Lab 14: Qualitative Organic Analysis

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Objectives

By the end of this laboratory, you should have developed the skills to do the following:

- Determine the functional group of an unknown compound by using classification tests.
- Understand the purpose of a synthetic derivative.

Background

Organic chemists often must identify unknown compounds. In some cases, such as a reaction, you may have a good idea of what the compound in question is. However in other cases, such as when you isolate a compound from a natural source, you may have no idea what the compound might be. In this experiment you will determine the identity of an unknown compound. First, you will need to purify your compound, then you will need to identify its functional group (it will contain only one), and finally you will need to make a derivative of the compound. You will confirm your results with boiling or melting point, IR, and NMR.

Impurities in your compound will make it extremely difficult to identify. Thus, before you do anything else, you will need to make sure your unknown compound is pure. Consider each of the following purification techniques you have learned over the course of the year.

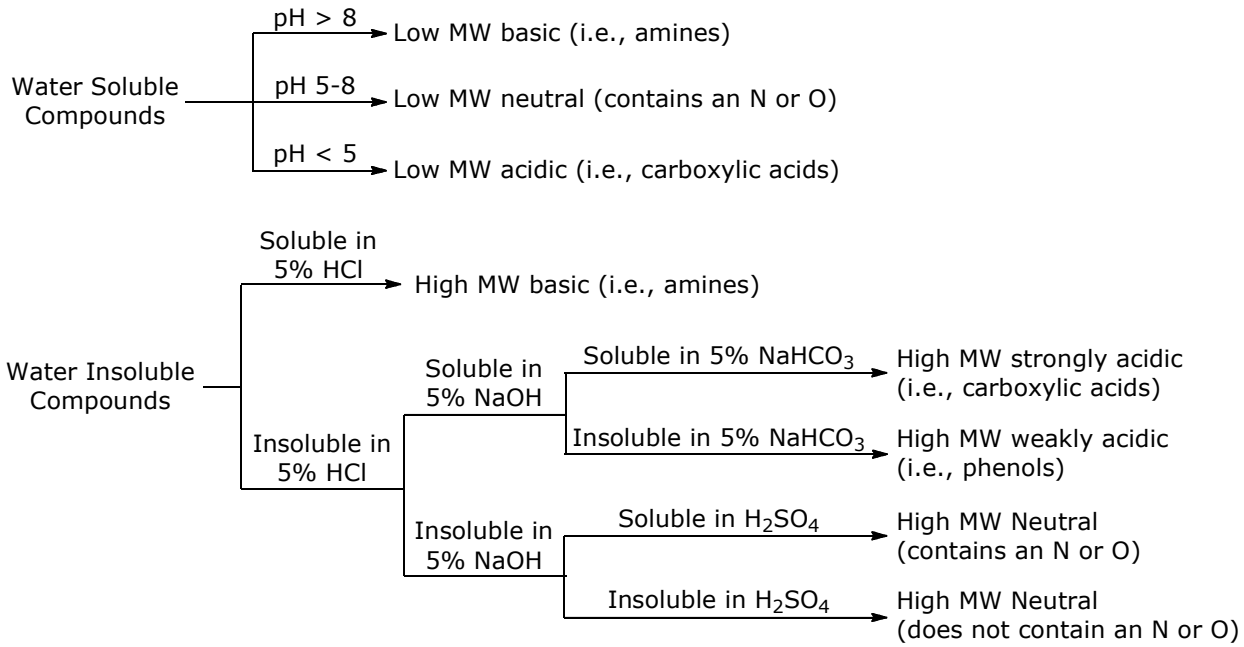
1. *Recrystallization:* Works well for solid compounds. You will need to find an appropriate recrystallization solvent. Consider a variety of solvents and mixed solvent systems.
2. *Distillation:* Works well for liquids that have a boiling point of <250 °C. (Note: Fractional distillation may be required if you suspect impurities close to the boiling point of your unknown.)
3. *Column Chromatography:* Works well for UV active compounds. You will need to use TLC to identify a solvent system that will separate your unknown from any impurities.

After you have purified your unknown, verify that it is pure enough to proceed by measuring the boiling or melting point. Note that while you will not know what the melting point or boiling point of your unknown should be, the narrowness is an excellent indicator of whether or not your product is pure. Also pay attention to the appearance of your unknown and see if it has changed (hopefully for the better) during the course of the purification process.

Once your unknown is pure, you will need to identify its functional group. Your unknown will have one major functional group (alcohol, ketone, aldehyde, amide, amine, carboxylic acid, or ester). Additionally, your unknown compound may or may not contain an aromatic ring. To determine the

functional group, it is recommended that you start with solubility tests, and then conduct functional group classification tests. IR spectroscopy may also be useful at this point.

Solubility can sometimes provide a surprisingly useful amount of information. First, you will test your unknown's solubility in water. Compounds with 4 carbons or less will easily dissolve in water, whereas compounds with 8 carbons or more will be insoluble. Compounds containing 5-7 carbons may or may not dissolve (often they will display "partial" solubility). If your compound dissolves in water, you will also want to check the pH of the solution. Amines will typically be basic, and carboxylic acids will typically be acidic. Most other compounds will be neutral. Compounds that are insoluble in water should then be subjected to a solubility test in 5% HCl. Typically, only amines will be soluble in HCl because they form water-soluble hydrochloride salts when they react with HCl. Compounds that are not soluble in HCl, should be subjected to testing in basic solutions (5% NaOH and 5% $NaHCO_3$). Both strong and weak acids (Carboxylic acids and phenols) will be deprotonated by NaOH to form water-soluble alkoxides. Only strong acids like carboxylic acids will react with $NaHCO_3$. Compounds that are not soluble in base should then be reacted with a very strong acid, sulfuric acid (note that in the case of sulfuric acid, "solubility" is also indicated by any type of reaction such as heat, gas generation, or a color change). Compounds that cannot become protonated by sulfuric acid at all (i.e., alkanes, alkyl halides, and aromatic carbons) will still remain insoluble. These solubility tests are summarized in the flow charts below.



The results from the solubility tests can significantly help in determining which classification tests should then be performed, or at least narrow down the list. By no means do you need to conduct all classification tests. In fact, you should do your best to select only tests that will provide you with additional information about your unknown and/or confirm results. Also, make sure that your glassware is clean and dry so you do not get any false positive or false negative results. Keep in mind that a negative result for a classification test provides useful information, so be sure to keep track of negative results as well as positive results. Also, for each classification test that you

perform, be sure to run a blank, and one or more controls. These will help you to determine if a reaction actually occurred. A blank includes everything but the unknown, and a control includes a compound for which the outcome is known in place of the unknown. Controls can be positive (a compound you know will react) or negative (a compound that you know will not react). The classification tests are summarized in the table below.

