Experiment 5.
The Synthesis and Analysis of Acetaminophen

Experimental Procedure

- Objectives
- Introduction
- Experimental Procedure

OBJECTIVES

- Experience the organic (drug) synthesize process
- Experience the purity confirmation experiment
  a. Thin Layer Chromatograph (“Retention Factor”)
  b. Melting point confirmation

Introduction

Acetanilide, phenacetin, and acetaminophen are mild analgesics (relieve pain) and antipyretics (relieve fever).

Preparation of acetaminophen involves treating an amine with an acid anhydride to form an amide. In this case, p-aminophenol, the amine, is treated with acetic anhydride to form an amide, acetaminophen.

The crude acetaminophen will be identified by using thin-layer chromatography method and by measuring melting point.

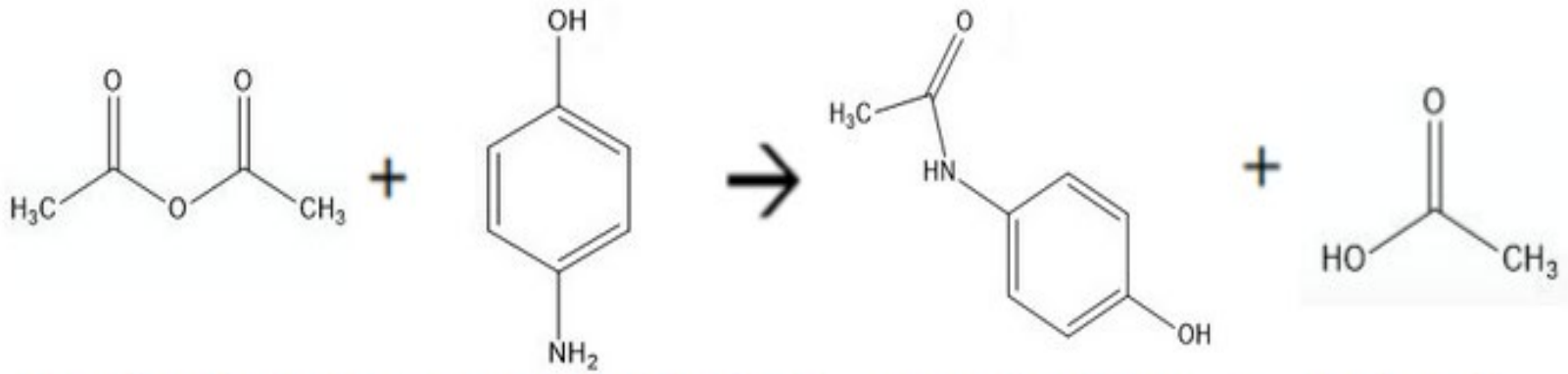


Figure 1: Chemical reaction for the synthesis of acetaminophen from acetic anhydride and p-amino phenol.

Parts 1 and 2

p-aminophenol (4-aminophenol)
Acetic anhydride,

Disposable pipette
Erlenmeyer flask, 50 mL
Beakers 400 mL, 100 mL
Graduated cylinders 10 mL and 25 mL
Watch glass
Stirring rod
Vial to hold acetaminophen sample
Buchner filtration funnel
Filter paper to fit Buchner funnel
Vacuum filtration flask
Silicone tubing for vacuum flask
Ice

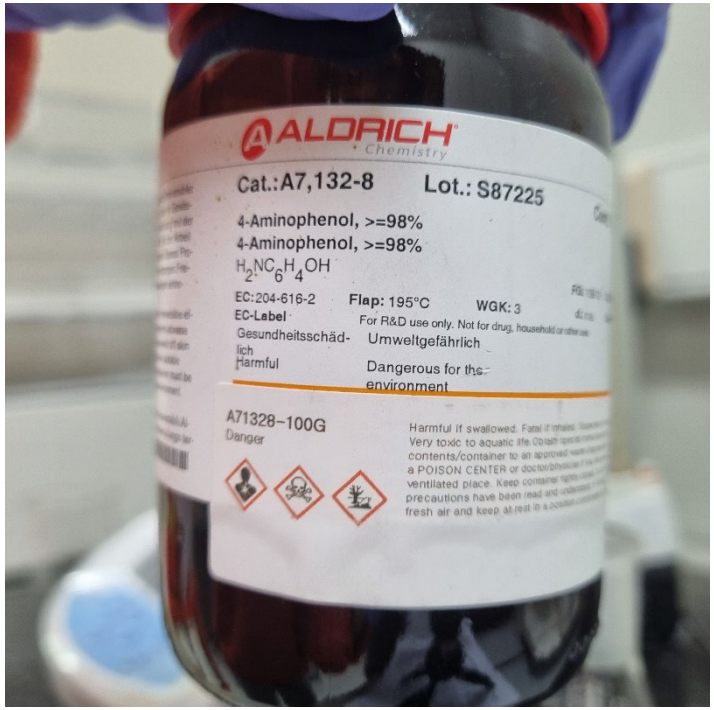
Experimental Procedure

SAFETY PRECAUTIONS

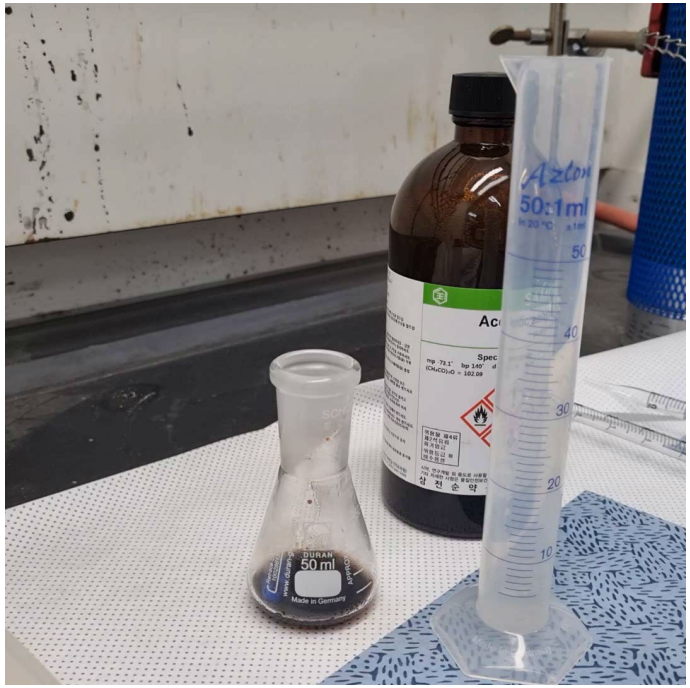
- ✓ Wear safety glasses or goggles at all times in the laboratory.
- ✓ Acetic anhydride is corrosive and its vapor is irritating to the respiratory system. Avoid skin contact and inhalation of the vapors. In the event of skin contact, rinse well with cold water. If the vapors are inhaled, move to an area where fresh air is available.
- ✓ *P*-aminophenol is harmful by inhalation and by contact with the skin. In the event of skin contact, rinse well with cold water. If the vapors are inhaled, move to an area where fresh air is available.

