SOP: CRYSTALLIZATION

1. Crystallization

Crystallization (or recrystallization) is the most important method for purification of organic compounds. The process of removing impurities by crystallization involves dissolving a compound in an appropriate hot solvent, allowing the solution to cool and become saturated with the compound being purified, allowing it to crystallize out of the solution, isolating it by filtration, washing its surface with cold solvent to remove residual impurities, and drying.

Crystallization is the process of precipitating crystals from a solution due to changes in solubility conditions of the solute in the solution. This is a separation technique which is similar to regular precipitation.

Precipitates are solids consisting of particles in a solution. Sometimes solids are a result of a chemical reaction in a solution. These solid particles will eventually settle down due to their density, and it is known as a precipitate. In centrifugation, the resulting precipitate is also known as the pellet. The solution above the precipitate is known as the supernatant. The particle size in the precipitate changes from occasion to occasion. Crystals can be easily filtered, and they are larger in size.

The difference in crystallization method from the normal precipitation is that, the resulted solid is a crystal. Crystalline precipitates are more easily filtered and purified. The crystal particle size can be improved by using dilute solutions and adding the precipitating reagent slowly while mixing. The quality of the crystal and the improvement in filterability can be obtained from the dissolution and recrystallization of the solid. Crystallization can be seen in nature too. It is most often carried out artificially for various types of crystal production and purification.

Crystallizations play an important role in the development of new systems, particularly in those cases where the structure function relationship is to be studied. As well as purification these techniques can be used to create new compounds. In this case the components to be reacted are dissolved separately and added together, using the principles outlined here.

2. Recrystallization

Recrystallization is a technique to purify the crystals obtained from crystallization method. Though crystallization separates the compound in almost pure form, when the crystals form some of the impurities may trap in it. By recrystallization method, these impurities can be removed to a greater extend.

Normally the crystals are dissolved in very little amount of hot solvent and allowed to dissolve completely. When this is allowed to cool slowly (may be after filtering), the crystals may appear again. These crystals are free of the impurity. The crystals can be separated by filtering the solution again. Recrystallization process can be carried out in several ways, and several times to increase the purity of the desired crystal.

What is the difference between Crystallization and Recrystallization?

- Recrystallization is done to crystals formed from a crystallization method.
- Crystallization is a separation technique. Recrystallization is used to purify the compound received from crystallization.

CRYSTALLIZATION
The process by which we prepare crystals of a substance is called crystallization.
In the process of crystallization following steps are involved.

1. Preparation of solution.
2. Filtration.
3. Crystal formation. (Cooling)
4. Drying of crystals

PURIFICATION OF SUBSTANCES
Substances can be purified by crystallization.

STEPS OF PURIFICATION

1. Preparation of solution
   - A definite amount of given substance is dissolved in a specific amount of solvent e.g. water in a beaker to prepare an aqueous solution of the substance.
   - The beaker is heated to dissolve the maximum amount of the solute.
   - The solution must be saturated.

2. Filtration of solution

In this step the solution is filtered while still hot. The insoluble impurities are separated.

3. Crystal formation

The filtered solution is cooled to produce crystals of the substance.

4. Drying of crystals

Crystals are still wet and are usually dried by solar heat or by placing them between the paper folds to remove excess moisture.

STEP 1: Choose the appropriate solvent. Remember "like dissolves like". For example, sugar and salt are water, but not oil, soluble -- and non-polar compounds such as hydrocarbons will dissolve in nonpolar hydrocarbon solvents such as hexane.

1. The ideal solvent has these properties:
   - It dissolves the compound when the solution is hot but not when it is cold.
   - It will either not dissolve the impurities at all (so they can be *filtered out* when the impure compound is dissolved), or such that it will dissolve them very well (so they will remain in solution when the desired compound is *crystallized out*).
   - It will not react with the compound.
   - It is non-flammable.
   - It is non-toxic.
   - It is cheap.
   - It is very volatile (so it can be removed easily from the crystals).
2. It is often difficult to decide upon the best solvent; the solvent is often chosen by experimentation, or by using the most non-polar solvent available.

