Experiment 5 – Preparation of Fruity Fragrances

Reading Assignment Mohrig Chapter 7.1 (Reflux), McMurry Chapter 13.11 (^{1}H NMR splitting), McMurry 21.3 (Fischer Esterification), & 21.10 (^{1}H NMR of esters)
Review Topics: Mohrig Chapter 5.3 (transferring liquids), 6.1-2 (heating)

Esters encompass a large family of organic compounds with broad applications in medicine, biology, and industry. Esters are represented by the structure R-CO-OR', in which R and R' are alkyl or aryl groups. Esters are widespread in nature, occurring naturally in plants and animals. Small esters, in combination with other volatile compounds, produce the pleasant aroma of fruits. In general, a symphony of chemicals is responsible for specific fruit fragrances; however, very often one single compound plays a leading role. For example, an artificial pineapple flavor contains more than twenty ingredients but ethyl butyrate is the major component. Some examples of ester flavors and fragrances are shown in **Figure 1**. In contrast to previous experiments where students isolated compounds from plants, in this experiment, students will synthesize these compounds in the lab.

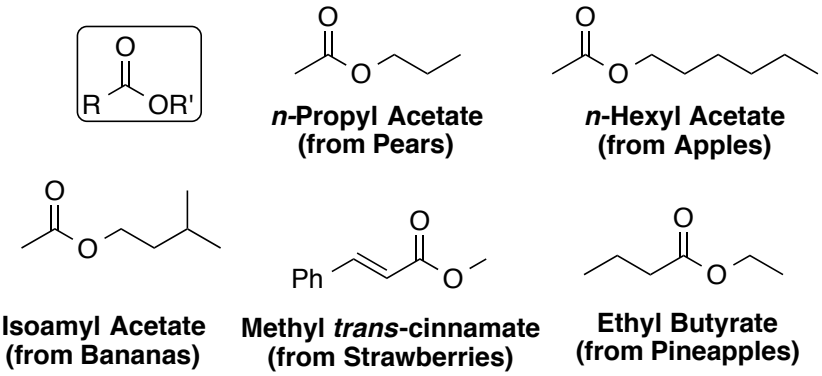

Figure 1. Examples of esters found in essential fruit oils.

Esters are carboxylic acid derivatives. They can be obtained by heating a carboxylic acid with an alcohol in the presence of catalytic amounts of mineral acids such as sulfuric or hydrochloric acids. This reaction is known as the Fischer esterification (**Figure 2**). This reaction is reversible and thus is limited by its equilibrium constant. A large excess of one of the reactants (which can be recovered at the end of the reaction) will push the equilibrium to favor products, thus increasing the yield. Constantly removing the product will also increase the yield. A Fischer esterification is only recommended with primary and secondary alcohols and unhindered carboxylic acids. Steric hindrance near the reaction center slows down the esterification.

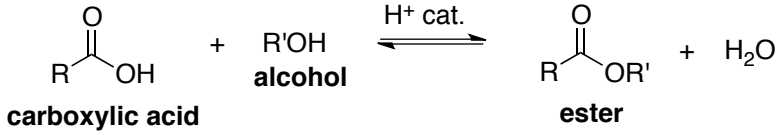

Figure 2. General scheme for a Fischer esterification.

In this experiment, you will prepare either isoamyl acetate (banana oil) from acetic acid and isoamyl alcohol or *n*-hexyl acetate (sour apple fragrance) from acetic acid and *n*-hexanol. Incidentally, isoamyl acetate is also the alarm pheromone of the honeybee and thus, it should

be kept away from beehives! You will perform the synthesis at the microscale level using a Fischer esterification under refluxing conditions. A round-bottom flask is topped with a water-cooled condenser. The contents of the flask are heated to the solvent's boiling point. The solvent vapors travel up and inside the reflux condenser, where they condense back to a liquid and fall back into the round-bottom flask. This allows the system to remain open while heating, but without losing any reaction components. This particular reaction is run "neat" or without solvent. The acid and alcohol are both liquids and act as the solvent. It is important that the reaction flask does not run dry and that *cold water* be running through the condenser at all times.