Part 1: Synthesis of acetaminophen

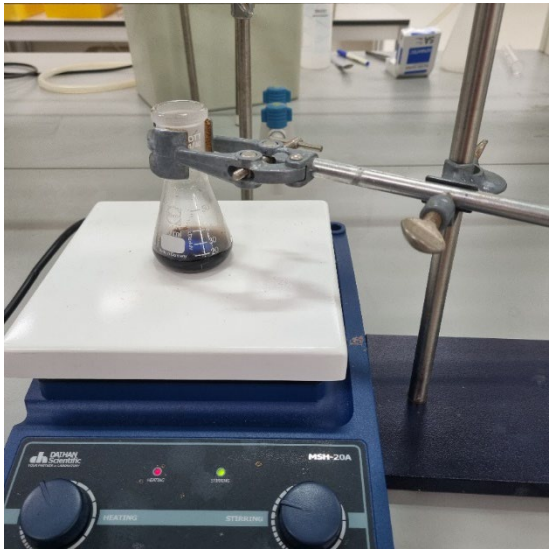
1. Weigh ~ 1.5 g of purified p-aminophenol (MW = 109.1 g/mol) and place this in a 50 mL Erlenmeyer flask. Using a graduated cylinder, add 1.7 mL of acetic anhydride (MW = 102.1 g/mol, d = 1.08 g/mL) and 5 mL of water.




SAMCH
Acetic anhydri
Special grade
SCHOTT
40
30
20

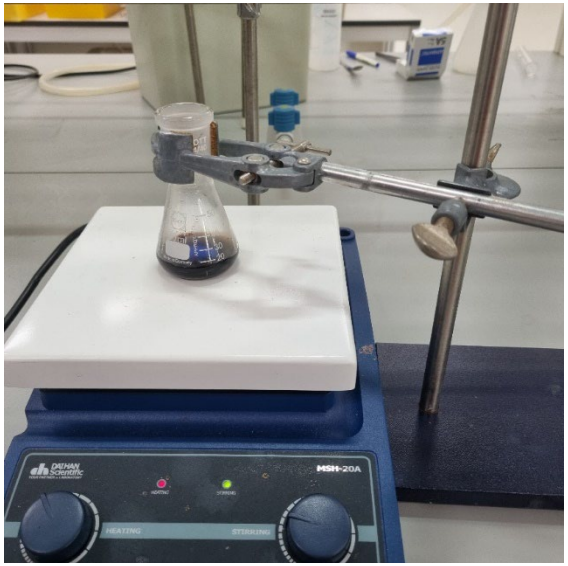

Azlon
50:1ml
DURAN
50 ml
Made in Germany

2. Place a magnetic stir bar in the flask and heat the reaction mixture on a hot place. Monitor the temperature of the reaction(While stirring the solution in the reaction vessel, the students must adjust the temperature dial on the hot plate to ensure that all the solvents in the reaction vessel do not evaporate while maintaining the reaction temperature of the vessel at ~ 100°C.)



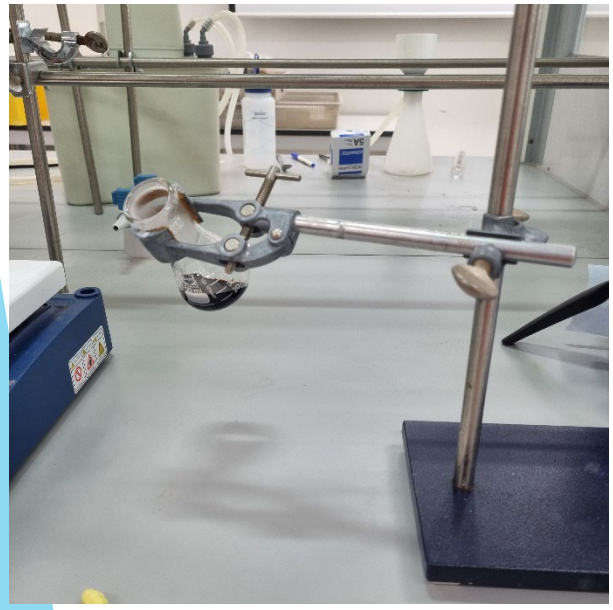
- p-aminophenol 1.5g
- Distilled water 5 mL
- 1.7 mL of Acetic anhydride

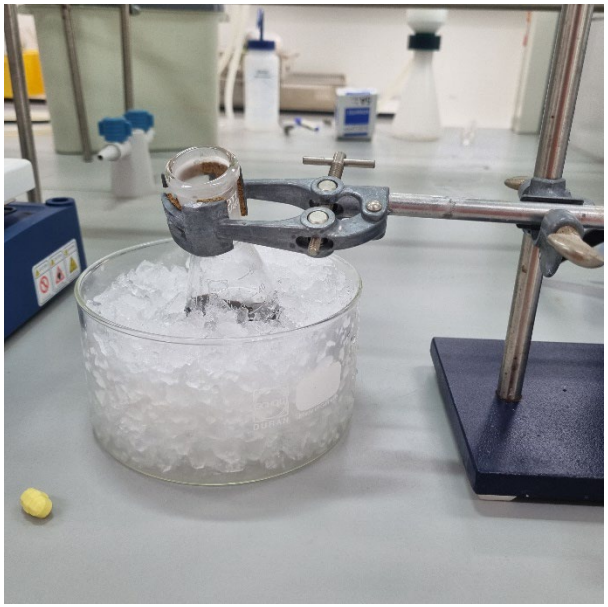
3. After the solid has dissolved (it may dissolve, precipitate and re-dissolve), heat the mixture for an additional 10 minutes to complete the reaction.

