UPRIGHT FLUORESCENT MICROSCOPE: Olympus BX60

Operating Instructions:

- 1. Turn on PC:
 - a. Press power switch on PC (if not already on)
 - b. Login with user= smic; pswd = B!01ogy
- 2. Turn on the microscope:
 - a. Switch on the back right corner to turn on transmitted light.
- 3. For fluorescence: turn on the Mercury lamp.
 - a. The power supply is located to the side of the microscope. Allow the bulb to warm up for approx. 2 min before beginning imaging.

**Note: it is helpful to first find and focus your specimen using transmitted light, then switch to fluorescence. Use this method (see below) until you are proficient with using fluorescence.

- 4. Load your slide onto the stage:
 - a. Lower the stage with the coarse focus knob, and then clip your slide onto the stage.
- 5. Select the desired objective lens (usually start with a lower objective 4, 10, or 20x):
 - a. Rotate the objective turret.
 - b. Then raise the stage with coarse focus knob (make sure you don't get too close to the lens!!)
- 6. Remove any filters/shutters (**See Figure 1):
 - a. Rotate the black 'wheel' above the objective turret until it says 0

take an image (see Imaging Instructions below) or switch to fluorescence.

Fluorescence Instructions:

- 1. Turn off transmitted light.
- 2. Rotate the filter wheel to your desired filter for your specific sample. (Green for CY3, TRITC; blue of Alexa488, FITC; UV for DAPI, etc.)
 - a. You should see a colored light coming through the objective lens (i.e. blue, green)
- 3. Find an area of your specimen that you want to image using the XY stage control and fine focus knob.

Imaging Instructions:

- 1. Pull out slider to block the light to the objective lens (right side level with the oculars)
 - a. Third stop, camera icon
- 2. Open the software on the computer: **cellSens Entry**
- 3. Click the **live** button to turn on the camera
- 4. Adjust the exposure by moving the slider to right until your image appears properly exposed
 - a. The exposure normally can be set as **auto** in bright field image mode and as **manual** in fluorescent image mode
- 5. Normalize background:
 - a. For Bright field images: on the top right toolbar, click the white dropper icon
 - b. For fluorescent images: on the top right toolbar, click the black dropper icon
 - c.



Fig. 1: Slider, Filter Wheel, Analyzer, Shutter and DIC