An acid-base extraction will be performed with an aqueous bicarbonate solution to separate the ester from the unreacted carboxylic acid. No organic solvent is necessary for the acid-base extraction because these esters are liquids and separate from the aqueous solution as an immiscible layer.

Time and quantity permitting, you will purify the ester product by microscale distillation using a Hickman still and a water-cooled condenser. The product will be analyzed by a chemical test and IR and compared to the alcohol standard to observe conversion to ester. Students will not obtain the IR of the starting materials, but instead will get these spectra from the TA.

Notebook Preparation

**Students will work individually on this experiment.*

Purpose – one-sentence description of the purpose plus the reaction scheme for the ester you will synthesize (**Figure 3**).

- Students in Rooms 257 (formerly 249) and 271 will make sour apple using *n*-hexanol.
- Students in Room 261 will make banana oil using isoamyl alcohol.

Reagent table – amount (mg or mL), mmol, equiv*, MW, bp or mp, density, and one-word hazards for each of the chemicals in the scheme above. Leave space in the table to write the actual amounts of starting materials used. Also include HCl and hydroxamic acid in the table.

* molar equivalents, described in experiment 3

Procedure – hand-written, step-by-step procedure in your own words. Draw a diagram of the reflux set up into your notebook. Include a list of materials.

Safety & Clean-up – copy the table at the end of the procedure into your notebook.

EXPERIMENTAL PROCEDURE

Preparation of *n*-Hexyl Acetate and Isoamyl Acetate

The statements in quotes are provided to give students guidance in writing the experimental methods section. One well-written sentence can explain an entire paragraph's worth of information! You will be expected to follow simple instructions such as these on the lab practical.

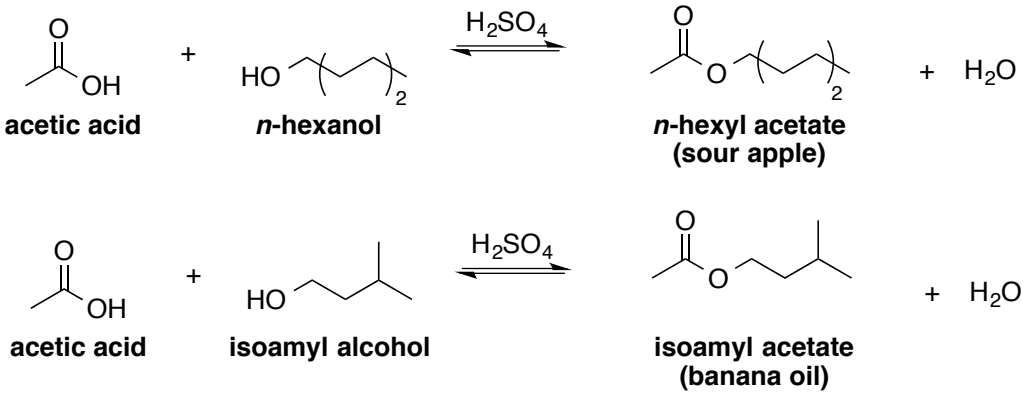

Figure 3. Reaction schemes for Fischer esterification

"*To a 15-mL RBF was added…[chemical name (mmol)]…and heated to reflux for 1 hour.*"

Pre-heat a sand bath on a hot plate at a medium setting (you may set this up as soon as you enter the lab, before the TA's pre-lab talk). Dispense 10 mmoles of the desired alcohol (isoamyl alcohol or *n*-hexanol) and 40 mmoles of glacial acetic acid into a 15-mL round-bottom flask (RBF) using a glass pipet and pluringe. *The conversion from mmoles to mL should be calculated and entered in the reagent table before lab.* Add 3 drops of sulfuric acid and magnetic stir bar then attach a microscale water-jacketed condenser. Be sure the water is running through the condenser and reaction is stirring before heating. Heat to reflux in the sand bath and allow the reaction to reflux for one hour. During this time, work on NMR spectroscopy problems in the McMurry text (Chapter 13 problems 19-21 and Chapter 21 problem 70). Your TA should check your solutions for credit before the end of lab – do not turn these in.