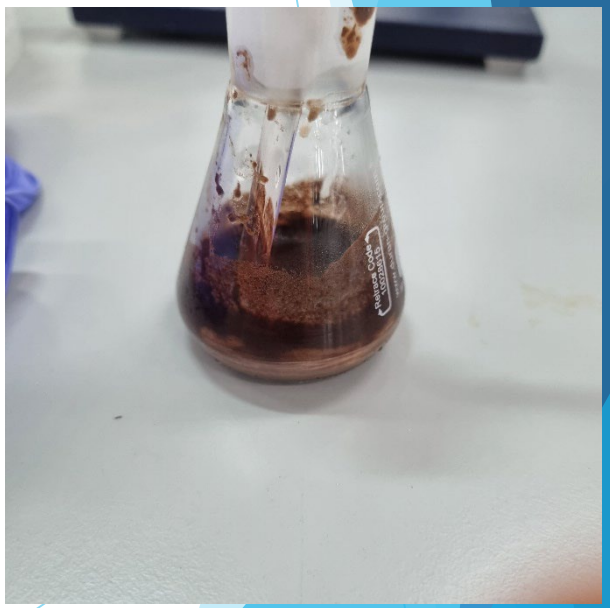


- p-aminophenol 1.5g
- Distilled water 5 mL
- 1.7 mL of Acetic anhydride

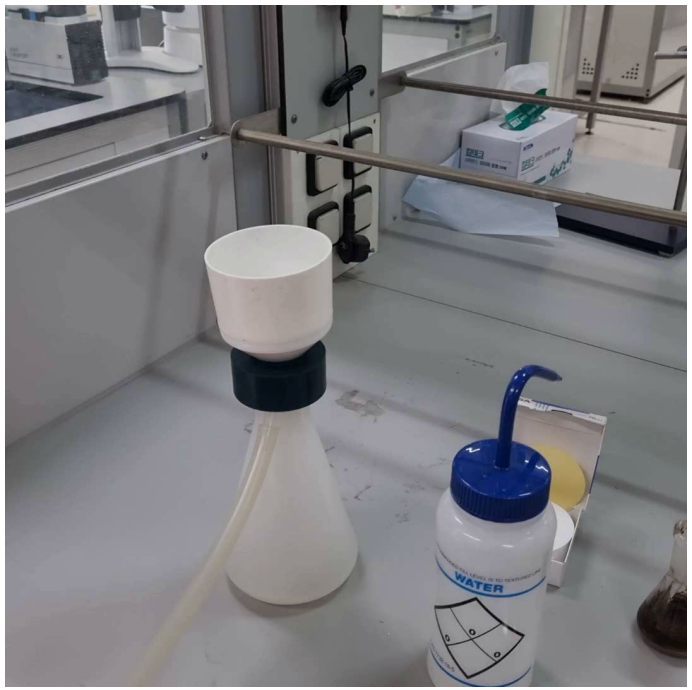
4. Remove the flask from the hot plate, and allow it to cool to room temperature slowly. If crystallization does not occur, scratch the inside walls of the flask and immerse in an ice bath for 10-15 minutes.



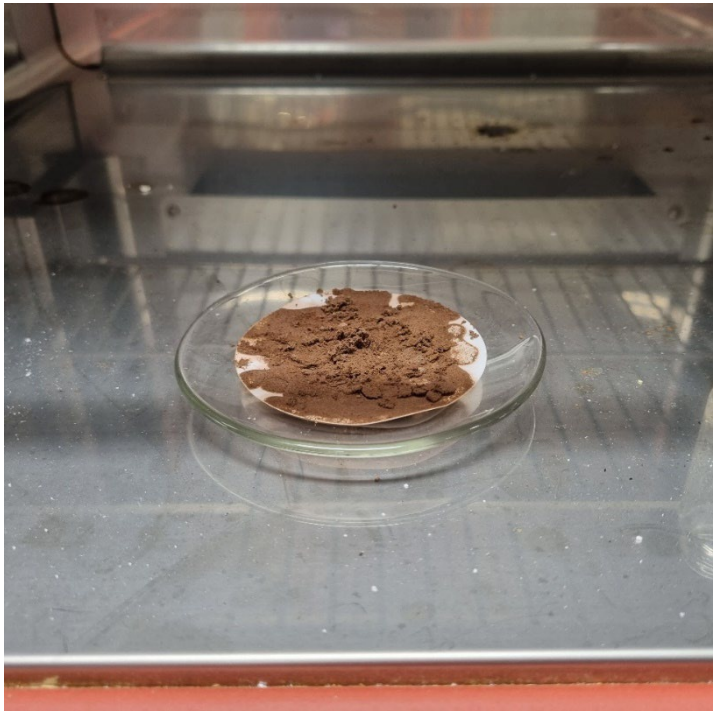




5. Collect the crystals by suction filtration on a Buchner funnel. Rinse the flask with two portions of 1 mL of ice-cold water and transfer this to the Buchner funnel. Dry the crystals for 10 minutes by allowing air to be drawn through them while they remain on the Buchner funnel. Transfer the crystals to a watch glass and record the weight of the crude product (MW = 51.2 g/mol). Calculate the crude percent yield.




WATER



Tips! Stick the wet filter paper very closely on the funnel by suction before the filtration for good filtration and dryness of your product !!!

ON/OFF

$$\%\ yield = \frac{Actual\ yield\ (grams)}{Theoretical\ yield\ (grams)} \times 100$$

Part 2: Thin Layer Chromatography

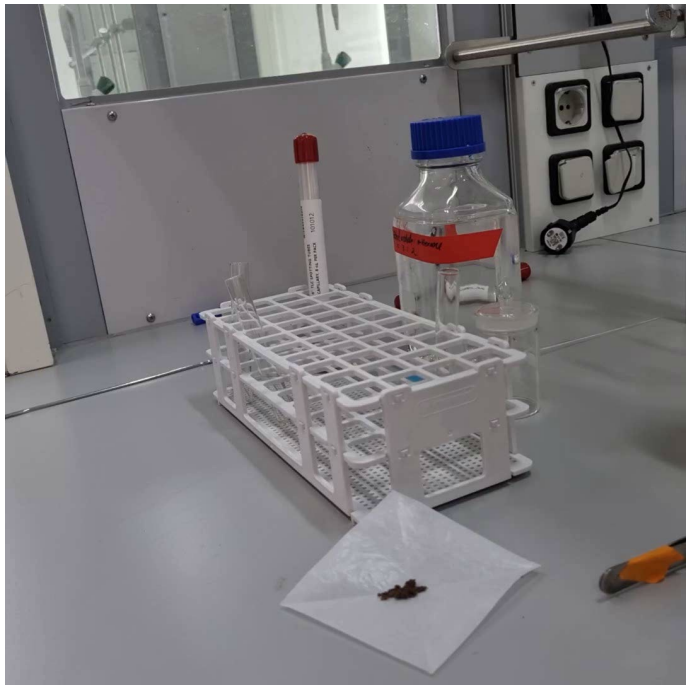
Introduction:

