Experiment 1

The Synthesis of 4-Amino-3-nitrobenzoic Acid Methyl Ester via Fischer Esterification

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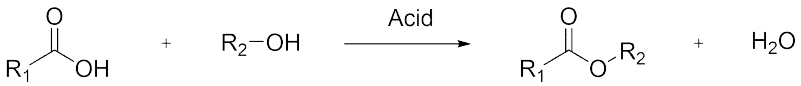
PURPOSE OF THE EXPERIMENT

Synthesize ester compounds via Fischer esterification.
Separate product from reaction mixture by work-up process.

BACKGROUND INFORMATION

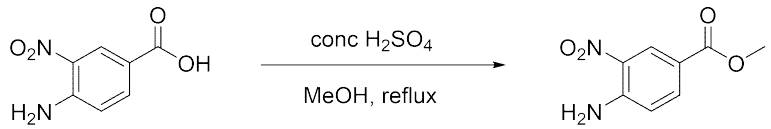
Fischer esterification

Fischer esterification is an important topic included in most introductory chemistry courses and examples of these reactions are abundant. The reaction is a condensation reaction in which a carboxylic acid reacts with an alcohol to produce an ester and water in an acidic condition.



Scheme 1. Schematic representation of Fischer esterification.

In this experiment, a carboxylic acid group of 4-amino-3-nitrobenzoic acid reacts with methanol heating under reflux to produce 4-amino-3-nitrobenzoic acid methyl ester via Fischer esterification with the catalytic amount of sulfuric acid.



Scheme 2. Synthesis of 4-Amino-3-nitrobenzoic Acid Methyl Ester from 4-Amino-3-nitrobenzoic Acid in Neat Methanol with the catalytic amount of sulfuric Acid.

Heating under reflux

Reflux is a technique involving the condensation of vapors and the return of this condensate to the system from which it originated.

In this experiment, a reaction mixture is placed in a round bottom flask, and this round bottom flask is connected to a water-cooled reflux condenser. While heating the reaction mixture for Fischer esterification, methanol (solvent) evaporate and produce vapor. Then, vapors are condensed by the condenser and return to the flask. The purpose of reflux is running the reaction at a controlled temperature without

losing large quantities of the solvent.

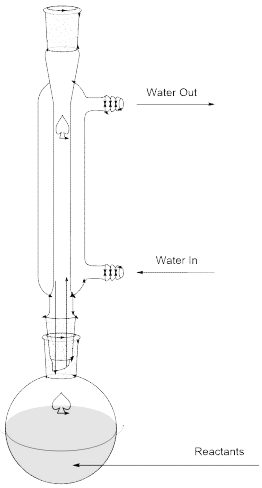


Figure 1. Laboratory reflux apparatus for heating a chemical reaction

Work-up

Work-up process refers to the series of manipulations required to isolate and purify the product of a chemical reaction.

Quenching

Quenching a reaction refers to the deactivation of any unreacted reagents. In this experiment, saturated sodium bicarbonate solution is added to the reaction mixture. When it is added to the reaction mixture, unreacted acid and starting material (4-amino-3-benzoic acid) ionized to prevent further reactions.

Liquid-Liquid Extraction

Liquid-liquid extraction is a method to separate compounds or metal complexes, based on their relative solubilities in two different immiscible liquids, usually water (polar) and an organic solvent (non-polar). Due to the different solubilities, there is a net transfer of one or more components from one liquid to another liquid phase. Once the transfer is finished, liquid layer which contains the target product is separated from the other liquid layer by using suitable apparatus such as a separatory funnel.

Separatory Funnel

Separatory funnel is a piece of laboratory glassware used in liquid-liquid extractions to separate the components of a mixture into two immiscible solvent phases of different densities. The more dense liquid, typically the aqueous phase, sinks and can be drained out through a valve away from the

less dense liquid, which remains in the separatory funnel. In this experiment ethyl acetate is the top layer and aqueous sodium bicarbonate is the bottom layer. Detailed usage is described in the liquid-liquid extraction process.

Removal of solvents by evaporation

After separating product from other components, usually product is dissolved in a solvent. Then we remove solvents from the final product by evaporating solvent using several ways, such as air-dry or using rotary evaporator.

Rotary Evaporator

Rotary evaporator is a device for the efficient and gentle removal of solvents from solutions by evaporation. Rotary evaporator lowers the pressure inside the flask to lower the boiling points of the solvents in it. Rotary evaporator is most often applied to separate “low boiling point” solvents.

Figure 2. Rotary Evaporator

Reference 1 *J. Chem. Educ.* **2020**, 97, 1997−2000.