Functional Group	**Test**	**Test No.**	**Notes**
Alcohols	Acetyl chloride	C-1	Tests for the presence of alcohols
	Chromic acid	C-9	Tests for the presence of 1° alcohols, 2° alcohols, & aldehydes
	Iodoform test	C-16	Tests for $-CH(OH)CH_3$ and $-COCH_3$ groupings
	Lucas's test	C-17	Used to classify alcohols as 1°, 2°, or 3°
Aldehydes	Benedict's test	C-6	Tests for the presence of aldehydes
	Chromic acid	C-9	Tests for the presence of 1° alcohols, 2° alcohols, & aldehydes
	2,4-Dinitrophenylhydrazine	C-11	Tests for the presence of aldehydes & ketones
	Tollen's test	C-23	Tests for the presence of aldehydes
Amides	Alkaline hydrolysis	C-2	Tests for the presence of amides & esters
	Elemental analysis	C-12	Tests for the presence of nitrogens and halogens
Amines	Basicity test	C-4	Used to distinguish alkyl amines from aromatic amines
	Bromine water	C-8	Tests for the presence of phenols & aromatic amines
	Elemental analysis	C-12	Tests for the presence of nitrogens and halogens
	Hinsberg's test	C-15	Used to classify amines as 1°, 2°, or 3°
	Quinhydrone	C-20	Used to classify amines as 1°, 2°, or 3°
Aromatics	Aluminum chloride & chloroform	C-3	Tests for the presence of aromatic rings
Carboxylic Acids	Neutralization equivalent	C-18	Provides information about the number of carboxylic acids and the MW
Esters	Alkaline hydrolysis	C-2	Tests for the presence of amides & esters
	Ferric hydroxamate	C-14	Tests for the presence of esters
Ketones	2,4-Dinitrophenylhydrazine	C-11	Tests for the presence of aldehydes & ketones
	Iodoform test	C-16	Tests for $-CH(OH)CH_3$ and $-COCH_3$ groupings
Phenols	Bromine water	C-8	Tests for the presence of phenols & aromatic amines
	Ferric chloride	C-13	Tests for the presence of phenols

At this point, you should be able to use your boiling or melting point data combined with the results of your functional group data to develop a hypothesis as to what your unknown might be or at least narrow down the list to only a few candidates. Note that due to the accuracy (or lack thereof) of our thermometers, your boiling or melting points may be up to 15 ˚C lower than the literature values.

Once your functional group has been determined, you will prepare a derivative of your unknown. To prepare a derivative, you will select a suitable reaction that converts your unknown into a different functional group for which the boiling or melting point is known. This is particularly useful because compounds that have similar boiling or melting points will often have derivatives that differ significantly in terms of boiling or melting point. You should then be able to identify your unknown using this information.

Finally, you can confirm the identity of your product using IR and NMR. Note that these measurements can be taken at anytime during the course of the lab after you purified your product. In fact, it is recommended that you conduct them sooner rather than later as they may provide valuable information as to the identity of your unknown (e.g., IR may reveal your functional group).

Lab Notebook Preparation A

Before coming to lab on the first day of this experiment, the following items must be in your lab notebook:

1. Title of experiment
2. Date the experiment is to be performed
3. Outline of your plan for determining the identity of your unknown
4. Hazards of and appropriate precautions for the safe handling of unknown organic compounds
5. References

Lab Notebook Preparation B

Before coming to lab on the day you plan to prepare a derivative, the following items must be in your lab notebook:

1. Title of experiment
2. Date the experiment is to be performed
3. List of possible unknowns
4. The chemical reaction(s) you are attempting (with skeletal structures…R groups are okay if you do not know the identity of your unknown yet)
5. For each reaction you are attempting, include a table with information about your starting materials. Include molecular weight, molar equivalents, and mmoles to be used. For solids include grams. For liquids, include grams, density, and volume. For solutions, include the concentration and volume. (Note: You will not be able to completely fill in the table if you do not know the identity of your unknown yet. If that is the case, list whatever data you can.)

6. Any relevant physical properties (i.e., melting points or boiling points of possible unknowns and their derivatives)
7. Hazards of and appropriate precautions for the specific reaction(s) you are conducting
8. References

Safety Notes

- Assume that all unknowns are flammable and harmful by inhalation, ingestion, and skin absorption. Do not inhale their vapors and avoid contact with eyes, skin and clothing.

Directions

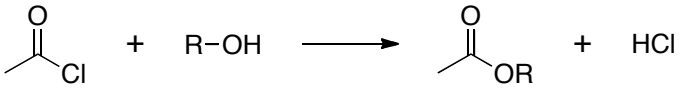
1. Purify your unknown using distillation, recrystallization, or column chromatography. It is recommended that purify the entire unknown provided so that you have enough pure material for all of the tests.
2. Measure the boiling or melting point of your unknown to confirm its purity.
3. Confirm with your instructor that the boiling or melting point you obtained for your unknown is within 15 ˚C of the reported literature value before proceeding.
4. Test the solubility of your unknown in water. (If your unknown is a solid, crush it into a fine powder.)
    a. Add approximately 30 mg of your unknown to a test tube or small vial.
    b. Add 1 mL of water and shake vigorously for approximately 30 seconds. If the unknown appears to be soluble, test the pH of the solution and then skip to step 9.
5. Test the solubility of your unknown in 5% HCl. (If your unknown is a solid, crush it into a fine powder.)
    a. Add approximately 30 mg of your unknown to a test tube or small vial.
    b. Add 1 mL of 5% HCl and shake vigorously for approximately 30 seconds. If the unknown appears to be soluble, skip to step 9.
6. Test the solubility of your unknown in 5% NaOH. (If your unknown is a solid, crush it into a fine powder.)
    a. Add approximately 30 mg of your unknown to a test tube or small vial.
    b. Add 1 mL of 5% NaOH and shake vigorously for approximately 30 seconds. If the unknown appears to be insoluble, skip to step 8.
7. Test the solubility of your unknown in 5% $NaHCO_3$. (If your unknown is a solid, crush it into a fine powder.)
    a. Add approximately 30 mg of your unknown to a test tube or small vial.
    b. Add 1 mL of 5% $NaHCO_3$ and shake vigorously for approximately 30 seconds.
    c. Note whether your unknown is soluble or insoluble and then skip to step 9.

8. Test the solubility of your unknown in concentrated H_2SO_4. (If your unknown is a solid, crush it into a fine powder.)
    a. Add approximately 30 mg of your unknown to a test tube or small vial.
    b. Add 1 mL of concentrated H_2SO_4 and shake vigorously for approximately 30 seconds.
    c. Note whether your unknown is soluble or insoluble. (Any indication of a reaction such as heat, gas generation, or a color change also indicates solubility.)
9. Conduct classification tests as needed. See directions for specific tests below.
10. Confirm the identity of your functional group with your instructor before proceeding.
11. Prepare one or more derivatives of your unknown. See directions for specific derivatives below.
12. Measure the melting point of any derivatives.
13. Confirm with your instructor that the melting point you obtained for your derivative is within 15 °C of the reported literature value.
14. Take the IR and NMR of your unknown.

Classification Tests

C-1 Acetyl Chloride

This reaction tests for the presence of alcohols. If you suspect that you have primary or secondary alcohol, use procedure A. If you suspect that you have a tertiary alcohol, use procedure B (many tertiary alcohols will not react with procedure A). Note that amines and phenols will also give a positive result for this test.



Safety Notes: Acetyl chloride is very corrosive and its vapors are irritating and toxic. It reacts violently with water and some alcohols. Work in a fume hood or under a snorkel and keep away from water. *N,N*-Dimethylaniline and ammonia are harmful if inhaled or allowed to contact the skin. Work under in a fume hood or under a snorkel.