Chromatography is a term that is widely used to describe a family of closely related separation methods. There are many separation methods, but the feature that distinguishes chromatography from other physical and chemical methods of separation is both a stationary and mobile phase; the sample to be separated interacts numerous times with these phases. The sample is carried through the system via the mobile phase, and the interactions that occur are due to the differences in the physical and chemical properties. These differing affinities govern the rate at which the individual components of the sample pass over the stationary phase under the influence of the mobile phase.

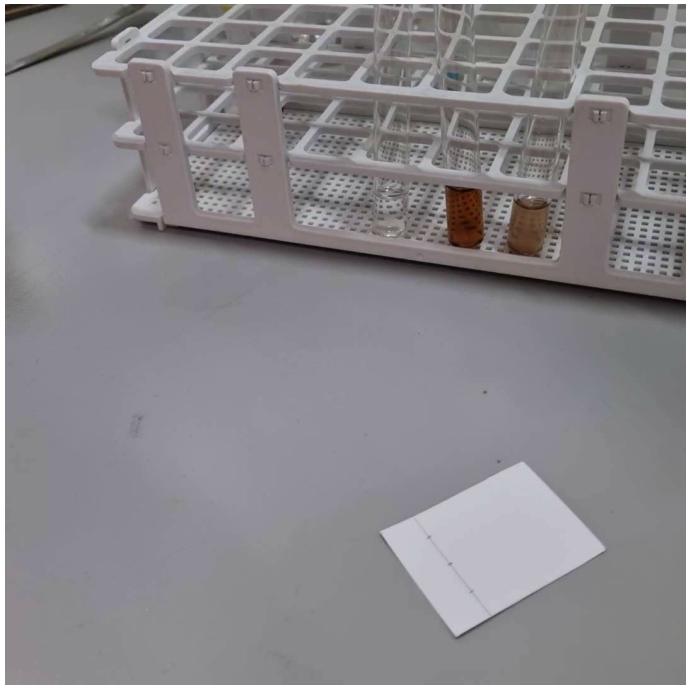
Thin layer chromatography (TLC) is one type of chromatography where the stationary phase is a thin layer of adsorbent particles attached to the solid plate. A small amount of sample is applied (spotted) near the bottom of the plate, and the plate is placed in the mobile phase. This solvent is drawn up by capillary action to a predetermined height. Each component, being different in chemical and physical composition, will interact with the stationary phase at a different time (retention time), thereby creating the individual bands on the plate. The retention time or retention factor (R_f) is used to characterize and compare components of various samples.

