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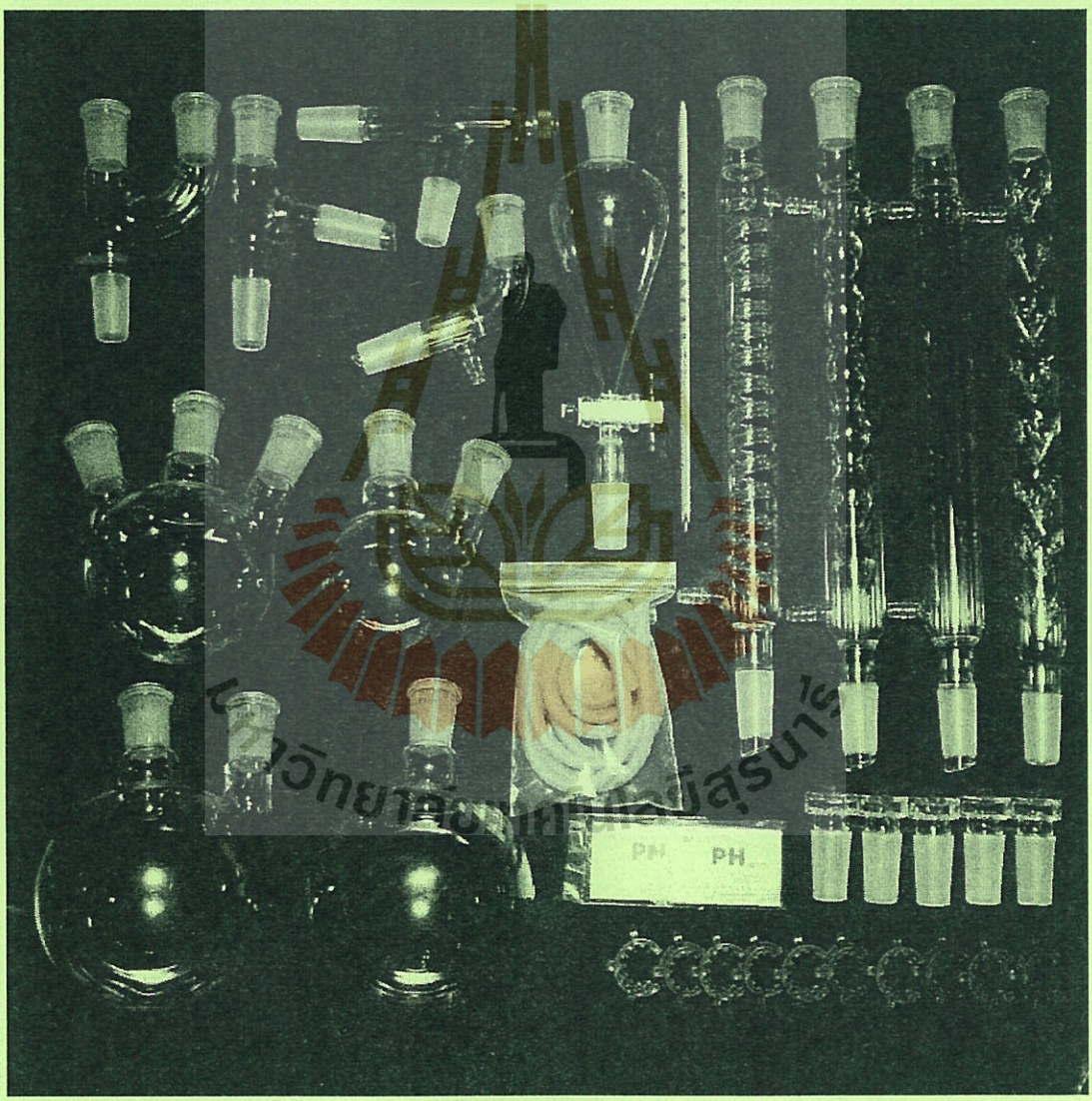

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สาขาวิชาเคมี สำนักวิชาวิทยาศาสตร์

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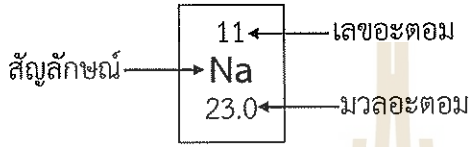

1A												3A	4A	5A	6A	7A	8A
1 H 1.0 hydrogen	2A																2 He 4.0 helium
3 Li 6.9 lithium	4 Be 9.0 beryllium											5 B 10.8 boron	6 C 12.0 carbon	7 N 14.0 nitrogen	8 O 16.0 oxygen	9 F 19.0 fluorine	10 Ne 20.2 neon
11 Na 23.0 sodium	12 Mg 24.3 magnesium	3B	4B	5B	6B	7B	8B			1B	2B	13 Al 27.0 aluminum	14 Si 28.1 silicon	15 P 31.0 phosphorus	16 S 32.1 sulfur	17 Cl 35.5 chlorine	18 Ar 39.9 argon
19 K 39.1 potassium	20 Ca 40.1 calcium	21 Sc 45.0 scandium	22 Ti 47.9 titanium	23 V 50.9 vanadium	24 Cr 52.0 chromium	25 Mn 54.9 manganese	26 Fe 55.8 iron	27 Co 58.9 cobalt	28 Ni 58.7 nickel	29 Cu 63.5 copper	30 Zn 65.4 zinc	31 Ga 69.7 gallium	32 Ge 72.6 germanium	33 As 74.9 arsenic	34 Se 79.0 selenium	35 Br 79.9 bromine	36 Kr 83.8 krypton
37 Rb 85.5 rubidium	38 Sr 87.6 strontium	39 Y 88.9 yttrium	40 Zr 91.2 zirconium	41 Nb 92.9 niobium	42 Mo 95.9 molybdenum	43 Tc (98) technetium	44 Ru 101.1 ruthenium	45 Rh 102.9 rhodium	46 Pd 106.4 palladium	47 Ag 107.9 silver	48 Cd 112.4 cadmium	49 In 114.8 indium	50 Sn 118.7 tin	51 Sb 121.8 antimony	52 Te 127.6 tellurium	53 I 126.9 iodine	54 Xe 131.3 xenon
55 Cs 132.9 cesium	56 Ba 137.3 barium	57 La* 138.9 lanthanum	72 Hf 178.5 hafnium	73 Ta 180.9 tantalum	74 W 183.9 tungsten	75 Re 186.2 rhenium	76 Os 190.2 osmium	77 Ir 192.2 iridium	78 Pt 195.1 platinum	79 Au 197.0 gold	80 Hg 200.6 mercury	81 Tl 204.4 thallium	82 Pb 207.2 lead	83 Bi 209.0 bismuth	84 Po (209) polonium	85 At (210) astatine	86 Rn (222) radon
87 Fr (223) francium	88 Ra (226) radium	89 Ac† (227) actinium	104 Rf (261) rutherfordium	105 Db (262) dubnium	106 Sg (263) seaborgium	107 Bh (264) bohrium	108 Hs (265) hassium	109 Mt (268) meitnerium	110 Ds (269) darmstadtium	111 Rg (272) roentgenium	112 Cn (285) copernicium	113 Uut (286) ununtrium	114 Fl (289) flerovium	115 Uup (289) ununpentium	116 Lv (293) Livermorium	117 Uus (294) ununseptium	118 Uuo (294) ununoctium

*Lanthanides	58 Ce 140.1 cerium	59 Pr 140.9 praseodymium	60 Nd 144.2 neodymium	61 Pm (145) promethium	62 Sm 150.4 samarium	63 Eu 152.0 europium	64 Gd 157.3 gadolinium	65 Tb 158.9 terbium	66 Dy 162.5 dysprosium	67 Ho 164.9 holmium	68 Er 167.3 erbium	69 Tm 168.9 thulium	70 Yb 173.0 ytterbium	71 Lu 175.0 lutetium
†Actinides	90 Th 232.0 thorium	91 Pa 231.0 protactinium	92 U 238.0 uranium	93 Np (237) neptunium	94 Pu (244) plutonium	95 Am (243) americium	96 Cm (247) curium	97 Bk (247) berkelium	98 Cf (251) californium	99 Es (252) einsteinium	100 Fm (257) fermium	101 Md (258) mendelevium	102 No (259) nobelium	103 Lr (260) lawrencium

ตัวเลขในวงเล็บคือมวลอะตอมของไอโซโทปที่เสถียรที่สุด

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Experiment 1	Determination of Melting Point

Objectives

1. Learn how to obtain an accurate melting point using Thiele tube and Griffin Melting point apparatus.
2. Determine the melting point of an unknown solid and use it to identify the unknown.
3. Identify an unknown solid using mixed melting point.

Introduction

The melting point of a compound is the temperature at which the solid phase is in equilibrium with the liquid phase. A solid compound changes to a liquid when the molecules acquire enough energy to overcome the forces holding them together in an orderly crystalline lattice. For most organic compounds, these intermolecular forces are relatively weak.

The normal melting point of a solid is defined as the temperature at which the solid and liquid are in equilibrium at a total pressure of 1 atmosphere. In contrast to the volume change that accompanies the vaporization of a liquid, the change in volume that takes place upon the melting of a solid is very small. This makes the melting point of a solid, unlike the boiling point of a liquid, practically independent of any ordinary pressure change.

The melting point range is defined as the span of temperature from the point at which the crystals first begin to liquefy to the point at which the entire sample is liquid. Most pure organic compounds melts over a narrow temperature range of 1-2 °C.

The presence of a soluble impurity almost always causes a decrease in the melting point expected for the pure compound and a broadening of the melting point range. In order to understand the effects of impurities on melting point behavior, consider the melting point-mass percent composition diagram for two different fictitious organic compounds, X and Y, shown in Figure 1.1. The vertical axis represents temperature and the horizontal axis represents varying mass percent compositions of X and Y.

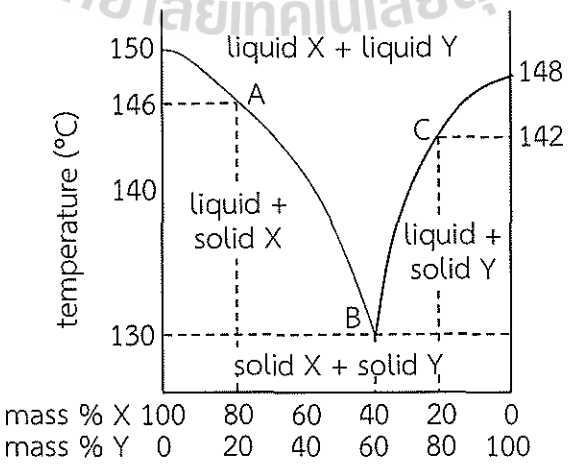


Figure 1.1 Melting point-mass percent composition diagram

Both compounds have sharp melting points. Compound X melts at 150 °C, as shown on the left vertical axis, and Y melts at 148 °C, as shown on the right vertical axis. As compound X is added to pure Y, the melting point of the mixture decreases along curve C-B until a minimum temperature of 130 °C is reached. Point B corresponds to 40 mass percent X and 60 mass percent Y and is called the eutectic composition for compound X and Y. Here, both solid X and solid Y are in equilibrium with the liquid. The eutectic temperature of 130 °C is the lowest possible melting point for a mixture of X and Y. At temperature below 130 °C, mixtures of X and Y exist together only in solid form.

Consider a 100-μg mixture composed of 20 μg of X and 80 μg of Y. In this mixture, X acts as an impurity in Y. As the mixture is heated, the temperature rises to the eutectic temperature of 130 °C. At this temperature, X and Y begin to melt together at point B, the eutectic composition of 40 mass percent X and 60 percent Y. The temperature remains constant at 130 °C until all 20 μg of X melts. At the eutectic temperature, X and Y will melt in the ratio of 40 parts X to 60 parts Y. If 20 μg of X melts, then 30 μg of Y also melts (20 μg X x 60/40 ratio = 30 μg Y). At this point, the remaining 50 μg of solid Y is in equilibrium with a molten mixture of the eutectic composition.

As more heat is applied to the mixture, the temperature begins to rise, and the remaining Y begins to melt. Y continues to melt as the temperature increases, shown by curve B-C.

Finally, at 142 °C, point C, where the liquid composition is 20 mass percent X and 80 mass percent Y, all of Y is melted. At temperatures higher than 142 °C, liquid X and liquid Y exist together with a composition at which the entire mixture liquefies is 142 °C, 6 degrees lower than the melting point of pure Y. Also, the melting point range 130-142 °C is quite broad.

If a mixture has exactly the eutectic composition of 40 mass percent X and 60 mass percent Y, the mixture shows a sharp melting point at 130 °C. Observing this melting point could lead to the false conclusion that the mixture is a pure compound. Addition of either pure X or pure Y to the mixture causes an increase in the melting point, as indicated by curve B-A or B-C, respectively. Observing this melting point increase indicates that the original sample is not pure.

Because the melting point of a compound is a physical constant, the melting point can be helpful in determining the identity of an unknown compound. A good correlation between the experimentally measured melting point of an unknown compound and the accepted melting point of a known compound suggests that the compound may be the same. However, many different compounds have the same melting point.

A mixed melting point can be useful in confirming the identity of an unknown compound. A small portion of a known compound, whose melting point is known from the chemical literature, is mixed with the unknown compound. If the melting point of the mixture is the same as that of the known compound, then the known and the unknown compounds may be identical. A decrease in the melting point of the mixture and a broadening of the melting point range indicates that the compounds are likely to be different.

Melting point can also be used to assess compound purity. Generally, a melting point range of 5 °C or more indicates that a compound is impure. Purification of the compound causes the melting point range to narrow and the melting point to increase. Repeated purification may be necessary before the melting point range narrows to 1-2 °C and reaches its maximum value, indicating that the compound is pure.

If large amounts of the solid are available (a gram or so), the most accurate method for determining the melting point is to heat the sample until it is melted and then allow it to cool slowly for crystallization. Keep track of the temperature of the sample as a function of time by means of an immersed thermometer as in a cooling curve. At first, the temperature falls as the liquid loses heat to the surroundings. When crystallization begins, however, the heat evolved during this process ($-\Delta H_{fus}$, the heat of fusion) will maintain the temperature at a constant value until crystallization is complete. At this point, the temperature will again fall as the solid loses heat to the surroundings. If the material is pure, the temperature of the sample remains constant during the entire process of solidification; this temperature is the melting point. This procedure is the one that should be used for calibrating a thermometer or checking the calibration of a thermometer by using solids of known melting points such as *p*-dichlorobenzene (mp. 53 °C), benzoic acid (mp. 122 °C), salicylic acid (mp. 159 °C) and anisic acid (mp. 184 °C) as reference compounds. In this case, any disagreement between the reading of the thermometer and the true melting point is attributed to an error in the calibration of the thermometer.

Capillary melting points, either in an oil bath or a melting-point apparatus, are most often used for the determination of the melting point of a solid. A finely powdered compound is packed into a thin-walled capillary tube 10-15 cm long, about 1 mm in inside diameter, and closed at one end, to a depth of 2-4 mm. The capillary, which contains the sample, and a thermometer are then suspended so they can be heated slowly and evenly. The temperature range over which the sample is observed to melt is taken as the melting point.

The thermometer and sample must be at the same temperature while the sample melts, so the rate of heating must be slow as the melting point is approached (about 1 °C per minute). Otherwise, the temperature of the thermometer bulb and the temperature of the compound in the capillary may not be the same. The transfer of heat energy by conduction takes place rather slowly.

If the melting point of the compound is unknown, it is convenient to first measure the approximate melting point of the compound, called the orientation melting point. The sample is heated at a rate of 10-15 °C per minute until it melts. Then the melting point apparatus is cooled to approximately 15 °C below the orientation melting point. A new sample is heated, increasing the temperature at a much slower rate of 1-2 °C per minute, to accurately measure the melting point. A slow heating rate is necessary because heating a sample too rapidly may cause the thermometer reading to differ from the actual temperature of the heat source.

If the melting point of the sample is known, the sample can be quickly heated to within 10-15 °C of its melting point. Then the heating rate can be slowed to increase 1-2 °C per minute until the sample melts.

Errors in observed melting points often occur due to a poor heat transfer rate from the heat source to the compound. One cause of poor heat transfer rate is the placement of too much sample into the capillary tube. Finely ground particles of the compound are also necessary for good heat transfer. If the particles are too coarse, they do not pack well, causing air pockets that slow heat transfer.

Sometimes slight changes, such as shrinking and sagging, occur in the crystalline structure of the sample before melting occur. Also, traces of solvent may be present due to insufficient drying and may appear as droplets on the outside surface of the sample. This phenomenon is called sweating and should not be mistaken for melting. The initial melting point temperature always corresponds to the first appearance of liquid within the bulk of the sample itself.

Some compounds decompose at or near their melting points. This decomposition is usually characterized by a darkening in the color of the compound as it melts. If the decomposition and melting occur over a narrow temperature range of 1-2 °C, the melting point is used for identification and as an indication of sample purity. The melting point of such compound is listed in the literature accompanied by *d* or *decomp*. If the sample melts over a large temperature range with decomposition, the data cannot be used for identification purposes.

Some compounds pass directly from solid to vapor phase without going through the liquid phase, a process called sublimation. When sublimation occurs, the sample at the bottom of the capillary tube vaporizes and recrystallizes higher up in the capillary tube. A sealed capillary tube is used to take the melting point of a compound that sublimes at or below its melting point. The literature reports the melting point for these compounds accompanied by *s*, *sub*, or *subl*.

Thiele Tube Method

The Thiele tube is a glass tube designed to contain heating oil and a thermometer to which a capillary tube containing the sample is attached. The shape of the Thiele tube allows for formation of convection currents in the oil when it is heated. These currents maintain a fairly uniform temperature distribution throughout the oil in the tube. The side arm of the tube is designed to generate these convection currents and thus transfer the heat from the flame evenly and rapidly throughout the heating oil. The sample, packed in a capillary tube is attached to the thermometer, and held by means of thread, a rubber band or a slice of rubber tubing as shown in Figure 1.2. It is important that this rubber band be above the level of the oil (allowing for expansion of the oil on heating). Otherwise, the oil softens the rubber and allows the capillary tubing to fall into the oil. Don't clamp the Thiele tube too tightly otherwise it might crack as it expands as it is heated, yet of course it needs to be secure.

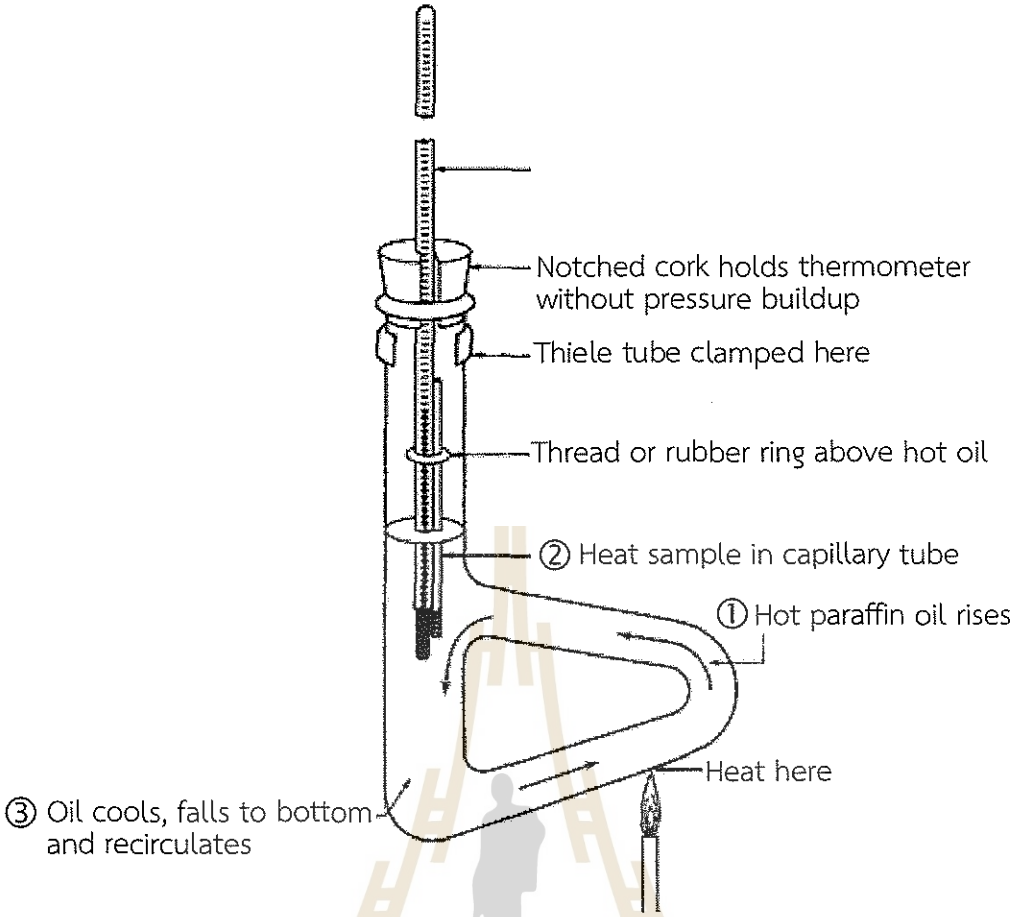


Figure 1.2 Arrangement of sample and thermometer and Thiele tube setup for melting point determination

The Thiele tube is usually heated using a microburner with a small flame but a Bunsen burner can also be used. When heating, the rate of temperature increase should be carefully controlled. Usually one holds the burner by its base and, using a small, gentle flame, moves the burner slowly back and forth along the bottom of the side arm of the Thiele tube. If the heating rate is too fast, the burner is removed for a few seconds before resuming the heating process. The rate of heating should be slow near the melting point (about 1-2 °C per minute) to ensure that the rate of temperature increase is not faster than the ability of the heat to be transferred to the sample being observed. At the melting point it is necessary that the thermometer bulb and the sample in the capillary tube be at thermal equilibrium.

Griffin Melting Point Apparatus

The Griffin Melting point apparatus, as shown in Figure 1.3, is for the determination of single or mixed melting points of solids up to a maximum temperature of 350 °C.

In use, the sample is contained in a capillary tube which is placed with a thermometer in a heating block within the apparatus. Internal illumination allows the melting process to be viewed through a magnifying lens on the front of the case and when this occurs, the melting point temperature is then read off from the thermometer.

The aluminum heating block has a through-hole with a glass window at each end to prevent air convection. Heating elements are clamped to either side of the block and are controlled via a solid-state circuit by a variable-heat control knob on the front panel. A rapid-heat switch is fitted to give full power when heating up and the variable-heat control enables fine adjustment of the heating rate as the melting point temperature is approached.

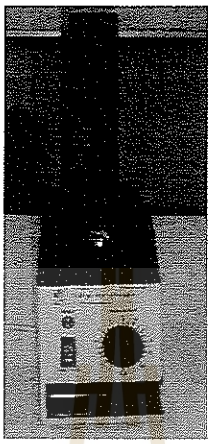


Figure 1.3 Griffin Melting point apparatus

The variable-heat control incorporates the mains ON-OFF switch and when in the ON position, both the mains indicator and sample illumination lamps will light. The control knob moves over an arbitrary 0 to 10 scale and an approximate guide to the setting temperatures is given in Figure 1.4.

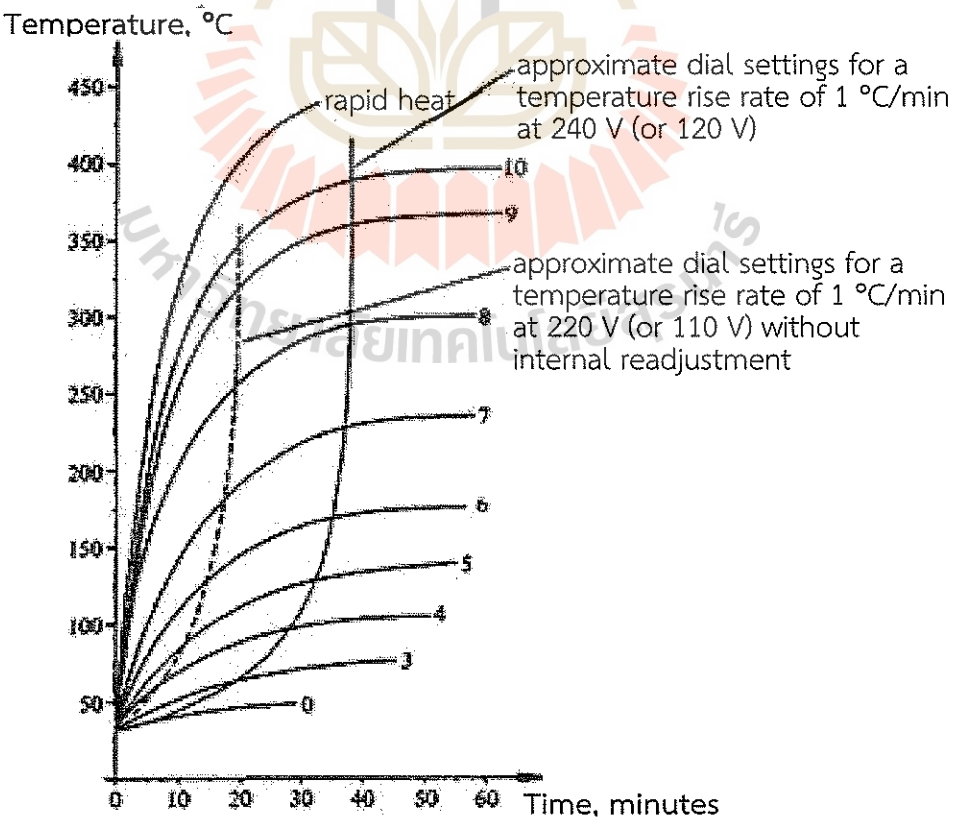


Figure 1.4 Approximate dial setting temperatures

Melting point determinations are straightforward in principle but optimum results will only be obtained through the exercise of care and correct technique.

Select a thermometer covering the required range and insert into the heating block. If the approximate melting point temperature is unknown, use a 0 to 360 °C range thermometer.

Place crystals of the material under investigation in a capillary tube to a depth of 2-4 mm and tap the tube gently on the bench a fixed number of times (say five). The depth and packing density of the sample can affect the results and care should be taken to achieve consistency. In addition, for optimum results the sample should ideally be in the form of a finely-divided powder which should be dried in a suitable desiccator for 24 hours if it is at all hygroscopic.

Insert the capillary tube into the heating block beside the thermometer and place empty capillary tubes in any holes not in use.

If the approximate melting point temperature is unknown, it should be determined by a trial run with the variable-heat control initially set to 6 or 7 on the scale. It must be remembered that, at high heating rates, a difference of up to 20 °C can exist between the temperature of the sample and the thermometer reading. Under these conditions, the indicated melting point temperature is usually a little higher than the true value.

When the approximate melting point temperature is known, the graph of Figure 1.4 can be used to determine the variable-heat control setting required to give a temperature rise rate of 1 °C/min at the given temperature.

Once the variable-heat control is set, the heating block will begin to heat up. If the expected melting point temperature is more than about 15 to 20 °C higher than the temperature of the heating block, then the rapid-heat switch may be used to increase the heating rate. There will be a short response time delay and, as mentioned previously, a difference between the temperature of the sample and the thermometer reading will develop. Care must therefore be taken to ensure that the required temperature is not exceeded. The correct technique will soon be acquired with practice.

Note. The rapid-heat switch must not be left on for prolonged periods.

When a suitable heating rate has been established, the melting process may be observed through the magnifying lens and the melting point temperature recorded.

After each determination, cool the heating block in readiness for the next sample. If this is a repeat or similar sample, it is only necessary to allow the temperature to fall a few degrees. However, if the next sample has a considerably lower melting point temperature, then the heating block can be cooled more quickly by using a water-cooled plug in place of the thermometer.

Experimental

You will first practice the melting point technique on benzoic acid (mp. 121-122 °C) using both Thiele tube and Griffin Melting point apparatus. You will be then given an unknown from Table

1.1. By determining the melting point of your unknown using Thiele tube, you will be able to narrow the choices to three, based on the melting point ranges. You will then use the mixed melting point method to identify your unknown by mixing it with each of the three possibilities and determining the melting point of each of the mixtures.

