

Aligning the Light Source of your Microscope

A properly aligned light source is essential for both your comfort and proper illumination using phase contrast objectives. Each microscope should be aligned by first focussing then centring the light coming from the illuminator at the bottom of the microscope.

1. Take a test slide and place it in the microscope slide holder, make sure the cover slip is on the top side.
2. Turn on the light source and focus on the specimen at 10X power. The phase contrast condenser should be in the **J position**. Turn the **lamp field stop** until you see a small circle or hexagon of light.
3. For optimum illumination this circle should have a crisp outline and be in the centre of your field of view. Check for the knob on the side of the microscope that raises and lowers the **condenser only** (do not use the fine or coarse focussing knobs for the objectives). Look through the eye pieces while moving the condenser up or down to obtain a sharp outline of the circle of light from the previous step. The condenser is now set at the correct depth for the thickness of slide you are using. To centre the light, use the **two screws** set in the side of the housing that holds the condenser. These move the condenser in the holder, left or right, up or down, to centre the circle of light in your field of view. Note: If you have to turn screws more than half a turn either way the top lense of the condenser may be slightly out of position. Place it in the upright position and start from the beginning.

Once you have aligned the light, open up the lamp field stop at the bottom of the microscope before moving on to the phase contrast portion.

For Phase Contrast

1. Move the phase condenser from the **J position** to the number indicated on the side of the objective you are using, for the 10X objective go to the **1 position**.
2. Remove one eyepiece and replace it with the **telescoping eyepiece**. When phase contrast is out of alignment, two rings, one grey and one white will be visible against a dark back ground.
3. Carefully check the front and side of the phase contrast condenser. There are two places for adjustment, one a **knurled knob** at the front and one a **set screw** at the side of the condenser that should be loosened to slide back and forth. Look through the telescoping eyepiece and move the rings until they are one on top of the other. The phase contrast portion of the microscope is now in alignment and the set screw should be retightened.
4. Be sure to move your condenser to the correct position each time you move between objectives. The correct position number is written on the side of each objective, 40X requires the number 2 setting.

Under phase contrast specimens should be much easier to visualize. Stained chromosomes will appear darker or even black, sometimes with a fluorescence around the edges. Over time you will be

able to tell when your microscope will need to be realigned. Once your cells lose the contrast or appear bubble like its time to realign. Depending on the age and wear on your microscope you may have to go through the whole procedure more than once during your lab session.