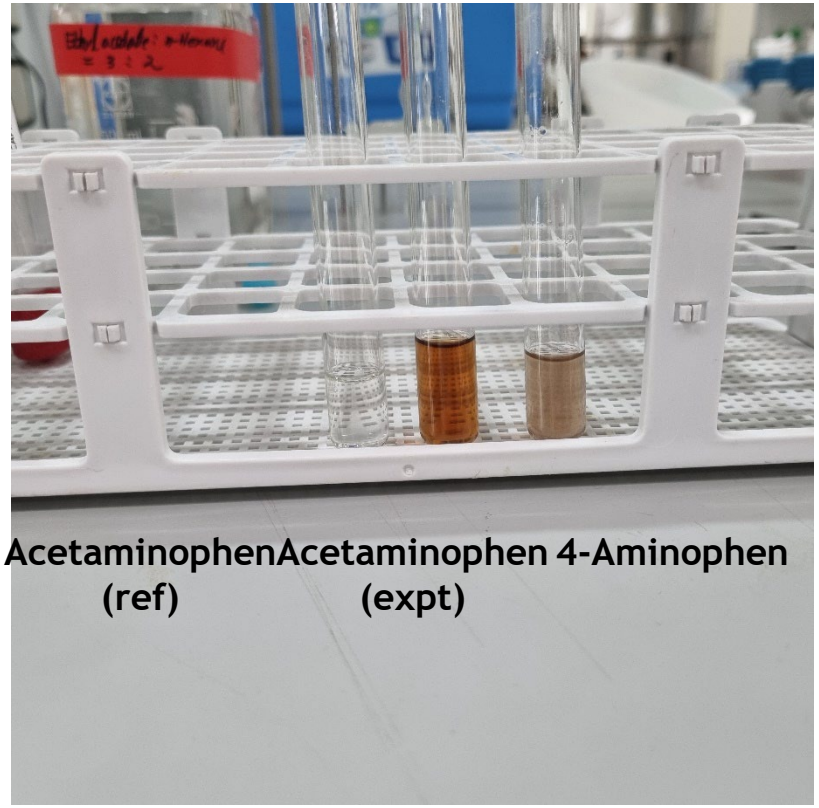
Experimental Procedure

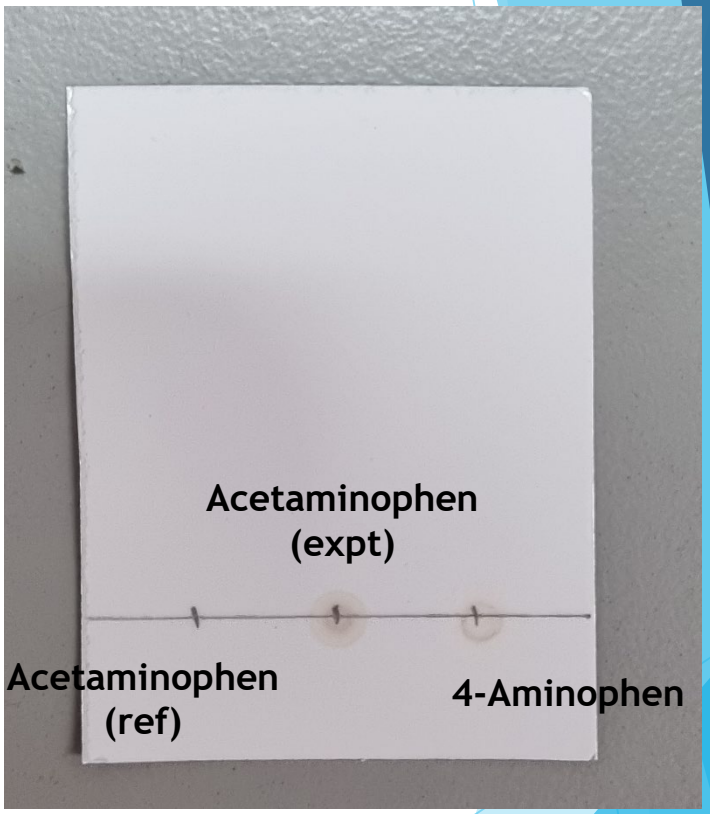
1. Obtain a couple of crystals of the crude product, dissolve them in acetone (bulk acetone is acceptable) and spot a TLC plate on the base line with the product.



2. Spot the starting material, crude acetaminophen and the authentic acetaminophen product (reference solutions are available!)



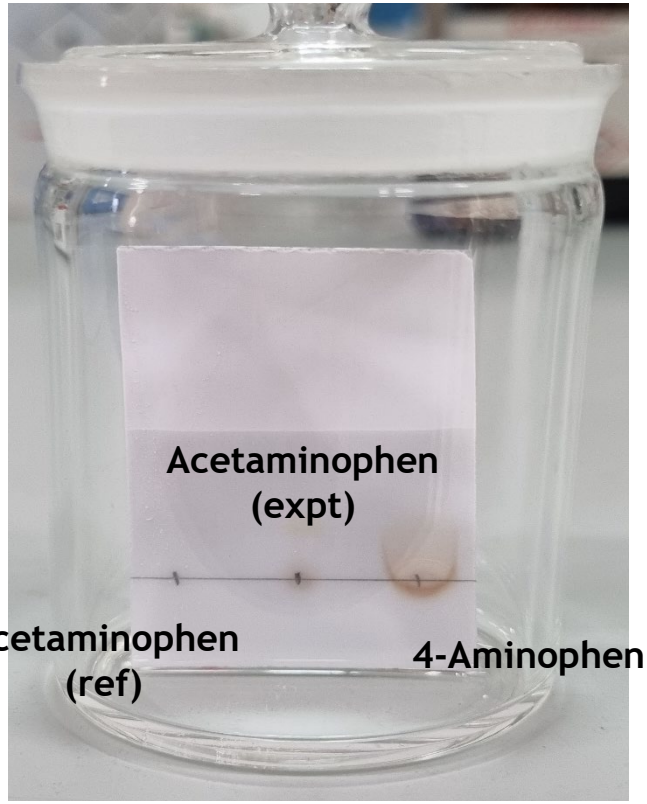

Acetaminophen
(ref)
Acetaminophen
(expt)
4-Aminophen

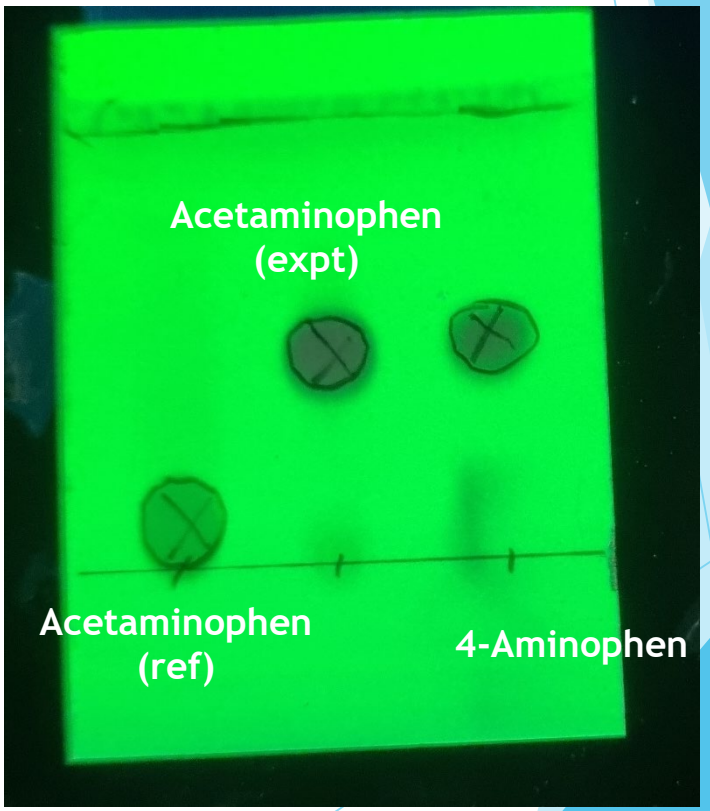

Acetaminophen
(expt)
Acetaminophen
(ref)
4-Aminophen

3. Prepare a developing chamber by adding about 0.5 cm of developing solution (55:35:10 mixture of ethyl acetate/petroleum ether/acetic acid) to the chamber. and then place the plate into the eluting chamber within proceed with elution. Stop the lid.