Experimental Procedure

A. Melting Point of Benzoic Acid

1. Obtain a sample of benzoic acid from a bottle (about a spatula-tip).
2. Grind the sample into a fine powder with the help of a spatula on a clean, dry watch glass as shown in Figure 1.5, and gather the powder into a small pile.

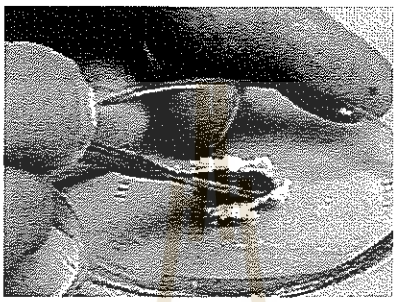


Figure 1.5 Grinding solid sample with a spatula

3. Fill a capillary tube with the finely powdered sample by pressing the open end into the pile to a depth of about 1 mm, then invert the capillary and gently dropping it through a glass funnel with a long stem onto a bench top to encourage the solid to drop to the bottom. Repeat this step until you have accumulated a sample 2-4 mm high in the bottom of the tube. If you have more solid than this in the tube, you should try to shake some out, then reseat the solid at the bottom of the tube.
4. Attach the capillary tube to a normal mercury thermometer with thread as shown in Figure 1.2.
5. Measure the melting point using Thiele tube as shown in Figure 1.2. Read the thermometer and record the temperature at which the sample first begins to melt and at which the entire sample completely melts. This span of temperature is the melting point range of the sample.
6. Repeat step 3 with a new capillary tube and measure the melting point using Griffin Melting point apparatus as shown in Figure 1.3 with the same thermometer. Record the melting point range.

B. Melting Point of an Unknown

1. Obtain a sample from your instructor. Record sample number
2. Follow steps 2-5 in section *A* to find a melting point of the sample using Thiele tube. It will be necessary to allow the hot bath to cool at least 15-20 °C below the suspected melting point before starting your experiment. Record the melting point range.

C. Identification of an Unknown

1. Based on the observed melting point, select 3 compounds in Table 1.1 below that are most likely to be your unknown.
2. Verify the identity of your unknown by measuring the mixed melting point with each of the selected known compounds using Thiele tube. The mixture can be made by putting together an equal quantity of the unknown and the known compounds on a watch glass. Use a spatula to thoroughly blend the mixture. Record the melting point range and identify the unknown compound.

Table 1.1 List of compounds and their melting points

Compound	Melting point (°C)
acetanilide	113-115
p-aminobenzoic acid	188-189
camphoric acid	183-186
trans-cinnamic acid	133-134
malonic acid	135-137
p-nitrophenol	113-115
resorcinol	110-113
succinic acid	187-189
urea	133-135

Laboratory Safety Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. Many chemicals are potentially harmful. Prevent contact with your eyes, skin, and clothing. Avoid ingesting any of the reagents.
3. Unknown may be flammable, toxic, and irritating.
4. Capillary tubes are fragile and easily broken.
5. Before using a Bunsen burner make sure all flammable materials are removed from the area around the Bunsen burner.
6. Take care to avoid burning yourself when using paraffin oil, a Bunsen burner, and the melting point apparatus.
7. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 1

Experiment 2	Determination of Boiling Point

Objectives

1. Learn how to obtain an accurate boiling point using micro method.
2. Determine the boiling point of a pure and impure liquids to study the effect of different impurities.
3. Determine the boiling point of an unknown liquid and use it to identify the unknown.

Introduction

Like the melting point of a solid, boiling point is useful for identification of a liquid and indicating its purity. Boiling point of a liquid (at a given pressure) is defined as the temperature at which its vapor pressure is equal to the atmospheric (surrounding) pressure. Though a pure liquid has a sharp boiling point, the converse is not always true. A sharp boiling point may sometimes be caused by a constant boiling mixture (azeotrope) of two or more liquids.

If a sample of a liquid is placed in an otherwise empty space, some of it will vaporize, and the pressure in the space above the liquid will rise to some constant value. The pressure under these conditions is due entirely to the vapor of the liquid, and is called the equilibrium vapor pressure.

The phenomenon of vapor pressure is interpreted in terms of molecules of liquid escaping into the empty space above the liquid. In order for the molecules to escape from the liquid phase into the vapor phase, the intermolecular forces (in order of increasing strength: van der Waals, dipole-dipole, hydrogen bonding) have to be overcome which requires energy. Since the nature of the intermolecular forces is determined by the molecular structure, then the amount of energy required to vaporize the sample also depends on the molecular structure, an example of the relationship between structure and properties. As the number of molecules in the vapor above the liquid becomes larger, the rate of return of the molecules from the vapor to the liquid increases until the rate of return is equal to the rate of escape. This is the equilibrium condition and the corresponding concentration of molecules in the vapor phase gives rise to the equilibrium vapor pressure. At higher temperatures, the greater average kinetic energy of the molecules in the liquid results in a greater constant rate of escape. Equilibrium is established at higher temperatures, and so larger numbers of molecules are present in the vapor phase and the pressure is higher.

When the vapor pressure of a liquid is equal to the atmospheric (or applied) pressure, then boiling occurs. The temperature at which this occurs, for a given pressure, is the boiling point. It should be noted, therefore, that the boiling point of a liquid decreases as the atmospheric (or applied) pressure decreases.

As a rule of thumb, the boiling point of many liquids will drop about 0.5 °C for a 10 mm decrease in pressure in the vicinity of 760 mmHg. At lower pressure, a 10 °C drop in boiling point is observed for each halving of the pressure.

A method for correcting boiling point requires knowing the atmospheric pressure (in mmHg), P_{obs}, when the boiling point, BP_{obs} is measured:

$$BP_{corr} = BP_{obs} - (P_{obs} - 760 \text{ mmHg}) \times 0.045 \text{ °C/mmHg}$$

Therefore, for the boiling point of water observed at 670 mmHg, based on 100 °C at sea level would be:

$$100 \text{ °C} = BP_{obs} - (670 \text{ mmHg} - 760 \text{ mmHg}) \times 0.045 \text{ °C/mmHg}$$

$$BP_{obs} = 96 \text{ °C}$$

At lower pressures, a boiling point nomograph or temperature-pressure alignment chart (Figure 2.1) can be used.

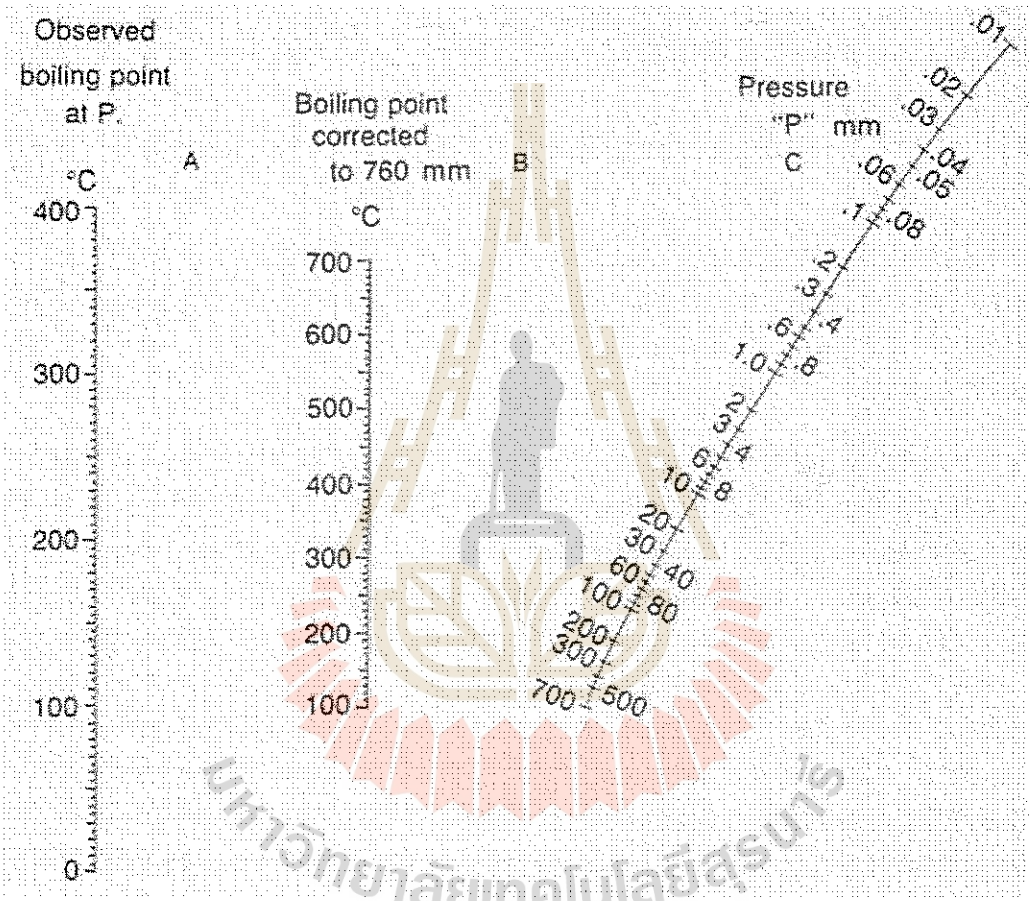


Figure 2.1 A pressure nomograph, used to correct boiling points

How to Use the Pressure Nomograph

The basic principle is that a line through two known points on any two different scales (A, B, C) can be used to read off the value on the third scale.

There are two ways in which a pressure nomograph can be used (i) to determine the boiling point at atmospheric pressure (760 mmHg) given the boiling point at a lower pressure and (ii) to determine the boiling point at a lower pressure given the boiling point at atmospheric pressure.

First, let's say we have a compound with a boiling point of 100 °C at 1 mmHg pressure. What is the boiling point at 760 mmHg?

To do this, we need to draw a line from 100 °C on scale A (left side, observed boiling point) to 1.0 mmHg on scale C (right side, pressure "P" mm) as shown in Figure 2.2. We can then read off the boiling point at 760 mmHg on line B, it is about 280 °C.

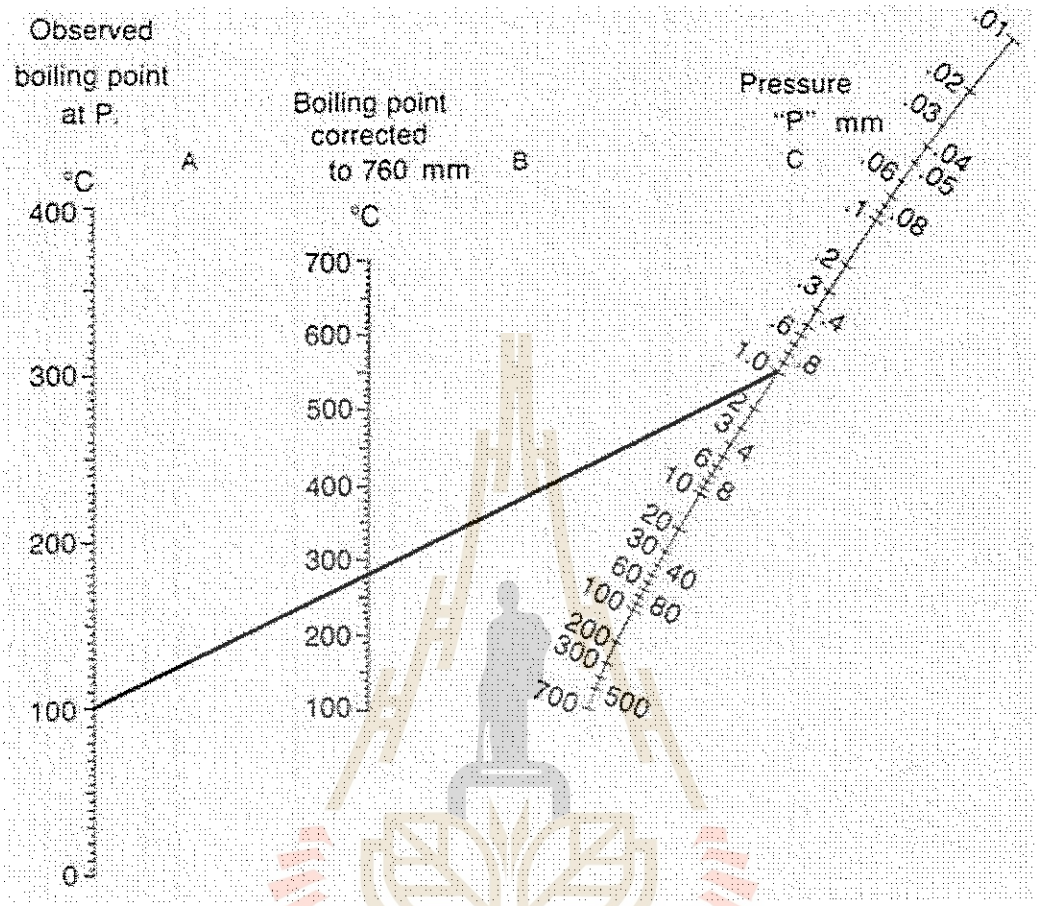


Figure 2.2 Using a pressure nomograph: Determining the boiling point at 760 mmHg (scale B) for a sample that boils at 100 °C (scale A) at 1 mmHg

Now what temperature would that same compound boil at 10 mmHg pressure? We draw a line that passes through 280 °C on scale B (middle scale, the boiling point at 760 mmHg) and to 10 mmHg on scale C. By extending that line to scale A, we can read off the new boiling point on scale A (left side) as being about 140 °C as shown in Figure 2.3.

Of course, you don't really have to "draw" the line, it can be done just using the edge of a ruler or something else straight.

In theory, when a liquid is at its boiling point one should observe bubbles of vapor forming as the liquid changes to the vapor phase. However, in practice this is usually not the case. Typically, the liquid becomes superheated as its temperature climbs above the true boiling point commences. Then the solution suddenly "bumps" or boils with tremendous vigor, bumping the hot liquid out of the container. Steps must be taken to guard against this process.

In order to promote smooth boiling, the solution can be stirred, or boiling stones (boiling chips) can be added to the liquid. These glassy type devices work by providing a sharp surface

upon which bubbles naturally forms which promote smooth generation of bubbles (prevent bumping and formation of large bubbles).

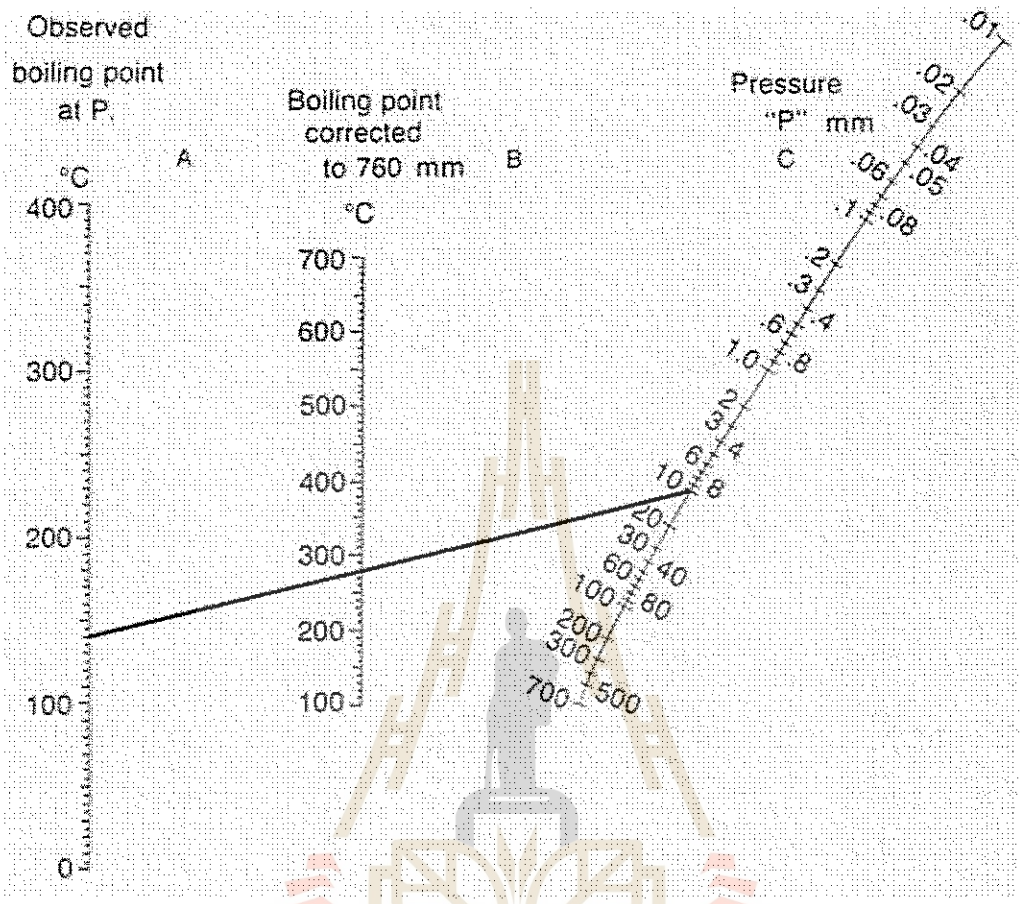


Figure 2.3 Using a pressure nomograph:
Determining the boiling point (scale A) at 10 mmHg (scale C) for a sample that boils at 280 °C at 760 mmHg (scale B)

Boiling point can be determined in a few different ways. If a reasonably large volume of sample is available, then the boiling point of the liquid can be determined using a simple distillation apparatus setup. However, when large volumes are not available, alternative experimental procedures have to be used. The micro method described below can be carried out with just a few mL of a liquid sample and is a reasonably accurate and convenient way to determine the boiling point.

Micro-Boliling Point Determination

If only small amounts of liquid material are available for boiling point determination, then micro boiling point apparatus based on Thiele tube should be used. The Thiele tube is a glass tube designed to contain heating oil and a thermometer to which a micro test tube containing the boiling point sample is attached. The shape of the Thiele tube allows for formation of convection currents in the oil when it is heated. These currents maintain a fairly uniform temperature distribution throughout the oil in the tube. The side arm of the tube is designed to generate these convection currents and thus transfer the heat from the flame evenly and

rapidly throughout the heating oil. Don't clamp the Thiele tube too tightly otherwise it might crack as it expands as it is heated, yet of course it needs to be secure.

The sample liquid is introduced by Pasteur pipette into a micro test tube with 0.5 cm in diameter (no more than 0.5 mL, which is about 10 mm depth in the small test tube), and a piece of melting point capillary tubing (sealed at one end) is dropped in with the open end down. The micro test tube assembly is then attached to a thermometer with thread, a rubber band or a thin slice of rubber tubing as shown in Figure 2.4. The whole unit is then placed in a Thiele tube.

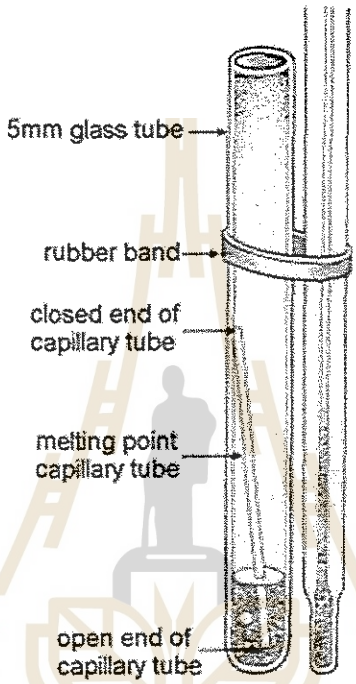


Figure 2.4 Arrangement of micro test tube and thermometer for boiling point determination

The placement of the test tube/thermometer unit in the Thiele tube is important:

i.. the micro test tube should be on the same side of the Thiele tube as the elbow as shown in Figure 2.5

ii. neither the test tube nor the thermometer bulb should be touching the glass walls of the Thiele tube

iii. the base of the micro test tube should be just below the joint to the upper part of the elbow (see Figure 2.5)

iv. the rubber band should be placed well above the level of the oil in the Thiele tube, and

v. the oil level should be just above the top of the top elbow joint. If the rubber band enters the oil, the band may soften and break in the hot oil allowing the micro test tube to fall into the oil. When positioning the band, one should bear in mind that the oil will expand when it is heated.

Once the setup has been complete, the lower part of the side arm of the Thiele tube is carefully heated with a small flame from the Bunsen burner moving the flame back and forth

along the arm. During the heating, there is an initial stream of bubbles as air is expelled and then, a little later, a rapid and continuous stream of bubbles emerges from the inverted capillary tube. At this point stop heating. Soon the stream of bubbles will slow down and stop. When they stop, the liquid sample will be drawn up in to the capillary tube. The moment when the bubbles stop and the liquid enters the capillary corresponds to the boiling point of the liquid, and the temperatures at both points should be recorded.

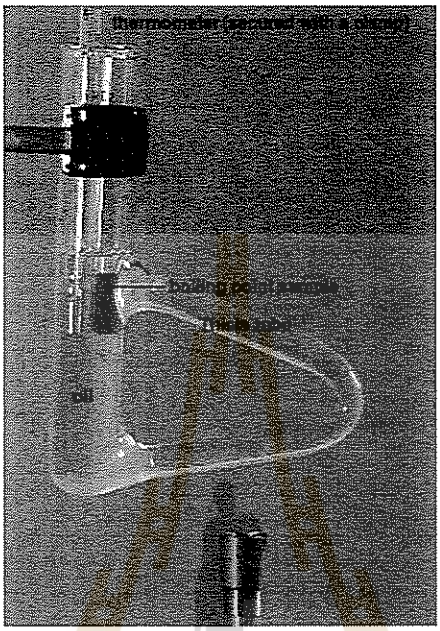


Figure 2.5 Setup for micro-boiling point determination

The explanation of this method is a reasonably simple one. During the initial heating, the air trapped in the capillary tube expands and leaves the tube and vapor from the liquid also enters the tube. There is always vapor in equilibrium with a heated liquid. This gives rise to the initial stream of bubbles. When the temperature reaches the boiling point, the vapor pressure inside the capillary tube equals the atmospheric pressure. As the temperature rises just above the boiling point then the vapor will start to escape: the second set of bubbles. Once the heating is stopped, the only vapor left in the capillary comes from the heated liquid which seals its open end. As the liquid cools, its vapor pressure will decrease and when the vapor pressure drops just below atmospheric pressure, the liquid will be drawn into the capillary tube (forced there by the higher atmospheric pressure).

Two problems are common to this method. The first arises when the liquid sample in the micro test tube is heated so strongly that it evaporates or is boiled away. Once the oil has cooled, add more liquid sample to the micro test tube with a new capillary tube and resume heating but more gently. The second arises when the liquid is not heated above its boiling point. If the heating is stopped at any point below the boiling point of the liquid, the liquid will enter the tube immediately. It will enter the tube because the trapped vapor will have a pressure less than that of the atmosphere.

Experimental

You will first practice the boiling point technique on distilled water (bp. 100 °C at 760 mmHg) using micro method. You will then determine the boiling points of water impure with ethanol, acetone and sodium chloride to study the effect of each impurity on the boiling point. Next, you will be given an unknown from Table 2.1. By determining its boiling point, you will be able to identify your unknown.

Experimental Procedure

A. Boiling Point of Pure Water

1. Firmly attach a micro test tube to a thermometer with thread.
2. Obtain a sample of distilled water from a bottle.
3. Add a small volume of distilled water into the micro test tube (about 1 cm depth) using Pasteur pipette.
4. Drop a capillary tube (sealed at one end) with the open end down into the water in the micro test tube.
5. Place the entire assembly in a Thiele tube already filled with paraffin oil up to the level slightly above the top of the top elbow joint by securing both Thiele tube and thermometer with clamps.
6. Carefully heat the lower part of the side arm of the Thiele tube with a small flame from a Bunsen burner moving the flame back and forth along the arm.
7. As the temperature is slowly increased, a rapid and continuous stream of bubbles from the inverted capillary tube begins. Continue heating for about 5-10 seconds to be sure that all of the air has been expelled from the capillary, and the vapor of the water remains in the capillary. Stop heating, but do not take the assembly out of the oil bath.
8. Carefully watch the capillary. Bubbles continue to be seen until the pressure exerted by the vapor of the water becomes equal to the atmospheric pressure. As the temperature decreases, the bubbles will slow down and stop and, at some point, the water will rise into the capillary. Read the thermometer and record the temperature at which the bubbles stop and at which the water begins to be drawn into the capillary tube. This span of temperature is the boiling point range of the water.
9. Don't forget to apply a boiling point correction due to the reduced atmospheric pressure in Korat.

B. Boiling Point of Impure Water with Ethanol

1. Obtain a small volume of a water-ethanol mixture (4:1 by volume) from a bottle.
2. Determine the boiling point of the mixture as done in section *A* using a new micro test tube and a new capillary tube. Be sure to allow the oil bath to cool at least 15-20 °C below the suspected boiling point before starting your experiment.

C. Boiling Point of Impure Water with Acetone

1. Obtain a small volume of a water-acetone mixture (4:1 by volume) from a bottle.
2. Determine the boiling point of the mixture as done in section *B*.

D. Boiling Point of Impure Water with Sodium Chloride

1. Dissolve 1 spatula measure of sodium chloride in 2 mL of distilled water obtained from a bottle.
2. Determine the boiling point of the mixture as done in section *B*.

E. Identification of an Unknown

1. Obtain a sample from your instructor. Record sample number.
2. Determine the boiling point of the sample as done in section *A* using a new micro test tube and a new capillary tube. Be sure to allow the oil bath to cool at least 15-20 °C below the suspected boiling point before starting your experiment. To ensure accuracy, you should carry out at least two separate boiling point determinations on your unknown sample. Each time you perform the procedure, you must use a new capillary tube.
3. Based on the observed boiling point, select one compound in Table 2.1 below that is most likely to be your unknown.

Table 2.1 List of compounds and their normal boiling points

Compound	Normal boiling point (°C)
acetone	55.8-56.3
methanol	64.0-65.0
ethyl acetate	76.9-77.4
ethanol	78.3-78.8
cyclohexane	80.2-81.2
n-propyl alcohol	96.0-98.0
toluene	110.1-111.1

Laboratory Safety Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. Unknown may be flammable, toxic, and irritating. Prevent contact with your eyes, skin, and clothing.
3. Capillary tubes are fragile and easily broken.
4. Before using a Bunsen burner, make sure all flammable materials are removed from the area around the Bunsen burner.
5. Take care to avoid burning yourself when using paraffin oil and a Bunsen burner.
6. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 2

Experiment 3	Recrystallization

Objectives

1. To purify phthalic acid by recrystallization.
2. To be able to select an appropriate recrystallizing solvent.

Introduction

When organic substances are synthesized in the laboratory or isolated from plants, they will obviously contain impurities. Several techniques for purifying these compounds have been developed. The most common method of purifying solid organic compounds is by recrystallization. In this technique, an impure solid compound is dissolved in a solvent and then allowed to slowly crystallize out as the solution cools. As the compound crystallizes from the solution, the molecules of the other compounds dissolved in solution are excluded from the growing crystal lattice, giving a pure solid.

Crystallization of a solid is not the same as precipitation of a solid. In crystallization, there is a slow, selective formation of the crystal framework resulting in a pure compound. In precipitation, there is a rapid formation of a solid from a solution that usually produces an amorphous solid containing many trapped impurities within the solid's crystal framework. For this reason, experimental procedures that produce a solid product by precipitation always include a final recrystallization step to give the pure compound.

The process of recrystallization relies on the property that for most compounds, as the temperature of a solvent increases, the solubility of the compound in that solvent also increases. For example, much more table sugar can be dissolved in very hot water (just below the boiling point) than in water at room temperature. What will happen if a concentrated solution of hot water and sugar is allowed to cool to room temperature? As the temperature of the solution decreases, the solubility of the sugar in the water also decreases, and the sugar molecules will begin to crystallize out of the solution. This is the basic process that goes on in the recrystallization of a solid.

