UNIT 3

EXPERIMENTING WITH SPECTROPHOTOMETER



SAFETY REMINDERS

Wear safety glasses at all times and don't wear nice clothes (you might ruin them!)



- 1. Look at Samples A, B, and C. Rank the samples based on observed color intensity from least intense to most intense. Record the data.
- 2. Use the absorbance setting at 540 nm.
- 3. Take a disposable pipette, squeeze the bulb and then place the tip of the pipette into the water. Slowly release the bulb to draw 1 mL of water into the pipette using the graduations on the pipette. Squeeze the bulb slowly to dispense the 1 mL of water directly into a cuvette. Repeat this process until you have added 4 mL of water into your cuvette. This cuvette with water will be used for comparison and is called the blank. Keep this sample throughout the entire activity because it will be your blank to zero the spectrophotometer between each reading. Always use lint-free wipes to remove any fingerprints from the cuvette before putting it in the spectrophotometer.
- 4. Put the cuvette with water (blank) into the spectrophotometer. Press and hold the zero button (0A/100%T). The spectrophotometer will flash "BLA" for a few seconds. Wait until the spectrophotometer reads 0.000 before removing the blank.
- 5. Put 4 mL of Sample A into a clean cuvette. Put the cuvette in the spectrophotometer. Measure the absorbance by looking at the number on the screen (you do not need to press any buttons) and record it.

Materials:

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- 1 spectrophotometer
- 4 cuvettes
- 3 beakers with 25 mL of various dilutions of food coloring labeled A, B, and C
- 1 beaker with 25 mL of water
- 4 disposable plastic
 1 mL pipettes
- Lint-free wipes



- 6. Repeat Steps 4-5 with Samples B and C.
- 7. Clean up by pouring out samples in the sink and rinsing the cuvettes with water. Return materials to the appropriate location.

Note:

- Cuvettes can be reused and rinsed with sterile water between uses.
- It will be calibrated with the same water initially.