Recommended Controls: 1-butanol, 2-methyl-2-propanol

Procedure A:

1. Add 10 drops of the unknown (0.4 g of a solid) to a test tube.
2. In the fume hood or under a snorkel, dropwise add 10 drops of acetyl chloride.
3. Observe any evolution of heat, and hold a piece of pH paper over the top of the test tube to determine if any HCI gas was generated.
4. After a minute or two, pour the mixture into about 2 mL of water, shake it, and note any phase separation.

Interpretation: Evidence of reaction (heat, HCI gas), especially if accompanied by phase separation indicates an alcohol.

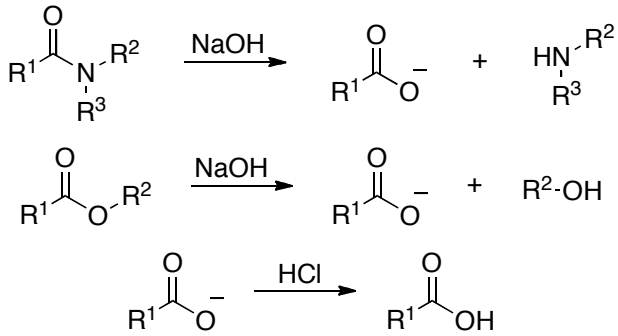
Procedure B:

1. In the fume hood or under a snorkel, mix 5 drops of acetyl chloride with 10 drops of *N,N*-dimethylaniline.
2. Cautiously add 5 drops of the unknown (0.2 g of a solid).
3. Warm the mixture on a 50 °C water bath for 15 minutes, and then cool the mixture to room temperature.
4. In the fume hood or under a snorkel, add 1 g of ice and 1 mL of concentrated ammonia, and mix.
5. Allow the mixture to stand for a few minutes. If a layer separates, remove it with a pipet and test it using the ferric hydroxamate test (C-14) to determine if an ester formed.

Interpretation: Formation of an ester as indicated by the ferric hydroxamate test (C-14) indicates a positive result.

C-2 Alkaline Hydrolysis

This reaction tests for the presence of amides and esters. If you suspect that you have an amide, use procedure A. If you suspect that you have an ester, use procedure B. (Note that esters with boiling points higher than 200 °C may be unreactive.)



Safety Notes: Sodium hydroxide is toxic and corrosive, causing severe damage to skin, eyes, and mucous membranes. Wear gloves and avoid contact.

Recommended Controls: benzamide

Procedure A:

1. Place 0.1 g of the unknown (3 drops of a liquid) in a test tube containing 4 mL of 6 M sodium hydroxide.
2. Secure a small piece of filter paper over the top of the tube and moisten it with 2 drops of 10% copper(II) sulfate.
3. Boil the mixture for a minute or two and note any color change on the filter paper.

4. Acidify the solution with 6 M HCI; if a carboxylic acid precipitates, save it for use as a derivative.

Interpretation: A blue color on the filter paper indicates an amide. This is caused by the reaction of the free amine with the copper (II) sulfate. Some amides will yield a precipitate or a separate liquid phase (the carboxylic acid) when the hydrolysis mixture is acidified. If the test is inconclusive, try increasing the heating time or repeating the reaction at 200 °C using 20% KOH in glycerine.

Recommended Controls: butyl acetate

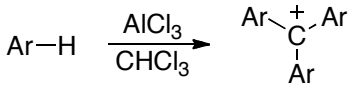
Procedure B:

1. Mix 1 mL of the unknown (1 g of a solid) with 10 mL of 6 M sodium hydroxide in a round bottom flask, and heat under reflux for 30 minutes (or until the solution is homogeneous).
2. If a separate organic layer or residue remains, heat the mixture longer until it disappears or until it is apparent that no reaction is taking place. (Most esters boiling under 110 °C will hydrolyze in 30 minutes; higher-boiling esters may take several hours.)
3. Cool the reaction mixture, remove any organic layer if there is one, and acidify the aqueous solution with 6 M sulfuric acid. If a carboxylic acid precipitates, save it for use as a derivative.

Interpretation: Evidence for an ester is indicated by disappearance of the organic layer (if any), and the appearance of a precipitate upon acidification.

C-3 Aluminum Chloride & Chloroform

This reaction tests for the presence of an aromatic ring. It can only be used for compounds that do not dissolve in sulfuric acid.



Safety Notes: Chloroform is harmful if inhaled or absorbed through the skin, and it is a suspected carcinogen in humans. Use gloves and handle in the fume hood or under a snorkel. Aluminum chloride reacts violently with water, skin and eye contact can cause painful burns, and inhaling the dust or vapors is harmful. Use gloves, handle in the fume hood or under a snorkel, and keep away from water.

Recommended Controls: toluene, biphenyl

Procedure:

1. In the fume hood or under a snorkel, prepare a solution containing 3 drops of the unknown (or 0.1 g of a solid) in 2 mL of chloroform. Save this solution for step 4.
2. Add about 0.2 g of anhydrous aluminum chloride to a dry test tube and heat it over a flame, angling the test tube so that the $AlCl_3$ sublimes onto the inner wall of the tube a few centimeters above the bottom.
3. Allow the tube to cool until it can be held comfortably in your hand.

4. Pipet a few drops of the solution from step 1 down the side of the tube so that it contacts the aluminum chloride. Note any color change at the point of contact.

Interpretation: An intense color at the point of contact such as yellow-orange, red, blue, green, or purple indicates the presence of an aromatic compound. A light yellow color is inconclusive or negative.

C-4 Basicity Test

This test is useful if you have already determined that you have an amine. It is used to distinguish alkyl amines from aromatic amines.

Recommended Controls: p-toluidine, dibutylamine

Procedure for water-soluble compounds:

1. Dissolve 4 drops of your unknown (0.10 g of a solid) in 3 mL of water.
2. Measure the pH of the solution using pH paper.

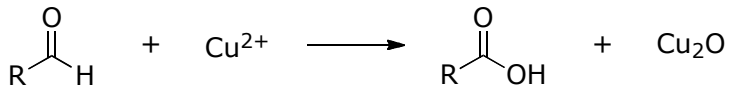
Procedure for water-insoluble compounds:

1. Dissolve 4 drops of your unknown (0.10 g of a solid) in 3 mL of a pH 5.5 acetate-acetic acid buffer.
2. Mix thoroughly.

Interpretation: Water-soluble alkyl amines give pH values above 11, whereas water-soluble aromatic amines have pH values below 10. Water-insoluble alkyl amines should dissolve in the buffer, but water-insoluble aromatic amines will not dissolve.

C-6 Benedict's Test

This reaction tests for the presence of aldehydes. Note that most ketones and aromatic aldehydes will not react.



Recommended Controls: butanal

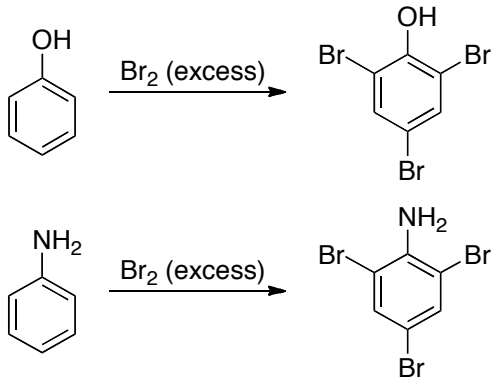
Procedure:

1. Add 2 drops of the unknown (80 mg if it is a solid) to 2 mL of water.
2. Add 2 mL of Benedict's reagent.
3. Heat the mixture to a boil.
4. Observe if a precipitate forms, and note its color.