To understand the recrystallization process, solubility behavior must first be considered. It is often stated that "like dissolves like". More correctly, it should be stated that, "compounds having similar structural features will be soluble in one another". Some obvious structural features that may affect solubility include polarity and the ability to hydrogen bond. For example, a compound having just a few carbons and an alcohol functional group would be expected to be soluble in solvents that have a few carbons and an alcohol functional group or in some other polar solvents, and to be less soluble in nonpolar solvents. Conversely an alkane would be expected to show the opposite solubility behavior. In most cases though it is not as this. If for example a compound has lots of carbons and hydrogens (> 6 C's) and just one alcohol group, the solubility will be dominated more by the alkyl part of the molecule than by the alcohol part, and the compound will show a solubility behavior more like that of an alkane. For known compounds, it is useful to consider the structure of the compound

when choosing a recrystallization solvent. An educated guess can save some time. Usually however, the structure of a compound may not be known so the solvent must be chosen by carrying out solubility tests.

The second part of this experiment involves carrying out solubility tests on known compounds. A compound usually exhibits one of three general solubility behaviors as shown in Figure 3.1: (1), the compound has a high solubility in both hot and cold solvent, (2), the compound has a low solubility in both hot and cold solvent, and (3), the compound has a high solubility in hot solvent and a low solubility in cold solvent. Solvents which exhibit the first two behaviors are not useful for recrystallizing a compound. A solvent showing the third behavior, that is, high solubility at high temperatures and low solubility at low temperatures, is one that is suitable for use as a recrystallization solvent.

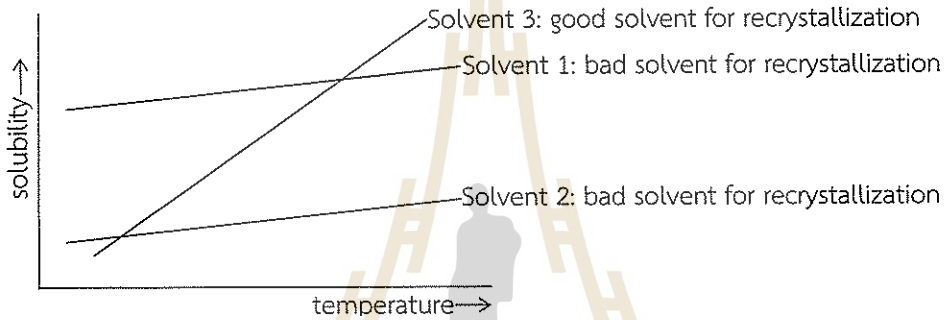
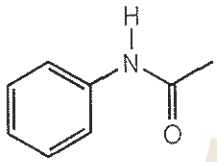


Figure 3.1 Three general solubility behaviors of solvents

Consider the three different types of impurities that may be present in a sample: soluble, insoluble, and colored. In theory, insoluble impurities can be removed from a compound fairly easily. The compound is dissolved in a solvent, the solution is filtered to remove the insoluble impurities, and the solvent evaporated to produce the solid compound. The insoluble impurities are left behind in the filter paper. Colored impurities can be removed in a similar way but with an additional step. The solid is dissolved in a solvent, activated charcoal is added, the solution is filtered as before, and the solvent is evaporated to produce the solid compound. The charcoal, which has adsorbed the colored impurities, is left behind in the filter paper. The third type of impurities, the soluble impurity, cannot be filtered out because it has solubility characteristics similar to those of the desired compound (hence the name soluble impurity). To remove soluble impurities, first, by doing solubility tests, a suitable solvent is chosen (high solubility in hot solvent, low solubility in cold solvent). The soluble impurities are then removed as follows: the desired compound along with the soluble impurities are dissolved in a minimum of near-boiling solvent. The solution is then allowed to cool slowly and without interruption. As the solution cools, the solubility of the compound (and of the soluble impurities) decreases, the solution becomes saturated with the desired compound, and the compound begins to crystallize. Because formation of crystals is a highly selective process that usually excludes foreign molecules, only crystals of the desired compound form. Because the soluble impurities are present in smaller amounts, the solution

never becomes saturated with the impurities, so the impurities remain in solution even after the solution has cooled. Removing the solution from the crystals thus removes the solvent and the soluble impurities from the desired crystals. A final rinse with a minimum of ice-cold solvent followed by its removal cleans off any residual soluble impurities clinging to the surface of the desired crystals. After allowing the solvent to evaporate, pure crystals of desired compound should remain. The weight and usually the melting point of the crystals would be determined and included in the report along with the percent recovery.

In practice, by following a set procedure, the same solvent is used throughout the whole recrystallization process, and the impurities are removed one by one. Note that in any recrystallization, some of the desired product is sacrificed and the recovery will be less than 100%. This is because even at the lower temperatures, the desired compound has some finite solubility in the recrystallization solvent and is thus lost when solvent and soluble impurities are removed. To illustrate this, look at the solubility of acetanilide in water.



Acetanilide

Solubility in 100 mL water:
0.5 g at 10 °C and 5 g at 99 °C

If 5 g were dissolved in 100 mL of water at 99 °C and then allowed to cool to 10 °C, 4.5 g would crystallize out and 0.5 g would remain in solution. The recovery then would be 90%. The 10% remaining in solution would be lost.

The Steps in the Recrystallization of a Compound

For our recrystallizations you may assume that insoluble and soluble impurities are present. The seven steps used here to recrystallize a compound are, (1) carry out solubility tests to determine a suitable solvent; (2) dissolve the solute in a minimum of near-boiling solvent; (3) remove any insoluble impurities by hot filtration; (4) allow the solution to cool slowly and undisturbed to room temperature then possibly to ice temperature; (5) collect the crystals by filtration; (6) rinse the crystals with a minimum amount of ice-cold solvent; and (7) allow the crystals to dry.

Choosing a Solvent

The choice of solvent is perhaps the most critical step in the process of recrystallization since the correct solvent must be selected to form a product of high purity and in good recovery or yield. Consequently a solvent should satisfy certain criteria for use in recrystallization.

(a) The desired compound should be reasonably soluble in the hot solvent, about 5 g/100 mL (5 mg/100 μL) being satisfactory, and insoluble or nearly insoluble in the cold solvent. Note that the reference temperature for determination of the solubility in "cold" solvent is often taken to be room temperature. This combination of solute and solvent will allow dissolution to occur in an amount of solvent that is not unduly large and will also permit recovery of the purified product in high yield. A solvent having this type of solubility

properties as a function of temperature would be said to have a favorable temperature coefficient for the desired solute.

(b) Conversely, the impurities should either be insoluble in the solvent at all temperatures or must remain at least moderately soluble in the cold solvent. In other words, if the impurities are soluble, the temperature coefficient for them must be unfavorable; otherwise the desired product and the impurities would both crystallize simultaneously from solution.

(c) The boiling point of the solvent should be low enough so that it can readily be removed from the crystals.

(d) The boiling point of the solvent should generally be lower than the melting point of the solid being purified.

(e) The solvent should not react chemically with the substance being purified.

The solvents commonly used in recrystallizations range widely in polarity, a property measured by the dielectric constants (ε) listed in Table 3.1. Those solvents with dielectric constants in the range of 2-3 are considered nonpolar, and those with constants above 10 as polar. Solvents in the 3-10 range are of intermediate polarity. Of the various solvents listed, petroleum ether deserves special mention because of its confusing common name. This solvent does not contain the ether functional group at all; rather it is a mixture of volatile aliphatic hydrocarbons, of which pentane is a chief component, obtained from the refining of petroleum. The composition and boiling point of the mixture depends on the particular distillation "cut" obtained. Thus, the boiling point range of this type of solvent is usually given, as in the description, "petroleum ether, b.p. 60-80 °C (760 mmHg)."

Table 3.1 Common recrystallization solvents

Solvent	Dielectric constant (ε)	Boiling point (°C, 760 torr)	Freezing point (°C)	Density (g/mL)	Water soluble	Flammable
petroleum ether	1.9	30-80			No	Yes
pentane	1.84	36	−130	0.63	No	Yes
hexane	1.89	69	−95	0.66	No	Yes
heptane	1.9	98	−91	0.68	No	Yes
ligroin	1.9	80-120			No	Yes
heptane	1.9	98	−91	0.68	No	Yes
octane	1.9	126	−57	0.70	No	Yes
nonane	1.9	151	−51	0.72	No	Yes
cyclohexane	1.965	80.7	6.6	0.78	No	Yes
1,4-dioxane	2.21	101	11.8	1.03	Yes	Yes
toluene	2.38	110.6	−95	0.87	No	Yes
diethyl ether	4.34	34.5	−116	0.71	Slightly	Yes
chloroform	4.81	61.7	−63.5	1.48	No	No
ethyl acetate	6.02	77.1	−84	0.90	Yes	Yes
tetrahydrofuran	7.60	65.6	−109	0.89	No	Yes
dichloromethane	9.08	40.6	−95	1.33	No	No
acetone	20.7	56	−95	0.79	Yes	Yes
ethanol (95%)	24.6	78	−117	0.79	Yes	Yes
methanol	32.6	65	−94	0.79	Yes	Yes
water	78.5	100	0	1.00	n/a	No

The chemical literature is a valuable source of information about solvents suitable for recrystallizing known compound. If the compound has not been prepared before, it is necessary to resort to trial-and-error techniques to find an appropriate solvent for recrystallization. First, test the solubility of tiny samples of the compound in test tubes with a variety of different solvents (water, ethanol, methanol, ethyl acetate, diethyl ether, hexane, toluene, etc.) at room temperature. If the compound dissolves in the solvent at room temperature, then that solvent is unsuitable for recrystallization. If the compound is insoluble in the solvent at room temperature, then the mixture is heated to the solvent's boiling point to determine if the solid will dissolve at high temperature, and then cooled to see whether it crystallizes from the solution at room temperature.

Sometimes no single satisfactory solvent can be found, so mixed solvents are used. The mixture is usually comprised of only two solvents; one of these dissolves the solute even when cold and the other one does not, and the two solvents must be miscible with each other. Some commonly used solvent pairs are listed in Table 3.2.

Table 3.2 Common solvent pairs

Solvent 1 (more polar)		Solvent 2 (less polar)	
Solvent	Dielectric constant (ε)	Solvent	Dielectric constant (ε)
water	78.3	ethanol	24.3
water	78.3	acetone	20.7
methanol	32.6	dichloromethane	9.08
ethanol	24.3	acetone	20.7
acetone	20.7	diethyl ether	4.34
acetone	20.7	ligroin	1.9
diethyl ether	4.34	hexane	1.89
ethyl acetate	6.02	cyclohexane	1.965
ethyl acetate	6.02	ligroin	1.9
dichloromethane	9.08	ligroin	1.9
toluene	2.38	ligroin	1.9

Dissolving the Solid

Once a suitable solvent is selected, the solid is weighed and placed in an Erlenmeyer flask, with a few crystals of impure material being retained as "seeds" to induce crystallization. (Erlenmeyer flasks are preferred over beakers for recrystallization because the conical shape of an Erlenmeyer flask decreases the amount of solvent lost to evaporation during heating, prevents the formation of a crust around the sides of the glass, and makes it easier to swirl the hot solution while dissolving the solid without splashing it out of the flask.) A boiling stone should be added to prevent bumping of the solution upon boiling. Alternatively, smooth boiling can be promoted by using a wooden boiling stick.

A small volume of solvent is added, and the mixture is heated on a hot plate or in a water bath on a hot plate (never heat organic solvents with a Bunsen burner) to the boiling point. Additional solvent, as needed, is added dropwise using a Pasteur pipette to the boiling mixture until the solid just dissolves. It is important to let boiling resume after each addition so that a

minimum amount of solvent is used to effect dissolution; using excessive amounts of solvent decreases the recovery of the solute. If it is necessary to perform a hot filtration, it is prudent to add an additional 2-5% of solvent to prevent premature crystallization during this operation.

If adding solvent fails to dissolve any more solid, it is likely that insoluble impurities are present. These can be removed by hot filtration. Thus, to avoid using too much solvent and risking poor recovery of the purified solute, you should observe the dissolution process carefully. This is particularly important when only a relatively small quantity of solid remains, as this may be the material that is insoluble.

The same general approach used for single-solvent dissolution is followed when mixed solvents are employed. However, there are two options for effecting dissolution. In one, the solid to be purified is first dissolved in a minimum volume of the hot solvent in which it is soluble; the second solvent is then added to the boiling solution until it turns cloudy. The cloudiness signals initial formation of crystals, caused by the fact that addition of the second solvent results in a solvent mixture in which the solute is less soluble. Finally, more of the first solvent is added dropwise until the solution clears.

Two further aspects of this option should be noted. First, the solution must be cooled slightly below the lower boiling point before the second solvent is added if this solvent has a boiling point lower than the first; otherwise the addition of this solvent could cause sudden and vigorous boiling of the mixture and hot solvent might spew from the apparatus. Second, hot filtration should be performed if needed before addition of the second solvent; this will prevent crystallization during the filtration step. A potential disadvantage of this method for mixed solvent recrystallization is that unduly large volumes of the second solvent may be required if excessive amounts of the initial solvent have been used.

In the second option, the solute is added to the solvent in which it is insoluble, and the mixture is heated near the boiling point of the solvent; the second solvent is then added in small portions until the solid just dissolves. As with recrystallization from a single solvent, it is generally wise to add 3-4% of additional solvent to prevent premature crystallization of the solute during hot filtration, if this step is necessary. The use of this approach to mixed solvent recrystallization also has the disadvantage that using too much of the first solvent will require the addition of undesirably large volumes of the second solvent.

Using Decolorizing Carbon

Colored impurities are sometimes difficult to remove from solid mixtures. These colored impurities, often due to the presence of polar or polymeric compounds, can cause a colorless organic solid to have a tint of color even after recrystallization. Decolorizing or activated carbon is used to remove the colored impurities from the sample. Decolorizing carbon is very finely divided carbon that provides high surface area to adsorb the colored impurities.

Very little decolorizing carbon is needed to remove the colored impurities from a solution. You must be judicious in your use of decolorizing carbon: if too much is used, it can adsorb the desired compound from the solution as well as the colored impurities. After the impure

solid sample is dissolved in hot solvent, a small amount of decolorizing carbon, about the size of a pea, is added to the hot but not boiling solution. This must be done carefully to avoid a surge of boiling from the hot solution. The solution is stirred and heated for a few minutes and then filtered hot to remove the decolorizing carbon. The resulting filtrate should be colorless and the recrystallization process continues as before.

Hot Filtration

Sometimes during a gravity filtration, crystals can start to grow in the filter funnel and may block the funnel, stopping filtration. This problem can be avoided by using a hot filtration where the whole filtration apparatus is heated in order to prevent the solution from cooling significantly.

The hot filtration process is best carried out using a fluted filter paper and a short-stemmed or stemless glass funnel as shown in Figure 3.2. Support the glass funnel on an iron ring clamp if it is at all insecure. If pressure builds up in the flask due to the presence of hot vapor, the filtering action will slow down or even stop. The use of the fluted paper reduces this problem. The hot solution should be filtered quickly through the fluted paper in a glass funnel into an Erlenmeyer flask. Alternatively, a small piece of wire or paper may be inserted between a funnel and an Erlenmeyer flask to prevent formation of a seal and subsequent pressure build-up. If crystals start to form in the paper or in the filter funnel, then the Erlenmeyer flask containing a few drops of pure solvent with the funnel and paper in place should be heated on a steam bath or a hot plate and the entire filtration procedure carried out on the steam bath or the hot plate. Hot solvent may be added to remove any crystals appearing in the filter paper.

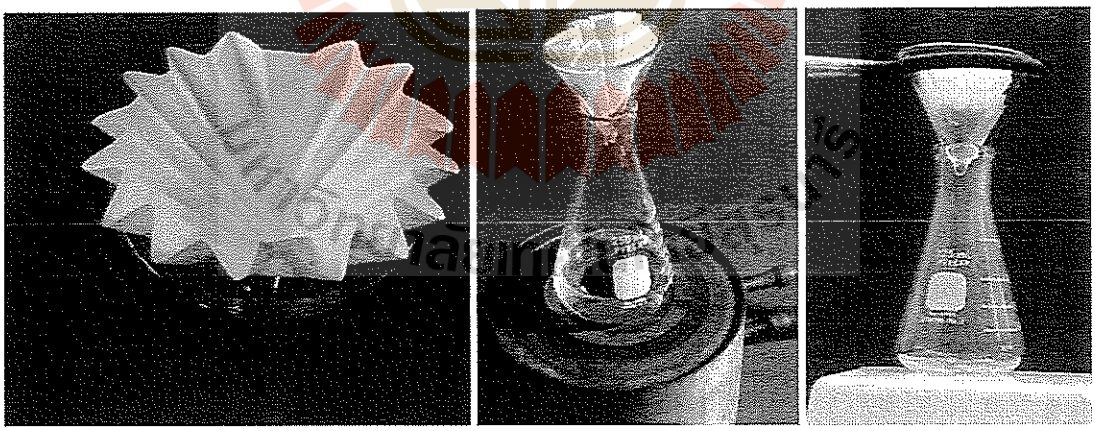


Figure 3.2 Setup for hot filtration

Do not use metal tongs or a test-tube holder to hold the hot Erlenmeyer flask while pouring the liquid since it is too easy to drop the flask. A better convenient simple holder may be fashioned from a piece of paper towel folded into a strip and pinched around the neck of the flask.

Crystallizing the Solid

After the insoluble impurities have been removed, cover the flask containing the hot filtrate with a watch glass and set it aside undisturbed to cool slowly to room temperature. As the solution cools, the solubility of the dissolved compound will decrease and the solid will begin to crystallize from the solution. After the flask has cooled to room temperature, it may be placed in an ice bath to increase the yield of solid. Do not rapidly cool the hot solution by placing the flask in an ice bath before it has cooled to room temperature. This is because the crystals formed tend to be very small, and their resulting large surface area may foster adsorption of impurities from solution. In this sense, the crystals are functioning like decolorizing carbon. Generally the solution should not be disturbed as it cools since this also leads to production of small crystals. The formation of crystals larger than about 2 mm should be avoided because some of the solution may become occluded or trapped within the crystals. The drying of such crystals is more difficult, and impurities may be left in them. Should overly large crystals begin to form, brief, gentle agitation of the solution normally induces production of smaller crystals.

Sometimes the dissolved compound fails to crystallize from the solution on cooling. It may be due to a condition known as supersaturation. If this happens, crystallization can be induced by various methods. One way to induce crystallization is by scratching the inner wall of the Erlenmeyer flask with a glass stirring rod. This is believed to release very small particles of glass which act as nuclei for crystal growth. Another method of inducing crystallization is to add a small crystal of the desired compound, called a seed crystal, to the solution, assuming a small amount of solid has been saved. Again, this seed crystal acts as a template on which the dissolved solid will begin crystallizing. If no solid is available and a volatile solvent is being used, it is sometimes possible to produce a seed crystal by immersing the tip of a glass stirring rod or metal spatula in the solution, withdrawing it, and allowing the solvent to evaporate. The crystals that form on the end of the rod or spatula are then reinserted into the solution to initiate crystallization. If neither of these two techniques results in crystallization, the compound was probably dissolved in too much hot solvent. If you believe that you may have too much solvent for the amount of dissolved compound, reheat the solution to boiling, boil off or distill some of the solvent, and then allow the solution to cool to room temperature again to effect crystallization.

Isolating the Solid by Suction Filtration

Once the compound has completely precipitated from the solution, it is separated from the remaining solution (also called the mother liquor) by filtration. Typically this is done by vacuum or suction filtration using a Büchner funnel as shown in Figure 3.3. Line the bottom of the Büchner funnel with a piece of filter paper that is large enough to cover the holes in the bottom plate of the funnel without curling up on the sides of the funnel. Place a neoprene adapter on the stem of the funnel and insert it in the top of a suction flask (a thick-walled Erlenmeyer flask with a side-arm) that has been securely clamped to a ring stand.

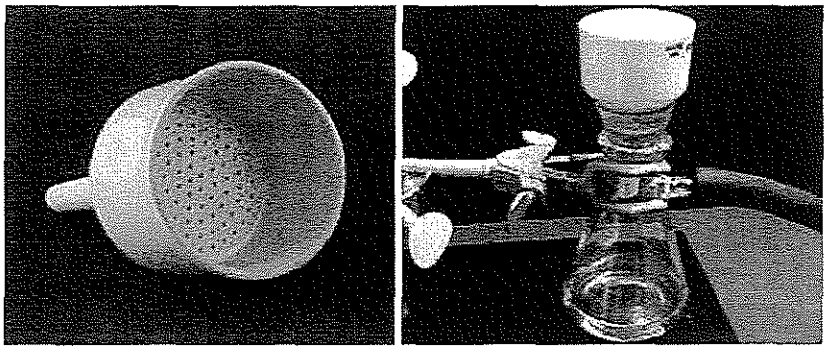


Figure 3.3 Setup for suction filtration

Using a piece of thick-walled vacuum tubing, connect the side-arm of the suction flask to a water aspirator. Turn the water to the aspirator on full force to create a vacuum through the system. If necessary, carefully adjust the piece of filter paper so that it covers all of the holes in the funnel, and then dampen it with a small volume of cold solvent; this will create a better seal between the filter paper and the plate in the funnel, preventing any solid from getting under the filter paper and passing through the funnel. Slowly pour the recrystallization solution into the funnel and allow the suction to pull the mother liquor through. Rinse the Erlenmeyer flask with a small volume of cold recrystallization solvent to remove any remaining solid. Add this solvent to the funnel and then wash the solid in the funnel, called the filter cake or residue, with a few milliliters of fresh, cold recrystallization solvent to remove any remaining mother liquor and dissolved impurities.

Leave the aspirator on for a few minutes and allow air to pass through the crystals to dry them. After pulling air through the crystals for a brief time, remove the vacuum from the system by disconnecting the vacuum tubing from the aspirator before turning the water off. If you turn the aspirator water off first, water can be sucked into the filter flask and may contaminate the product. The filter cake is removed from the funnel by carefully prying it from the filter using a spatula. The cake of crystals will still be slightly wet with solvent and should be allowed to dry thoroughly before measuring the weight or melting point of the solid material.

What Can Go Wrong in a Recrystallization?

Care must be taken at each step to obtain good results, which can be measured by % recovery and purity of the desired compound. Recrystallizations require careful technique and practice. Some loss, resulting from transferring solids from one container to another and leaving a little material behind, cannot be avoided. Such losses should however be minimized as much as possible by using careful technique. Also, because of the finite solubility of the solid in the recrystallization solvent, even at low temperature, any unnecessary prolonged contact with recrystallization solvent, especially if the solvent is not ice-cold will result in loss of product. For that reason, the following problems commonly occur: if too much solvent is added in the recrystallization, a poor or no yield of crystals will result. If the solid is dissolved below the boiling point of the solution, too much solvent will be needed, resulting in a poor yield. If too

much rinse solvent or rinse solvent that is not chilled to ice temperature is used, some of the product will be redissolved and lost. In summary, to obtain optimal results, a minimum of near-boiling solvent should be used for the recrystallization, and a minimum of ice-cold solvent should be used for the rinse.

Occasionally the solute will separate from solution as an oil rather than a solid. This type of separation, which is sometimes called oiling out, is undesirable for purification of solutes because the oils usually contain significant amounts of impurities. Two general approaches are helpful in solving this problem. (1) Oil may persist on cooling with no evidence of crystallization. These may often be induced to crystallize by scratching the oil against the side of the flask with a glass stirring rod at the interface of the oil and the solution. If this fails, several small seed crystals of the original solid may be added to the oil, and the mixture allowed to stand for a period of time. Failure of these, alternatives may necessitate separation of the oil from the solution and crystallization of it from another solvent. (2) Oils may form from the hot solution and then solidify to an amorphous mass at lower temperatures; in the meantime, crystals of the solute may precipitate from the mother liquor. Because the oil is not a pure liquid, the solid mass produced from it will be impure. In a case such this, the usual remedy is to reheat the entire mixture to affect dissolution, add a few milliliters of additional pure solvent, and allow the resulting solution to cool.

The Theoretical Percent Recovery from Recrystallization

The following formulas are used in recrystallization problems.

% lost in cold solvent = (solubility in cold solvent / solubility in hot solvent) x 100

% recovery of solid = [g (solid) – g (solid lost)] x 100 / g (solid)

Example 3.1 The solubility of solid "X" in hot water (5.50 g/100 mL at 100 °C) is not very great, and its solubility in cold water (0.53 g/100 mL at 0 °C) is significant. What would be the maximum theoretical percent recovery from recrystallization of 5.00 g of solid "X" from 100 mL water? Assuming the solution is chilled at 0 °C.

percent solid lost in cold water = (solubility in cold water / solubility in hot water) x 100
= (0.53/5.50) x 100 = 9.64 %

grams solid lost in cold water = grams of original solid x percent lost
= 5.00 g x 9.64% = 0.482 g

g (solid recovered) = g (solid) – g (solid lost) = 5.00 – 0.482 = 4.52 g

% recovery = g (solid recovered) x 100 / g (solid)
= (4.52/5.00) x 100 = 90.4 %

Example 3.2 The solubility of compound "X" in ethanol is 0.80 g per 100 mL at 0 °C and 5.00 g per 100 mL at 78 °C. What is the minimum amount of ethanol needed to recrystallize a 12.00 g sample of compound "X"? How much would be lost in the recrystallization, that is, would remain in the cold solvent?

amount of ethanol needed at 78 °C = (12.00 g)(100 mL / 5.00 g) = 240 mL

amount of sample remaining in the cold solvent at 0 °C = (240 mL)(0.80 g / 100 mL)

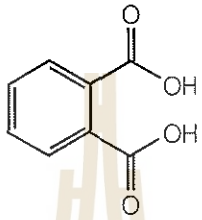
= 1.9 g

or % lost = (0.80/5.00) x 100 = 16 %

amount of sample remaining in the cold solvent at 0 °C = 12.00 x 16% = 1.9 g

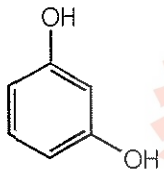
Experimental

In the first part of the experiment, you will be given an impure sample of phthalic acid that you will purify by recrystallization. Phthalic acid is very soluble in boiling water, 18 g/100 mL, and is much less soluble in chilled (14 °C) water, 0.54 g/100 mL and in ice-cold (0 °C) water, 0.30 g/100 mL. Thus, water is a very good recrystallization solvent for phthalic acid.

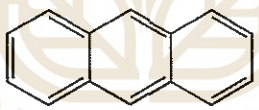


Phthalic acid

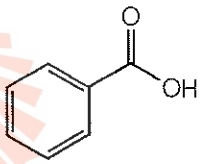
In the second part of the experiment, you will test the solubility properties of resorcinol, anthracene and benzoic acid in three different solvents: water, toluene and petroleum ether. Knowing the solubility properties, you will be able to draw some conclusions regarding an appropriate recrystallization solvent or solvents for each compound. You should perform these tests while your phthalic acid is crystallizing.



Resorcinol



Anthracene



Benzoic acid

Experimental Procedure

A. Recrystallization of an Impure Phthalic Acid

1. Flute a filter paper. First, as shown in Figure 3.4, fold in half and fold in half again; open and make new folds bisecting the previous folds until a fan-like arrangement is obtained. Pleat into a fan by folding each segment in the opposite direction to its neighbor, in accordion-like fashion. When opened out, the fan-like fluted paper results. Fluting the filter paper maximizes the rate at which the liquid may flow through the filter paper by increasing the surface area and by allowing air to enter the flask along its sides to permit rapid pressure equalization.