UV lamp

4. Remove the plate from the chamber, let it dry, and trace the spots under UV light (254 nm). Calculate the R_f values.





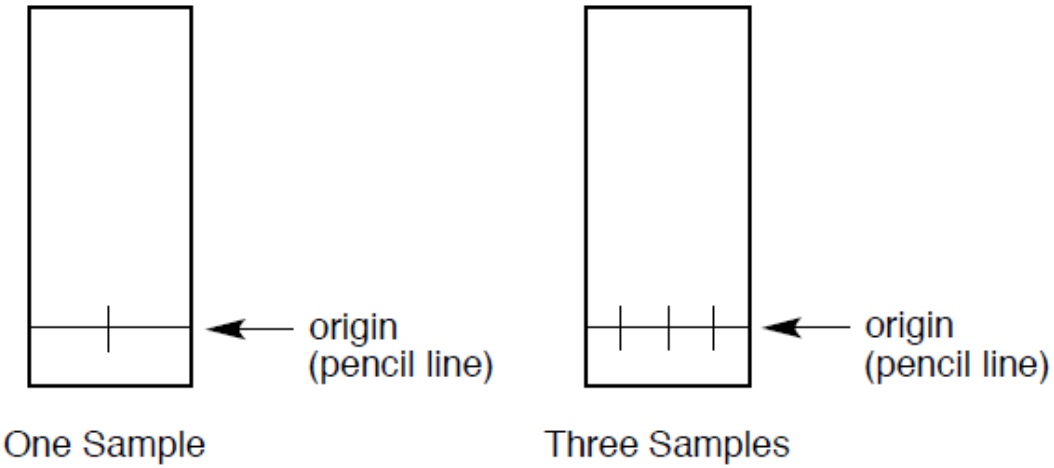
Spotting of the sample:

1. Obtain two TLC plates. Handle the plates by the edges only, making sure to avoid touching the white silica layer.

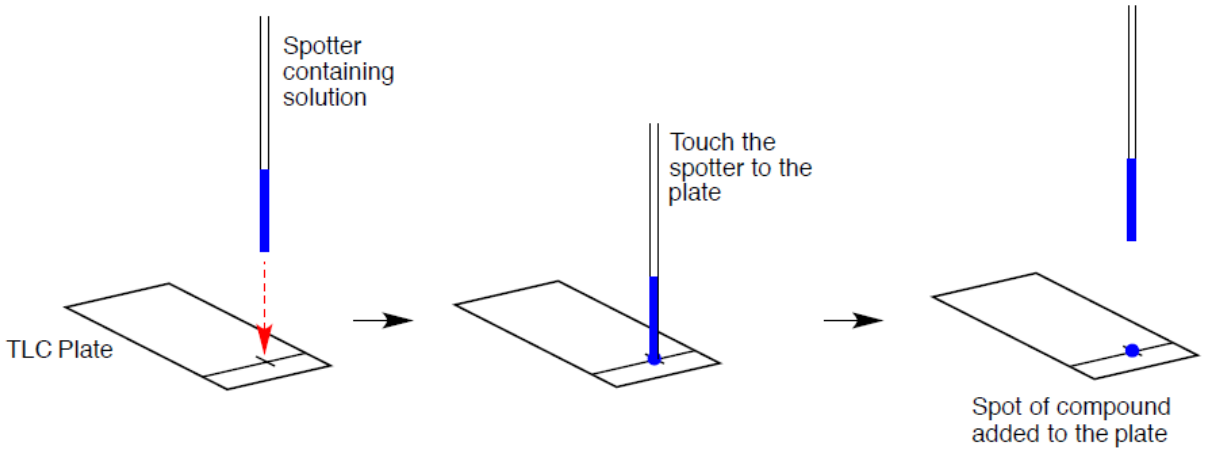
2. With a pencil (not pen), use the ruler to *gently* draw a line about 1.0 cm from the bottom of the plate. Be careful not to chip off the silica layer. Beginning about 0.5 cm from the side edge of the plate, draw 3 evenly spaced crosshairs–one for each sample.

3. Immerse the capillary tube into the sample tube until some of the liquid is drawn into the capillary.

4. Very gently press the small end of the capillary tube at a crosshair on the TLC plate. Keep the spot small by touching the capillary quickly to the plate and then removing it immediately. Allow the spot to dry and repeat the spotting *directly* over your original spot two more times.



TLC Plate Setup

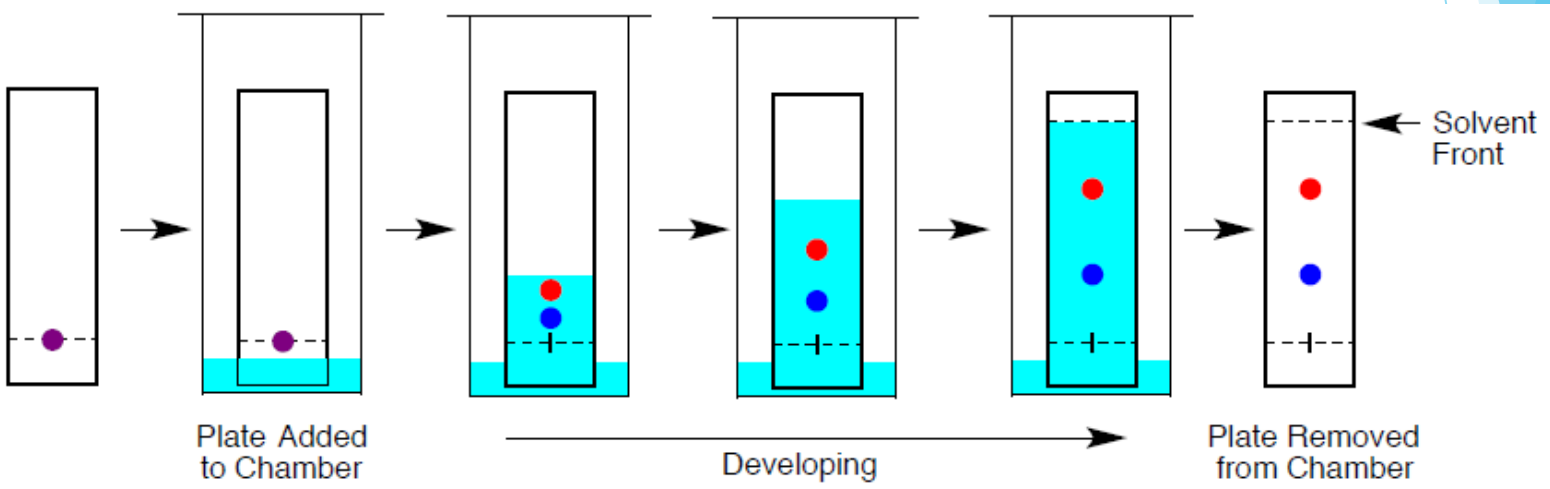


Spotting the TLC Plate

Development of the TLC plates

1. Prepare a developing chamber by adding about 0.5 cm of developing solution to the chamber.
2. Place the TLC plates in the chambers (one in each) so that your spots are aligned along the bottom and so the plate is not touching the sides of the chamber. The plate should also be facing *on a diagonal up and sideways*. Allow the solvent to rise to within one cm of the top of the plate. **WATCH—don't let the solvent rise to the top of the plate!**
3. Remove the TLC plate and immediately mark where the solvent stopped rising with a pencil. This is called the solvent front. Allow the TLC plate to dry.
4. Visualize the spots by illumination under the UV lamp.
5. Mark the center of each spot with a pencil, then remove it from the UV lamp. At your table, measure the distance traveled by each component. Using the TLC plate diagram below, draw what your plate looks like. In the table on the following page record the distances from the origin (original location of the spot) to the solvent front and the distance from the origin to the center of each spot.