Interpretation: Benedict's reagent contains copper(II) sulfate, sodium citrate, and sodium carbonate. Aldehydes will react with the Cu^{2+} from the copper(II) sulfate to form copper(I) oxide which appears as a yellow or orange precipitate (it may look a little green in the blue reaction solution). Note that most ketones and aromatic aldehydes will not react.

C-8 Bromine Water

This reaction tests for the presence of phenols and aromatic amines.



Recommended Control: phenol

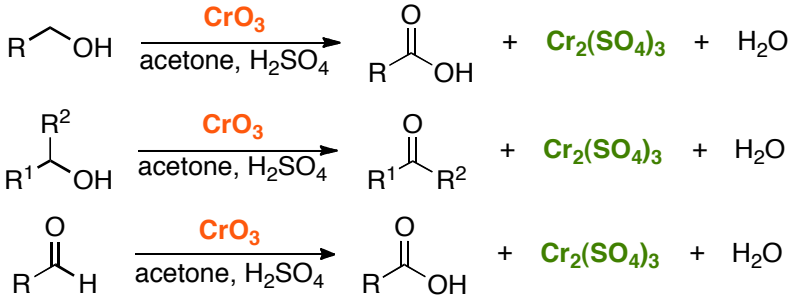
Procedure:

1. Dissolve 3 drops of the unknown (0.1 g of a solid) in 10 mL of water. (If you know based on the results of your solubility tests that the unknown is insoluble in water, add ethanol until your unknown just dissolves.)
2. Measure the pH of the solution with pH paper.
3. Under the hood, add saturated bromine water drop by drop until the bromine color persists. Watch for evidence of a precipitate.

Interpretation: Decolorization of the bromine, accompanied by simultaneous formation of a white (or nearly white) precipitate, indicates a phenol or aromatic amine. If the unknown is a phenol, the pH of the initial solution should be less than 7.

C-9 Chromic Acid

This reaction tests for the presence of primary alcohols, secondary alcohols, and aldehydes.



Safety Notes: The chromic acid reagent is corrosive and carcinogenic. Avoid contact.

Recommended Controls: 1-butanol, butanal

Procedure:

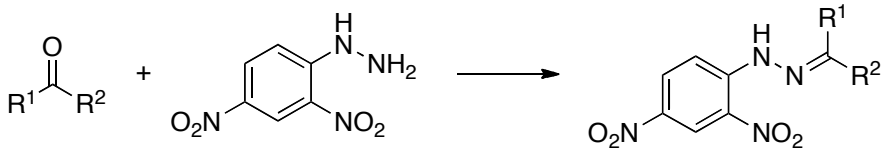
1. Dissolve 1 drop of the unknown (40 mg of a solid) in 1 mL of reagent grade acetone.

2. Add 1 drop of the chromic acid reagent and swirl.
3. Note the time it takes for the formation of a cloudy blue-green suspension to occur (this should be accompanied by the disappearance of the orange colored reagent).

Interpretation: A reaction within 2-3 seconds indicates a primary or secondary alcohol. With aliphatic aldehydes, the solution turns cloudy in about 5 seconds and the blue-green suspension forms within 30 seconds. Aromatic aldehydes require 30-90 seconds or longer to form the suspension. Note that the formation of another dark color (particularly with the color of the liquid remains orange) should be considered a negative test.

C-11 2,4-Dinitrophenylhydrazine

This reaction tests for the presence of aldehydes and ketones.



Safety Notes: 2,4-Dinitrophenylhydrazine (DNPH) is harmful if absorbed through the skin. Wear gloves and avoid contact.

Recommended Controls: cyclohexanone, benzaldehyde

Procedure:

1. Dissolve 1 drop of the unknown (40 mg of a solid) in 1 mL of 95% ethanol (use more ethanol if necessary to completely dissolve the unknown).
2. Add this solution to 2 mL of the DNPH reagent.
3. Shake and let the mixture stand for 15 minutes or until a precipitate forms. (If a precipitate is observed at this point, this is considered a positive result.)
4. Scratch the inside of the test tube and observe if a precipitate forms, and note its color.

Interpretation: Formation of a crystalline yellow or orange-red precipitate indicates an aldehyde or ketone. The color of the precipitate may give a clue to the structure of the carbonyl compound (unconjugated aliphatic aldehydes and ketones usually yield a yellow precipitate, while aromatic and α,β-unsaturated aldehydes and ketones yield a orange-red precipitate).

C-12 Elemental Analysis

This reaction tests for the presence of nitrogens and halogens.

Safety Notes: Sodium can cause serious burns and the sodium-lead alloy may react violently with some substances. Wear gloves, avoid contact, and keep the sodium-lead alloy away from other chemicals.

Recommended Controls: butylamine, acetamide, bromobenzene

Procedure:

1. In the fume hood or under a snorkel, place 0.25 g of 10% sodium-lead alloy in a small, dry test tube held vertically by a clamp.
2. Melt the alloy with a Bunsen burner flame and continue heating until the sodium vapor rises about 1 cm up the tube.
3. Using a Pasteur pipet, add 2 drops of the unknown (or 10 mg of a solid) directly onto the molten alloy so that it does not touch the sides of the tube.
4. Heat gently to start the reaction, remove the flame until the reaction subsides, then heat the tube strongly for a minute or two, keeping the bottom a dull red color.
5. Let the tube cool to room temperature.
6. Dropwise add 1.5 mL of water and heat gently for a minute or so until the excess sodium has decomposed and gas evolution ceases.
7. Filter the solution through a Pasteur pipet with a small plug of cotton, wash the cotton with 1 mL of water, and combine the wash water with the filtrate. (Use a rubber bulb to expel any liquid that adheres to the cotton.) The filtrate should be colorless or just slightly yellow. If it is darker, repeat the fusion with stronger heating or more of the alloy.

To test for nitrogen:

1. Put 5 drops of the sodium fusion solution into a small test tube.
2. While stirring, add enough solid sodium bicarbonate, to saturate it (a little excess solid should be present).
3. Add 1 drop of this solution to a test tube containing 10 drops of PNB reagent (*p*-nitrobenzaldehyde in dimethyl sulfoxide) and note any color change.

To test for halogens:

1. Acidify 10 drops of the sodium fusion solution with dilute nitric acid.
2. Boil it gently under the hood for a few minutes.
3. Add a drop or two of 0.3 M aqueous silver nitrate, and note the color and volume of any precipitate that forms. (If a voluminous precipitate forms, let the precipitate settle and then remove the solvent using a pipet.)
4. Add 2 mL of 3 M aqueous ammonia to the solid, shake vigorously, and note your observations.
5. To test further for bromine and iodine, acidify 1 mL of the original sodium fusion solution with 1 M sulfuric acid, boil for a few minutes, and add 0.5 mL of dichloromethane and a then a drop of freshly prepared chlorine water. Shake and look for a color in the dichloromethane layer.

Interpretation: In the PNB test, a purple color indicates the presence of nitrogen (green indicates sulfur). In the halogen tests, formation of a voluminous precipitate on addition of silver nitrate indicates that a halogen is present, and the color of the precipitate (a silver halide) may suggest which halogen: white for chlorine, pale yellow for bromine, and yellow for iodine. If only a faint turbidity is produced, it may be caused by traces of impurities or by incomplete sodium fusion. If

the precipitate is silver chloride, it will dissolve in aqueous ammonia; silver bromide is only slightly soluble and silver iodide is insoluble. In the chlorine water test, a red-brown color is produced by elemental bromine and a violet color by elemental iodine.