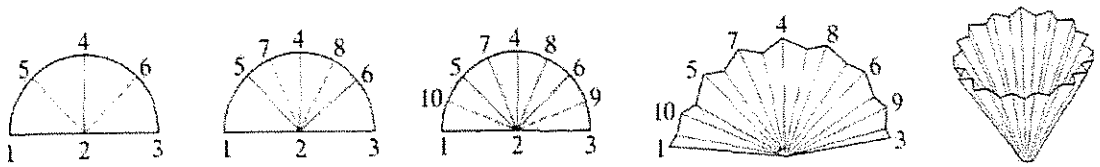


Figure 3.4 Folding a fluted filter paper

2. Set up a short-stemmed or stemless glass funnel in a small metal ring clamped to a ring stand above a hot plate. Fit the glass funnel with a piece of fluted filter paper (see Figure 3.2). Add 10 mL of distilled water in a 50 mL Erlenmeyer flask and place the flask on the hot plate, beneath the stem of the funnel. Cover the funnel with a watch glass and heat the water to boiling until the filter paper is wet. This step is to preheat the funnel and the filter paper to prevent the phthalic acid from crystallizing in the funnel upon gravity filtration.
3. Set up a 50 mL beaker, about three-quarters filled with distilled water, on the hot plate. Cover the beaker with a watch glass and heat the water to boiling. This water will be used later to remove any phthalic acid crystals appearing in the filter paper during hot filtration.
4. Calculate the volume of boiling water required to dissolve 1.0 g of phthalic acid (use the data given above).
5. Obtain a sample of impure phthalic acid for recrystallization. The phthalic acid has been contaminated with charcoal, which is not soluble under either temperature condition. Place 1 g to of phthalic acid sample (to a precision of the fourth decimal place) in a 50 mL Erlenmeyer flask.
6. Place 10 mL of water in a graduated cylinder. Add 4-5 mL of the water to the solid in the flask along with 2-3 pieces of boiling chips, and then begin heating the mixture on a hot plate.
7. As soon as the water begins to boil, continue to add water dropwise from the graduated cylinder until the solid completely dissolves. If 10 mL of water has been added and the solid still has not completely dissolved, refill the graduated cylinder, and continue to add water, but at a faster rate. Wear double-layer cloth gloves to protect your hands. Take the flask off the hot plate. Cover the flask with a watch glass. Record the total volume of water required to dissolve the crystals.
8. Skip the decoloration step since the resulting solution is colorless.
9. When the preheated flask and funnel are ready, wear double-layer cloth gloves to protect your hands. Reheat the flask containing the phthalic acid mixture on the hot plate until the mixture just begins to boil again. Swirl the mixture to make sure that the phthalic acid dissolves to the greatest extent possible.
10. Discard any boiling water remaining in the preheated flask and place it back on the hot plate. Use a glass stirring rod as a guide for running the hot mixture of phthalic acid into the preheated funnel. Do not fill the funnel more than half-full at a time, to prevent the solution being lost over the rim of the filter paper. If any crystals appear in the filter paper, add a small amount of hot water prepared in the beaker to dissolve them.
11. Rinse the flask with hot water and pour it onto the filter paper. If excessive amount of water is added, boil off the solution until the volume is reduced to the original volume in step 9.
12. Take the flask with the filtrate off the hot plate and allow the filtrate to cool undisturbed to room temperature. During this slow cooling, crystals of the solid will form. When the flask has reached room temperature, place it in an ice bath for at least 10 minutes.

13. Set up a vacuum filtration apparatus (see Figure 3.3 by using a water pump instead of a water aspirator). Place a disk of filter paper in the Büchner funnel and collect the crystals by vacuum filtration. You may use a spatula to assist in transferring the crystals to the funnel, then wash the phthalic acid in the funnel with small amounts of ice cold water.
14. Transfer the crystals along with the filter paper from the funnel to a watch glass, place another piece of filter paper on top of the crystals and press firmly to remove some of the water. Leave them in a desiccator to dry until your next lab period. The filtrate in the suction flask may be poured into the sink.
15. You will obtain the weight of your recovered product next week. Calculate the percent recovered using the following written formula.

$$\% \text{ Recovery} = \frac{\text{weight of phthalic acid obtained after recrystallization}}{\text{weight of phthalic acid before recrystallization}} \times 100$$

16. Submit product in a properly labeled container.

B. Solubility Tests

1. Use about 10 mg of each compound for the tests. It is not necessary to use a balance to measure out the solids accurately. Samples of about 10 mg of solid in a test tube will be on display near the compounds. Simply use an amount which approximates that. The solvent does not have to be measured exactly either. Use a disposable pipet to add solvent (20 drops per 1 mL).
2. To heat solutions in a test tube, use steam bath as the heat source and a glass stirring rod to promote smooth boiling.
3. Test the three solid compounds, resorcinol, anthracene and benzoic acid, in each of the three solvents (water, toluene and petroleum ether). The most efficient strategy is to test one compound with the three solvents before going on to the next compound.
4. Test solubilities as follows. First see if 10 mg of the compound is soluble in 0.25 mL of the solvent at room temperature. After adding solvent always flick the tube with a finger to swirl the contents and to help ensure complete mixing, and give it 30 seconds to dissolve. If the solid completely dissolves, it is considered to be very soluble at room temperature, in which case nothing more has to be done with the solid/solvent combination.
5. If the compound is insoluble at room temperature, heat the tube to the boiling point of the solution. If the compound is still insoluble at the boiling point (give it 30 seconds), it is considered to have a very low solubility at all temperatures, in which case nothing more has to be done with that solid/solvent combination.
6. If it seems that some of it has dissolved in 0.25 mL of boiling solvent, add another 0.25 mL and reheat it to see if it will all dissolve.
7. If the compound has dissolved in boiling solvent (either 0.25 or 0.5 mL), cool the tube in cold water to see if crystallization occurs. Scratching the tube with a glass stirring rod at the air-liquid interface may be necessary to induce crystallization.

8. If crystals form, redissolve the solid by heating and allow the tube to cool more slowly, without cooling in cold water. Slow cooling may allow larger crystals to form.
9. If the crystals are large enough, record the crystal form (e.g., needles, plates, prisms). The relative amount of the crystals, compared to the starting amount, should be noted.

Laboratory Safety Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. To avoid short circuiting or melting of wires, make sure that the electrical wire of the hot plate is fully uncoiled from the metal part.
3. Take care to avoid burning yourself when using the electric hot plate.
4. Use care in heating low-boiling point solvents such as petroleum ether on the steam bath. Such solvents evaporate easily.
5. Petroleum ether and toluene are flammable.
6. Never point the heating tube at yourself or at anyone else because the tube contents may become overheated and shoot out of the tube.
7. Dispose of all solutions in the ORGANIC LIQUID WASTE container in the hood.
8. Place used boiling chips into the "boiling chip" waste container, NOT into the trash.
9. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 3

Experiment 4	Extraction of Caffeine from Tea

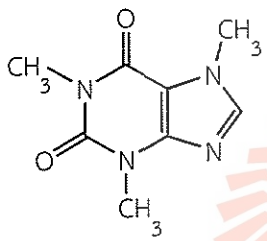
Objectives

1. To extract caffeine from tea leaves using liquid/liquid extraction technique.
2. To purify crude caffeine by sublimation.

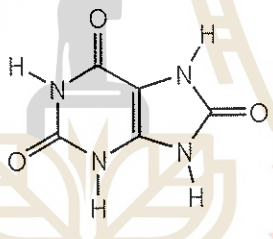
Introduction

Caffeine is a commonly encountered mild stimulant and a diuretic; it is widely used in proprietary drugs for the stimulant effect to prevent drowsiness. Caffeine is naturally present in the leaf, fruit and bark of a number of plants, including tea, coffee and cacao. Tea contains about 30-75 mg and coffee 80-125 mg in a typical 150 mL (cup) serving. The amount of caffeine in tea varies by brand but the average in common brands is typically about 30-40 mg caffeine/tea bag.

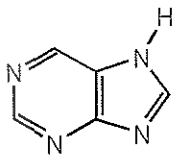
Caffeine belongs to a very important class of compounds called purine alkaloids, a major component of nucleic acids. Other purine systems are found in naturally occurring compounds, including uric acid, which is the form in which nitrogen is excreted in non-mammalian animals. The presence of methyl groups in caffeine is a result of biochemical methylation, a common process in plant metabolism.



Caffeine



Uric acid



Purine

The extraction of caffeine from coffee is commercially important as the effects of caffeine are considered undesirable by some people. Decaffeination used to be carried out by treating the green coffee beans with a small amount of hot water and then exposing to a solvent (trichloroethylene) until 97% of the caffeine was removed. Residual solvent was removed after extraction by steam distillation. The process also removes wax from the beans, which are then roasted in the usual way. In these solvent based methods, the coffee beans are extracted as many as 10 times, maybe for as long as 10 hours until the required level of decaffeination has been reached. A more common method today is to use supercritical carbon dioxide.

Tea has been consumed as a beverage for almost 2,000 years, starting in China. It is a beverage produced by steeping in freshly boiled water the young leaves and leaf buds of the tea plant, *Camellia sinensis*. Today, two principal varieties are used, the small-leaved China plant (*C. sinensis sinensis*) and the large-leaved Assam plant (*C. sinensis assamica*). Hybrids of these two varieties are also grown. The leaves may be fermented or left unfermented. Fermented

teas are referred to as black tea, unfermented teas as green tea, and partially fermented teas as oolong.

Tea leaves consist mostly of cellulose, a water-insoluble polymer of glucose, which is a simple sugar (a monosaccharide). Along with the cellulose are found a number of other compounds including caffeine, tannins (phenolic compounds, compounds that have an –OH directly bonded to an aromatic ring) and a small amount of chlorophyll.

The idea in this experiment is to extract the water soluble materials in the tea leaves into boiling water. The solubility of caffeine in water is 2.2 g/100 mL at 25 °C, 18 g/100 mL at 80 °C, and 67 g/100 mL at 100 °C. The hot solution is allowed to cool and the caffeine is then extracted from the water with dichloromethane (methylene chloride), which is an organic solvent that is insoluble in water. Since caffeine is more soluble in dichloromethane (14 g/100 mL) than it is in water (2.2 g/100 mL), it readily dissolves in the dichloromethane. However, the tannins are also slightly soluble in the dichloromethane and we want to separate the caffeine from the tannins by having the caffeine dissolve in the dichloromethane and the tannins remain in the water. We can do this by taking advantage of the fact that phenols are acidic enough to be converted to their salts (deprotonation of the –OH group) by reaction with sodium carbonate. So, we will add sodium carbonate to the water and the tannins will be converted to phenolic anions which are not soluble in the dichloromethane but are soluble in highly polar water. The dichloromethane–caffeine mixture can then be separated on the basis of the different densities of dichloromethane and water because dichloromethane is much denser than water and insoluble in it. Residual water is separated from dichloromethane by drain out the dichloromethane through a separatory funnel, thus dichloromethane passed through the funnel while water still remains in the funnel. Water and dichloromethane are slightly soluble in each other. So, after separating the solvents, residual water will remain in the organic layer. Mainly anhydrous sodium sulfate or magnesium sulfate is used for the removal of water from organic layer.

Extraction

In a chemical sense, extraction is the general term for recovery of a substance from a crude solid or a solution by bringing it into contact with a solvent that preferentially dissolves the desired material. A familiar example of the first case, so called solid/liquid extraction, is making a cup of tea or coffee – the soluble flavor and odor chemicals and caffeine are extracted from the solid tea leaves or ground coffee beans into hot water (the solvent). Insoluble plant material is left behind in the tea bag or coffee filter. An example of the second case, referred to as liquid/liquid extraction, is using an organic solvent to extract the caffeine from an aqueous tea or coffee solution, leaving the more water-soluble compounds behind in the aqueous solution.

By far liquid/liquid extraction is the most common technique used in the organic laboratory. Organic reactions often yield a number of by-products, some inorganic and some organic. Also, since they do not go to 100% completion, some starting material is often present at the end of an organic reaction. The real “work” in organic chemistry is not running the reaction, but

rather in what is aptly called the "work-up" of the reaction mixture, that is, the separation and purification of the desired product from the mixture of by-products and residual starting material. Liquid/liquid extraction is often used as the initial step in the work-up of a reaction before final purification of the product by recrystallization, distillation or sublimation.

In a technical sense, liquid/liquid extraction is based on the principle of the equilibrium distribution of a substance (solute) between two immiscible phases, one of which is usually a solvent. The solvent does not need to be a pure liquid but may be a mixture of several solvents or a solution of some chemical reagent that will react with one or more components of the mixture being extracted to form a new substance soluble in the solution. By shaking the substance together with the two liquids, it can be transferred from one liquid to the other. For example, acetanilide is partly soluble in both water and diethyl ether. If a solution of acetanilide in water is shaken with a portion of diethyl ether (which is immiscible with water), some of the acetanilide will be transferred to the ether layer. The ether layer, being less dense than water, separates out above the water layer and can be removed and replaced with another portion of ether. When this in turn is shaken with the aqueous solution, more acetanilide passes into the new ether layer. This new layer can be removed and combined with the first. By repeating this process enough times, virtually all of the acetanilide can be transferred from the water to the ether.

It has been found that when two immiscible solvents are shaken together, the solute distributes itself between them in a ratio roughly proportional to its solubility in each. The ratio of the concentration of the solute in each solvent at equilibrium (C_1/C_2) is a constant at a given temperature called the distribution coefficient or partition coefficient (K).

Let us consider the extraction of a compound whose solubilities in ether and water are 10 g/100 mL and 2 g/100 mL, respectively. If a solution of 1 g of the compound in 100 mL of water is extracted with 100 mL of ether, and the amount that transfers to the ether layer is x g, then the fraction of the compound transferred to the ether phase can be calculated as follows:

$$\text{Distribution coefficient, } K_{e/w} = \frac{C_{ether}}{C_{water}} = \frac{10 \text{ g}/100 \text{ mL}}{2 \text{ g}/100 \text{ mL}} = 5$$

If, originally, there was 1 g of compound in 100 mL water, and the amount of material in ether in g at equilibrium = x, then there must be 1 – x g of compound in the water at equilibrium. Using this, the distribution coefficient equation can be written as:

$$K_{e/w} = \frac{C_{ether}}{C_{water}} = \frac{x \text{ g}/100 \text{ mL}}{(1-x) \text{ g}/100 \text{ mL}} = 5$$

Rearranging this gives: $x = 5 - 5x$

Which solves as: $x = 0.83$ g in ether and $1 - x = 0.17$ g in water

$$\% \text{ Extraction} = \frac{\text{g of compound extracted}}{\text{g of compound dissolved in original solvent}} \times 100 = \frac{0.83 \text{ g}}{1 \text{ g}} \times 100 = 83\%$$

If the extraction is carried out with the same amount of ether in two equal portions of 50 mL each, then we have for the first extraction:

$$\frac{x_1 \text{ g}/50 \text{ mL}}{(1 - x_1) \text{ g}/100 \text{ mL}} = 5;\ x_1 = 0.71 \text{ g in ether and } 1 - x_1 = 0.29 \text{ g in water}$$

and for the second extraction:

$$\frac{x_2 \text{ g}/50 \text{ mL}}{(0.29 - x_2) \text{ g}/100 \text{ mL}} = 5;\ x_2 = 0.21 \text{ g in ether and } x_1 + x_2 = 0.92 \text{ g}$$

The total amount extracted by 100 mL as 2 x 50 mL portions of ether is thus 0.92 g (compared to only 0.83 g using 1 portion of 100 mL).

$$\% \text{ Extraction} = \frac{0.92 \text{ g}}{1 \text{ g}} \times 100 = 92\%$$

Or use the following formula;

$$\% \text{ Extraction} = [1 - (\frac{V_{water}}{V_{water} + K_{e/w}V_{ether}})^n] \times 100, \text{ where n is the number of extractions}$$

$$= [1 - (\frac{100}{100 + (5)(50)})^2 \times 100 = 92\%$$

These values show that many smaller extractions are more efficient than one large extraction. Virtually complete removal of the compound can therefore be effected, even if the distribution coefficient is very low, by repeated extractions with small volumes of solvent. In practice this is accomplished by use of an apparatus in which the solution to be extracted is continuously treated with fresh solvent. Most applications of liquid/liquid extraction in the laboratory require only a few contacts with fresh portions of solvent, using a simple separatory funnel.

Example 4.1 Given at 20 °C only 0.24 g of an organic acid A dissolves in 100 mL of water, but 2.70 g of A dissolves in 100 mL of ether.

(a) Calculate the value of distribution coefficient.

$$K_{e/w} = \frac{C_{ether}}{C_{water}} = \frac{2.70 \text{ g}/100 \text{ mL}}{0.24 \text{ g}/100 \text{ mL}} = 11.25$$

(b) Calculate the percentage of extraction if 0.12 g of acid remains in 100 mL of aqueous solution after extraction with 100 mL of ether.

$$K_{e/w} = 11.25 = \frac{x \text{ g}/100 \text{ mL}}{0.12 \text{ g}/100 \text{ mL}}$$

$$x = 11.25 \times 0.12 = 1.35 \text{ g of acid extracted in ether}$$

g of acid originally in water = 0.12 + 1.35 = 1.47 g

$$\% \text{ Extraction} = \frac{1.35 \text{ g}}{1.47 \text{ g}} \times 100 = 91.8\%$$

(c) Calculate the volume of ether required to extract 85.0% of a 3.00 g of acid A in 100 mL of water.

g of acid extracted in ether = 0.850 x 3.00 = 2.55 g

g of acid remained in water = 3.00 – 2.55 = 0.45 g

$$K_{e/w} = 11.25 = \frac{2.55 \text{ g}/V_{ether}}{0.45 \text{ g}/100 \text{ mL}}$$

$$V_{ether} = 50.4 \text{ mL}$$

(d) Calculate the total amount of acid extracted by a double extractions of 50 mL ether in each extraction in (c).

First extraction (first 50 mL):

$$K_{e/w} = 11.25 = \frac{x_1 \text{ g}/50 \text{ mL}}{(3.00 - x_1) \text{ g}/100 \text{ mL}}$$

x_1 = 2.55 g acid extracted in first extraction

and $3.00 - x_1$ = 0.45 g acid remained in aqueous solution after first extraction

Second extraction (second 50 mL):

$$K_{e/w} = 11.25 = \frac{x_2 \text{ g}/50 \text{ mL}}{(0.45 - x_2) \text{ g}/100 \text{ mL}}$$

x_2 = 0.38 g acid extracted in second extraction

and $0.45 - x_2$ = 0.07 g acid remained in aqueous solution after second extraction

$x_1 + x_2 = 2.55 + 0.38 = 2.93$ g acid extracted by two extractions

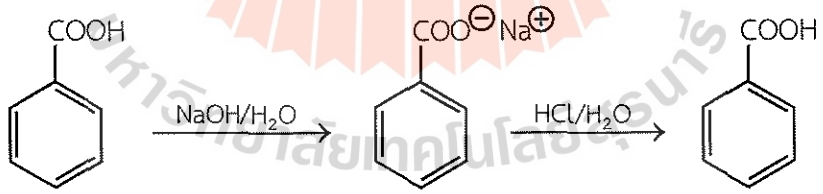
$$\% \text{ Extraction} = \frac{2.93 \text{ g}}{3.00 \text{ g}} \times 100 = 97.7\%$$

Or

$$\% \text{ Extraction} = [1 - (\frac{V_{water}}{V_{water} + K_{e/w} V_{ether}})^n] \times 100$$

$$= [1 - (\frac{100}{100 + (11.25)(50)})^2 \times 100 = 97.7\%$$

Another type of extraction is an acid/base extraction. It is a modification of liquid/liquid extraction which can provide a further level of fine-tuning. If one or more of the compounds in the mixture to be separated is acidic or basic, the solubilities of these acidic and basic components can be manipulated to our advantage by applying simple acid-base reactions. Using such a manipulation, an acidic or basic compound that may be ether soluble and water insoluble can be changed to be ether insoluble and water soluble by carrying out an acid-base reaction. The solubilities of acidic and basic compounds can thus be changed at will. Once the layers are separated as before, the neutral acid or base can be regenerated by yet another acid-base reaction. An example of such a manipulation of solubilities is shown below for a carboxylic acid.



Carboxylic acid	Salt of carboxylic acid	Carboxylic acid
– high solubility in ether	– low solubility in ether	– high solubility in ether
– low solubility in water	– high solubility in water	– low solubility in water

Extraction Solvents

One important aspect when choosing a solvent system for extraction is to pick two immiscible solvents. Some common liquid/liquid extraction solvent pairs are water-dichloromethane, water-ether and water-hexane. Notice that each combination includes water. Most extractions involve water because it is highly polar and immiscible with most organic solvents. In addition, the compound you are attempting to extract, must be soluble in the organic solvent, but insoluble in the water layer. An organic compound like benzene is simple to extract from water because its solubility in water is very low. However, solvents like ethanol and methanol

will not separate using liquid/liquid extraction techniques because they are soluble in both organic solvents and water.

Diethyl ether is the most common extraction solvent. It has a very low boiling point (34.5 °C) and can dissolve a large number of organic compounds, both polar and nonpolar. However, diethyl ether must be used with great care since it is extremely flammable and tends to form explosive peroxides on standings.

Dichloromethane (methylene chloride) has most of the advantage of diethyl ether; in addition, it is nonflammable and denser than water. However, it has a tendency to form emulsions, which can make it difficult to separate the layers cleanly. Other useful solvents and their properties are listed in Table 4.1. Various grades of petroleum ether (a mixture of low boiling hydrocarbons) can be used in place of pentane.

Table 4.1 Properties of commonly used extraction solvents

Solvent	Boiling point (°C)	Density at 20 °C (g/mL)	Solubility in water at 20 °C (g/100 mL)	Comments
chloroform	61.2	1.479	0.795	Can form emulsion, easy to dry and remove, health hazard, suspected carcinogen
dichloromethane	39.8	1.326	1.32	Good general solvent, easy to dry and remove, suspected carcinogen
diethyl ether	34.5	0.713	7.5	Good general solvent, absorb some water, easy to remove, very flammable, its vapor should not be inhaled
ethyl acetate	77	0.895	8.7	Absorbs much of water
hexane	69	0.659	0.014	Easily dried
pentane	36.1	0.626	0.04	Easy to dry and remove, very flammable
1,1,2-trichlorotri-fluoroethane	47.7	1.575	0.017	Maybe substituted for carbon tetrachloride

From the foregoing discussions some of the desirable properties of an organic extraction solvent become apparent. (1) It must readily dissolve the substance being extracted but must not dissolve to any appreciable extent in the solvent from which desired substance is being extracted. (2) It should extract neither the impurities nor other substances present in the original mixture. (3) It should not react with the substance being extracted. (4) It should be readily separated from the desired solute after extraction. (5) It should be reasonably safe to work with and relatively inexpensive. Few solvents will meet all of these criteria, and in some cases a completely satisfactory solvent cannot be found. Therefore, the scientist must select a solvent system that most nearly approaches the ideal.

Some of the solvents commonly used for extracting aqueous solutions or mixtures include diethyl ether, methylene chloride, chloroform, carbon tetrachloride, benzene, *n*-pentane, *n*-hexane, and various mixtures of saturated hydrocarbons from petroleum (petroleum ether, ligroin, etc.). Each of these has a relatively low boiling point so that it may be fairly easily separated from the solute by evaporation or distillation. Methanol and ethanol are not good

solvents for extracting aqueous solutions or mixtures because of their solubility in water; however, if an aqueous solution can be saturated with potassium carbonate without affecting the solute, ethanol can be used to extract polar solutes from the solution.

Use of a Separatory Funnel

Once the two layers are mixed, you will need to separate them. You could separate the two layers by pouring off the less dense layer into a separate container. You would find that it is difficult to do this cleanly, however. With a water-ether mixture, you would undoubtedly end up with some ether left on top of the water or some water poured off with the top ether layer. To make the separation of two liquids, chemists use a separatory funnel as shown in Figure 4.1.

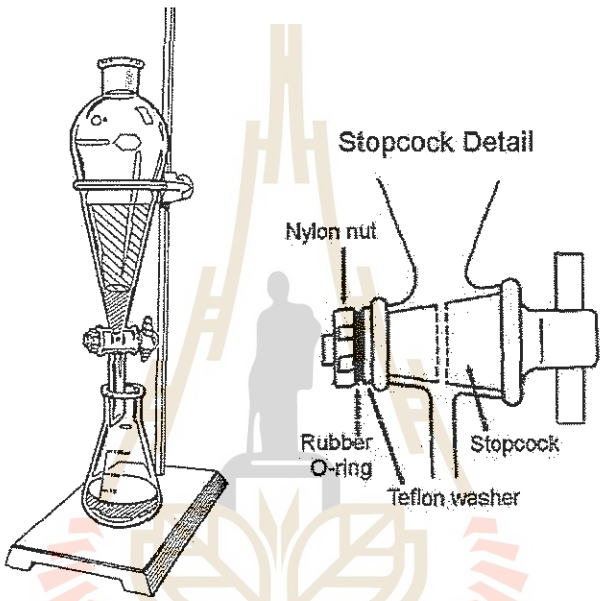


Figure 4.1 Separatory funnel for extractions

The separtory funnel is a tapered vessel with a stopcock at the bottom which permits a sharp separation of two liquid layers in a liquid/liquid extraction or in any situation requiring the separation of an organic liquid from the aqueous layer. A separatory funnel is expensive and fragile, and when full, it is top heavy. The funnel should be supported on a ring of the proper size at a convenient height; don't prop it up on its stem. Before each use, check that the stopcock is seated and rotates freely. A clip or leash should be used to prevent the stopcock from falling out if it is accidentally loosened. In case of glass stopcock, a very light film of stopcock lubricant should be applied around the stopcock in bands on each side of the hole (Teflon stopcock requires no lubricant). Excess grease will be washed away by organic solvents and contaminate the solution.

The separatory funnel should be filled to no more than about three-quarters of the total depth, so that thorough mixing is possible. After filling (check first that stopcock is closed!), stopper securely with a properly fitting glass, plastic or rubber stopper. Before using the funnel for the first time, it is a good idea to shake with a few milliliters of solvent to make sure that stopcock and stopper are tight.

Hold the funnel with the stopcock end tilted up; the stopper is kept in place securely with the heel of one hand and the stopcock end is supported in the other hand. Now gently invert the funnel. As soon as the funnel is inverted, open the stopcock to release any pressure (see Figure 4.2) by pointing the stem of the funnel away from your face and your neighbors (volatile solvents such as ether and dichloromethane develop considerable pressure). Then close the stopcock and shake the funnel in a horizontal position two or three times. Stop and slowly open the stopcock to vent any pressure as before. Repeat this process until opening the stopcock causes no further pressure release. Close the stopcock and shake the funnel 15-20 times. Stop and slowly open the stopcock to release the pressure. Replace the funnel in the ring and remove the stopper. Allow the liquids to stand until the layers have completely separated. Draw off the lower layer through the stopcock into a flask of proper size. Do not draw the liquid through the stopcock too rapidly. Slow the flow carefully as the boundary between the two layers approaches the stopcock. Stop the flow of liquid completely just as the upper layer enters the hole in the stopcock.

Figure 4.2 Method for holding and venting the separatory funnel

In the extraction of an aqueous solution, the solvent may be either lighter than water (e.g. ether or hexane) or heavier than water (e.g. chloroform or dichloromethane). In the first case, if several portions of solvent are used, the aqueous layer must be drained into a receiver (usually the flask in which it was orginally contained), and the organic solution is transferred to a second flask by pouring from the top of the separatory funnel. The aqueous phase is then returned to the separatory funnel for further extraction as needed. With a solvent denser than water, the aqueous solution is simply retained in the funnel and shaken with successive portions of solvent.

It is sometimes not immediately obvious which layer in the separatory funnel is the organic phase and which is the aqueous. If in doubt, withdraw a small sample of the lower phase in a test tube and add a few drops of water to see whether or not two layers form. If you see two layers then this indicates that the lower phase in the separatory funnel is the organic layer. Alternatively add a few drops of water to the top of the separatory funnel and watch

to see which layer the drops end up in. If they sink through the upper layer into the bottom layer, then the lower layer is the water layer.

In many cases the separatory funnel is used simply as a means of recovering an organic product from a large amount of water with minimum mechanical loss. A small volume of ether or other solvent is added to the mixture to permit sharp separation of layers. Even though the compound may have negligible solubility in water, a second portion of solvent should be used to rinse the aqueous layer and the separatory funnel.

A common application of extraction is the removal of water-soluble impurities from an organic solution, e.g. an ether solution may contain dissolved hydrogen chloride. This is removed by "washing" the ether in a separatory funnel with aqueous carbonate or hydroxide solution and then water.