TLC Developing Chamber



Developing a TLC Plate

Analyze your data

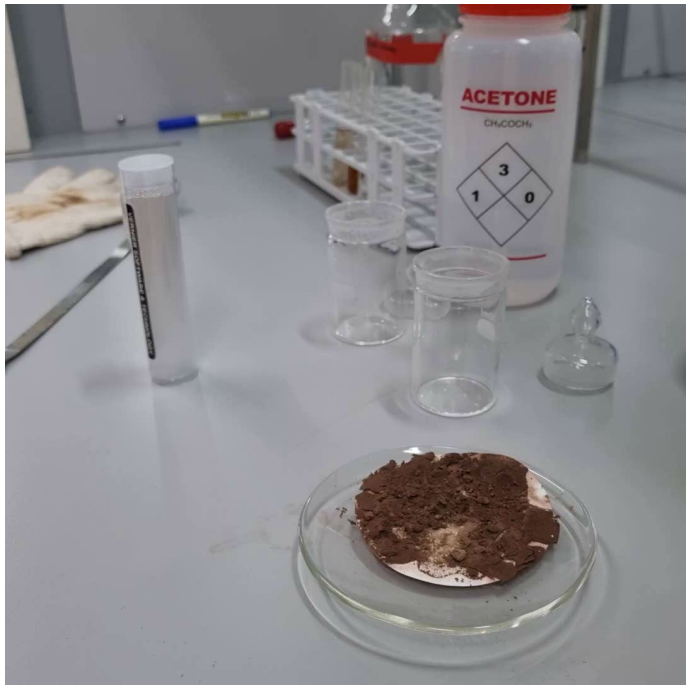
1. Determine the Rf for *every* band on your TLC plate.

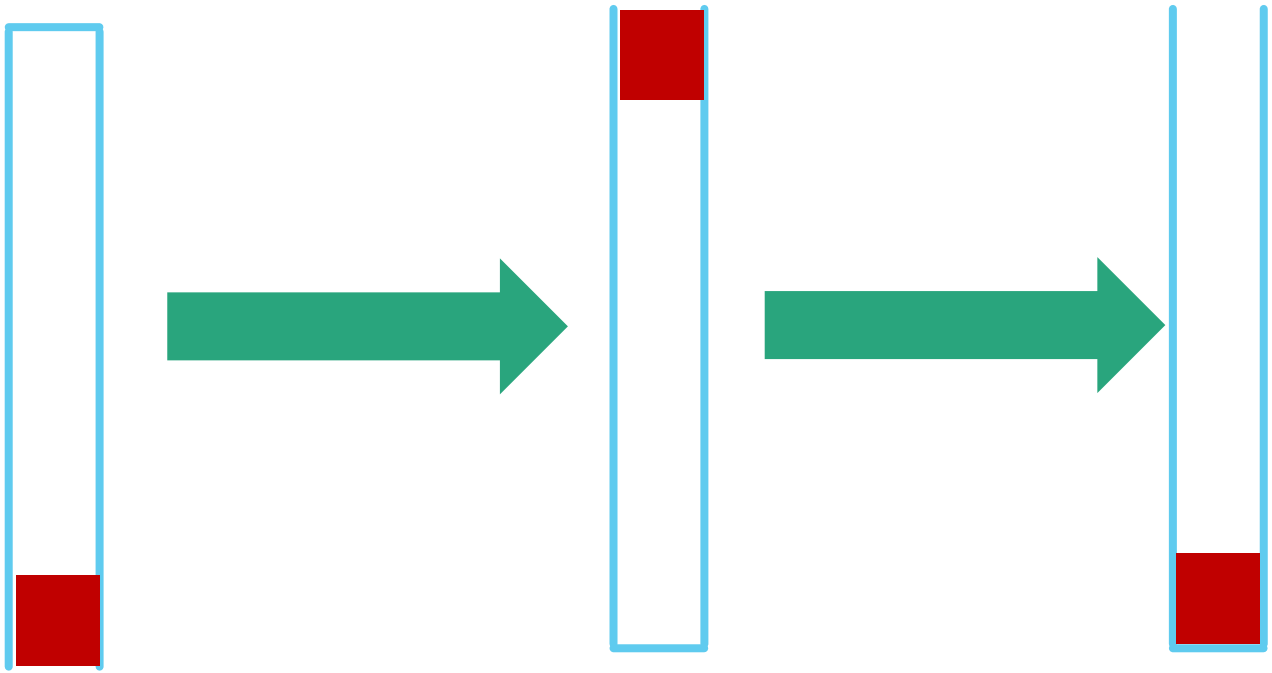
$$R_f = \frac{\text{distance from origin to center of spot}}{\text{distance from origin to solvent front}}$$

1. Record the R_f for each band in the table.

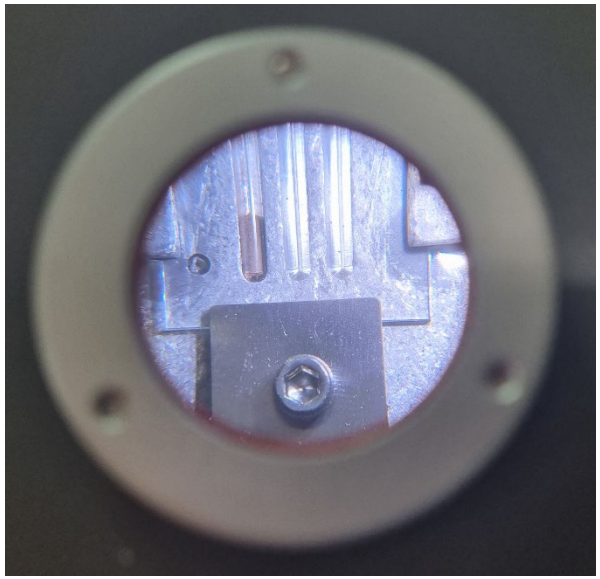
Part 3: Determining the melting point of the acetaminophen sample