C-13 Ferric Chloride

This reaction tests for the presence of phenols.

Recommended Control: phenol

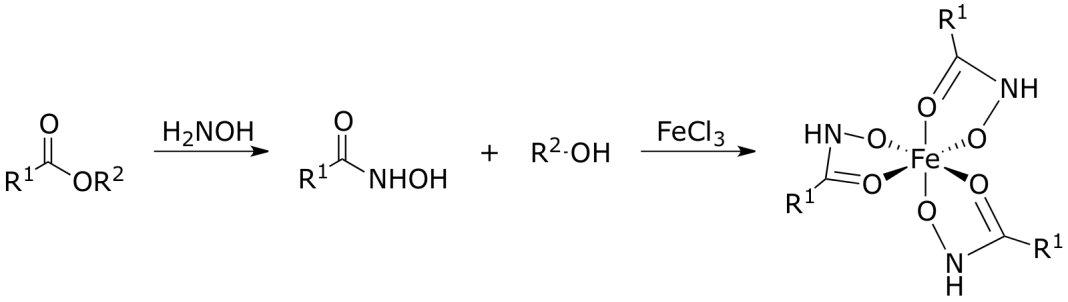
Procedure:

1. Dissolve 1 drop of the unknown (40 mg of a solid) in 1 mL of water. (If you know based on the results of your solubility tests that the unknown is insoluble in water, use 0.5 mL of water and 0.5 mL of methanol instead of 1 mL of water.)
2. Add two drops of 2.5% ferric chloride solution.

Interpretation: Formation of an intense red, green, blue, or purple color suggests a phenol or an easily enolizable compound (such as an aldehyde or ketone). Some phenols do not react under these conditions.

C-14 Ferric Hydroxamate Test

This reaction tests for the presence of esters.



Safety Notes: Hydroxylamine hydrochloride is toxic and mutagenic and it can cause a form of anemia. Avoid contact with its solution. Sodium hydroxide is toxic and corrosive, causing severe damage to skin, eyes, and mucous membranes. Wear gloves and avoid contact with the NaOH solution.

Recommended Controls: butyl acetate

Procedure:

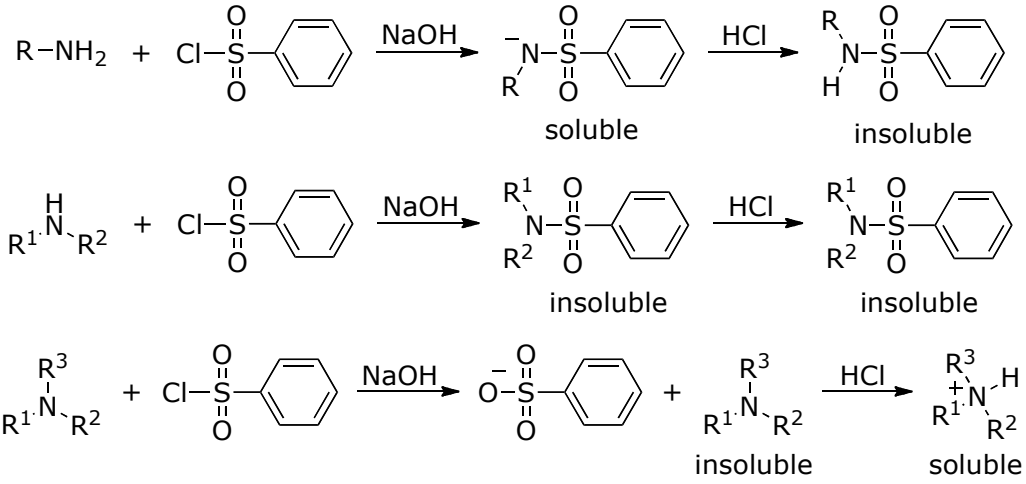
1. Before you use the ferric hydroxamate test, perform the following preliminary test. Dissolve 1 drop of the unknown (40 mg of a solid) in 1 mL of 95% ethanol. Add 1 mL of 1 M hydrochloric acid; then add two drops of 2.5% ferric chloride. If a definite color other than yellow results, the ferric hydroxamate test cannot be used. This test eliminates those phenols and enols that give colors with ferric chloride in acidic solution and that would therefore give a false positive result in the ferric hydroxamate test.
2. Mix 1 mL of 0.5 M ethanolic hydroxylamine hydrochloride with 5 drops of 6 M sodium hydroxide.

3. Add 1 drop of the unknown (40 mg of a solid) and heat the solution to boiling.
4. Allow it to cool slightly and add 2 mL of 1 M hydrochloric acid. (If the solution is cloudy at this point, add enough 95% ethanol make it clear.)
5. Add 5 drops of 2.5% ferric chloride solution and observe any color produced. If the color does not persist, add more ferric chloride solution drop by drop until the color becomes permanent.

Interpretation: A burgundy or magenta color that is distinctly different from the color obtained in the preliminary test indicates an ester.

C-15 Hinsberg's Test

This test is useful if you have already established that you have an amine. It is useful for classifying amines as primary, secondary, or tertiary. It is important to record all observations carefully during this test.



Safety Notes: p-Toluenesulfonyl chloride is toxic and corrosive. Use gloves and work in a fume hood or under a snorkel.

Recommended Controls: butylamine, dibutylamine, tributylamine

Procedure:

1. In the fume hood or under a snorkel, mix 3 drops of the unknown (0.1 g of a solid) with 5 mL of 3 M sodium hydroxide in a test tube.
2. Add 0.2 g of *p*-toluenesulfonyl chloride.
3. Stopper the tube and shake it intermittently for 3-5 minutes.
4. Remove the stopper and warm the solution in a hot water bath, with shaking, for 1 minute.
5. Use pH paper to check if the solution is basic (if it is not basic, add more NaOH until it is).
6. If there is a solid or liquid residue in the test tube, separate it from the solution by vacuum filtration (if it is a solid) or with a Pasteur pipet (if it is a liquid).

7. Test the solubility of the solid or liquid residue in water and (if it's insoluble) in 5% hydrochloric acid.
8. Acidify the original solution with 6 M hydrochloric acid and, if no precipitate forms immediately, scratch the sides of the test tube and cool.

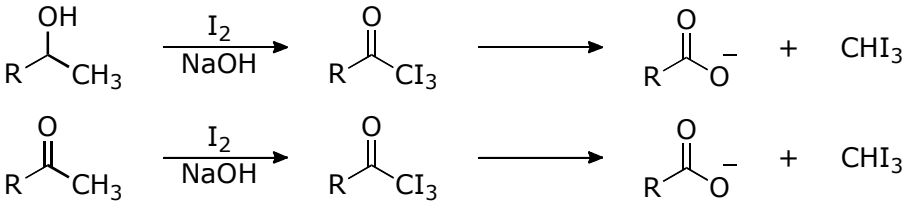
Interpretation: Formation of a white precipitate (a *p*-toluenesulfonamide) when the reaction mixture is acidified indicates a primary amine. Most primary amines yield a clear solution after the initial reaction, but some form sodium salts or disulfonyl derivatives that precipitate during the reaction. The salts should dissolve in water, and amines that form disulfonyl derivatives should yield additional precipitate when the reaction mixture is acidified.