Emulsions

An emulsion is a suspension of tiny droplets of one solvent mixed in the other, leading to a poor separation. Gentle shaking and swirling the separatory funnel is the best technique to avoid emulsions. However, if an emulsion occurs, there are several simple methods to destroy it. The first is time. Over time the layers will eventually separate. With a severe emulsion, you may not have time during a three hour lab period to wait. Another method is to add brine or salt water to the mixture. Since organic solvents are less soluble in a highly ionic solution such as salt water, the organic layer and aqueous layer will be forced to separate. This method works well with small emulsions. If you have a more difficult emulsion, separate the layers as much as possible and dry the organic layer with a drying agent. The water will be removed from the organic layer along with the drying agent. Subsequent extractions should proceed without further trouble.

In this experiment, when the tannins in tea are converted to their salt, they will become anionic surfactants. Detergents and soap are surfactants and these cause materials that do not dissolve in water (like oil, grease and dichloromethane) to form an emulsion with water. As we want to be able to separate the aqueous phase from the dichloromethane phase, it is better not to let the emulsion to occur during the extraction. Consequently, as you extract the caffeine from water into dichloromethane, do not shake the separatory funnel too vigorously.

Drying Agents

One significant problem with liquid/liquid extraction is that no solvent is completely insoluble in another solvent. In practice, one additional step is usually carried out before evaporating the organic solvent: drying over anhydrous sodium sulfate or other drying agents. Drying a liquid might seem like a peculiar concept, since we normally think of all liquids as being wet. Drying an organic liquid in the organic lab has a special meaning to chemists. It means to remove all traces of water. Even water and hexane are slightly soluble in each other. After separating the two solvents, residual water will remain in the hexane or other organic layer. This will remain and stick to the solid product when we remove the more volatile solvent.

Therefore, chemists remove the water from the organic layer by adding an insoluble inorganic solid to the solution which will absorb the water, thus "drying" it. Granular anhydrous sodium sulfate is the drying agent most often used although other drying agents are also available (see Table 4.2). All of the inorganic solids work by reacting with the water to form hydrates, which is their preferred form if water is available.

Table 4.2 Drying agents commonly used for drying solutions in organic solvents

Substance	Capacity	Speed	Intensity	Cost	Convenience	Suitability
calcium chloride	high	medium	high	low	high	a
calcium sulfate (drierite)	low	very high	very high	medium	high	b
magnesium sulfate	high	high	medium, high	low	medium	c
molecular sieves, 4Å	high	high	high		high	c
potassium carbonate	medium	medium	medium		medium	d
sodium sulfate	very high	low	low	low	medium	e

a. Combines with alcohols, phenols, amines, amino acids, amides, ketones, and some aldehydes and esters. It should not be used to dry solutions containing compounds of these types unless it is desired to remove them also. Some calcium hydroxide may be present that will combine with acids. The hexahydrate is unstable above 30 °C.
b. Generally useful. The hemihydrate is stable to at least 100 °C.
c. Generally useful.
d. Combines with acids and phenols. It should not be used to dry solutions containing acids unless it is desired to remove them also.
e. Generally useful. The decahydrate is unstable above 32 °C.

These drying agents do not dissolve in the solvent; they are "drying". They may change somewhat, for example, sodium sulfate will clump together as it reacts with water, but they will remain solids in normal extraction solvents. This makes them easy to remove by decantation (pouring off) of the liquid or by gravity filtration. Usually the organic solvent will go from cloudy to clear in the process of being "dried". You should be careful to remove all of these solid drying agents before solvent evaporation or you might think they are your produtct. When you take a metling point and the product doesn't melt by 300 °C, you probably have isolated your drying agent.

It is recommended that the drying agent you choose be in a grannular form. After the drying agent has removed the residual water, it is easier to remove large grannular particles. Drying a solvent however, is not an exact science. An excess of drying agent should be used to ensure that all the water is removed. If the water remains after the materials are collected, it could interfere with the analysis. Add drying agent until there are no longer clumps of drying agent stuck to the sides or bottom of the flask. The drying agents should be free floating in the flask, like snow as shown in Figure 4.3.

(a) (b) (c)

Figure 4.3 (a) Clumps of drying agent that has absorbed water (b) after adding a little more drying agent, the clumps become smaller and (c) after obtaining dry solution, free-floating crystals are visible.

There are many other choices for drying agents including molecular sieves and sodium metal. There are benefits and disadvantages to each one. Sodium, for example, is an excellent drying agent, however it violently decomposes in water to create NaOH and H_2 gas and may ignite spontaneously. Therefore it should be used with caution and only when removing very small amounts of water. Many times a particular drying agent wil work better than others in a certain situation.

Sublimation

Another easy and inexpensive purification technique is sublimation. It will be used as the final purification step in the isolation of caffeine from tea in this experiment.

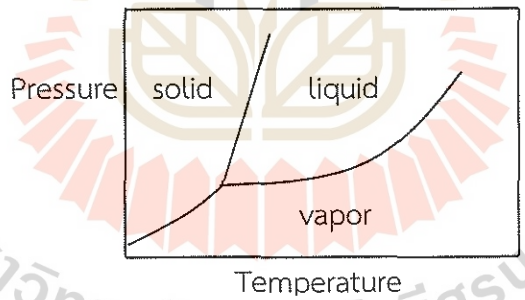


Figure 4.4 Phase diagram

Sublimation is the phase change from a solid directly into the gaseous phase without first melting. Even though it may be low, a solid does exert a vapor pressure, and vaporization directly to the gaseous phase does occur. Figure 4.4 is a typical phase diagram which can be used to determine whether a substance exists in the solid, liquid or gaseous state at a particular temperature and pressure. The boundary lines between these three phases are determined experimentally for individual compounds. The phase changes of melting, boiling and sublimation, and the reverse processes of solidification (crystallization) and condensation for constant pressure systems are shown on the diagram. Note that one can also move between phases by changing pressure at constant temperature. The triple point is the location where all three phases exist coincidentally.

There are a number of relatively high melting compounds that exert an appreciable vapor pressure at 1 atmosphere, and if this pressure exceeds 760 mmHg at a temperature below the melting point, no liquid state will exist at that pressure e.g. solid CO_2. Such solids at temperatures near or below their melting point can be separated from non-volatile substances by sublimation. This process will be favored by reducing the pressure below 1 atmosphere, and vacuum sublimation is a useful method of purifying a volatile solid if the contaminants are high molecular weight materials.

An apparatus used that incorporates a cold surface of fairly high surface area, a so-called "cold finger" held close to the material to be sublimed. The system is evacuated and the solid material warmed gently. After the sublimation is complete, the solid is scraped from the cold finger with a spatula. Sublimation is most useful in the case of very small samples, as mechanical losses can be kept very low. In practice a suction or vacuum flask is used so that it has a vent if it is heated or it can be evacuated if it is connected to a vacuum line or an aspirator. Remember that you should never heated a sealed vessel – that could cause an explosion.

(a) (b)

Figure 4.5 (a) The sublimation set up and (b) cold finger after sublimaltion

Experimental

In this experiment you are going to make a small but strong cup of tea and extract the caffeine from it by liquid/liquid extraction using dichloromethane. Then, you are going to purify the crude caffeine by sublimation and calculate the mass percent of caffeine in the tea sample.

Experimental Procedure

1. Place 5 g of dry tea leaves from tea bags into a tared clean 150 mL beaker. Record the exact mass of tea leaves.
2. Add 2.0 g of sodium carbonate and 30 mL of distilled water into the beaker.
3. Gently boil the contents for 10 minutes on a hot plate. Place a watch glass on top of the beaker to prevent excessive evaporation.
4. Filter the hot tea through a glass funnel plugged with a small piece of cotton and keep the hot tea in a 50 mL Erlenmeyer flask.

5. Transfer the tea leaves back into the beaker. Add 20 mL of distilled water and again bring the contents to boiling.
6. Filter and combine the hot tea into the 50 mL Erlenmeyer flask. Discard the tea leaves.
7. Cool the tea solution in an ice-water bath until the tea is cooled to room temperature.
8. Add 10 mL of dichloromethane to a 125 mL separatory funnel that is supported by a ring on a stand (see Figure 4.1). Then, transfer the tea solution into the separatory funnel using a glass funnel.
9. Stopper the separatory funnel. Release any pressure that may have built up inside the funnel, gently shake the funnel horizontally a few times and vent any pressure. Repeat this shaking and venting process several times. Remember not to shake the funnel too vigorously to prevent any emulsion from forming. Close the stopcock and swirl the content in the funnel. Place the funnel in the ring and remove the stopper.
10. Allow the contents in the separatory funnel to settle. There should be two distinct mostly clear layers. If there is an emulsion (cloudy) layer between two clear layers, it is sometimes possible to break the emulsion by swirling the contents of the funnel or stirring the contents using a glass rod. If the emulsion persists, seek your instructor's help.
11. Carefully drain the lower (dichloromethane) layer into a clean 50 mL Erlenmeyer flask. Try not to include any of the aqueous (upper) layer. If there is a lot of emulsion, keep it in the Erlenmeyer flask.
12. Repeat steps 9 to 11 using another 10 mL portion of dichloromethane.
13. Add half a spatula of anhydrous magnesium sulfate to the combined dichloromethane extracts in the Erlenmeyer flask. Swirl the contents of the flask and allow it to settle. The anhydrous magnesium sulfate will absorb the small amount of water that is dissolved in the dichloromethane and small amounts of water from the aqueous layer that may have gotten into the flask by accident. If there is fine powdery magnesium sulfate visible then the solution is dry (if not, add more magnesium sulfate). Leave it for 10 minutes. If you collect a substantial amount of emulsion in the Erlenmeyer flask, the magnesium sulfate should help to "break" the emulsion. You may need to separate the aqueous content from the dichloromethane solution at this point. Seek your instructor's help.
14. Decant the liquid from the flask into a clean, dry and pre-weighed 50 mL beaker. Rinse the magnesium sulfate with a small amount of dichloromethane and add this to the beaker. Place the beaker on a steam bath to evaporate dichloromethane. Wipe the moisture from the bottom of the beaker. Weigh the beaker containing the crude caffeine and determine your crude yield before sublimation.
15. Carefully transfer the crude caffeine into a pre-dried 125 mL Erlenmeyer flask. Add a few drops of dichloromethane to dissolve the leftover crude caffeine if necessary to completely transfer it. Rinse the beaker with additional drops of dichloromethane and add this to the Erlenmeyer flask. Place the flask on the steam bath to evaporate dichloromethane. Wipe the moisture from the bottom of the flask.
16. Loosely fit a "cold finger" (insert a test tube containing ice into the mouth of the Erlenmeyer flask containing the crude caffeine, and loosely seal the "cold finger" by

means of the small cone filter adapter) into the flask so that the end of the tube is about 3 cm above the base of the flask.

17. Gently heat the crude caffeine in the flask in a heating mantle. At atmospheric pressure, caffeine melts at 238 °C and sublimes at 178 °C. If a lot of water droplets collect on the "cold finger", it may be necessary to remove it and wipe it dry. Replace the "cold finger". Be careful of hot flask and allow sublimate to accumulate. As the ice in the "cold finger" melts, water may be removed with a pipette.
18. After the sublimation is complete, leave the "cold finger" in the flask until your next lab period. Scrape the pure caffeine with a spatula onto pre-weighed weighing paper. Determine the mass of the pure caffeine and calculate the mass percent of caffeine in the tea sample based on the initial amount of the tea used.
19. Submit the caffeine in a properly labeled container.

Laboratory Safety Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. To avoid short circuiting or melting of wires, make sure that the electrical wire of the hot plate is fully uncoiled from the metal part.
3. Take care to avoid burning yourself when using the electric hot plate.
4. When handling dichloromethane, avoid skin contact and avoid breathing its vapors. Keep it in the fume hood whenever possible.
5. Never point the stem of the separatory funnel at yourself or at anyone else when releasing any pressure built up inside the funnel.
6. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 4

Experiment 5	Simple and Fractional Distillation

Objectives

1. To separate a mixture of two miscible liquids using both simple and fractional distillation techniques.
2. To compare the efficiencies of separation of the two liquids by simple and fractional distillation.

Introduction

Distillation is one of the oldest and still most common methods for both the purification and identification of organic liquids. It is a physical process used to separate chemicals from a mixture by the difference in how easily they vaporize. As the mixture is heated, the temperature rises until it reaches the temperature of the lowest boiling substance in the mixture, while the other components of the mixture remain in their original phase in the mixture. The resultant hot vapor passes into a condenser and is converted to the liquid, which is then collected in a receiver flask. The other components of the mixture remain in their original phase until the most volatile substance has all boiled off. Only then does the temperature of the gas phase rises again until it reaches the boiling point of a second component in the mixture, and so on.

The boiling point of a substance – determined by distillation – is a useful physical property for the characterization of pure compounds.

At any given temperature a liquid is in equilibrium with its vapor. This equilibrium is described by the vapor pressure of the liquid. The vapor pressure is the pressure that the molecules at the surface of the liquid exert against the external pressure, which is usually the atmospheric pressure. The vapor pressure is a very sensitive function of temperature. It does not increase linearly but in fact increases exponentially with temperature. The vapor pressure of a substance roughly doubles for every increase in 10 °C as shown in Figure 5.1.

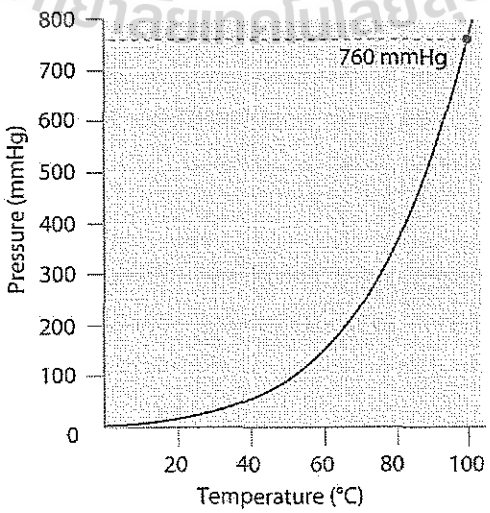


Figure 5.1 Vapor pressure dependence on temperature for water

When the vapor pressure of the liquid equals the applied pressure, the liquid boils. Thus, the boiling point of a liquid is the temperature at which the vapor pressure equals the applied pressure. The normal boiling point of a liquid is the temperature at which the vapor pressure of a liquid equals atmospheric pressure (1 atm). Thus, the boiling point of a liquid is a measure of its volatility.

The successful application of distillation techniques depends on several factors. These include the difference in vapor pressure (related to the difference in the boiling points) of the components present, the size of the sample, and the distillation apparatus. Distillation relies on the fact that the vapor above a liquid mixture is richer in the more volatile component in the liquid, the composition being controlled by Raoult's law:

In an ideal solution the partial pressure (P_A) of component A at a given temperature is equal to the vapor pressure P_A^o of pure A multiplied by the mole fraction of A (X_A) in solution.

Consider an ideal solution of A and B:

$$X_A = \frac{n_A}{n_A + n_B}, \quad X_B = \frac{n_B}{n_A + n_B} \quad \text{and} \quad X_A + X_B = 1$$

n_A and n_B represent the number of moles of components A and B.

$$P_A = X_A P_A^o, \quad P_B = X_B P_B^o \quad \text{and} \quad P_T \text{ (total vapor pressure)} = P_A + P_B$$

This relationship, derived from Raoult's law, is capable of describing the boiling point behavior of compound A in a mixture of compounds under a variety of different circumstances. The boiling point of the solution is reached when P_T is equal to the pressure applied to the surface of the solution.

There two major types of distillation are to be considered:

Simple distillation – used frequently in the organic chemistry teaching labs. It is often considered when:

(a) the liquid is relatively pure to begin with (e.g., no more than 10% liquid contaminants)
(b) essentially a pure material is separated from non-volatile or from a solid contaminant
(c) the liquid is contaminated by a liquid with a boiling point that differs by at least 70 °C.

Simple distillation uses an apparatus that consists of a distillation flask or a pot, a distillation head, a condenser, an adapter, and a receiver (see Figure 5.2). This distillation involves a single equilibrium between the liquid and vapor which is referred to as involving one theoretical plate.

Fractional distillation – The principle of fractional distillation is based on the establishment of a large number of theoretical vaporization-condensation cycles (theoretical plates): the apparatus of a simple distillation is modified by inserting a fractionating column between the distillation flask and the distillation head as shown in Figure 5.3. Typically, any one of a variety of materials, including glass beads and metal sponge, fill the fractionating column providing a large surface area in which the initial distillate is redistilled and condensed again. This process continues as the vapors rise up the column until the vapors finally make it into the condenser.

These vapors and the final distillate will contain a greater percentage of the lower boiling liquid. Continuous repetition of redistillation process in fractional distillation gives good separation of the volatile liquid components.

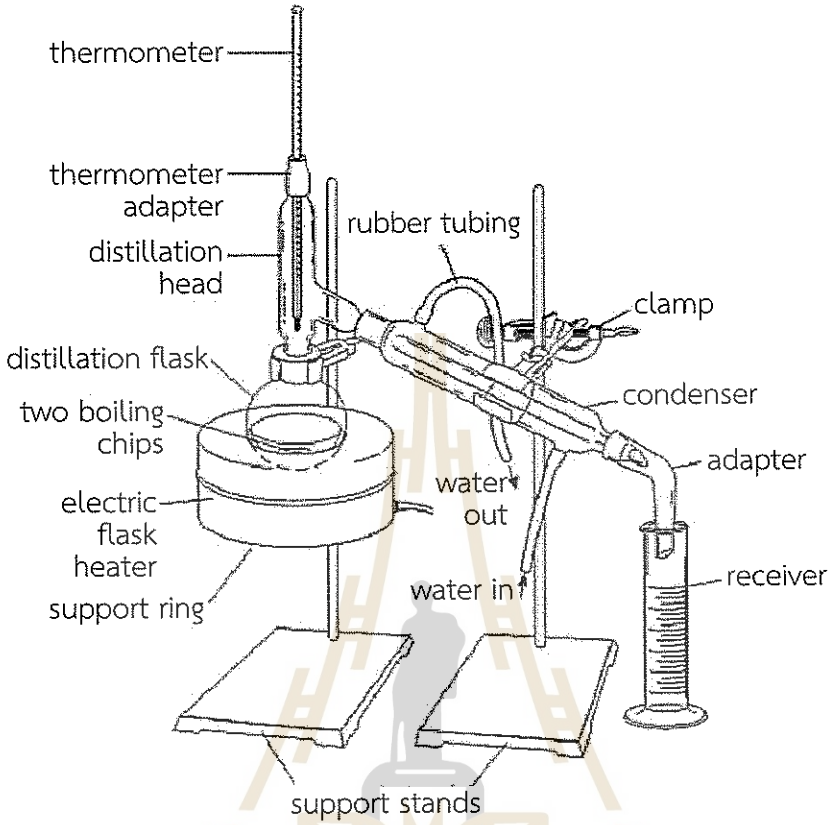


Figure 5.2 An apparatus for simple distillation

The fractionating column must be positioned vertically so that condensed liquid can drip down through the rising hot vapors. This dripping promotes equilibrium between the liquid and vapor phases, a condition that allows the column to operate at maximum efficiency and provide an optimum separation. An equally important factor affecting separation of the compounds is the distillation rate. If the distillation is conducted too rapidly, liquid-vapor equilibria will not be established in the fractionating column, and poor separation of the compounds will result.

As the liquid boils, a condensation line of vapor can be observed as it moves up the distillation head. Once these vapors reach the thermometer bulb, a dramatic temperature increase is observed. The temperature of the vapors in the distillation head provides information regarding the progress of the distillation. Initially, the vapors are rich in the more volatile compound, and the observed temperature is close to the boiling point of that compound. In a distillation with an efficient separation, the initial temperature remains relatively constant until all of the compound is collected. After the compound with the lower boiling point is completely distilled, the temperature rises sharply as the vapors of the higher boiling compound reach the thermometer bulb. At this time, the boiling point of the higher boiling compound is observed as it distills into the receiver.

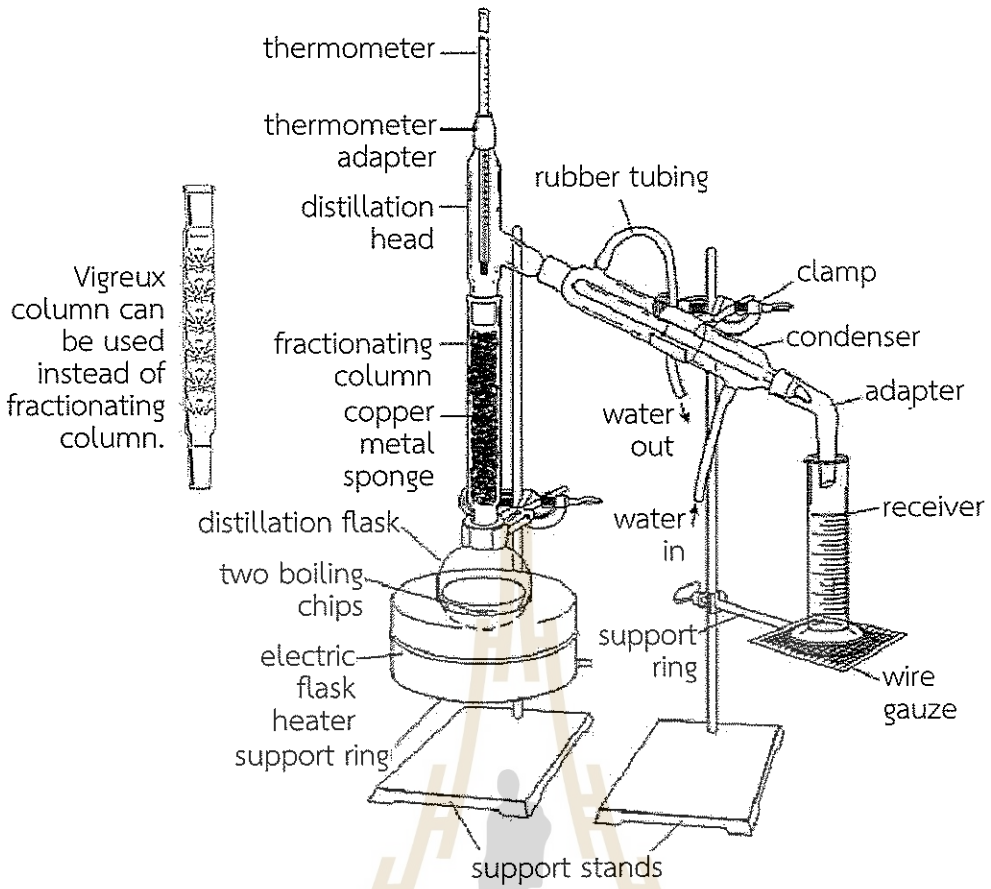


Figure 5.3 An apparatus for fractional distillation

When no fractionating column is used, or when the fractionating column is inefficient, mixtures of the distilled compounds are incompletely separated. This inefficiency is indicated by a very gradual increase in the temperature measured during the distillation. Samples collected at temperature between the boiling points of the two compounds will consist of mixtures of the two compounds.

Not all mixtures of liquids obey Raoult's law, such mixtures; called azeotropes, mimic the boiling behavior of pure liquids. These mixtures when present at specific concentrations usually distill at a constant boiling temperature and cannot be separated by distillation. Examples of such mixtures are 95% ethanol-5% water (b.p. 78.1 °C).

To understand the nature of simple distillation, fractional distillation and azeotropes, we need to look at vapor/liquid diagrams for pairs of solvents. The graph below (Figure 5.4) shows such a diagram for 2 solvents, A and B. A is the lower boiling material. The bottom of the graph shows the liquid state and the top of the graph shows the vapor state. The area in between the two curves shows what is happening in the distillation column. If we start with a mixture of A and B that corresponds to the letter C1 (concentration 1) on the graph. The mixture is vaporized (distilled). Follow the horizontal dash line until it reaches the vapor curve. This is concentration C2 in the diagram. This process has improved the concentration of A, the lower boiling component. The material is then condensed. Follow the vertical dash line down to the liquid curve. If this was simple distillation, we could stop now. It can be seen that the

purification effected by the simple distillation of such a mixture of volatile liquids is very imperfect.

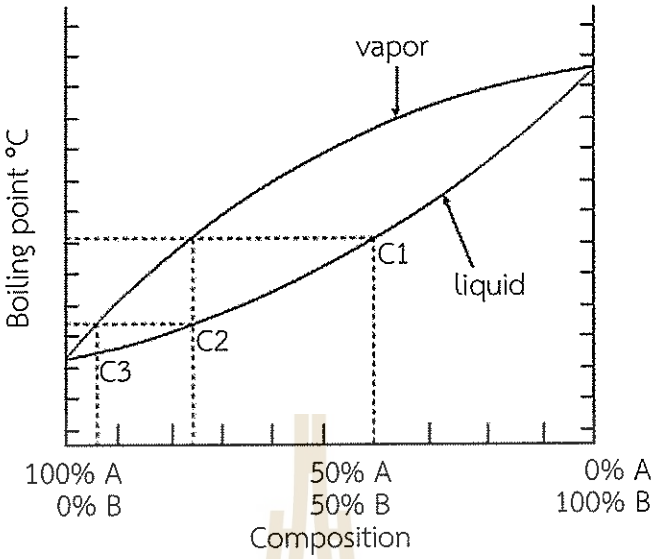


Figure 5.4 A boiling point-composition curve

However, in fractional distillation the distillation process continues. The condensed material is vaporized again. Follow the horizontal dash line across from the liquid curve at C2 to the vapor curve. There is another improvement in the concentration of the lower boiling component A. The vapor is condensed again. Follow the vertical dash line down to the liquid curve.

This number of times that the process of vaporization and condensation occurs depends on the efficiency of the distillation column. The more efficient the distillation column, the more times this happens and the purer the final product will be.

With azeotropes, the vapor liquid curves (Figure 5.5 and 5.6) are not ideal and have a point where the vapor curve meets the liquid curve. This point is called the azeotrope point.

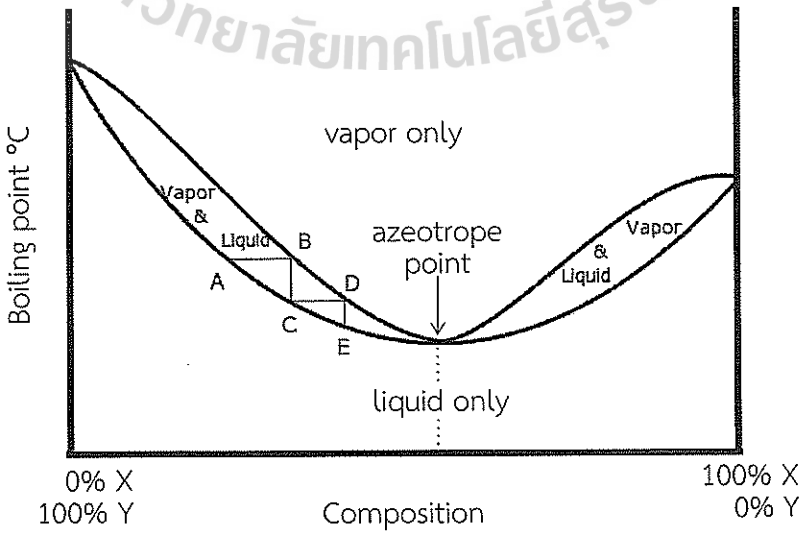


Figure 5.5 A boiling point-composition curve for a minimum-boiling azeotrope

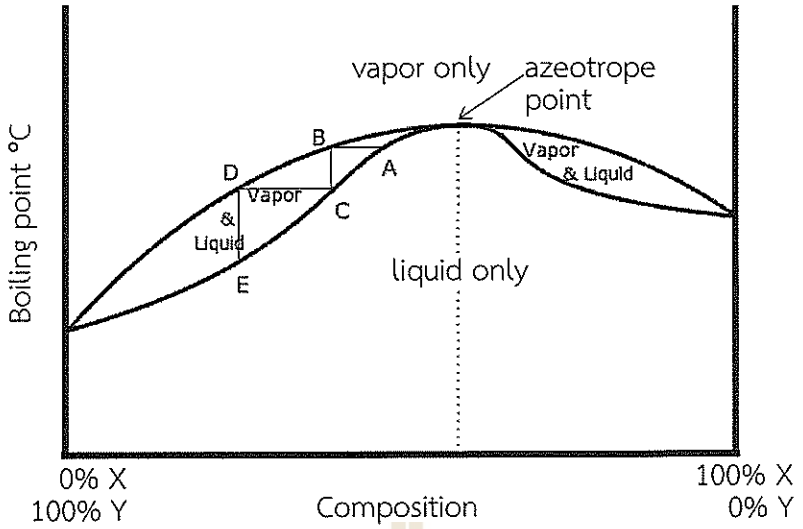


Figure 5.6 A boiling point-composition curve for a maximum-boiling azeotrope

Experimental

In this experiment you will compare distillations of a mixture of methanol (b.p. 64-65 °C) and propan-1-ol (b.p. 96-98 °C) using the glassware setup for a simple distillation and one for fractional distillation.