"The reaction was quenched and washed with…"

Carefully lift the apparatus from the heat and allow the mixture to cool to ambient temperature. Disassemble the apparatus and turn off (clamp) the water, but keep the hoses attached. Do not wash the condenser at this stage, as it will be used later. Transfer the liquid to a 16 x 125 screw-cap test tube with a pipet. Rinse the RBF with 2 mL of 5% $NaHCO_3$ in 15% NaCl solution. Slowly transfer the rinse to the screw-cap test tube. Stir the mixture with a microspatula until gas evolution (carbon dioxide) has subsided. Cap the tube and invert it several times to mix the layers. Frequently vent the system to release the pressure by momentarily unscrewing the cap. Let the system settle for about 10 minutes.

Use a pipet to transfer the lower (aqueous) layer to a labeled test tube. Keep this until the end of the experiment then discard it. Wash the organic layer remaining in the test tube twice with 1 mL (each time) of the $NaHCO_3$-NaCl solution. Invert and vent well in each wash. Collect the aqueous washes in the same labeled test tube as before.

"The combined organic layers were dried (Na_2SO_4) and filtered to afford…The crude reaction mixture was purified via distillation (or not) and products analyzed by IR."

Dry the organic layer by adding a microspatula-ful of anhydrous Na_2SO_4. Note that this drying agent is more granular than $MgSO_4$ and will create a similar but not identical snow-globe effect when sufficient drying agent is added. It may be necessary to add more, however, this may effect how much liquid can be obtained after filtration. After 5 minutes, remove the liquid from the drying agent using a pipet loosely packed with a small piece of cotton. Approximate whether you have at least 0.4 mL of product by comparing it to a test tube of the same size containing 0.4 mL of water. If there is less than 0.4 mL of product, transfer it to a pre-weighed vial, weigh the product, and proceed to IR analysis and chemical tests.

If there is over 0.4 mL of product, transfer the product to a 5 mL RBF. Assemble the distillation apparatus using a Hickman still, boiling chip, and microscale condenser (condenser goes on top of the still). Distill by heating in a sand bath on a hot plate at a medium setting until two or three drops of liquid remain in the flask. If the Hickman still is full before the distillation is complete, carefully remove the condenser and empty the still with a pipet. Transfer the distillate to a pre-weighed vial, weigh the product, and analyze the product by IR.

"Product formation was confirmed (or not) by the hydroxamic acid test for esters."

Performs this test with product alongside the starting materials (alcohol and acetic acid) as well as ethyl acetate (an ester). Prepare four labeled test tubes. Add one drop of sample to 1 mL of 0.5 M hydroxylamine hydrochloride (NH_2OH-HCl) in 95% ethanol in a test tube. Add 0.2 mL of a 6 M NaOH solution drop-wise and a boiling chip. Bring the mixture to a boil by heating in a water bath. Let the system cool and add 2 M HCl drop-wise until the pH is 2-3. If cloudiness develops, add 2 mL of 95% ethanol. Add 2 drops of 3% ferric chloride solution. A red-violet color is a positive test.

Table 1. Clean-up & Safety

Clean-up	**Safety**
Liquid waste: aqueous layers and solutions from chemical test	H_2SO_4, HCl, hydroxamic acid, NaOH, and acetic acid are *corrosive*
Solid waste: Na_2SO_4, pipets, filter pipets	Acetic acid, ethyl acetate, and ethanol are *flammable*
After analysis, dispose of your product in the liquid waste using a *very small amount* of ethanol from a wash bottle to aid the transfer.	Isoamyl alcohol is an *irritant*
Wash all glassware and wipe down counters; return shared glassware to reagent counter	Wear gloves & goggles throughout the experiment.
Clean IR plates with acetone saturated with NaCl. Return plates to the dessicator after use.	

