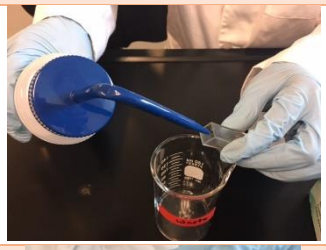
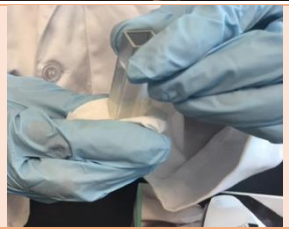
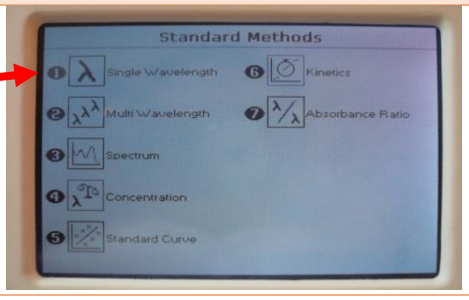
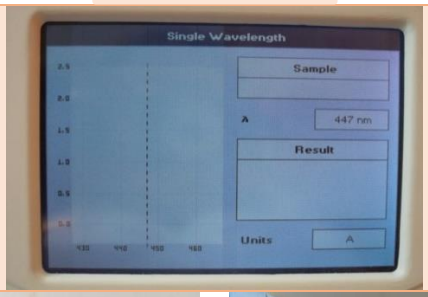
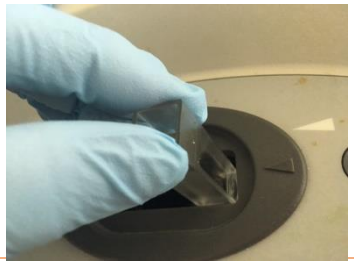
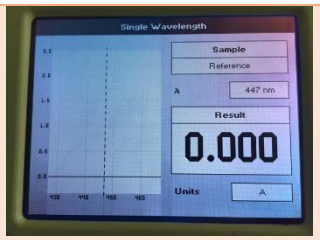
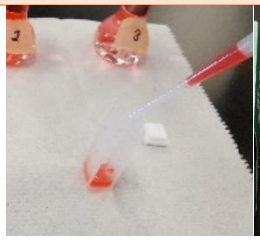
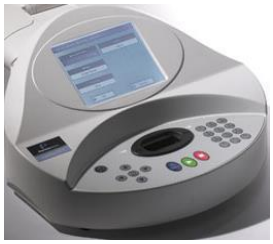
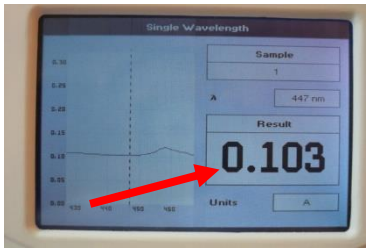
VALENCIA COLLEGE

Chemistry

Lab Technique 21b: Using a UV-Vis Spectrophotometer – Lambda XLS

Turn on the instrument. Let it warm up for 15 minutes to allow the lamp and detector to stabilize.	
Prepare your Blank (Reference) in a Cuvette:	
Condition first by rinsing the cuvette two times with a small portion of the blank ensure that your solution is the only species in the cuvette. Collect rinse in a waste beaker. Pour the solution into the cuvette to approximately ¾ full.	
Make sure that the cuvettes are clean and dry. Wipe any fingerprints with a KimWipe, not a paper towel, to avoid scratching the cuvette.	
Calibrate the Instrument for Single Wavelength Use:	
From the home page, using the keypad, select 1 to open the Standard Methods folder.	 
In the Standard Methods screen, press 1 to open the Single Wavelength method.	

In the Single Wavelength – Method Settings screen, use the arrows (1) on the instrument to navigate through the screen: a) Wavelength – enter the wavelength stated in the experiment using the keypad numbers (2) b) Mode – select Absorbance unless stated otherwise in the experiment (%Transmission is the other option)	 
Press OK  .	
The Single Wavelength results screen should now be displayed.	
Remove the cell chamber cover. Place in a safe place nearby. Do not use the cover while the instrument is in use. The light beam is directed from RIGHT to LEFT through the cell chamber as indicated by the arrows next to the chamber.	 
Notice that the cuvette has two frosted sides (left side of the picture) and two clear sides (right side of picture).	
Insert the cuvette containing the blank in the cuvette chamber: place it flush to the right side, frosted side of cuvette facing you so the clear sides are aligned with the direction of the light beam.	 

Press 0A / 100%T to zero out the instrument. The instrument should display an absorbance value of 0.000 for the blank.	 
Remove the cuvette and leave it filled with the blank, just in case you need to recalibrate or want to verify the instrument is reading correctly .	
Prepare your Sample in a Cuvette:	
Condition first by rinsing the cuvette two times with a small portion of sample to ensure that your solution is the only species in the cuvette. Collect rinse in a waste beaker. Pour the solution into the cuvette to approximately ¾ full.	 
Make sure that the cuvette is clean and dry. Wipe any fingerprints with a KimWipe.	
Insert the cuvette in the cuvette chamber and press ▶. The result at the selected wavelength is displayed on screen. Repeat this section until you have measured the absorbance of all of your samples.	 
When completely done: • Empty and rinse both cuvettes. • Press ■ to return to the Standard Methods folder after all your samples have been measured. • Replace the cell chamber cover.	
Note: This instrument is only available on the West campus. If you are on the Lake Nona campus, use Technique 21a.	