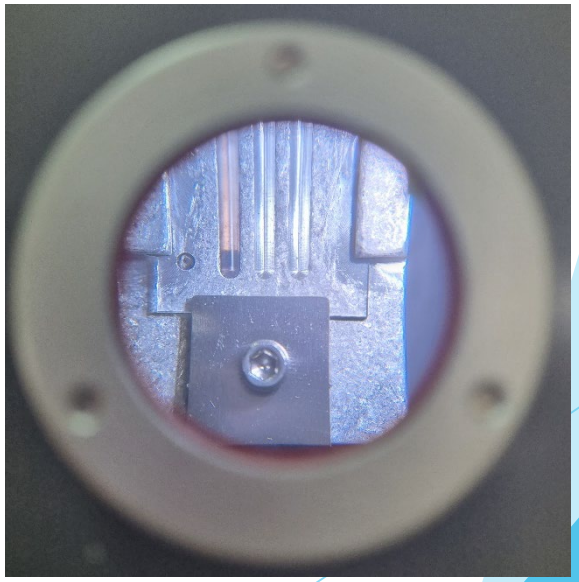
1. Fill a capillary melting point tube to a depth of 0.2 cm with the recrystallized acetaminophen.


ACETONE
CH3COCH3
3
1
0



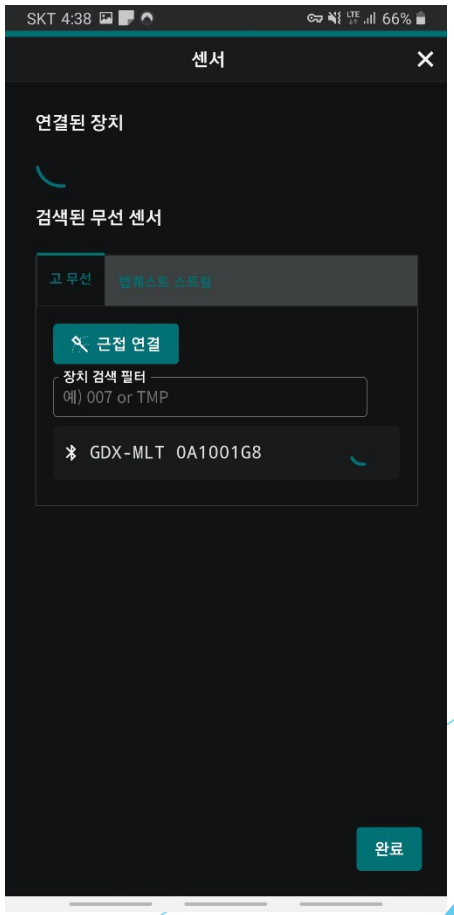
2. Place the capillary tube in the melting point apparatus. **The melting point range should be recorded from first droplets forming to complete liquidation of compound.**

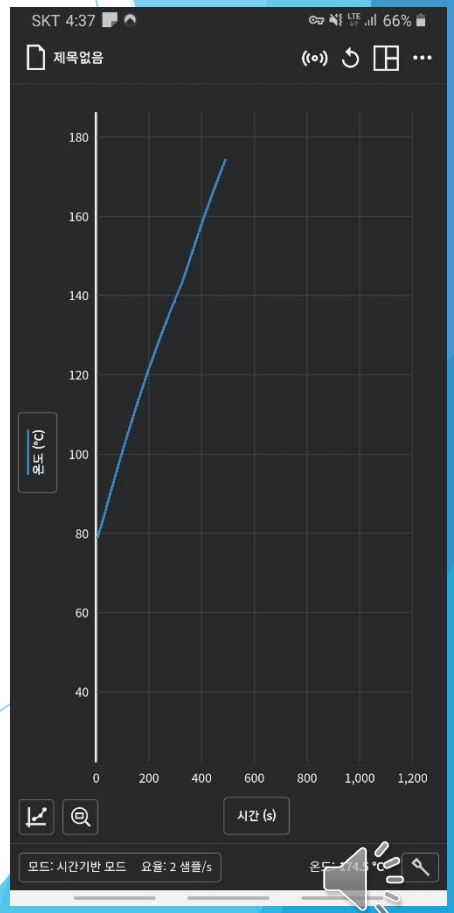




Measure the temperature

a. Install Graphical Analysis™ on your smart phone.
b. Launch Graphical Analysis™ on your smart phone.
c. Touch sensor data collection mode.
d. Wireless connection to the sensor and your smartphone
e. Record temperature.





1. Load a small portion of a solid substance into a capillary tube.
2. Carefully place the capillary tube of solid into one of the three slots in the aluminum heating block of the Melt Station. You can tilt the Melt Station toward you slightly for a better look at the heating block.
3. Tilt the Melt Station up or down slightly to get the best view of the solid sample through the viewing lens.
4. Click Collect to begin data collection. On the Melt Station, turn the control knob to the Rapid Heat area. The red LED will come on, indicating the Melt Station is heating. Rapid Heat will warm your solid sample at a rate of >10° C/min.
5. Observe the temperature vs. time graph. When the temperature is within about 10° C of the expected melting temperature of your solid sample, turn the control knob to that temperature, slowing the heating rate to ~1.5° C/min.

6. Carefully observe your sample. At the first indication of the solid melting, note and record the temperature. When the entire solid has melted, note and record the temperature. The examine line can be used to help mark these spots while monitoring the substance melting. Text can be added using the Text Annotation feature in Graphical Analysis.
7. Stop data collection. The run is automatically stored. On the Melt Station, turn the control knob to the Fan/Cooling setting. The blue LED will come on, indicating that the Melt Station is cooling.
8. Prepare a second solid sample to test. Observe the temperature of the heating block in the meter. After the heating block cools to a suitably low temperature, you can begin heating the Melt Station again.

RESULTS

Mass of p-aminophenol _____g

Mass of crude acetaminophen _____g

Mass of purified acetaminophen _____g

Theoretical yield of acetaminophen _____g

Percent yield of acetaminophen _____%

Melting point of acetaminophen _____°C

TLC Solvent System:	
	R_f Value
4-Aminophenol	
Crude Acetaminophen	
Authentic Acetaminophen	