Introduction: Pre-lab Questions

1. Describe the Fischer esterification reaction in one-to-two sentences, including functional groups involved and reaction conditions. Show the full arrow-pushing mechanism for the ester you will synthesize.

2. Why is the reaction mixture extracted with sodium bicarbonate and sodium chloride solution? What role does each salt play? Draw the reaction that sodium bicarbonate takes place in.

3. Calculate the mass (mg) and volume (mL) of alcohol and acid that will be mixed from the mmol given in the procedure. Include these values in the reagent table in your notebook. Show your work.

4. Calculate the theoretical yield of the synthesis in mg (assuming the reaction goes to completion). Show your work.

5. How is Le Chatelier's Principle used to ensure the reaction will go to completion?

Results: In-Lab Questions

1. Calculate the percent yield of the synthesis and discuss any source of product loss. List the sources of intrinsic error including the percent intrinsic error used in each measurement.

2. Based on the techniques you have learned in the organic chemistry lab, how would you separate any unreacted alcohol from the ester? This is assuming that distillation did not do an adequate job of purifying the product. Briefly discuss how you would go about performing such a separation (*hint: the 8M syllabus contains a schedule of experiments, one of which is applicable here).*

3. Interpret the IR of the starting materials and products. Briefly discuss how you know your reaction went to completion (or not).

4. Report and interpret your hydroxamic acid test results. What do the results tell you about the success of your experiment?

5. Interpret the ^{1}H NMR spectra provided in lecture for *both* banana oil and sour apple, regardless of which was synthesized. Spectra provided in lecture and posted online. Use the following table format and add as many rows as needed to analyze each signal. Create separate tables for each ester and be sure to draw the structures with each set of H's labeled (A, B, C, etc.). You may use the lecture handout for the NMR analysis of banana oil, as long as it's legible!

Signal	Integration (# of H's)	Expected Chemical Shift (ppm)	Observed Chemical Shift (ppm)	Multiplicity/ Splitting (m = n+1)	# of 3-bond Neighbors (n)

Experimental Methods

Guidelines and sample experimental methods section are available online. Remember the sample experimental contains way more information than is pertinent to CHEM 8 students! Use the bold headings in the Exp 5 procedure to get an idea of the level of detail to include in the experimental methods section (you may use those words). You will need to fill in your own data and descriptions in place of "…" Simply report whether the presence of an ester was confirmed by the hydroxamic acid test (no details). IR is the only form of characterization to report, as you are not directly analyzing your sample by NMR.

Adapted from…Palleros, D. R. "Preparation of Fruity Fragrances," *Experimental Organic Chemistry*, **2000**. Wiley: Hoboken. p. 473 – 485.

Exp 5 – Fruity Fragrances Name _____

Section Day _____ Time _____ TA Name _____

CHEM 8M GRADING RUBRIC - Use as cover page for report

SECTION	INSTRUCTOR COMMENTS	POINTS ASSIGNED
IN-LAB QUIZ		**/ 5**
LAB REPORT		
ABSTRACT One paragraph, four sentences: Purpose, procedure, main result(s), and conclusion(s).	NONE	/ 0
INTRODUCTION Original responses to pre-lab questions with TA initials		/ **30**
RESULTS The main results are stated, as outlined in the in-lab questions, using complete sentences.		/ **40**
EXPERIMENTAL SECTION The experimental details (including final amount used and obtained) are *briefly* described in a few sentences. See writing guidelines and sample experimental online.		**/ 15**
NOTEBOOK PAGES Proper format: reaction scheme, chemical info table, procedure, waste and clean-up procedure.		/ **20**
NEATNESS AND ORGANIZATION Proper order and format (see syllabus for full descriptions of each section).		**/ 10**
DRAWER CLEAN-UP All equipment clean, no extra items.		**/ 5**
NMR PROBLEMS McMurry 13.19-21 and 21.70. Completed during reaction reflux or shown to TA in office hours.		Up to 5 extra credit points ☺
	LAB REPORT TOTAL	**/ 125**