Most secondary amines yield a white solid that does not dissolve in water or 5% HCl. A liquid residue that is more dense than water and insoluble in 5% HCI may be a secondary amine's arenesulfonamide that has failed to crystallize.

Tertiary amines should not react; any residue will be the original liquid or solid amine, which should dissolve in dilute HCI. Water-soluble tertiary amines yield a clear solution that does not form a separate phase on acidification.

C-16 Iodoform Test

This test is useful if you have already established that you have an alcohol or a ketone. It indicates if you have –CH(OH)CH_3 and –$COCH_3$ groupings.



Recommended Controls: 2-butanol, 2-butanone

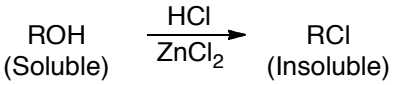
Procedure:

1. Dissolve 3 drops of the unknown (or 0.1 g of a solid) in 2 mL of water. (If you know based on the results of your solubility tests that the unknown is insoluble in water, use methanol as a solvent instead of water.)
2. Add 1 mL of 3 M sodium hydroxide solution.
3. Add the 0.5 M iodine-potassium iodide reagent drop by drop until the brown iodine color persists after shaking. (If a yellow precipitate is observed at this point, this is considered a positive result.)
4. Place the test tube in a 60 °C water bath and add more iodine-potassium iodide solution as necessary until the brown color remains after 2 minutes of heating.
5. Add 3 M NaOH drop by drop until the brown color just disappears (a light yellow color may remain).
6. Remove the test tube from the water bath, add 10 mL of cold water, and let it stand for 15 minutes.

Interpretation: Formation of a yellow precipitate (iodoform) indicates a positive test. If you wish to confirm the identity of the precipitate, measure its melting point (121°C). The test is positive for methyl ketones and acetaldehyde, and for alcohols that contain a $-CH(OH)CH_3$ grouping.

C-17 Lucas's Test

This test is useful if you have already established that you have an alcohol. It is useful for classifying alcohols as primary, secondary, or tertiary. However, note that this test cannot be used for solid alcohols or alcohols that are not soluble in the Lucas reagent (typically alcohols that have boiling points higher than ~140 °C).



Safety Notes: The Lucas reagent ($ZnCl_2$ in concentrated HCl) can cause serious burns, and is harmful if inhaled. Use gloves and handle in a fume hood or under a snorkel.

Recommended Controls: 1-butanol, 2-butanol, 2-methyl-2-propanol

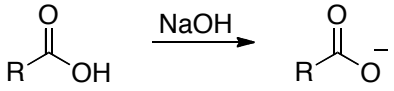
Procedure:

1. In the fume hood or under a snorkel, add 2 mL of the Lucas reagent to a test tube.
2. Add 4 drops of the unknown, then stopper the tube immediately, and shake vigorously.
3. Allow the mixture to stand for 15 minutes or more, observing it periodically for evidence of reaction (cloudiness or layer separation). If the alcohol appears to be secondary or tertiary, repeat the test using concentrated HCl in place of the Lucas reagent.

Interpretation: Tertiary alcohols that are soluble in the Lucas reagent should turn the reagent cloudy almost immediately and quickly form a separate layer of alkyl chloride. Secondary alcohols usually turn the clear solution cloudy in 3-5 minutes and form a distinct layer within 15 minutes. Primary alcohols do not react under these conditions. (Most allylic and benzylic alcohols give the same result as tertiary alcohols, except that the chloride formed from allyl alcohol is itself soluble in the reagent and separates out only upon addition of ice water.) High-boiling alcohols that are insoluble in the Lucas reagent cannot be tested because they will form a separate layer immediately. When tested with concentrated HCl, a tertiary alcohol should react within minutes, and a secondary alcohol should not react at all.

C-18 Neutralization Equivalent

This test is useful if you have already established that you have a carboxylic acid. It is useful for providing information about the molecular weight of your unknown and the number of carboxylic acids present in your unknown.



Recommended Control: adipic acid

Procedure:

1. Accurately weigh (to 3 decimal places) about 0.2 g of an unknown carboxylic acid and dissolve it in 50-100 mL of water, ethanol, or a mixture of the two, depending on its solubility.
2. Titrate this solution with a standardized solution of 0.1 M sodium hydroxide using phenolphthalein as the indicator (or bromothymol blue if the solvent is ethanol).
3. Calculate the neutralization equivalent (NE) of the acid using the formula shown.

$$NE = \frac{\text{mass of sample in mg}}{\text{mL of NaOH X molarity of NaOH}}$$

Interpretation: The neutralization equivalent (equivalent weight) of a carboxylic acid is equal to its molecular weight divided by the number of carboxyl acids it has. For instance, the NE of adipic acid [$HOOC(CH_2)_4COOH$; MW = 146] is 73. A solid carboxylic acid that has an unusually low neutralization equivalent for its melting point probably contains more than one carboxylic acid.

C-20 Quinhydrone

This test is useful if you have already established that you have an amine. It is useful for classifying amines as primary, secondary, or tertiary. Note that his test will not work for diaminobenzenes or nitro-substituted aromatic amines. This test should be run in conjunction with test **C-4** or with an infrared spectrum that shows whether the amine is aliphatic or aromatic. Controls should be run for comparison since the colors are difficult to identify.

Safety Notes: The components of quinhydrone (hydroquinone and benzoquinone) are toxic and irritating. Avoid contact with the reagent.

Recommended Controls: butylamine, dibutylamine, tributylamine, aniline, *N*-methylaniline, *N,N*-dimethylaniline

Procedure:

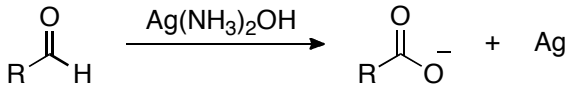
1. Shake 1 drop of an unknown alkylamine (30 mg of a solid) or 6 drops of an unknown arylamine (0.2 g of a solid) with 6 mL of water in a test tube. If the amine dissolves, add 6 mL more of water; if not, add 6 mL of ethanol.
2. Shake the mixture, add 1 drop of 2.5% quinhydrone in methanol and let it stand for 2 minutes or more.
3. Compare the color of this solution with those of the controls.

Interpretation: Most amines from the following classes give the colors indicated. It is best to use this test in conjunction with Hinsberg's test or an IR spectrum for confirmation.

Amine Type	1°	2°	3°
Aliphatic	violet	rose	yellow
Aromatic	rose	amber	yellow

C-23 Tollen's Test

This reaction tests for the presence of aldehydes.



Safety Notes: Silver nitrate is corrosive and toxic. Wear gloves and avoid contact. Use the reagent immediately after preparation and be sure to dispose of the Tollen's reagent as indicated (explosive silver salts may form if Tollen's reagent is stored or improperly disposed of).

