize bacteria)

* Focus the slide as before under the scanning, low power, and high power objectives. Now the stage and lighting are set for the best resolution of the specimen.
* DO NOT lower stage!
* Rotate the revolving nosepiece back the way you came to high (back to low, then to scanning). Do not drag the long oil immersion objective over the cover slip.
* Place a drop of immersion oil (from your drawer) on the cover slip where the light is passing through the slide. Be careful not to allow any oil to flow over the edge of the slide onto the condenser lens or onto the stage.
* Looking from the side of your scope, visually confirm that the objective will not touch the cover slip of the slide. Rotate the revolving nosepiece DIRECTLY from Scanning (4X) objective to the 100X objective. You can see the oil come into contact with the 100X objective.
* Focus using the fine adjustment knob.
* You may need more light. Move the iris diaphragm lever to allow more light on the slide.
* After study of specimen is complete, turn the revolving nosepiece DIRECTLY from 100X objective to the 4X objective. This avoids bringing other long lenses in contact with the oil.
* Open the spring stage clip and remove slide forward toward the edge of the stage.
* CLEAN the slide – Remove most of the oil by blotting cover slip with a Kimwipe, add some 70% ethyl alcohol on a clean Kimwipe and remove any remaining oily residue. Return clean slide to where you obtained it (your slide box or side counter)
* CLEAN the 100X objective if you are through using oil for this lab session. Blot (do NOT rub) the objective with a clean Kimwipe. Use lens paper to polish the 100X lens until no oily residue is observed on the Kimwipe. Use lens paper to check other objectives to be sure no oil is on them.

**\*\*\*Never use any liquid to clean you microscope lenses.\*\*\***

**Using phase contrast optics**:

**This type of microscopy is used when live, unstained specimens are to be viewed. (Micro Lab)**

* Focus the specimen as you have been instructed above on 4X, 10X, and 40X.
* When you are in best focus on high power (40X objective), push the phase ring holder (under stage) into the path of light. Make sure the condenser is raised to its highest position. Also, make sure the lever controlling the amount of light entering the condenser is fully open. You may also have to turn the light source on full.
* Only the high power (40X) objective may be used with the phase contrast optics.

**Using dark field optics:**

**This type of microscopy is used when studying diatoms and algae. (Micro Lab)**

* Obtain a dark field adapter. Your lab instructor who will show you where to obtain the adapter numbered for your microscope.
* Remove the blue filter and snap it onto the bottom of the adapter.
* Snap the top of the adapter to the bottom of the condenser.
* Focus normally. Your best resolution will be at low power. Note the different colors.

**Measurement of a specimen using the microscope**:

**Use the ocular micrometer – the small “ruler” in the right eyepiece.**

For ***small specimens*** which will fit under the ocular micrometer:

* Position ocular micrometer over specimen. Move the slide on stage to position the specimen. You can move the ocular micrometer by rotating the ocular. The size of the specimen can be determined by multiplying the number of ocular micrometer spaces covered by the specimen by the conversion factor for that objective as given in the following:
	+ **OBJECTIVE OCULAR MICROMETER CONVERSION FACTOR**
* Scanning 25 microns (micrometers)
* Low-power 10 microns
* High-power 2.5 microns
* Oil immersion 1.0 microns