3. Here is a list of solvents (from most polar to least polar). Note that solvents adjacent to one another are miscible (they will dissolve in each other). Commonly used solvents are in bold.
   - **Water (H_2O)** is non-flammable, non-toxic, cheap, and will dissolve many polar organic compounds; its drawback is high boiling point (100^0C), making it relatively non-volatile and difficult to remove from crystals.
   - **Acetic acid (CH_3COOH)** is useful for oxidation reactions, but will react with alcohols and amines, and is therefore difficult to remove (boiling point is 118^0C).
   - **Dimethyl sulfoxide (DMSO), methyl sulfoxide (CH_3SOCH_3)** is used mainly as solvent for reactions; rarely for crystallizations.
   - **Methanol (CH_3OH)** is a useful solvent that will dissolve compounds of higher polarity than will other alcohols.
   - **Acetone (CH_3COCH_3)** is an excellent solvent; its drawback is low boiling point of 56^0C.
   - **2-Butanone, methyl ethyl ketone, MEK ($CH_3COCH_2CH_3$)** is an excellent solvent with boiling point 80^0C.
   - **Ethyl acetate ($CH_3COOC_2H_5$)** is an excellent solvent with boiling point 78^0C.
   - **Dichloromethane, methylene chloride (CH_2Cl_2)** is useful as a solvent pair with ligroin, but its boiling point, 35^0C, is too low to make it a good crystallization solvent.
   - **Diethyl ether ($CH_3CH_2OCH_2CH_3$)** is useful as a solvent pair with ligroin, but its boiling point, 40^0C, is too low to make it a good crystallization solvent.
   - **Methyl *t*-Butyl ether ($CH_3OC(CH_3)_3$)** is cheap, good replacement for diethyl ether given its higher boiling point, 52^0C.
   - **Dioxane ($C_4H_8O_2$)** is easy to remove from crystals; mild carcinogen; forms peroxides; boiling point 101^0C.
   - **Toluene ($C_6H_5CH_3$)** is an excellent solvent for crystallization of aryl compounds and has replaced the once commonly used benzene (a weak carcinogen); a drawback is its high boiling point of 111^0C, making it difficult to remove from crystals.
   - **Pentane (C_5H_{12})** is widely used for nonpolar compounds; often used as solvent pair with another solvent.
   - **Hexane (C_6H_{14})** is used for nonpolar compounds; inert; often used in a solvent pair; boiling point 69^0C.
   - **Cyclohexane (C_6H_{12})** is similar to hexane, but cheaper, and has boiling point 81^0C.
   - Petroleum ether is a mixture of saturated hydrocarbons of which pentane is a chief component; cheap, and used interchangeably with pentane; boiling point 30-60^0C.

Steps for choosing the solvent:

1. Put a few crystals of the impure compound in a test tube and add a single drop of the solvent, allow it to flow down by the side of the tube.
2. If the crystals dissolve immediately at room temperature, *reject the solvent* because too much of the compound will remain dissolved at low temperature, and try another solvent.
3. If the crystals do not dissolve at room temperature, warm the tube on a hot sand bath and observe the crystals. Add a drop more solvent if they do not dissolve. If they dissolve at the boiling point of the solvent and then crystallize again when cooled to room temperature, you have found an appropriate solvent. If not, try another solvent.
4. If after a process of trial and error -- and no single satisfactory solvent is found -- use a solvent pair. Dissolve the crystals in the better solvent (the one in which they are readily dissolved), and add the poorer solvent to the *hot* solution until it becomes cloudy (the solution is saturated with the solute). The solvent pair must be miscible with each other.

5. Some useful solvent pairs are:
    a) acetic acid-water,
    b) ethanol-water,
    c) acetone-water,
    d) dioxane-water,
    e) acetone-ethanol,
    f) ethanol-diethyl ether,
    g) methanol-2Butanone,
    h) ethyl acetate-cyclohexane,
    i) acetone-ligroin, ethyl acetate-ligroin,
    j) diethyl ether-ligroin,
    k) dichloromethane-ligroin,
    l) toluene-ligroin.

STEP 2:
Dissolve the impure compound: To do so, place that compound in a test tube.

1. Crush large crystals with a stirring rod to promote dissolving.
2. Add the solvent drop by drop.
3. To remove non-soluble, solid impurities, use excess solvent to dilute the solution and filter out the solid impurities at room temperature (see step 4 for filtration procedure), then evaporate the solvent.
4. Prior to heating, place a wood applicator stick; or alternatively, porous porcelain boiling chips may be used, in the tube to avoid super heating (heating of the solution above the solvent's boiling point without actually boiling).
5. The air trapped in the wood/boiling stone will come out to form *nuclei* to allow even boiling.
6. After solid impurities have been removed and the solvent evaporated, add solvent drop by drop, while stirring the crystals with a glass rod and warming the tube on a steam bath or sand bath, until the compound is completely dissolved with the minimal amount of solvent.

STEP 3:
Decolorize the solution.

1. Skip this step if the solution is colourless or has only a light shade of yellow.
2. If the solution is coloured (which results from production of high-molecular weight by-products of chemical reactions), add excess solvent and activated charcoal (carbon), and boil the solution for a few minutes.
3. The coloured impurities will adsorb onto the surface of activated charcoal, due to its high degree of microporosity.
4. Remove the charcoal with adsorbed impurities by filtration, as described in the next step.

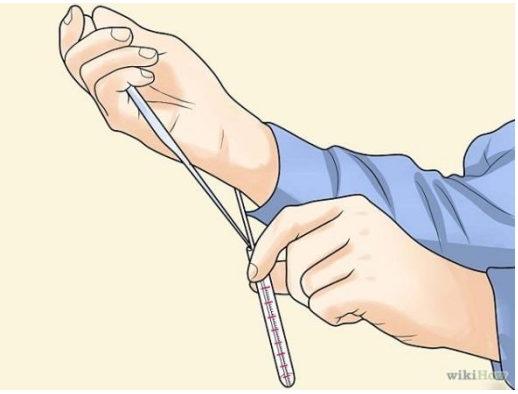
STEP 4:
Remove solids by filtration. Filtration can be done by gravity filtration, decantation, or removal of solvent using a pipette. Generally, do not use vacuum filtration, as the hot solvent will cool during the process, allowing the product to crystallize in the filter.