Recommended Controls: benzaldehyde

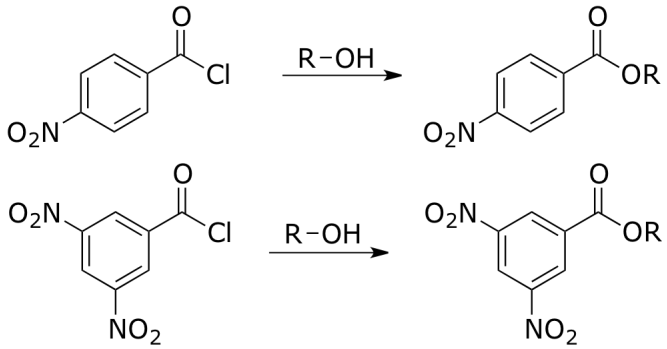
Procedure:

1. Measure 2 mL of 0.3 M aqueous silver nitrate into a test tube and add 1 drop of 3 M sodium hydroxide.
2. Add 2 M aqueous ammonia drop by drop, with shaking, until the precipitate of silver oxide just dissolves (avoid an excess of ammonia).
3. Add 1 drop of the unknown (40 mg of a solid) to this solution, shake the mixture, and let it stand for 10 minutes. (If a silver mirror is observed at this point, this is considered a positive result.)
4. Heat the mixture in a 35 °C water bath for 5 minutes.
5. Immediately after the test has been completed, dissolve any solid residue in 1M nitric acid and then dispose of the solution in the designated waste container.
6. *Interpretation:* Formation of a silver mirror on the inside of the test tube is a positive test for an aldehyde. (Note that if the tube is not sufficiently clean, a black precipitate or a suspension of metallic silver may form instead.)

Derivatives of Alcohols

D-1 p-*Nitrobenzoates and 3,5-Dinitrobenzoates*

For these derivatives, it is extremely important to ensure that your glassware and your alcohol are dry (i.e., free of water). Water can easily react with the acid chlorides to form carboxylic acids to form the respective carboxylic acids rather than the desired esters. (Note: The *p*-nitrobenzoic acid has a melting point of 237 °C. and the 3,5-dinitrobenzoic acid has a melting point of 205-207 °C.) If necessary, dry your glassware in an oven before proceeding.



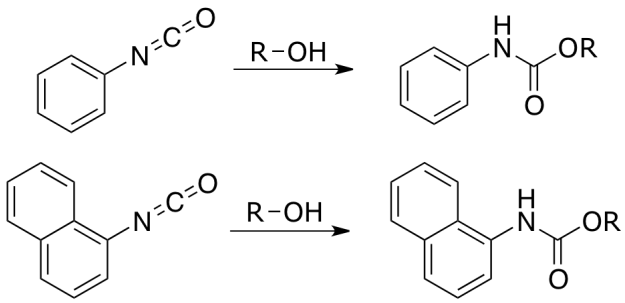
Safety Notes: Acid chlorides are corrosive and lachrymators. Avoid contact with these reagents and use in a fume hood or under a snorkel.

Procedure:

1. Dry your unknown alcohol with magnesium sulfate or sodium sulfate.
2. Filter to remove the drying agent.
3. If you are making the *p*-nitrobenzoate derivative, add 0.20 g of *p*-nitrobenzoyl chloride to a small round bottom flask. If you are making the 3,5-dinitrobenzoate derivative, add 0.20 g of 3,5-dinitrobenzoyl chloride to a small round bottom flask.
4. In the fume hood or under a snorkel, dropwise add 0.10 g of your unknown alcohol to the acid chloride while stirring.
5. Heat the mixture in a 60-70 °C water bath. If your alcohol has boiling point of < 160 °C, heat the mixture for 5 minutes. If your alcohol has boiling point of > 160 °C, heat the mixture for 15 minutes.
6. Stir in 4 mL of 0.2 M sodium carbonate.
7. Heat the mixture to 50-60 °C for 30 seconds.
8. Cool to room temperature and then in an ice bath.
9. Collect the precipitate by small-scale vacuum filtration. Wash with cold water.
10. Recrystallize the precipitate from ethanol or an ethanol-water mixture.

D-2 Phenylurethanes and 1-Naphthylurethanes

For these derivatives, it is extremely important to ensure that your glassware and your alcohol are dry (i.e., free of water). Water can easily react with the isocyanates to form the respective ureas rather than the desired carbamates. (Note: The dipheylurea has a melting point of 241 °C, and the di-1-naphthylurea has a melting point of 297 °C.) If necessary, dry your glassware in an oven before proceeding.



Safety Notes: Isocyanates are irritants and lachrymators. Avoid contact with these reagents and use in a fume hood or under a snorkel.

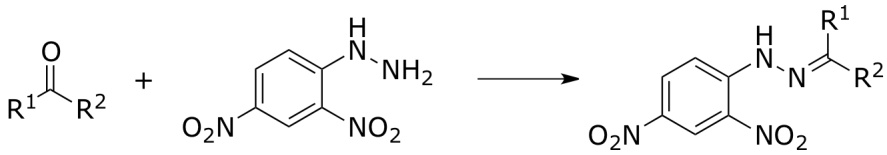
Procedure:

1. Dry your unknown alcohol with magnesium sulfate or sodium sulfate.

2. Filter to remove the drying agent.
3. If you are making the phenylurethane derivative, add 5 drops of phenyl isocyanate to a small round bottom flask. If you are making the 1-naphthylurethane derivative, add 5 drops of 1-naphthyl isocyanate to small round bottom flask.
4. In the fume hood or under a snorkel, dropwise add 5 drops of the dry alcohol to the isocyanate. If no reaction is apparent, heat the mixture in a 60-70 °C water bath for 15 minutes.
5. Cool to room temperature and then in an ice bath.
6. Collect the precipitate by small-scale vacuum filtration.
7. Recrystallize the precipitate from petroleum ether or heptane. (If necessary, perform a hot gravity filtration.)

Derivatives of Aldehydes and Ketones

D-3 2,4-Dinitrophenylhydrazones

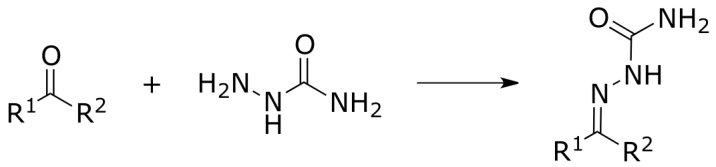


Safety Notes: 2,4-Dinitrophenylhydrazine is toxic and sulfuric acid is corrosive. Avoid contact with these reagents and use in a fume hood or under a snorkel.

Procedure:

1. In a small round bottom flask, dissolve 0.10 g of the unknown aldehyde or ketone in 1 mL of ethanol. (If your unknown is not completely dissolved, add ethanol drop by drop until it goes into solution).
2. In the fume hood or under a snorkel, dropwise add 3 mL of the 2,4-dinitrophenylhydrazine-sulfuric acid reagent.
3. Allow the solution to stand at room temperature until crystallization is complete. If no crystals form, heat the mixture in a 60-70 °C water bath for 15 minutes, and then it cool again. If there is still no precipitate, add cold water drop by drop to the solution until precipitate is observed.
4. Collect the precipitate by small-scale vacuum filtration. Wash once with 5 mL of cold 5% $NaHCO_3$ and once with cold water.
5. Recrystallize the precipitate from ethanol or an ethanol-water mixture. (Note: If more than 6 mL of ethanol is needed for recrystallization, add ethyl acetate drop by drop to the hot solution until everything is dissolved.)

D-4 Semicarbazones

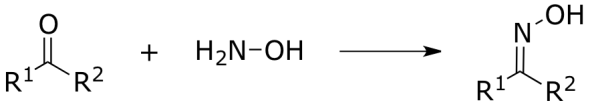


Safety Notes: Semicarbazide hydrochloride is a suspected carcinogen. Avoid contact with the reagent.