EXPERIMENT 1 The Synthesis of 4-Amino-3-nitrobenzoic Acid Methyl Ester via Fischer Esterification

Reagents and Properties

substance	*quantity*	*molar mass (g/mol)*	*mp (°C)*	bp (°C)	*density (g/mL)*
4-Amino-3-nitrobenzoic acid	150.00 mg	182.13	280		
Concentrated sulfuric acid	3 drops	98.079	10.31	337	1.84
Methanol	10 mL	34.04	-97.6	64.7	0.791
Saturated Sodium bicarbonate aqueous solution	60-80 mL	84.007	50		1.1
Ethyl acetate	20 mL	88.11	-83.6	77.1	0.9

PROCEDURE

Caution: Wear lab coats and safety goggles at all times while in the lab. Many chemicals are potentially harmful. Prevent

contact with your eyes, skin, and clothing. Wearing contact lens is strictly prohibited.

Apparatus

2 of 100 mL round bottom flask, funnel, thermowell, hot plate, magnetic bar, reflux condenser (Liebig condenser), Clamps, rubber tubes, 125 mL separatory funnel, separatory funnel cap, rubber septum, needle, 125 mL Erlenmeyer flask.

A. Methylation of 4-amino-3-nitrobenzoic acid

Caution: Perform experiment in a **fume hood**, 4-amino-3-nitrobenzoic acid is irritant and may cause skin, eye, and respiratory irritation. Sulfuric acid is corrosive.

A.1 Preparing the reaction mixture

1. Measure 150.00 mg of 4-amino-3-nitrobenzoic acid.
2. Add a magnetic bar to the reaction flask.
3. Transfer 4-amino-3-nitrobenzoic acid to 100 mL round bottom flask using a funnel.

Figure 3. 4-Amino-3-nitrobenzoic acid in 100 mL round bottom flask.

4. Add 10 mL of methanol to the the reaction flask.
5. Shake the reaction flask until 4-amino-3-nitrobenzoic acid dissolves into methanol.
6. Add 3 drops of concentrated sulfuric acid to the reaction flask. [Note 1]

Note 1. Release dropper slowly to prevent sulfuric acid splashing.

Figure 4. Prepared reaction mixture

A.2 Fischer esterification of 4-amino-3-nitrobenzoic acid

1. Equip a reflux condenser to the reaction flask.
2. Put a thermowell on the hot plate.
3. Fit the flask with thermowell and fix it with clamps.
4. Insert two tubes to top and bottom port.
5. Connect the end of bottom port tube to the water pipe and the end of top port tube to the sink.
6. Open the water pipe with an appropriate flow rate so that the tube does not bounce off.
7. Cover the end of reflux condenser with a rubber septum.

Figure 5. Heating under reflux

Note 2. Measure 1 hour from starting reflux (dropping solvent from reflux condenser)

8. Put a pressure release needle to the septum.
9. Heat to reflux the reaction mixture at 75℃ with stirring for 1 hour. [Note 2]
10. After 1 hour, turn off the thermowell, hot plate, and water flow.
11. Cool down and separate the reaction flask with reflux condenser.

Figure 6. Reaction mixture (after 1 hour)

B. Reaction work-up

B.1 Quenching

1. Fix the separatory funnel with clamps and turn the

stopcock to close the end of funnel.

Figure 7. Fixed separatory funnel

2. Add 20 mL of saturated sodium bicarbonate solution to the reaction flask.

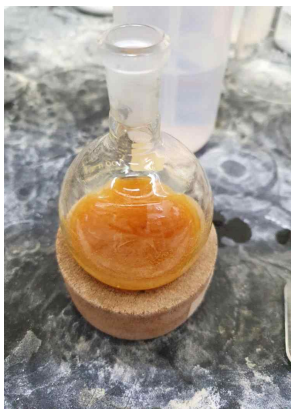


Figure 8. reaction mixture quenched with 20 mL of saturated sodium bicarbonate solution

3. Trasfer the resulting mixture into the separatory funnel.

Figure 9. Trasfer mixture into the separatory funnel.

B.2 liquid-liquid Extraction

Caution: During the pressure releasing work, **don't point the**

end of the funnel to a person. Erupted solution can splatter on a person.

1. Using funnel, add 20 mL of ethyl acetate to the separatory funnel.

Figure 10. Add 20 mL of ethyl acetate to the funnel.

Note 3. Grab a funnel cap tightly when you shake separatory funnel.

2. Cap the separatory funnel.
3. Shake the separatory funnel and slowly open the cock to release the gases from mixing. [Note 3]
4. Close the cock

Figure 11. Shake the separatory funnel

5. Repeat 3-4 for 4 times.
6. Fix the funnel with clamp and open the cap.
7. Wait for the mixture to separate into two layers.
8. Set 125 mL Erlenmeyer flask under the funnel.
9. Open the cock until the bottom layer (aqueous layer) falls to the flask.
10. Close the cock.
11. Add 20 mL of saturated sodium bicarbonate solution to the separatory funnel.
12. Repeat shaking, removing bottom layer, and add 20 mL of saturated sodium bicarbonate solution until the bottom layer (aqueous layer) remains clear.