Experimental Procedure

A. Simple Distillation

1. The apparatus should be assembled as shown in Figure 5.2 starting with a 50 mL round-bottom flask, as a distillation flask, fitting into a heating mantle and working outwards toward a receiver. Make sure that the bottom of the distillation flask touching the heating surface of the heating mantle. In addition, it is useful to support the heating mantle on a wooden block or an iron ring so that heating may be discontinued immediately in case that the heating becomes too vigorous. Note that clamping of the distillation flask, which is advisable, becomes essential if the heating mantle is to be removed.
2. Place two boiling chips into the empty distillation flask. Boiling chips must be added to the cold liquid to ensure smooth distillation and prevent "bumping". Without boiling chips, bubbles of vapor do not escape easily from the body of the liquid. As a result, the liquid becomes superheated and the vapor is expelled periodically in a sudden uncontrollable burst that causes the apparatus to bump. Never add boiling chips to the heated liquid as you will run the risk of releasing large amounts of vapor at once. Since the pores of the boiling chips fill with liquid as soon as boiling ceases, the chips cannot be reused.
3. Add 15 mL of methanol and 15 mL of propan-1-ol into the distillation flask using a glass funnel. Be careful not to spill any chemicals onto the heating mantle.
4. Attach a distillation head fitting with a thermometer to the distillation flask. Do not grease the joints when setting up the distillation apparatus as it may contaminate the liquids you are distilling. Note that the position of the thermometer bulb must be right below the arm of the distillation head.

5. Connect a condenser to the distillation head and securely attach a piece of condenser tubing to each condenser outlet. Securely connect the other end of the "water in" tubing to a water faucet nearby and place the other end of the "water out" tubing in a drain. Note the direction of the flow of water in the condenser as indicated in Figure 5.2. Water enters the condenser at the tubule nearest the receiver (you can try hooking up the hoses "backwards" to observe the difference).
6. Attach an adapter to the condenser using an elastic band to prevent it from dropping.
7. Use a 25 mL graduated cylinder as a receiver. Position it such that the lip of the cylinder is as close as possible to the end of the adapter. Use wooden blocks to raise the gradated cylinder, if necessary.
8. Make sure all connections in the apparatus are tight.
9. Plug in the heating mantle and before heating your distillation apparatus or turning on the water for cooling the condenser, have your instructor check your distillation apparatus.
10. After your instructor has checked your apparatus, slowly turn on the water for the condenser. Because of the large heat capacity of water, only a small stream is needed; too much water pressure will cause the tubing to pop off.
11. Begin heating the mixture to boiling. Once the boiling starts, adjust the heater to maintain a distillation rate of one drop per second. Record the temperature after the first drop is collected and again after every 1 mL of distillate is collected. Continue the distillation until fewer than 5 mL of liquid remains in the distillation flask. NEVER distill to dryness, as this creates a potential risk of explosion.
12. Check that the joints remain tightly sealed periodically throughout the distillation; gap, partially in the distillation head, would lead to a serious loss of material.
13. As the lower boiling component is distilled, the boiling point of the mixture in the distillation flask will increase.
14. Turn off the heater and lower it from the distillation flask. Allow the flask to cool for a few minutes. Then turn off the water to the condenser.
15. Make a plot between the temperature (Y axis) and the volume of the distillate (X axis).

B. Fractional Distillation

1. Assemble a fractional distillation apparatus as shown in Figure 5.3, using the 50 mL round-bottom flask from section *A* as the distillation flask and Vigreux column instead of a fractionating column.
2. Add two new boiling chips into the flask.
3. Transfer all the distillate from section *A* into the flask using the glass funnel, taking care not to spill any chemicals onto the heating mantle.
4. Use the 25 mL graduated cylinder from section *A* as the receiver.
5. After your instructor has checked your apparatus, slowly turn on the water for the condenser.
6. Heat the mixture to boiling. Once boiling starts, turn down the heater. You should see a ring of condensate which gradually rises up the length of the Vigreux column, such that

the column eventually acquires a uniform temperature gradient. Increase the heat slightly if the ring of condensate stops rising.

7. When the vapors reach the top of the column, reduce the heating rate so that vapor condensation line remains just above the column and below the side arm of the distillation head.
8. Maintain the vapor condensation line in this position for 5 minutes to allow the vapor and liquid in the column to reach equilibrium.
9. Wrap the Vigreux column and distillation head with aluminum foil to minimize the temperature fluctuations.
10. Adjust the heating rate to produce distillate at a rate of 1-2 drops per 5 seconds.
11. Record the temperature after the first drop is collected and again after every 1 mL of distillate is collected. Continue the distillation until fewer than 5 mL of liquid remains in the distillation flask. NEVER distill to dryness, as this creates a potential risk of explosion.
12. Turn off the heater and lower it from the flask. Allow the flask to cool for a few minutes, then turn off the water to the condenser.
13. Make a plot between the temperature and the volume of the distillate on the same graph as in section *A*, using different symbols or colors. Label the curves.

Laboratory Safety Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. Take care to avoid burning yourself when using the heating mantle.
3. Never add boiling chips to the hot liquid. The larger surface area of the boiling chips can cause the hot liquid to foam out of the apparatus.
4. Never distill the distillation flask to dryness as there is a risk of explosion and fire.
5. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 5

Experiment 6	Isolation of Clove Oil by Steam Distillation

Objectives

1. To isolate clove oil from cloves by steam distillation.
2. To characterize the isolated clove oil using simple chemical reactions.

Introduction

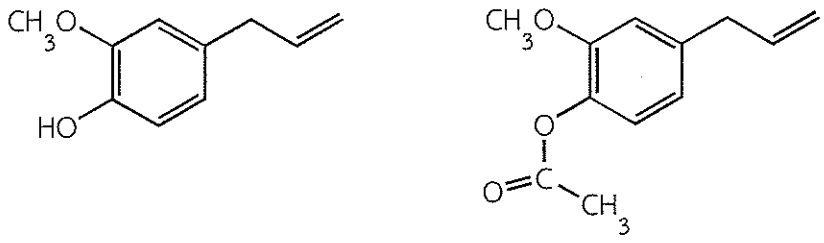
The flora and fauna of the world contain a vast array of organic molecules that can be isolated, studied, and put to use in improving the quality of our lives. Many of these natural products find applications as flavors, fragrances, dyes, and pharmaceuticals. Beneficial compounds that are obtained from plants and animals can also be modified by chemical means, for instance, through the alteration of one or more functional groups, to obtain molecules with even better properties. The essential oils of seeds, leaves, and flowers are examples of sources of natural products that have proven to be of use to humans. The flavor of vanilla, the antiseptic agent camphor, the solvent turpentine, and the insect repellant citronella are all essential oils that have been extracted from plant material.

Figure 6.1 Fresh and dried flower buds of clove

The clove tree produces flower buds (Figure 6.1) which, when harvested and dried, become the familiar spice of the same name. It has long been known that cloves contain an ingredient with an anesthetic property that is particularly effective against dental pain. Before modern medicine developed better remedies, people often chewed on cloves to dull the pain of a toothache. The active compound responsible for this effect is found in the essential oil of cloves, which makes up approximately 16-18% of the clove by weight. Steam distillation of freshly ground cloves results in clove oil, which consists of several compounds. Eugenol is the major component, comprising 85-90%. Eugenol acetate comprises 9-10%. Their structures are shown in Figure 6.2.

Eugenol has a boiling point of 254 °C. It would be difficult to isolate this oil by simple distillation since this high temperature could lead to its decomposition as well as the decomposition of other components of the cloves. However, eugenol can be isolated relatively easily from cloves using the technique of steam distillation. Once the aqueous

distillate containing the clove oil has been obtained, the oil can be separated from the water by extraction into the organic solvent dichloromethane.



(a) (b)

Figure 6.2 Structures of (a) eugenol and (b) eugenol acetate

Steam Distillation

Distillation is a common method for the separation and purification of organic compounds. These are usually liquids at room temperature although numerous lower melting solids can be distilled at higher temperatures, for example, 150 °C. However, many organic substances decompose at these higher temperatures. By distilling at low pressure (~1 torr or 1/760 of an atmospheric pressure), the technique known as vacuum distillation, the material boils at lower temperatures and decomposition is minimized.

Steam distillation is another way to distill high boiling substances and is useful for the isolation of oils, waxes, and some complex fats. In steam distillation, as shown in Figure 6.3, the distillation flask is infused with steam, which carries the oil's vapor into the distillation head and then into the condenser, where the oil and water co-condense. As an alternative, steam is generated *in situ* in the distillation flask.

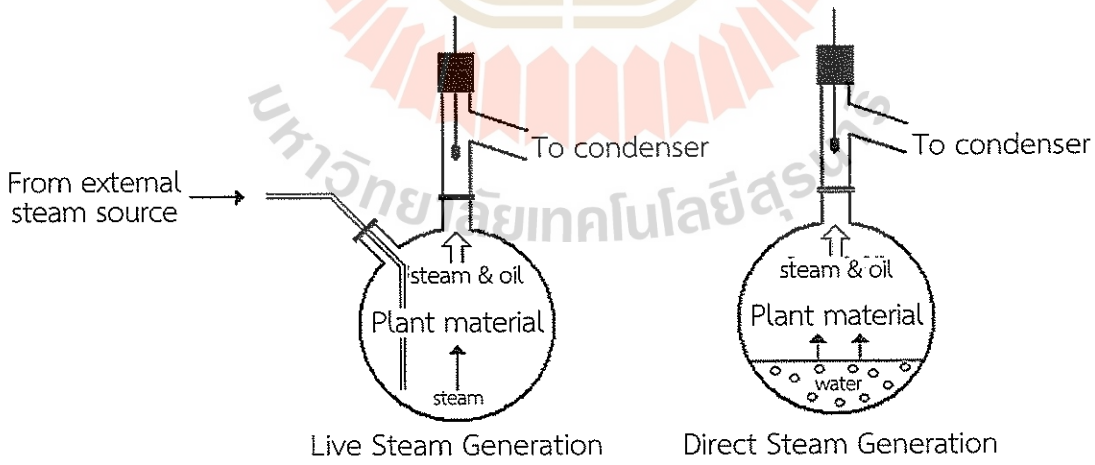


Figure 6.3 Two methods of steam distillation

Steam distillation works because the water and the oil are immiscible. Hence, they boil independent of each other as illustrated in Figure 6.4.

Figure 6.4 Each component of an immiscible liquid mixture contributes to the total vapor pressure as if the other component were not present

So, boiling occurs when the sum of the pure vapor pressures equals the atmospheric pressure:

$$P_{atm} = P^{\circ}_{water} + P^{\circ}_{oil}$$

Thus, a mixture of two immiscible liquids boils at a temperature lower than the normal boiling point of either component of the mixture. Because the vapor pressure of the water is much higher than that of the oil, the mixture will boil at a temperature slightly less than the normal boiling point of water. This means the oil will vaporize under very mild conditions.

However, this methodology leaves us with one complication; the distillate is a mixture of oil and water. The relative numbers of moles (*n*) of each component of the distillate will be given by:

$$\frac{n_{oil}}{n_{water}} = \frac{P^{\circ}_{oil}}{P^{\circ}_{water}}$$

If we know the average molecular weight (*M*) of the oil, then we can estimate the mass (*m*) of oil that will be obtained in a given amount of water:

$$\frac{m_{oil}}{m_{water}} = \frac{P^{\circ}_{oil}\, M_{oil}}{P^{\circ}_{water}\, M_{water}}$$

The vapor pressure of water is 760 torr at 100 °C. The presence of any immiscible organic material contributes to the total vapor pressure and thus causes the boiling point of water to drop below 100 °C While eugenol only has a vapor pressure of about 20 torr at 100 °C, the eugenol vapor is swept out of the boiling flask by the boiling water vapor or steam into the condenser where everything condenses, separating into liquid water with small amounts of immiscible eugenol condensing out also. This yields a cloudy distillate of water and eugenol (plus other volatile organic substances). Since it takes a lot of water vapor to sweep out the higher boiling organic oils, it is often necessary to add water to the distilling flask at frequent intervals throughout the distillation until the organic compounds have been removed from the mixture. The liquid in the condenser will become clear when the organic compounds have been removed from the mixture. The water is added via separatory funnel, burette, or pipet. The addition rate should be equal to the collection rate of drops.

Chemical Tests for Eugenol

Eugenol contains a carbon-carbon double bond and an aromatic hydroxyl group called a phenol. These functional groups provide the basis for simple chemical tests used to

characterize the clove oil. A solution of bromine (Br_2) in dichloromethane decolorizes as Br_2 reacts with the double bond to form a colorless compound, as shown in Figure 6.5. A positive test is the disappearance of the Br_2 color.

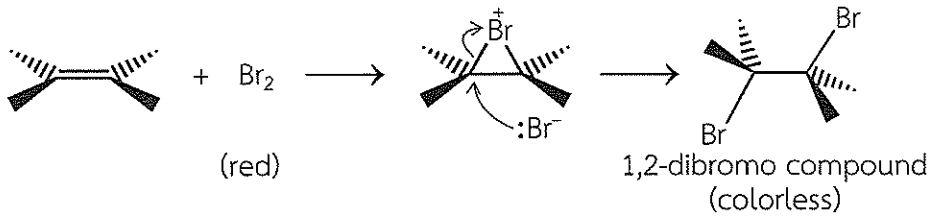


Figure 6.5 Addition of Br_2 to a carbon-carbon double bond

A potassium permanganate ($KMnO_4$) solution can oxidize a carbon-carbon double bond at room temperature to form a 1,2-diol with the simultaneous reduction of Mn^{7+} in manganese(IV) oxide (MnO_2), as shown in Figure 6.6. A positive test is the disappearance of the purple $KMnO_4$ and the appearance of MnO_2 as a brown precipitate.

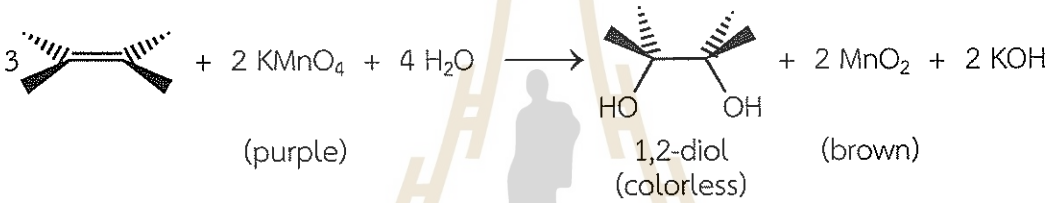


Figure 6.6 Oxidation of a carbon-carbon double bond

Phenols (ArOH) react with Fe^{3+} ion in iron(III) chloride ($FeCl_3$) to give complexes that are blue, green, red, or purple, as shown in Figure 6.7. The color may last for only a few seconds or for many hours, depending on the stability of the complex.

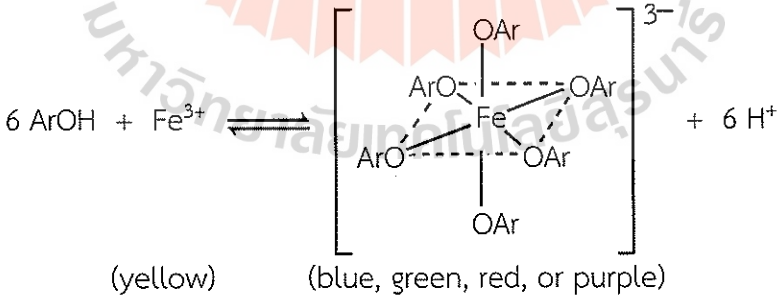


Figure 6.7 Complexation of phenols and iron(III) ion

Experimental

In this experiment, you will steam distill clove oil from freshly ground cloves. Following the distillation, clove oil and water will be present in the receiving flask. Since clove oil will be a minor fraction of the distillate, it must be extracted from the water into an organic solvent such as dichloromethane. Removing dichloromethane then leaves clove oil as the product.

Experimental Procedure

A. Isolation of Clove Oil

1. Weigh 5 g of dry cloves. Grind them to a coarse powder using a mortar and pestle. Reweigh the powder and record the mass.
2. Transfer 5 g of ground cloves to a 100 mL round-bottom flask. Add 50 mL of distilled water and a few boiling chips.
3. Assemble the distillation apparatus as shown in Figure 5.2 (the apparatus for simple distillation). Use the 100 mL round-bottom flask as a distillation flask and a 50 mL Erlenmeyer flask as a receiver.
4. Ask your instructor to inspect your equipment setup. Start the flow of water through the condenser. Turn on the heating mantle. When the mixture boils, adjust the heat to maintain a distillation rate of approximately 1 drop per second. Record the temperature of the distillation.
5. Stop the distillation when approximately 30-40 mL of distillate has been collected. Record the temperature at this point. (What will be the temperature of the distillation if the clove oil is all distilled out?)
6. Allow the distillate to cool to room temperature. Carefully pour the distillate into a separatory funnel. Add 10 mL of saturated NaCl solution. Rinse the inside of the condenser and the receiver with 5-10 mL of dichloromethane into the separatory funnel.
7. Cap the separatory funnel and gently swirl the contents for several seconds. Vent the separatory funnel frequently. After the pressure has been vented, shake the contents vigorously to thoroughly mix the two layers.
8. Allow the layers to separate. Drain the dichloromethane layer into a 50 mL Erlenmeyer flask.
9. Repeat the extraction of the aqueous layer twice, each time with 5 mL portion of dichloromethane. Combine organic layer in the same Erlenmeyer flask.
10. Dry the combined dichloromethane solution with anhydrous $MgSO_4$.
11. Decant the dichloromethane solution into a pre-weighed ceramic evaporating dish, making certain that no $MgSO_4$ is transferred with the solution.
12. Place the evaporating dish on a steam bath to remove dichloromethane.
13. When all of the dichloromethane has been evaporated, allow the evaporating dish to cool to room temperature. Weigh it to the nearest 0.0001 g and record the mass. Subtract the mass of the empty dish to obtain the mass of the clove oil.
14. Report the mass and percent yield of the clove oil.

B. Characterization of the Isolated Clove Oil

1. Dissolve all of the clove oil in 2-3 mL of methanol.
2. Obtain 6 test tubes and label them 1-6. Label tubes 2, 4, and 6 as “blank”. Add 1 mL of methanol to all 6 test tubes.
3. Add 5 drops of the clove oil solution to test tubes 1, 3, and 5. Gently swirl each tube.
4. Add 5 drops of bromine in dichloromethane to test tubes 1 and 2. Gently swirl and record your observation.

5. Add 5 drops of $KMnO_4$ solution to test tubes 3 and 4. Gently swirl and record your observation.
6. Add a few drops of $FeCl_3$ solution to test tubes 5 and 6. Gently swirl and record your observation.

Laboratory Safety and Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. Take care to avoid burning yourself when using the heating mantle.
3. Dichloromethane is irritating and toxic. Prevent eye, skin, and clothing contact.
4. Methanol is flammable and toxic. Keep away from flames or heat sources.
5. Bromine is a highly corrosive substance that causes severe burns and gives off irritating fumes. Carry out the test in a fume hood and wear gloves when handling the reagent.
6. Potassium permanganate are toxic and irritating. Prevent eye, skin, and clothing contact.
7. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 6

Experiment 7	Chromatography

Objectives

1. To use thin-layer chromatography to select a suitable solvent system for use in separation of plant pigments.
2. To determine R_f values of the plant pigments.
3. To separate the plant pigments by column chromatography.

Introduction

The term "chromatography" was invented in 1906 by a Russian botanist named Mikhail Tswett from the Greek words for color (chroma) and writing (graphe). It is derived from the original use of this method for separating yellow and green plant pigments. Since then, chromatography has become a cornerstone of separation science, that branch of chemistry devoted to separating compounds from mixtures. There are two main categories of chromatography: preparative and analytical.

Analytical work (which may be used in an environmental lab to look for pollutants) uses small sample sizes; the objective is to separate compounds in order to identify them. Preparative work (which may be used in the pharmaceutical industry) uses large quantities of sample and collects the output in bulk; the point of the chromatography here is to remove impurities from a commercial product.

All chromatographic techniques have a two-part operation in common. A stationary phase (usually a solid, thick liquid, or bonded coating) stays fixed in one place, and a mobile phase or eluent (usually a liquid or gas) moves through it or across it.

A sample to be separated, when placed on the stationary phase, will gradually move along in the same direction as the mobile phase. If a sample compound (or analyte) has no interaction with the stationary phase, it will run right through and come out of the system (elute) at the same rate as the mobile phase. On the other hand, if an analyte has no interaction with the mobile phase, it will stick directly to the stationary phase and never elute. Neither of these are good outcomes.

In a well-designed chromatography process, the chemist will choose stationary and mobile phases that will both have at least some interaction with the analytes. Any individual sample molecule will interact first with one phase and then the other, back and forth repeatedly, but the fraction of each analyte overall in each phase will remain constant. This distribution or partition coefficient, K_c, given by:

$$\text{Analyte in mobile phase} \rightleftharpoons \text{Analyte in stationary phase}$$

$$K_c = \frac{[\text{analyte in stationary phase}]}{[\text{analyte in mobile phase}]} = \frac{C_s}{C_m}$$

among the selected phases must differ for each analyte in order for them to separate. (Compounds will not separate chromatographically if they have the same distribution coefficient on a particular system.)

Analytes with greater attraction to the stationary phase (greater K_c) will still flow through the system because they spend part of their time moving in the mobile phase, but they will tend to lag behind those analytes that have a greater interaction with and thus spend more time in the mobile phase (smaller K_c). Gradually, as they progress through the system, the analytes separate from each other (usually in order of their K_c's), and can be captured or at least detected in relatively pure form as they elute.

Chromatography can be carried out in many ways. In thin-layer chromatography (TLC) and in adsorption column chromatography, a solution of the mixture flows over a solid adsorbent. Separation occurs as molecules are adsorbed and desorbed while passing over the surface. In paper chromatography applications, the mixture is partitioned between water molecules adsorbed on the paper and a solvent that moves over the paper. In gas chromatography (GC), a mixture of volatile compounds is separated by passing the vapor over an adsorbent packing in a long heated tube.

Chromatographic methods have high "resolving power", i.e. they are capable of sharp separations of closely related compounds, particularly when very small samples are used. The separated compounds can be collected or detected, depending on the particular chromatographic technique involved.

Thin-Layer Chromatography

TLC is an extremely valuable analytical technique in the organic laboratory. It provides a rapid separation of compounds, and thereby gives an indication of the number and nature of the components of a mixture. TLC can also be used to select the proper conditions for a column chromatographic separation, to identify compounds by comparison with known samples, to check the purity of a compound, or to monitor the progress of a reaction, an extraction, or a purification procedure. TLC is a sensitive technique – microgram quantities can be analyzed by TLC – and it takes little time for an analysis (about 5-10 minutes).

TLC is carried out on glass plates or strips of plastic or metal coated on one side with a thin layer of adsorbent – the most common are silica gel ($SiO_2 \cdot xH_2O$) and alumina (Al_2O_3). The adsorbent contains a small amount of gypsum ($CaSO_4$) which acts as a binder to give an adherent coating. For routine work, small TLC plates can be prepared by dipping microscope slides in a slurry of the adsorbent in chloroform. More uniform plates are obtained by mixing the adsorbent with an equal weight of water, spreading the mixture on a glass plate and allowing it to set dry. Precoated TLC plates are commercially available with various adsorbents in very uniform layers.

To "load" the plate, very small samples of the sample mixture in some volatile solvent are applied as spots near one end and the volatile application solvent is allowed to evaporate. The plate is then placed, with the sample end down, in a closed vessel containing a shallow

pool of the "developing solvent". The solvent rises on the plate by capillary action, passing over the sample and causing the compounds to move at varying rates depending on their relative affinities for the adsorbent and the solvent. When the solvent front has risen to just below the top of the plate, the plate is removed and the solvent is allowed to evaporate. After the separation is complete, the TLC plate is called a chromatogram as shown in Figure 7.1. The zones or spots containing various components of the mixture are then detected at various points along the plate. If the compounds are colorless, they are made visible by treating the plate with a reagent that causes color to develop.

Because TLC adsorbents are typically very polar, the more polar is a compound in the mixture, the more strongly it adheres to the adsorbent and the more slowly it moves. Similarly, intermolecular attractions between the solvent and the compounds determine the solubility of the compounds in the mobile phase. In general, the more polar the solvent, the more rapidly a given compound moves. Polar compounds, which are strongly attracted to the adsorbent, require polar solvents to attract them away from the adsorbent.

Determination of Retention Factor or Rate of Flow (R_f) Value

The ratio of the distance that a compound moves to the distance that the solvent moves is called the retention factor or rate of flow, denoted as R_f.

$$R_f = \frac{\text{distance traveled by the compound}}{\text{distance traveled by the solvent}}$$

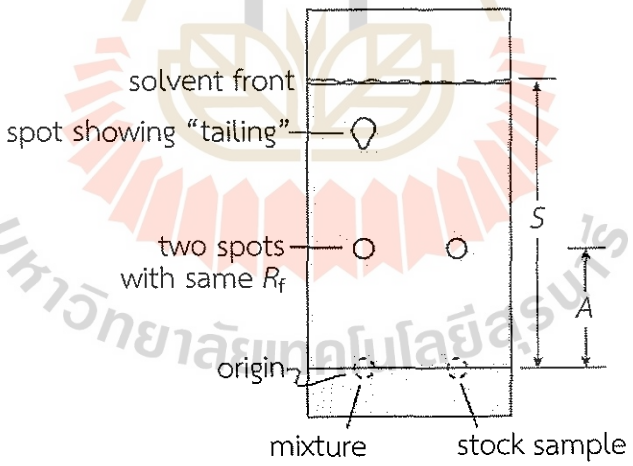


Figure 7.1 A chromatogram showing measurements for R_f calculation

For example, in Figure 7.1 the stock sample compound moved distance *A* while the solvent traveled distance *S*. If distance *A* is 2.50 cm and distance *S* is 5.50 cm, then the R_f is calculated as shown below.

$$R_f = \frac{A}{S} = \frac{2.50\text{ cm}}{5.50\text{ cm}} = 0.455$$

R_f value is always between zero and one. A TLC analysis might be summarized something like, "Using a silica gel plate and ethyl acetate as the developing solvent, unknown mixture X

showed three spots having R_f's of 0.120, 0.250, and 0.870". Note that observing three spots means only that there are at least three components in the mixture. Some components may have such similar polarities that they appear under one spot after development.

If a developing solvent of too high polarity is used, all components in the mixture will move along with the solvent and no separation will be observed (R_f's will be too large). If the solvent is of too low polarity, the components will not move enough, and again separation will not occur (R_f's will be too small). In practice, different solvents or mixtures of solvents are tried until a good separation is observed. Typically an effective solvent is one that gives R_f's in the range of 0.3-0.7. Common solvents used in chromatography, both thin-layer and column, are listed in Table 7.1. The higher a solvent's dielectric constant the more polar it is.