Procedure:

1. Mix together 0.20 g of semicarbazide hydrochloride, 0.30 g of sodium acetate, and 2 mL of water in a small round bottom flask.
2. If your unknown aldehyde or ketone is water soluble, add 0.20 g of it directly to the flask and stir to dissolve. If your unknown aldehyde or ketone is not water soluble, add a minimum amount of ethanol to the mixture until your unknown goes into solution.
3. Stir the mixture for two minutes.
4. Cool the mixture in an ice bath. If no crystals form, heat the mixture in a 60-70 °C water bath for 5 minutes, and then it cool again.
5. Collect the precipitate by small-scale vacuum filtration. Wash with cold water.
6. Recrystallize the precipitate from ethanol or an ethanol-water mixture.

D-5 Oximes



Safety Notes: Hydroxylamine hydrochloride is toxic and mutagenic. Avoid contact with the reagent.

Procedure:

1. Mix together 0.125 g of hydroxylamine hydrochloride, 0.30 g of sodium acetate, and 2 mL of water to a small round bottom flask.
2. If your unknown aldehyde or ketone is water soluble, add 0.20 g of it directly to the flask and stir to dissolve. If your unknown aldehyde or ketone is not water soluble, add a minimum amount of ethanol to the mixture until your unknown goes into solution.
3. Stir the mixture for two minutes.
4. Cool the mixture in an ice bath. If no crystals form, heat the mixture in a 60-70 °C water bath for 15 minutes, and then it cool again. If there is still no precipitate, add cold water drop by drop to the solution until precipitate is observed.
5. Collect the precipitate by small-scale vacuum filtration. Wash with cold water.
6. Recrystallize the precipitate from ethanol or an ethanol-water mixture.

Reporting Your Results

Write your report according to the guidelines described in "Topic 4: Writing an Organic Chemistry Lab Report". Work by yourself on this report.

References & Additional Resources

1. Lehman, J. W. *Operational Organic Chemistry: A Problem-Solving Approach to the Laboratory Course*, 3rd ed.; Prentice Hall: Upper Saddle River, NJ, 1999; pp 529-572.

Table 1. Possible Alcohol Unknowns

Alcohol	BP (°C)	3,5-Dinitro benzoate MP (°C)	4-Nitro benzoate MP (°C)	1-Naphthyl urethane MP (°C)	Phenyl urethane MP (°C)
methanol	65	108	96	124	47
ethanol	78	93	57	79	52
2-propanol	83	123	110	106	88
2-methyl-2-propanol	83	142	-	-	136
2-propen-1-ol	97	49	28	108	70
1-propanol	97	74	35	105	57
2-butanol	99	76	26	97	65
2-methyl-2-butanol	101	116	85	72	42
2-methyl-1-propanol	108	87	69	104	86
3-pentanol	116	101	17	95	48
1-butanol	118	64	36	71	61
2-pentanol	119	62	24	74	-
3-methyl-3-pentanol	123	94 (62)	69	104	43
3-methyl-1-butanol	132	61	21	68	57
4-methyl-2-pentanol	132	65	26	88	143
1-pentanol	137	46	11	68	46
cyclopentanol	141	115	62	118	132
2-ethyl-1-butanol	148	51	-	60	-
1-hexanol	157	58	5	59	42
cyclohexanol	161	113	50	129	82
furfuryl alcohol	172	80	76	130	45
1-heptanol	177	47	10	62	60
2-octanol	174	32	28	63	114
1-octanol	195	61	12	67	74
1-phenylethanol	202	95	43	106	92
benzyl alcohol	204	113	85	134	77
2-phenylethanol	219	108	62	119	78
1-decanol	231	57	30	73	60
3-phenyl-1-propanol	236	45	47	-	92
1-dodecanol	259	60	45 (42)	80	74

*A dash indicates that no information is reported in the literature
**Melting points in parenthesis represent conflicting literature values

Table 2. Possible Aldehyde Unknowns

Aldehyde	BP (°C)	MP (°C)	2,4-Dinitro phenylhydrazone MP (°C)	Semicarbazone MP (°C)	Oxime MP (°C)
ethanal	21		168 (157)	162	47
propanal	48		148 (155)	154	40
propenal	52		165	171	-
2-methylpropanal	64		187 (183)	125 (119)	-
butanal	75		123	106	-
3-methylbutanal	92		123	107	48
pentanal	103		106 (98)	-	52
2-butenal	104		190	199	119
2-ethylbutanal	117		95 (30)	99	-
hexanal	130		104	106	51
heptanal	153		106	109	57
2-furaldehyde	162		212 (230)	202	91
2-ethylhexanal	163		114 (120)	254d	-
octanal	171		106	101	60
benzaldehyde	178		239	222	35
phenylethanal	195	33	121 (110)	153 (156)	99
4-methylbenzaldehyde	204		234	234 (215)	80
3,7-dimethyl-6-octenal	207		77	84 (91)	-
2-chlorobenzaldehyde	209		213 (209)	229 (146)	76 (101)
4-methoxybenzaldehyde	248		253d	210	133
2-methoxybenzaldehyde		38	254	215	92
4-chlorobenzaldehyde		48	265	230	110 (146)
3-nitrobenzaldehyde		58	290	246	120
4-nitrobenzaldehyde		106	320	221 (211)	133 (182)

*A dash indicates that no information is reported in the literature
**Melting points in parenthesis represent conflicting literature values
***If the substance changes color and smokes, this is considered decomposition (d = decomposes)

Table 3. Possible Ketone Unknowns

Ketone	BP (°C)	MP (°C)	2,4-Dinitro phenylhydrazone MP (°C)	Semicarbazone MP (°C)	Oxime MP (°C)
acetone	56		126	187	59
2-butanone	80		118	146	-
3-methyl-2-butanone	94		124	113	-
2-pentanone	101		143	112 (106)	58
3-pentanone	102		156	138	69
3,3-dimethyl-2-butanone	106		125	157	75 (79)
4-methyl-2-pentanone	117		95 (81)	132	58
2,4-dimethyl-3-pentanone	124		95 (88)	160	34
2-hexanone	128		110	125	49
4-methyl-3-penten-2-one	130		205	164 (133)	48
cyclopentanone	131		146	210 (203)	56
4-heptanone	144		75	132	-
2-heptanone	151		89	123	-
cyclohexanone	156		162	166	91
2,6-dimethyl-4-heptanone	168		92	122	210
2-octanone	173		58	124	-
cycloheptanone	181		148	163	23
acetophenone	202	20	238	198 (203)	60
2-methylacetophenone	214		159	205	61
propiophenone	218	21	191	182 (174)	54
3-methylacetophenone	220		207	203	57
4-methylacetophenone	226	28	258	205	88
2-undecanone	228		63	122	44
4-phenyl-2-butanone	235		127	142	87
3-methoxyacetophenone	240		-	196	-
2-methoxyacetophenone	245		-	183	83 (96)
4-methoxyacetophenone		38	228	198	87
4-phenyl-3-buten-2-one		42	227 (223)	187	117
benzophenone		48	238	167	144
2-acetonaphthone		54	262d	235	145
3-nitroacetophenone		80	228	257	132
9-fluorenone		83	283	234	195

*A dash indicates that no information is reported in the literature
**Melting points in parenthesis represent conflicting literature values
***If the substance changes color and smokes, this is considered decomposition (d = decomposes