

Spec 20 General Instructions

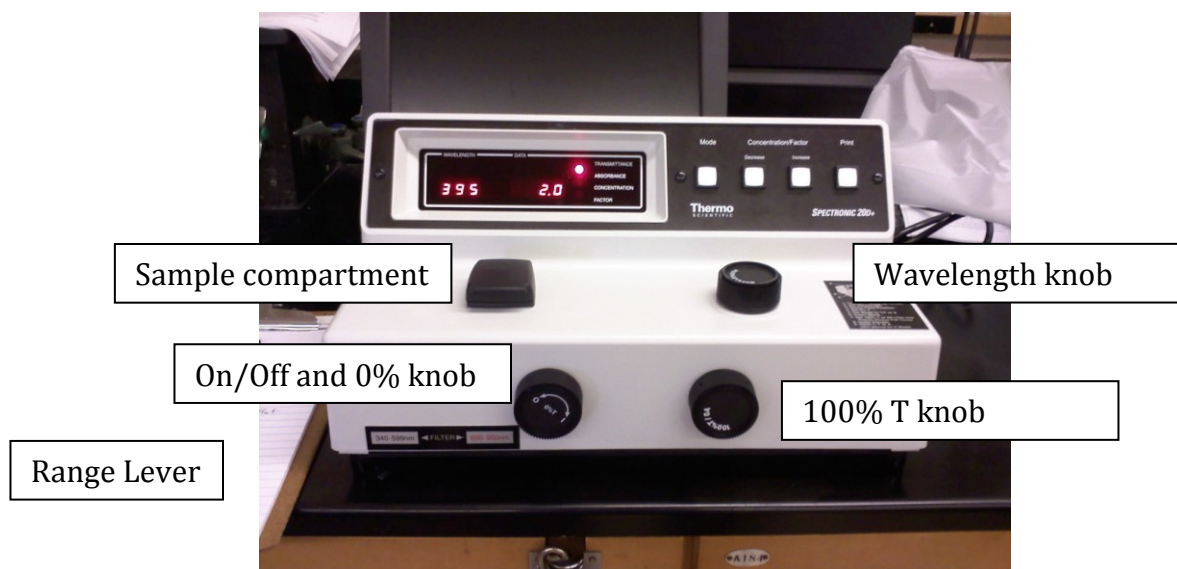
Setup – This part will be done before Gen Chem lab. Students should NOT do it themselves without explicit permission. If something seems wrong, ask an instructor.

Done before
the start of lab.
Do not do
without
permission

Step 1: Turn on the spectrophotometer (left front knob), and allow the lamp to warm up for at least 15 minutes.

Step 2: Confirm that the black lever located on the front left panel far left is set appropriately (left 340-599 nm for blue/green/UV wavelengths, right 600-950 for orange/red/near IR).

Step 3: On the digital display, confirm that Transmittance is selected. If transmittance is not highlighted, press the MODE button until it is.



Collect background – This corrects for the absorption by the cuvette and solvent. It needs to be done for each set of samples.

Step 4: Adjust the wavelength knob (top) so that the digital display reads the desired wavelength.

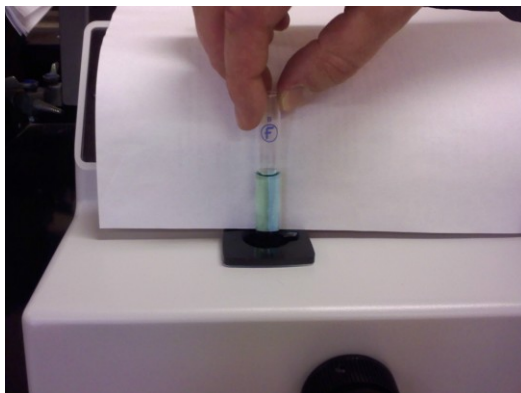
Step 5: Adjust the 0% transmittance knob (front panel, left knob) so that the transmittance reads 0.0. Be careful not to turn too far and switch the instrument off.

Step 6: Fill a blank cuvette no more than two thirds full with the solvent (often DI water). See picture on back for appropriate level.

Step 7: Dry the outside of the cuvette with a Kim-wipe to remove moisture and smudges. Insert cuvette into spectrophotometer so that the label faces outward, and is aligned with the raised line. **NEVER TWIST THE CUVETTE ONCE IT HAS BEEN PLACED!** Close the lid.

Step 8: Adjust the 100% transmittance knob (front panel, right knob) so that it reads 100%.

The spectrophotometer is now zeroed and ready to collect data. If you are prepared to analyze your samples, steps 4-8 will not need to be repeated unless the knobs are moved.



Data Collection.

Step 9: Fill a cuvette no more than two thirds full with your sample (see picture above). Wipe the outside of the cuvette to remove moisture and smudges.

Step 10: Insert your sample ensuring that the label on the cuvette faces out and is aligned with the line on the spectrophotometer. Do not twist the cuvette once it's been inserted. Close the lid. Record the transmittance. Although not allowed for CHM 111 (you have to do the calculations), pressing the mode button will provide Absorbance values.

Step 11: Repeat steps 9&10 for all samples.

Important notes

- Do not turn the spectrophotometer off unless specifically instructed to do so. If it is switched off, allow 5 minutes before turning it back on. Then wait 15 minutes for the lamp to warm up.
- Be careful not to get any liquid in the sample compartment. If a spill occurs, notify an instructor immediately.
- The hardest part is often keeping track of which sample is which. NEVER LABEL CUVETTES. Instead, label the test tube rack.
- Prepare all samples before you start measuring anything on the spectrophotometer.
- If the % T is a negative number, your sample is too concentrated for the instrument (or you didn't calibrate properly (steps 4 – 8). Dilute the sample and try again.