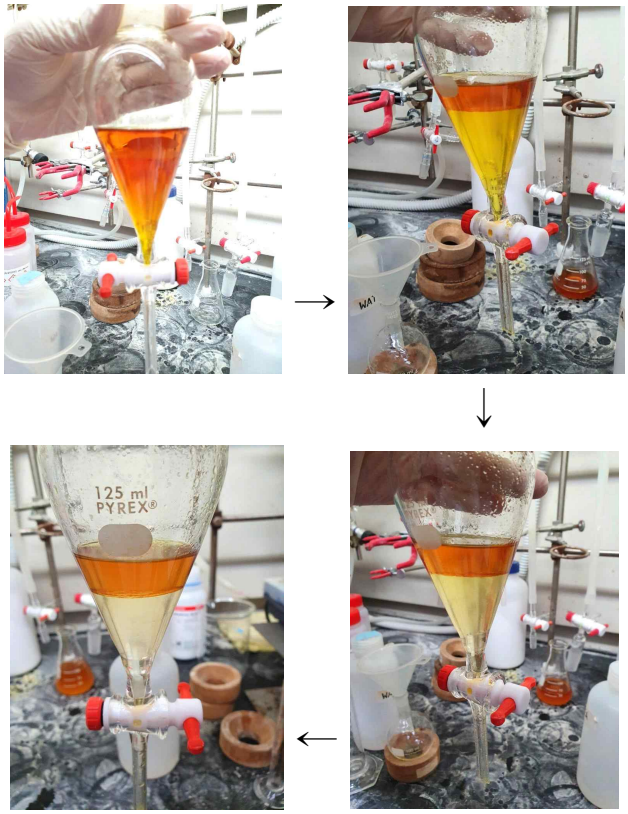


Figure 12. Color change of sodium bicarbonate layer

B.3 Drying by rotary evaporator

1. Measure the mass of 100 mL round bottom flask.
2. When the bottom layer of the funnel remains clear, remove the bottom layer and transfer remaining layer (ethyl acetate layer) into a pre-weighed 100 mL round bottom flask.

Figure 13. Collected ethyl acetate (top) layer

3. Connect the 100 mL round bottom flask to rotary evaporator and fix with keck clip.
4. Set the temperature of heating bath 30~50℃.
5. Turn on the rotary evaporator.
6. Control the pressure control valve to maintain the inner space of rotary evaporator to be almost in a vacuum state. When the solution boils, slightly open the pressure control

valve.

7. When the solvent evaporate completely, turn off the rotary evaporator and fully open the pressure control valve.
8. Separate the 100 mL round bottom flask from the rotary evaporator.

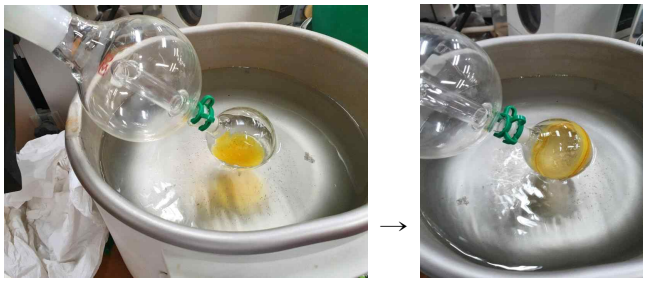


Figure 14. Removal of solvents by rotary evaporator.

9. Measure the mass of the round bottom flask with final solid product.

Figure 15. Final product (4-amino-3-nitrobenzoic acid methyl ester)

10. Calculate percentage yield of the reaction.

C. Characterizing the Product

NMR analysis

Assign the structure of the product based on the ^{1}H NMR spectra provided by TA. (Use DMSO-d_6 as a solvent)

Pre-Laboratory Questions

1. Summarize all the MSDS of chemicals used in this experiment.

2. Draw an arrow-pushing mechanism for the formation of the product in this experiment.

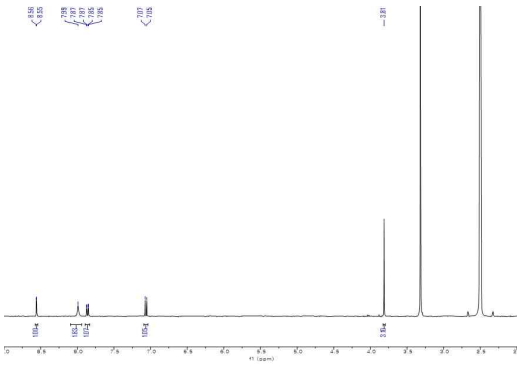
3. Write a net transfer of one or more components from one liquid to another liquid phase during liquid-liquid extraction step.

Post-Laboratory Questions

1. Explain why saturated sodium bicarbonate solution was used in the liquid-liquid extraction.

2. Assign the ^{1}H NMR spectra of the product (4-amino-3-nitrobenzoic methyl ester).

^{1}H NMR (400MHz; DMSO-d6)



3. Suggest at least one way to improve the reaction yield.