Table 7.1 Common solvents used in TLC and column chromatography

Solvent	Dielectric constant (ε)
hexane	1.9
petroleum ether	2.0
cyclohexane	2.0
carbon tetrachloride	2.2
benzene	2.3
toluene	2.4
diethyl ether	3.4
chloroform	4.8
ethyl acetate	6.0
acetic acid	6.2
isopropyl alcohol	18.3
acetone	20.7
ethanol	24.3
methanol	32.6
water	78.5

In general, these solvents have low boiling points and low viscosities, so they migrate rapidly along a TLC plate. They are listed in order of increasing polarity. Mixture of these solvents are often used to "fine-tune" the separation of analytes. Some suggested mixtures for specific classes of organic compounds to be separated on silica gel are shown in Table 7.2. Similar solvent mixture suggestions are available for TLC plates made of alumina.

Table 7.2 Mixtures of solvents for different classes of organic compounds

Compound class	Solvent
alcohol	cyclohexane : ethyl acetate (1:1 or 1:2) or petroleum ether : diethyl ether (10:1)
amides	ethyl acetate : methanol (5:1)
amines and amino acids	methanol : chloroform (2:3) + 1% (vol) of 33% ammonia
carboxylic acids	chloroform + 90% formic acid to saturation
esters	cyclohexane : ethyl acetate (1:1 or 1:2)
hydrocarbons	petroleum ether or cyclohexane or benzene : diethyl ether (2:1) or cyclohexane : ethyl acetate (3:1)
ketones	cyclohexane : ethyl acetate (1:1 or 1:2) or diethyl ether : hexane (1:9)

The R_f for a compound is a constant from one experiment to the next only if the chromatography conditions below are also constant:

– solvent system
– adsorbent
– thickness of the adsorbent
– amount of material spotted
– temperature

Since these factors are difficult to keep constant from experiment to experiment, relative R_f values are generally considered. "Relative R_f" means that the values are reported relative to a standard, or it means that you compare the R_f values of compounds run on the same plate at the same time.

The R_f can provide corroborative evidence as to the identity of a compound. If the identity of a compound is suspected but not yet proven, an authentic sample of the compound, or standard, is spotted and run on a TLC plate side by side (or on top of each other) with the compound in question. If two substances have the same R_f value, they are likely (but not necessarily) the same compound. If they have different R_f values, they are definitely different compounds. Note that this identity check must be performed on a single plate, because it is difficult to duplicate all the factors which influence R_f exactly from experiment to experiment.

Since two different compounds can have the same R_f in a given solvent. Additional evidence that two samples are the same compound can be obtained by comparing their mobility in several solvent systems of varying polarities. Two different compounds are unlikely to have the same R_f in solvents of different polarities, while two different samples of the same compound will have the same R_f in every solvent.

Preparing to Run TLC

Capillaries or TLC applicators for applying the samples to the chromatographic plate can be prepared by heating and drawing out a soft glass pipette or melting point or capillary tubes. Soften a 1 cm section in the center of the glass tube by heating in a low Bunsen burner flame, then remove the tube from the flame and draw it out to a thread-like thickness. Allow the glass to cool then carefully break the glass in the thin portion gives you the capillaries.

The developing chamber (Figure 7.2) can be prepared from a beaker with watch glass as a lid or a small screw-capped jar. The developing chamber is usually lined with a piece of filter paper (cut to shape and size) to adsorb the solvent and ensure that, when the chamber is closed, its atmosphere is saturated with solvent vapor, minimizing evaporation from the plate. The solvent system is added to the developing chamber which is then carefully turned at an angle to saturate the filter paper. The solvent depth should be no more than 5 mm (typically 2-5 mm).

TLC plates or sheets should be handled by the edges, and you should avoid touching the coated surface otherwise fingerprints will ruin the experiment. Samples of the test solutions should be spotted on the coated side about 1 cm from one end of the sheet, and about 0.5

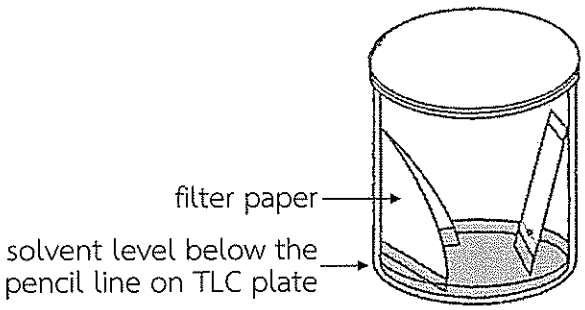


Figure 7.2 A developing chamber

cm apart, with the outer two spots about 0.5 cm from the edge of the sheet. It is common practice to draw a light pencil line about 1 cm up from the bottom edge of the plate (this line defines the origin) and then divide this line at about 5 mm intervals to create the lanes as shown in Figure 7.3.

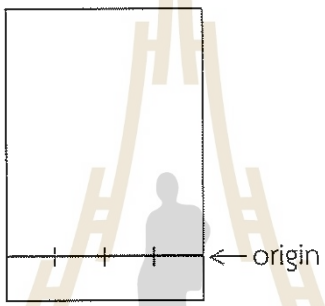


Figure 7.3 A TLC plate labeled for the origin

A TLC experiment consists of three general steps – spotting, developing, and visualizing.

Spotting a Plate

First the sample to be analyzed is dissolved in a volatile solvent such as acetone or dichloromethane. Sample of unknowns are typically spotted in the middle lanes of the plate. It is important to avoid applying too large an amount of sample since it can lead to "tailing" and poor separation as shown in Figure 7.1. The spot after application should be about 1-2 mm in diameter. It is a good idea to practice applying spots to a small scrap of the sheet. You can check to see how well you are doing by examining the spots under the UV lamp. If the spots are too pale at this stage, then you will need more applications to the plate by applying additional sample solution to the same spot after the solvent evaporates, or you should concentrate your samples. If spots are too large, another plate can be made. To apply the samples, touch the end of a capillary tube to the solution and then touch this gently to the plate at the proper place.

Developing a Plate

When the samples have been applied, place the TLC plate with the spotted side at the bottom into a developing chamber containing a pool of solvent no more than 5 mm deep. Keep the cap on the chamber at all times except when placing the plate into or taking the plate out of the chamber. Watch out for the following: the solvent must be below the level of the spots or the spotted material will dissolve in the developing solvent; the plate must

stand vertically in the chamber and the adsorbent must not touch the filter paper; the solvent must not be allowed to run all the way to the top of the plate. The development takes little time so if a plate comes out poorly, another can easily be done.

It is important to handle the plates without touching the front surface of the silica gel or alumina covered plate or fingerprints will result and the plate may be spoiled. Use clean forceps for transferring the plates in and out of the developing chamber. Once the plate is in place, cap the chamber securely and develop the chromatogram until the solvent has risen to within 5 mm of the top of the plate. The point that the solvent has reached is called the solvent front (or eluent front). When the solvent front is close to the top of the plate remove the plate from the chamber, and mark the solvent front with a small scratch (using a spatula or forceps) or a pencil mark. Remove the plate and recap the developing chamber. Allow the TLC plate to dry horizontally. If the solvent front is allowed to reach the top of the plate, the mixture compound may continue to move along the plate. An R_f obtained under these circumstances is not valid. Always allow the developing solvent to run almost completely to the top of the plate. This will use the entire plate and allow for the best possible separation.

Visualizing the Compound

Upon development, a successful separation of colored compounds will reveal distinct spots, indicating that the mixture compounds have separated, as shown in Figure 7.1. It is a good idea to draw circles around the spots with a pencil as a permanent record of the visualization. To make separated colorless compounds observable to the eye, the spots are treated in some way to make them visible. The process is called visualization.

Some compounds are fluorescent and can be visualized by viewing the TLC plate under a UV lamp. Frequently, the adsorbent contains a chemically inert fluorescent material. When viewed under UV light, compounds that absorb UV light appear as dark spots that may be outlined with a pencil.

Another simple method for visualizing organic compounds is to place the chromatogram in a chamber containing iodine (I_2) crystals and vapor. The I_2 vapor forms a colored complex with many compounds and allows detection of their spots. The spot location must be marked immediately because the I_2 will eventually sublime from the plate.

In some instances, a reagent such as phosphomolybdic acid, potassium permanganate, or ninhydrin solution is sprayed on the plate. These reagents can react with the colorless compound on the plate and give a colored product.

It is good practice to use at least two visualization techniques in case a compound does not show up with one particular method. The R_f value for a spot can now be calculated and recorded along with the color (if any) of the spot.

When looking at a developed TLC plate, it is a good idea to review the "quality" of the analysis or note if there are any problems with the plate that means it might need to be rerun. A poor quality plate would mean that it is difficult to obtain the desired information from the plate.

Some common problems are illustrated in Figure 7.4.

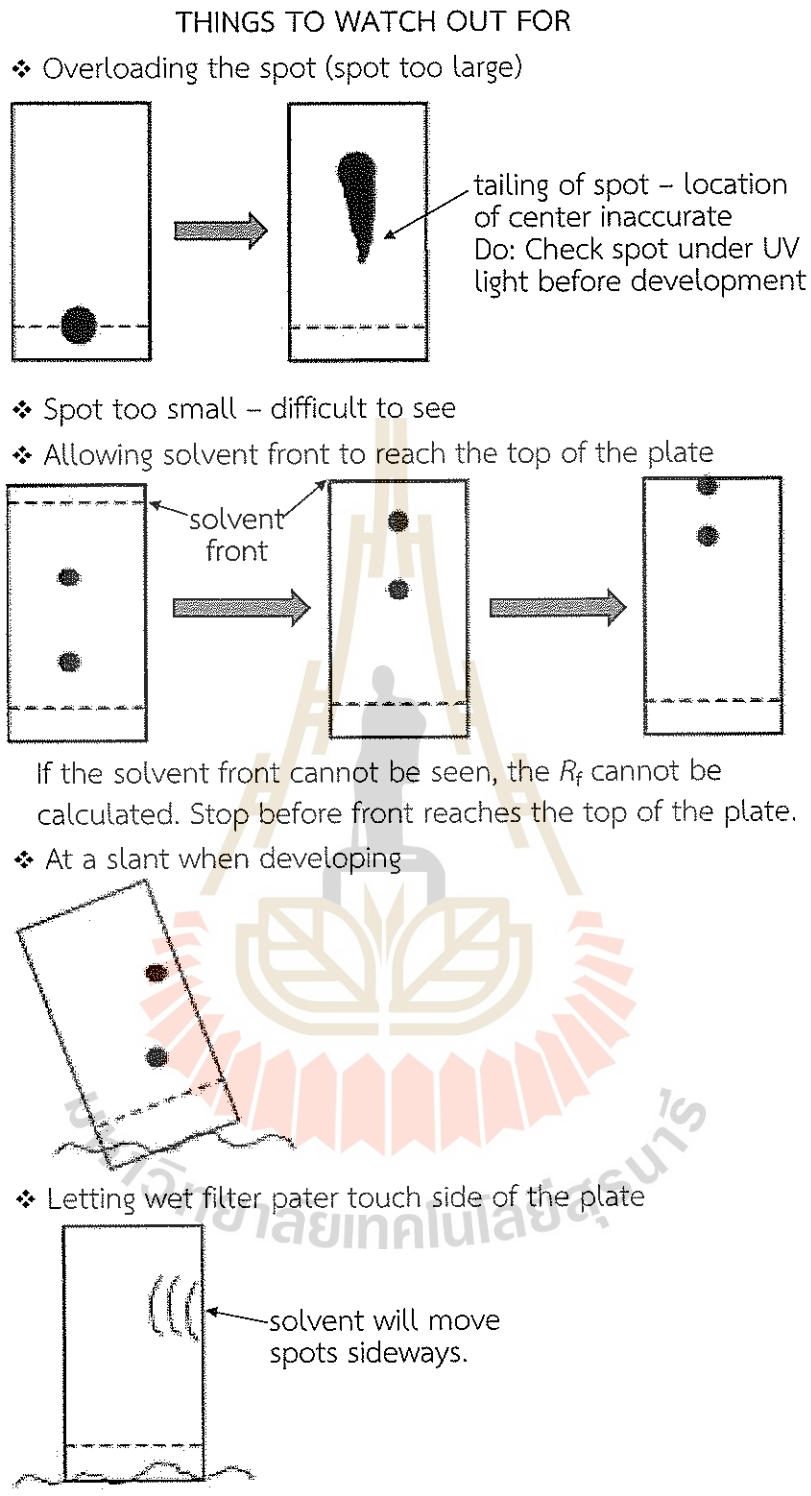


Figure 7.4 Common problems with the TLC plate

Adsorption Column Chromatography

Adsorption column chromatography is another common and useful separation technique in organic chemistry. This separation method involves the same principles as TLC, but can be applied to separate larger quantities than TLC. Column chromatography can be used on both

a large and small scale. The applications of this technique are wide reaching and cross many disciplines including biology, biochemistry, microbiology and medicine. Many common antibiotics are purified by column chromatography.

The differences in principles of adsorption column chromatography and TLC are as follows:

(i) The stationary phase is contained inside a column, rather than applied as a coating on a plate. The column is commonly made of glass, but some are made of metal or other materials.

(ii) The sample to be separated is loaded from the top. The eluting solvent, or mobile phase, is also added from the top.

(iii) The solvent flows down the column by gravity, carrying with it the components of the sample. By the same principles that apply in TLC, the components travel at different rates effecting the separation.

As in TLC, the less polar components travel down the column faster than the more polar ones. Once the less polar components are out of the column, the polarity of the solvent can be increased to speed up the rate of travel of the more polar components. The process of washing a compound through a column using a solvent is known as elution. Thus, the solvent is sometimes called the eluent.

Choosing a Stationary Phase

As with TLC, alumina and silica gel are the two most popular stationary phases in adsorption column chromatography. For these common phases, the partitioning works in an analogous manner. The more polar sample will be retained on the stationary phase longer. Thus the least polar compound will elute from the column first, followed by each compound in order of increasing polarity.

Although the interactions between the mobile and stationary phase are based on the same principles for column chromatography and TLC, be careful when predicting the order of elution. Since the direction of the solvent flow in TLC moves up and in column chromatography the solvent flows down, it appears that the order is "upside-down". In TLC the more polar molecules will have lower R_f values, but in column chromatography they will be retained longer on the column. Remember this when considering the polarities of the stationary phase as well as the polarity of the compounds being separated when predicting the order of elution.

Stationary phases for column chromatography can come in a variety of sizes, activities, acidic and basic variations for both alumina and silica gel. The types of stationary phase chosen are determined experimentally, or often based on results from a previous TLC experiment. The type of adsorbent, the size of the column, the polarity of the mobile phase as well as the rate of elution all affect the separation. These conditions can be manipulated to get the best separation for your mixture.

Choosing Solvents

Solvent systems for use as mobile phases in column chromatography can be determined from

previous TLC experiments, the literature, or experimentally. Normally, a separation will begin by using nonpolar or low polarity solvent, allowing the compounds to adsorb to the stationary phase, then slowly switching the polarity of the solvent to desorb the compounds and allow them to travel with the mobile phase. The polarity of the solvents should be changed gradually. On a macroscale, the mixing of two solvents can create heat and crack the column leading to a poor separation.

Some typical solvent combinations are ligroin-dichloromethane, hexane-ethyl acetate, and hexane-toluene. Often an experimentally determined ratio of these solvents can sufficiently separate most compounds. Solvents such as methanol and water are normally not used because they can destroy the integrity of the stationary phase by dissolving some of the silica gel.

Apparatus

Column can be as thin as pencil to a diameter of several feet in industrial processes. They can separate milligram to kilogram quantities of materials. In this experiment, we will be separating a mixture of small quantity, so a small column can be used. Figure 7.5 shows the typical setup we will be using during this experiment. It is essential to have several clean tared Erlenmeyer flasks, reaction tubes, beakers, test tubes or vials available to collect the solvent and compounds as they elute. Once you have the general setup prepared, you can move on to packing the stationary phase in the column.

Figure 7.5 Typical microscale column chromatography setup

Packing the Column

There are several acceptable methods when packing a column. These include dry packing (there are two versions of dry packing) and the slurry method. The slurry method normally achieves the best packing results, but there are several occasions when the dry packing method works just as well if not better.

Dry packing is the method of choice for a microscale column. Begin by placing a small piece of cotton in the bottom of the column to prevent the adsorbent from leaking out. Fill the

column with a nonpolar solvent. Slowly add the powdered alumina or silica gel while gently tapping the side of the column so that the adsorbent will pack tightly into the column. The solid should "float" to the bottom of the column. Try to pack the column as evenly as possible; cracks, air bubbles, and channels will lead to a poor separation.

For the second dry packing method, the stationary phase is deposited in the column before the solvent. In this case fill the column to the intended height with the stationary phase and then slowly add the nonpolar solvent. The solvent should be added slowly as to avoid uneven channeling. This method is typically used with alumina only, since silica gel expands and does not pack well with this dry method.

The slurry method is often used for macroscale separations. Plug the column with a small piece of cotton. Combine the solid stationary phase with a small amount of nonpolar solvent in a beaker. Thoroughly mix the two until a consistent paste is formed, but is still capable of flowing. Pour this homogeneous mixture into the column as carefully as possible using a spatula to scrape out the solid as you pour the liquid. The slurry method normally gives the best column packing, but is also a more difficult technique to master. Whether the dry or slurry method is chosen, the most important aspect of packing the column is creating an evenly distributed and packed stationary phase. As mentioned, cracks, air bubbles and channeling will lead to a poor separation.

Once the column is loaded, open the stopcock and allow the solvent level to drop to the top of the packing, but do not allow the solvent layer to go below this point. Allowing this solvent level to go below the stationary phase, (known as letting the column to "run dry,") should always be avoided since it allows air bubbles and channel formation to occur leading to a poor separation.

Adding the Sample

Once the packing is complete, the sample can be loaded directly to the top of the column. Normally, a minimum amount of a polar solvent, 5-10 drops, is used to dissolve the mixture. The solution is then carefully added to the top of the column using a pipet without disrupting the flat top surface of the column. A thin horizontal band of sample is best for an optimal separation. After the sample is loaded, a small layer of white sand is added to the top of the column. This will help to keep the top of the column level when adding solvent eluent. Once the mixture is added and the protective layer of sand is in place, continuously add the solvent eluent while collecting small fractions at the bottom of the column. Using a pipet to add the first bit of solvent on top of the packing, sample, and sand will minimize disturbance of the column and diluting the sample. Collecting small fractions (1-3 mL) is important to the success of your column separation. Fractions that are too small can always be pooled together; however, if the collected fractions are too large, you may get more than one compound in any particular fraction. If this occurs, the only way to complete the separation is to redo the chromatography. Since column chromatography is time consuming, collecting large fraction is discouraged.

Monitoring the Column

If the mixture to be separated contains colored compounds, then monitoring the column is very simple. The colored bands will move down the column along with the solvent and as they approach the end of the column, collect the colors in individual containers. Use the color as your guide. However, most organic molecules are colorless. In this case, the reaction must be monitored by TLC. Spot each fraction on a TLC plate. Four or five fractions can be spotted on a single TLC plate. Develop the plate and use the observed spot or spots to determine which compound is in each of the collect fractions. Spotting some of the starting material or the product (if available) on the TLC plate as a standard will help in the identification.

Isolating the Separated Compounds

Once you believe all the materials have been removed from the column, the colors of the materials or TLC results should indicate which fractions contain the compound(s) you are interested in isolating. Combine the like or same fractions and evaporate the solvent. The pure separated compound will be left behind. Recrystallization may be used to further purify a solid product. However, on a milligram scale, there is usually not enough material to do this.

Plant Pigments

Plants use a number of different pigments in their light-harvesting systems. These compounds belong to the chlorophyll and carotenoid classes. Representative members of these groups, chlorophyll *a* and *b* and *β*–carotene are shown below.

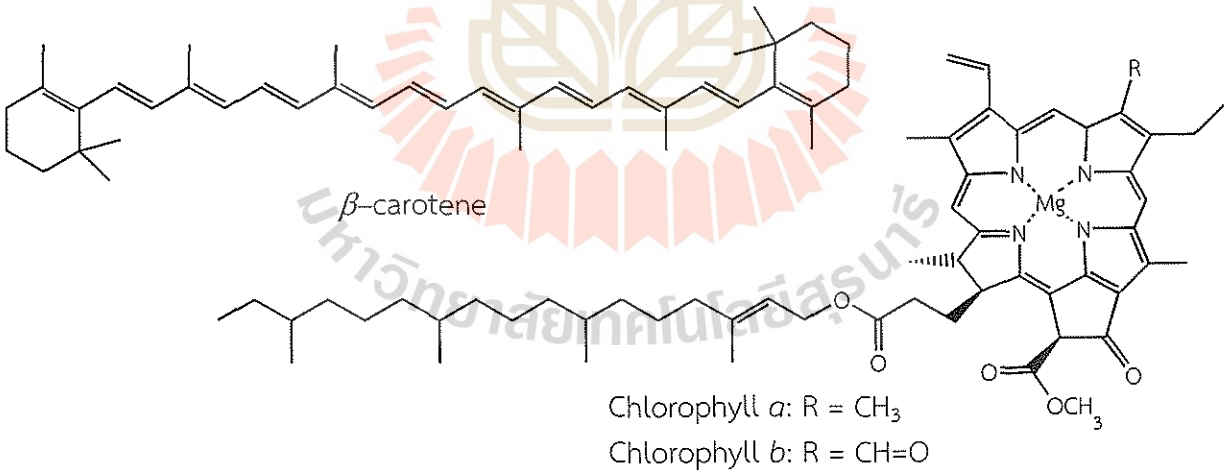


Figure 7.6 Structures of *β*–carotene and chlorophyll *a* and *b*

TLC will allow you to separate these pigments in a sample of leaf extract. You should be able to see spots from several carotenes, including *β*–carotene, *α*–carotene, whose endocyclic double bond is shifted one position (out of conjugation) relative to the β isomer, and several oxygen-containing carotene derivatives called xanthophylls. All should appear as yellow or orange spots on the TLC plate. In addition, you should see spots corresponding to the green chlorophylls *a* and *b* as well as gray spots for pheophytins *a* and *b*. Pheophytins are just the chlorophylls with the Mg^{2+} replaced by two H^{+}'s.

Experimental

In this experiment you will run TLC's of mango leaf extract using three different developing solvent systems, 1:1 cyclohexane:toluene mixture, 7:3 hexane:acetone mixture, and 2:1 hexane:ethanol mixture. From the resulting chromatograms, select the best solvent system that gives the best pigment separation. Find out how many pigments there are and what their R_f values are from the best chromatogram. You will then run a chromatographic column using the selected eluting solvent to separate these pigments. You should be able to obtain three fractions from the chromatographic column, one yellow, one green, and one yellowish green.

Experimental Procedure

A. Extraction of Mango Leaves

1. Transfer 5 g of ground mango leaves to a mortar. Add 10 mL of acetone and use a pestle to press the ground leaves to remove water from the leaves. Discard this solution.
2. Add 10 mL of hexane to the mortar and again press the ground leaves to obtain dark green solution. Use a Pasteur pipette to transfer the resulting extract into a small test tube.
3. Dry the leaf extract with anhydrous Na_2SO_4 and decant the extract into a small screw-capped vial.

B. Mobile Phase Determination by TLC Method

1. Obtain three TLC plates (silica gel with pore size of 60 Å coated on aluminum sheet, 2 x 10 cm, which have been activated to fluoresce under UV_{254} radiation) from the supply room. Handle the plates only by the edges. Using a pencil, not pen, very very lightly draw a line across the short side of each plate, on silica gel side approximately 1 cm from the bottom as shown in Figure 7.7. Be careful not to scratch the silica gel as you are drawing the line. Also lightly label at the top of each plate to indicate the developing solvent that is going to use, C:T for cyclohexane:toluene (1:1), H:A for hexane:acetone (7:3), and H:E for hexane:ethanol (2:1).

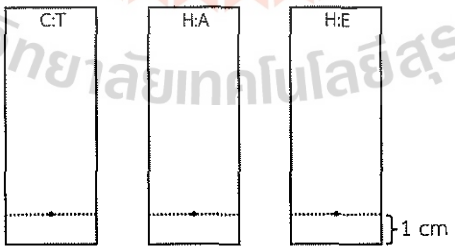


Figure 7.7 Position of the pencil line and the spot on the TLC plates

2. Set up three developing chambers as shown in Figure 7.2 using three screw-capped jars. Put a piece of filter paper cut in rectangular in each jar. Label each: C:T, H:A, and H:E. Add 1:1 mixture of cyclohexane and toluene in the jar labeled C:T, 7:3 mixture of hexane and acetone in the one labeled H:A, and 2:1 mixture of hexane and ethanol to the one labeled H:E. The solvent depth should be no more than 0.5 cm. Cap each jar and gently swirl to soak the filter paper inside.

3. Using a small capillary tube, spot your leaf extract from section *A* onto each TLC plate. (Before you spot a real TLC plate, practice on a piece of filter paper – try to make a spot as small as possible.) Make the first spot by touching the tip of the capillary tube filled with sample to the surface of the plate. Do not allow the tip of the capillary tube to touch the surface for more than a fraction of a second. If it does, it will produce a spot that is too large. Give the solvent a few seconds to evaporate, spot again, and repeat the process a few times to build up the concentration without widening the spot excessively. It is important the spot be reasonably concentrated and small.
4. Develop each TLC plate according to the solvent labeled on the plate in three separate jars at the same time. Use forceps to carefully insert each TLC plate, cap the jar, and allow the solvent to rise until it gets close to the top of the plate. Be careful not to disturb the jar. Remove the plate with forceps, mark the position of the solvent front with a pencil, and allow the solvent to evaporate. (Why doesn't it matter exactly how close the solvent gets to the top? Why does it matter that you mark exactly where the solvent front ended up immediately after you remove the plate?)
5. If something goes seriously awry (compounds all run to the edge, for example), try it again. If you can't easily see the spots, use more; if everything runs together in a big smear, you may have spotted too much, so use less. Seek advice from your instructor as necessary.
6. Circle the spots that are visible (in case they disappear due to exposure to light and air). Accurately sketch the TLC chromatograms in your report, and note the colors of the various spots. Next, expose each plate to 254-nm UV light using a UV cabinet. The silica TLC plates contain a fluorescent indicator that will glow green when exposed to 254-nm light. Compounds that absorb UV light appear as dark spots. Circle any new spots that show up.
7. From the results, decide which solvent system would be most appropriate for the separation of the pigments. Report the result to your instructor.
   Which solvent system of the three gives the best separation?
8. From the best chromatogram, calculate the R_f values of the chlorophylls, pheophytins, and carotenes/xanthophylls that you can identify based on spot color. How do the R_f values change with solvent polarity? Perhaps certain compounds are more sensitive to the polarity change than others. What does this tell you about those compounds?

C. Separation of the Pigments by Column Chromatography

1. Using a long glass rod, carefully pack a small piece of cotton in the bottom of a chromatography column. Take care not to use too much cotton or pack it too tightly since that will result in a much slower rate of solvent flow. You just need enough to prevent the adsorbent from leaking out.
2. Clamp the column to a stand and make sure that the column is securely fastened and in a vertical position, close the stopcock, and add hexane until the column is approximately 1/3 full.