Gravity filtration: this is the method of choice for removing fine charcoal, dust, lint, etc.

1. Get three Erlenmeyer flasks heated on a steam bath or hot plate: one containing the solution to be filtered, another one containing a few millilitres of solvent and a stemless funnel, and the third containing several millilitres of the crystallizing solvent to use for rinsing.
2. Place a fluted filter paper (useful since you are not using vacuum) in a stemless funnel (stemless to prevent the saturated solution from cooling and clogging the stem with crystals) over the second Erlenmeyer flask.
3. Bring the solution to be filtered to a boil, grasp it in a towel/heat resisting gloves, and pour the solution into the filter paper.

4. Add the boiling solvent from the third Erlenmeyer flask to any crystals already formed on the filter paper.
5. Rinse the first Erlenmeyer flask that contained the solution being filtered, adding the rinse to the filter paper.
6. Remove excess solvent by boiling the filtered solution.

Decantation: This is used for large solid impurities. Simply pour off (decant) the hot solvent, leaving the insoluble solids behind.
Removal of solvent using a pipette: This is used for a small amount of solution and if the solid impurities are large enough. Insert a pipette with square tip into the bottom of the test tube (rounded bottom), and remove the liquid using suction, leaving solid impurities behind.



STEP 5:
Crystallize the compound of interest. This step assumes that any coloured impurities and insoluble impurities have been removed by the appropriate steps above.

1. Remove any excess solvent by boiling it off or blowing it off with a gentle stream of air.
2. Start from a solution saturated with the solute at the boiling point.
3. Allow it to cool *slowly* to room temperature.
4. Crystallization should begin. If not, initiate the process by adding a seed crystal or scratching the inside of the tube with a glass rod at the liquid-air interface.
5. Once crystallization has begun, take care not to disturb the container to allow formation of large crystals.
6. To promote slow cooling (which allows larger crystals to form), you can insulate the container with cotton or paper towels.
7. Larger crystals are easier to separate from impurities.
8. Once the container has completely cooled to room temperature, cool it in ice for about five minutes to maximize the amount of crystals.

STEP 6:
Collect and wash the crystals: To do this, separate the crystals from the ice-cold solvent by filtration. This can be done using the Hirsch funnel, the Buchner funnel, or by removal of the solvent using a pipette.

Filtration using the Hirsch funnel:

1. Place the Hirsch funnel with *non-fluted* filter paper in a vacuum tight fitted flask.
2. Place the filter flask in ice to keep the solvent cold.
3. Wet the filter paper with the crystallization solvent.
4. Hook the flask to an aspirator, turn on the aspirator, and ascertain that the filter paper is pulled down onto the funnel by the vacuum.
5. Pour and scrape the crystals onto the funnel, and break the vacuum as soon as all liquid is removed from the crystals.

6. Use a few drops of ice-cold solvent to rinse the crystallization flask and pour that onto the funnel while reapplying vacuum, and break the vacuum as soon as all liquid is removed from the crystals.
7. Wash the crystals a few more times with ice-cold solvent to remove any residual impurities. At the end of washing, leave the vacuum on to dry the crystals.

Filtration using the Buchner funnel:

1. Place a piece of *non-fluted* filter paper in the bottom of the Buchner funnel, and wet it with solvent.
2. Fit the funnel tightly against a filter flask via a rubber or synthetic rubber adapter to allow vacuum suction.
3. Pour and scrape the crystals onto the funnel, and break the vacuum as soon as the liquid is removed into the flask as the crystals are left on the paper.
4. Rinse the crystallization flask with ice-cold solvent and add this to the washed crystals
5. Reapply the vacuum and break vacuum when the liquid is removed from the crystals.
6. Repeat and wash the crystals as many times as needed.
7. Leave the vacuum on to dry the crystals at the end.
8. Wash using a pipette: used for small amount of crystals to be washed.
9. Insert a pipette with square tip into the bottom of the test tube (rounded bottom), and remove the liquid, leaving the washed solids behind.

<u>STEP 7</u>:

Dry the washed product: Final drying for a small amount of crystallized product can be done by squeezing the crystals between sheets of filter paper and allowing them to dry on a watch glass.

HEALTH AND SAFETY ISSUES

- The key health and safety area is being aware of the hazards associated with the chosen solvent such as flash points and toxicity.
- Appropriate gloves should be worn e.g. latex if working with aqueous solvents and nitrile when dealing with organic. These details are given in the MSDS if one is unsure of the correct gloves to use.
- The usual laboratory safety measures such PPE should be observed.
- Crystallizations should be set up in a fumehood if one is working with a particularly hazardous or smelly chemical such as diethyl ether, pyridine and lutidine.
- When using a Buchner funnel it should be clumped to a stand or the grid in the fumehood.