3. Weigh approximately 7 g of alumina (neutral form) into a 50 mL beaker. Using a dry spatula and a dry short-stem glass funnel, slowly sprinkle the dry alumina into the hexane in the column while you tap the column.
4. After all of the alumina has been added to the column, let the alumina settle and gently tap the column so that the alumina will pack tightly into the column and the surface of the alumina will be level. Place a beaker under the column, open the stopcock, and continue to tap the column as you allow the hexane to drain slowly. Using a Pasteur pipette, add more hexane as necessary to remove alumina that may stick to the inner wall of the column and allow the hexane to drain into the beaker.
5. When finish packing, the hexane is drained out until it just barely covers the surface of the alumina, which should be perfectly flat. Using a Pasteur pipette, slowly add 1 mL of your leaf extract to the top of the surface of the alumina by placing the tip of the pipette against the inside wall of the column and letting the sample to flow gently into the column. The flat surface of the alumina should be minimally disturbed. Use a few drops of fresh hexane to transfer the remainder of your sample onto the column. Drain some hexane from the column until the sample just barely covers the surface of the alumina. Then, add a few drops of fresh hexane to rinse the inside wall of the column, and drain out some hexane until the liquid just covers the alumina. Repeat until the sample is seen as a narrow band at the top of the column.
6. Using a dry spatula and a dry short-stem glass funnel, carefully add a 4 to 5 mm layer of sand into the column, and use hexane to remove sand that may stick to the inner wall of the column. Drain the hexane into the beaker until the hexane level is just even with the surface of the alumina.
7. Change the solvent from hexane to the solvent of choice (from step 7 in section *B*) by carefully adding the solvent into the column, and drain the solvent into the beaker. Now you are ready to begin the eluting process.
8. Add fresh solvent as necessary. Do not disturb the sand by adding carefully the solvent into the column using a Pasteur pipette by placing the tip of the pipette against the inside wall of the column and letting the solvent to flow gently into the column. After a couple of inches of solvent have been added to the column, fill the rest of the way by pouring solvent into the column using a glass funnel. Never let the solvent level drop below the top of the alumina.
9. The colored bands will travel down the column as the pigments are eluted. As soon as the colored compound begins to elute from the column, the collection beaker is changed to a 50 mL Erlenmeyer flask. Label the flask the name or class of the pigment separated. (The process is complicated if the compound is not colored. In such experiments, equal sized fractions are collected sequentially and carefully labeled for later analysis.)
10. After the first pigment is off the column, change the collection flask to a 50 mL beaker. When the second pigment begins to elute from the column, change the collection beaker to a new 50 mL Erlenmeyer flask.
11. Repeat the process until all pigments are off the column.

12. Drain the remaining solvent in the column into a beaker. Invert the column upside down, open the stopcock and shake the column so that the alumina will come out of the column into a waste container provided. If some alumina still remains in the column, insert a pipette rubber bulb into the tip of the column and quickly press the bulb to force the alumina to come out.
13. Submit all of the pigment solutions in the Erlenmeyer flasks to your instructor.

Laboratory Safety Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. Hexane, cyclohexane, toluene, and acetone are irritating and flammable. Avoid breathing their vapors.
3. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 7

Experiment 8	Williamson Ether Synthesis

Objectives

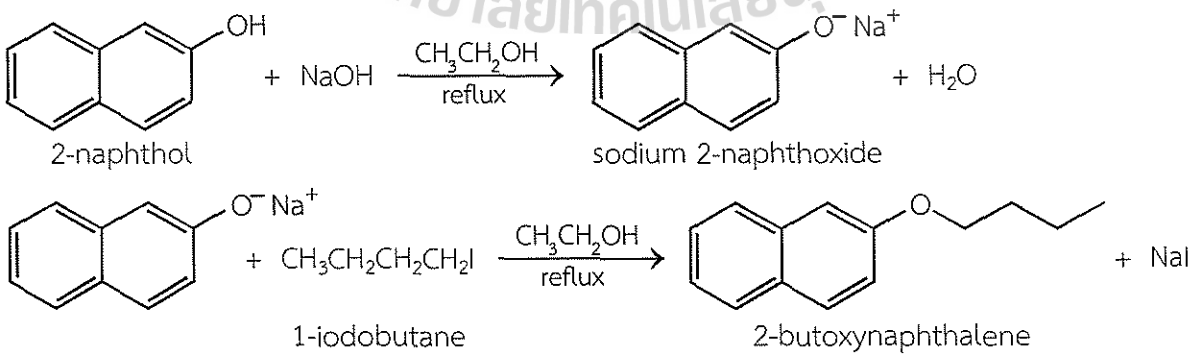
1. To demonstrate an example of an S_N2 reaction, the Williamson ether synthesis.
2. To prepare 2-butoxynaphthalene from 2-naphthol and 1-iodobutane.

Introduction

Many of the reactions used in organic chemistry are described as being named reactions. The Fischer esterification reaction was a named reaction, referring to Emil Fischer who discovered and popularized it as a method to produce esters. Likewise, the Grignard reaction was named after its discoverer. In this experiment, the Williamson ether synthesis is another named reaction, developed by Dr. Alexander W. Williamson who was a professor at University College in London in the latter part of the 1800's. This reaction has been around for a long time and has been used successfully to synthesize many different ethers.

Ethers can be produced from two alcohols. However, unless you want to have symmetrical ethers (e.g. diethyl ether ($CH_3CH_2OCH_2CH_3$) derived from ethanol), ether synthesis from different alcohols in the same reaction mixture will produce a variety of products. To produce an unsymmetrical ether (e.g. *t*-butyl methyl ether, $(CH_3)_3COCH_3$), the Williamson ether synthesis is employed. One component of the reaction is an alkyl halide and the other component is the alkoxide (or phenoxide) ion produced from an alcohol. For this reaction to occur at a high yield, the alcohol portion can be either primary, secondary, or tertiary, which can then be converted into an alkoxide ion, a better nucleophile than the alcohol, using basic conditions. The alkoxide ion then reacts via an S_N2 reaction mechanism with a primary alkyl halide. If the alkyl halide was either secondary or tertiary, an E2 reaction would likely take place instead of substitution.

In this experiment, you will be using the Williamson reaction to make 2-butoxynaphthalene as shown in Scheme 8.1.



Scheme 8.1 The Williamson ether synthesis of 2-butoxynaphthalene

The first step of the reaction is an acid-base reaction, where the acidic proton of 2-naphthol is removed with a strong base, sodium hydroxide. As you know, sodium hydroxide is not

sufficiently strong to remove the hydrogen from alcohols; however, 2-naphthol is a different story. The proton of 2-naphthol is nearly one million times more acidic than alcohols, due to the resonance stabilization of the 2-naphthoxide ion. The extra stability of the 2-naphthoxide ion compared to an alkoxide ion makes reaction of 2-naphthol with NaOH favorable. Because 2-naphthol is soluble in ethanol and to minimize the use of highly volatile organic solvents, ethanol is the preferred solvent. Although polar aprotic solvents are optimal for S_N2 reactions, the ideal conditions may not always be the conditions chosen.

The exothermic nature of the acid-base reaction causes the ethanol to boil. To prevent the ethanol from boiling away, an upright condenser, called a reflux condenser, is attached to the reaction flask. Ethanol vapors condense in the condenser and the liquid runs back into the flask.

The ethanol is used in a small volume so that the rate of reaction of 2-naphthoxide ion with 1-iodobutane is fast enough to permit the entire reaction sequence to be carried out in a single laboratory period.

Once the sodium 2-naphthoxide has been formed, 1-iodobutane is added dropwise. As the reaction proceeds, sodium iodide precipitates. After all the 1-iodobutane has been added, the reaction mixture is heated at reflux for 1 hour to complete the reaction. Refluxing is a term used to describe boiling the reaction for a period of time with a condenser on top of the round-bottom flask to capture the vapor by cooling and then allow the condensed solvent to drop back into the flask. This allows the reaction mixture to heat at a constant temperature (equal to the boiling point of the solvent), thus allowing the reaction to proceed with heat added but without the loss of solvent over time.

After the reaction has been carried out, the product must be isolated from the reaction mixture and purified. The general procedure is termed work-up. A specific work-up procedure is dictated by the physical and chemical properties of the products and by-products in the mixture. In this experiment, the first step in the work-up is the addition of the reaction mixture into ice water to dissolve the sodium iodide and to reduce the solubility of the 2-butoxynaphthalene in the reaction mixture (2-butoxynaphthalene is a solid and not soluble in water).

The next step in the work-up procedure is to separate the solid product ether from the solution by suction or vacuum filtration using ice cold water to wash the crystals. The resulting solid is allowed to dry. The purity of the product can then be checked by TLC analysis and its identity by melting point determination.

The 2-butoxynaphthalene is attractive for several reasons, including the following:

1. It is easily prepared crystalline solid whose melting point can be used to confirm its identity.
2. It is a flavoring agent that has a strong fruity taste reminiscent of raspberry and strawberry.
3. It is a precursor to photoacid generators, which are compounds that absorb photons and then release protons.

Experimental Procedure

1. To a 100 mL round-bottom flask, add 3 boiling chips, 5-6 pellets of sodium hydroxide, 1.0 g of 2-naphthol, and 20 mL of ethanol.
2. Set up the flask and its contents for refluxing as follows:
   a. Put the flask in a heating mantle.
   b. Insert a reflux condenser (with rubber hoses attached) into the round-bottom flask and clamp the flask so that it is sitting snugly in the heating mantle. The reflux condenser should be in an upright position, as shown in Figure 8.1.

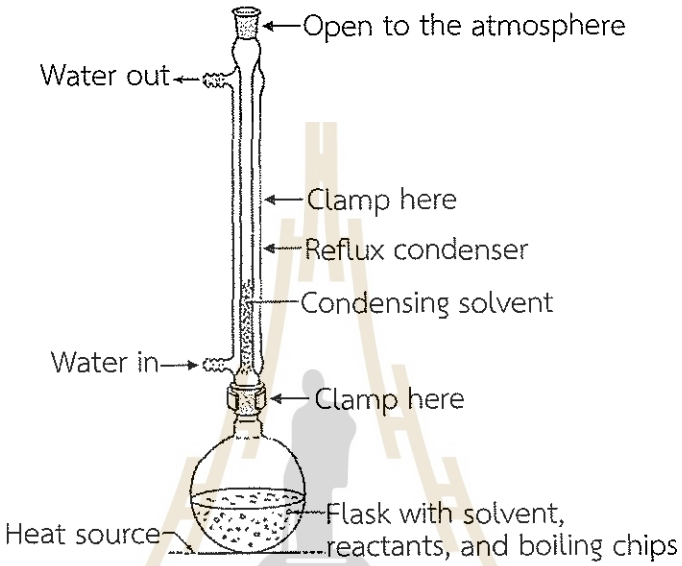


Figure 8.1 Reflux by round-bottom flask

   c. Ask your instructor to inspect your apparatus setup.
   d. Turn on the water to the reflux condenser (carefully!) and make sure that the water in the condenser is cold before you start to heat the flask.
   e. Heat the mixture to reflux for 15 minutes (start timing once the reaction starts to boil).
   f. Raise the flask from the heating mantle and allow the solution to cool for 2 minutes. All the solids should be dissolved by now.
   g. Remove the condenser and add 1.0 mL of 1-iodobutane (d = 1.610 g/cm^3) dropwise into the round-bottom flask.
   h. Replace the condenser and reflux the solution for an additional hour. Remember to start timing when you first see the liquid dropping from the reflux condenser.
3. After 1 hour of reflux period is over, remove the heating mantle and allow the reaction mixture to cool for 1 minute.
4. Check the composition of the reaction mixture by TLC analysis using a silica gel TLC plate (2 x 10 cm) and a developing system of ethyl acetate:hexane (1:9) in the following manner.
   a. Prepare a developing chamber using a small screw-capped jar containing a filter paper and the 1:9 mixture of ethyl acetate and hexane no more than 5 mm in depth.

b. Using a TLC spotting capillary tube, place a spot of the 2-naphthol starting material dissolved in ethyl acetate (this TLC reference solution is available in the hood) on the TLC plate about 0.5 cm from the edge of the plate.
c. Using a clean TLC spotting capillary tube, place a spot of the reaction mixture next to the starting material on the TLC plate about 1 cm apart and 0.5 cm from the edge of the plate.
d. Develop the TLC plate, and view the plate under 254-nm UV light.

5. While the TLC plate is developing. Pour the reaction mixture onto 25 g of ice in a 250 mL beaker. The product will rapidly crystallize. Rinse the round-bottom flask with about 20 mL of cold water and add this to the beaker containing the ice and the product. To maximize yield, keep the solution in the ice water for several minutes after the crystallization begins.
6. Collect the solid product on a Büchner funnel using vacuum filtration and pre-weighed filter paper. Remember to use cold water to rinse the solid product. If any solid product appears crystallizing in the filtrate, filter a second time to collect the remainder of the product.
7. Allow the solid to dry in a desiccator until your next laboratory period.
8. After drying, weigh the product to determine your percent yield. Also take a TLC (use acetone as a solvent for sample preparation and 1:9 mixture of ethyl acetate and hexane as a developing solvent) and melting point of your product to check its purity and identity, respectively. The literature value for the melting point of 2-butoxynaphthalene is 33-35 °C.
9. Hand in the product in a properly labeled container.

Laboratory Safety Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. 2-Naphthol and 1-iodobutane are irritants and harmful to the environment. In addition 1-iodobutane is flammable.
3. Sodium hydroxide is corrosive to both clothing and skin. If you spill any on yourself, rinse it immediately with water.
4. Ethanol causes eye and respiratory tract irritation and is flammable.
5. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 8

Experiment 9	Qualitative Analysis

Objectives

1. To study positive and negative results of each qualitative chemical test.
2. To identify 8 unknown compounds according to their qualitative chemical tests.

Introduction

Chemists frequently use qualitative patterns of reactivity to identify the functional groups of unknown compounds. This technique, called qualitative analysis, was an especially important tool for structure determination in the early days of organic chemistry. An alkene, for example, can be identified by its reaction with Br_2 – disappearance of the red-brown color of the bromine provides clear visual evidence that a reaction has occurred. Similarly, upon treatment with chromic acid, certain functional groups are oxidized, and this is accompanied by reduction of the orange Cr(VI) to the blue-green Cr(III) – an obvious change.

Since the development of powerful spectroscopic methods for structure determination, including infrared (IR), and especially nuclear magnetic resonance (NMR, which we will study later), most of these qualitative chemical tests have become less important than they once were. However, these tests are quick and easy, and they are still useful for identifying functional groups or confirming the presence of functional groups identified spectroscopically. Perhaps more important for our purposes, these tests will familiarize us with the chemical behavior of a variety of different compounds, and they will give us a chance to learn some new chemistry.

In this lab you will use qualitative chemical tests to identify eight compounds. These have been placed in plain brown bottles labelled "A" – "H" to protect their identities. Fortunately, you will not have to determine the entire structure of each compound (you could not do that at this point in the course even if you wanted to). The compounds are butan-1-amine, butan-2-one, butan-1-ol, ethanal, butanoic acid, 1-bromobutane, hexane, and cyclopentene. Since each compound has a different functional group (or none), you will be able to determine which compound is in which bottle by determining what functional group is present.

Chemical Tests

A combination of physical and chemical tests will be used to determine what functional groups are present. Each of these tests should give an easily visible result. The different tests are explained in detail and the procedures are described below. The section that follows the individual tests will guide you through the process of putting it all together and developing a scheme that you can use to identify each of the compounds. If you work this out before you come to the lab, the experiment should go very quickly and you will be out early.

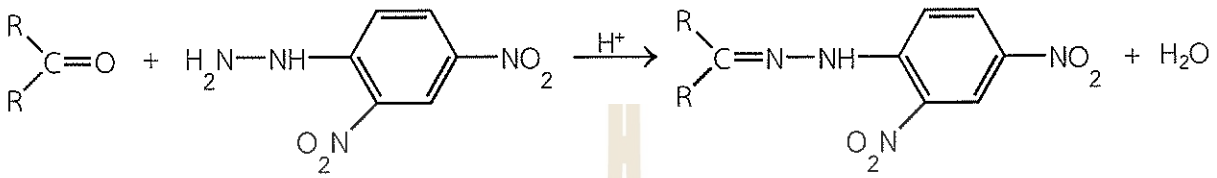
Water Solubility and Litmus Tests

Because the molecules in our set of 8 are relatively small, a polar functional group – especially one capable of hydrogen bonding – will probably make the compound miscible with, or at

least soluble in, water. For water soluble compounds, test the pH of the solutions. Note that it is not necessary for a compound to be completely protonated or deprotonated by water to alter the pH.

2,4-Dinitrophenylhydrazine Test

Ketones and aldehydes react with hydrazines to form compounds called hydrazones. For example, the reaction of 2,4-dinitrophenylhydrazine (2,4-DNPH) with a generic ketone is shown in Scheme 9.1.



2,4-dinitrophenylhydrazine (2,4-DNPH)

2,4-dinitrophenylhydrazone (2,4-DNP)

Scheme 9.1 A reaction for 2,4-dinitrophenylhydrazine test

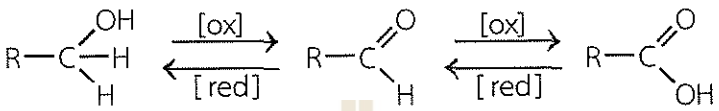
Although this is a reaction you have not seen before, you can write the mechanism. There are two steps and some "proton shuffling". The N attached to the aromatic ring is not nucleophilic because its lone pair is delocalized by resonance; the "end" nitrogen is the nucleophilic one. This N attacks the carbonyl C, displacing the π–electrons onto O – that is step 1 – formation of the CN bond. Next, the negative O is protonated and the positive N is deprotonated (H^+ is transferred to the solvent). The resulting "carbinolamine" then undergoes an acid-catalyzed dehydration (you already know that mechanism) to form the CN π–bond of the product – O is protonated, then water is lost (that is "step 2" – cleavage of the CO bond), the resonance-stabilized cation spits out a proton and a hydrazone results.

2,4-Dinitrophenylhydrazones ("2,4-DNP's") are typically yellow or orange crystalline solids. The hydrazones derived from simple aldehydes and ketones are usually yellow; those derived from conjugated carbonyl compounds are usually orange or red-orange. Hydrazones of simple aldehydes and ketones normally form quite easily. (Complications are sometimes encountered as a result of impurities, formation of colored complexes with certain compounds, and side-reactions that can give false positive tests, but these complications should not be a major concern for us. However, this is why we also need to run control experiments so we can see what authentic positive and negative results look like.)

Chromic Acid Test

Certain alcohols, aldehydes, and a few other types of compounds can be oxidized with aqueous chromic acid, H_2CrO_4. Oxidation of the organic compound is accompanied by reduction of the chromium from the soluble orange Cr(VI) reagent to an insoluble blue-green Cr(III) product.

Most oxidations of organic compounds involve loss of hydrogens and/or gain of oxygens or gain of bonds to oxygen. For example, a primary alcohol (see Scheme 9.2) – one whose –OH group is attached to a primary C – can be oxidized by Cr(VI) (as well as other reagents) to an aldehyde; most oxidants, including aqueous chromic acid, will further oxidize the aldehyde to a carboxylic acid. At this stage the C has no more H's, so that is the end of the road – carboxylic acids cannot (easily) be oxidized further. Note the exchange of C–H bonds for C–O bonds in each step. (To recognize oxidations and reductions, you need to focus on what is happening to the C not the O.)



Scheme 9.2 Oxidation of a primary alcohol to an aldehyde and to a carboxylic acid

Secondary alcohols are oxidizable (to ketones), but tertiary alcohols are not. Draw these and you will see why – the key is whether the C bearing the hydroxyl group has an H or not – if not, that C is not oxidizable. To take this idea one step further, unlike all other carboxylic acids, formic acid, HCO_2H can be oxidized to CO_2 (or to the "hydrate" of CO_2, H_2CO_3). Formic acid is the only oxidizalbe carboxylic acid because it is the only one with an H on the carboxyl C.

Alcohols are normally oxidized rapidly with chromic acid, H_2CrO_4; aldehydes are oxidized a bit more slowly, possibly because their oxidation occurs via an intermediate hydrate ($RCH(OH)_2$, formed by addition of water to the CO π–bond). In addition, chromic acid will oxidize phenols and may also oxidize amine nitrogens and even alkenes, alkynes, and certain ketones under more extreme conditions. The amine should be out of the picture by the time you get to this test, but you should run controls on the other functional groups.

Permanganate Test

In basic solution at room temperature $KMnO_4$ will hydroxylate an alkene (i.e. add two –OH groups to the π–bond, like the OsO_4 reaction we learned in lecture); in neutral or acidic solution, it will chop an alkene into two pieces by cleaving both σ– and π–bonds (like ozonolysis, but messier). Both of these reactions are oxidations of the organic compound; the manganese is reduced from Mn(VII) to Mn(IV) in the process. This reduction changes the purple permanganate solution to a brown precipitate of MnO_2.

Under the relatively mild conditions of our test (dilute, neutral permanganate solution), only alkenes and alkynes should react. However, other oxidizable compounds may react as well, e.g. amines, phenols, aldehydes, and alcohols (surprisingly, most alcohols are not oxidized under these conditions). Also, be aware that a tiny amount of oxidizable impurity might cause a false positive with the first drop of permanganate added. Again, these are the reasons that we run controls – so we can see what authentic positive and negative results look like.

Beilstein Test

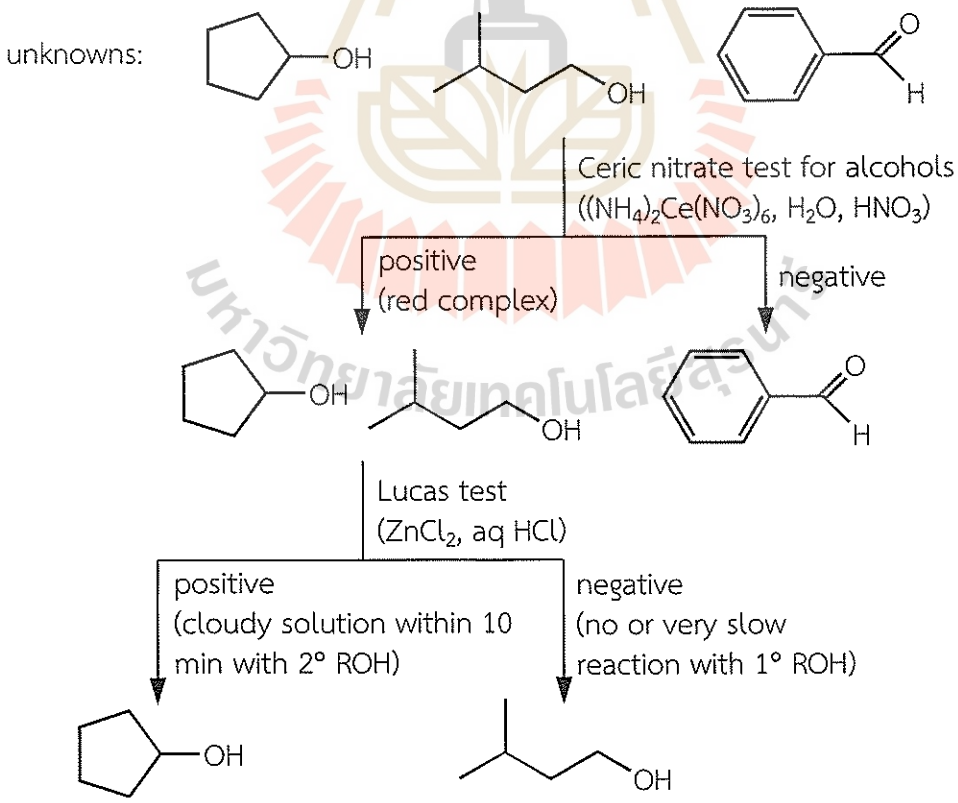
This is a classic test for halogenated compounds. When red-hot Cu is placed in contact with an organohalide, some CuX is formed. Cuprous halides are volatile enough that placing them in a flame will generate a green color in the flame from the vaporized CuX. The green is a characteristic of the copper atoms.

Putting It All Together

To do this lab quickly and sensibly you need to do a little preparation before you start. At this point you have written down the structures of the eight unknowns and the five knowns and categorized them according to functional groups. If you have not done that yet, now would be a good time.

Experimental

Write a flow chart that shows which compounds should give positive and negative results for each of the tests in the following sequence: litmus test (here there are three options, neutral, acidic, or basic), 2,4-DNPH test, H_2CrO_4 test, $KMnO_4$ test, and Beilstein flame test. Once you have positively identified a compound, you can set that aside. Do not continue running tests. For example, if one compound tests acidic with litmus, you know which one that is. Do not bother running the other tests on that one. As an example, a flow chart for a completely different series of compounds and tests is shown in Scheme 9.3 below.



Scheme 9.3 Sample flow chart

Your flow chart is the most important part of your pre-lab write-up. You must do this carefully and thoughtfully before you can start the experiment. Your instructor will check the flow charts early in the lab so potential points of confusion can be identified before anyone gets completely off track.

Each partner should participate in each part of this experiment and independently record observations. Do not split the tasks and exchange data later!

Experimental Procedure

A. Water Solubility and Litmus Tests

1. To get an idea of the water solubility of each compound, add about 1 mL of water to 10 drops of compound in a test tube, stopper the tube and shake it vigorously. If a homogeneous solution results, the compound is extremely soluble or miscible in water. If the compound does not completely dissolve, dilute the mixture with water to a total volume of 10 mL and shake. If the compound completely dissolves, we will call it "soluble"; if not, we will call it "insoluble".
2. Based on their structures, which compounds do you expect to be water-soluble and which insoluble? This may give you some ideas about which is which, but you will still need to carry on with the litmus test and then follow the sequence dictated by your flow chart.
3. Test the pH of all the homogeneous solutions by placing a drop on pH paper (do not dip the paper into the solution. One of the compounds in the set should test acidic; one should test basic. Which ones? Now for the compounds that does not dissolve at all, it is pointless to test the pH of the water, so do not do that. Discard these solutions and mixtures when you are finished – the remaining tests must be done starting with the pure compounds.

B. 2,4-Dinitrophenylhydrazine Test

1. Dissolve 1 drop of compound in about 1 mL of 95% aqueous ethanol, and add 2 mL of the 2,4-dinitrophenylhydrazine reagent (this is a solution of the hydrazine (about 2%) in ethanol (50%), H_3PO_4 (40%), and H_2O (10%)). Gently shake the mixture and let it stand.
2. Run two controls: benzaldehyde (known positive) and propan-1-ol (known negative).

C. Chromic Acid Test (Jones Reagent)

1. Dissolve 1 drop of compound in about 1 mL of reagent grade acetone. Add one drop of the chromic acid reagent (this is made by dissolving 5.0 g of CrO_3 in 5 mL of concentrated H_2SO_4 and diluting with 15 mL of water). Gently swirl the mixture and let it stand. Note the time required for reaction. A positive test should be visible within a few minutes.
2. Run four controls: benzaldehyde (known positive), propan-1-ol (known positive), 2-methylpropan-2-ol (known negative), and cyclohexene (known negative).

D. Permanganate Test

1. Dissolve 1 drop of the pure compound in 2 mL of 95% aqueous ethanol, and add 5 drops of 1% aqueous $KMnO_4$ solution. Remember, decolorization of just the first drop may be due to an oxidizable contaminant rather than the compound of interest. If a reaction does

not take place immediately, shake the mixture and let it stand for 5 minutes. This should be sufficient time for even the most stubborn alkene to decide to react; anything that happens after this time is probably due to oxidation of a less reactive functional group. Even alcohols tend not to react within 5 minute under these conditions (strange, but true).

2. Run four controls: cyclohexene (known positive), cyclohexane (known negative), benzaldehyde, and propan-1-ol (Do they turn brown? Green? Start smoking? Grow hair?)

E. Beilstein Test

1. Get about 10 cm copper wire and make a small loop at the end. Heat the loop in a flame until it is glowing red.
2. Plunge the red-hot copper loop into about 0.5 mL of the compound to be tested (use a small vial as a container), then return it to the flame and watch carefully.
   Note that Bunsen burner has an air intake that may need to be adjusted – a yellow flame (too much gas, not enough air) is too cool and too bright; you need a blue flame for this experiment. Be careful not to set fire to the compounds, your notebook, paper towels, yourself, your partner, or anything else.
3. Do not run controls this time; at this point you should be down to two compounds, one that creates fireworks and one that does not.

Laboratory Safety Precaution

1. Wear safety goggles and lab coat at all times while working in the laboratory.
2. Most of the unknown liquids are flammable, and their vapors may be harmful. Some can cause irritation on contact with eyes or skin. Keep them away from open flames and avoid unnecessary contact or inhalation of vapors.
3. The 2,4-dinitrophenylhydrazine reagent and the chromic acid reagent are toxic and corrosive. Chromic acid is a suspected carcinogen.
4. The aqueous potassium permanganate solution is likely corrosive.
5. Wash your hands thoroughly with soap or detergent before leaving the laboratory.

NOTE FOR EXPERIMENT 9