Hydrogenation and Transesterification of Vegetable Oils with GC Analysis

During a hydrogenation reaction, the number of carbon double bonds in a molecule is decreased by reacting a sample with hydrogen using a metal catalyst. During this lab experience, we will perform a hydrogenation reaction on vegetable cooking oil. The vegetable oil will be transformed into a vegetable fat or shortening. Reducing the number of double bonds within the carbon chain of triglyceride oil changes the number and strength of intermolecular interactions. We will see the macroscopic result of these molecular changes during this lab. Commercially, vegetable fats are used instead of vegetable oils to prepare certain foods where a fat gives a superior product compared to an oil (i.e., cookies, pie crust, biscuits). The production, transportation, and storage of vegetable-derived fats is more economical than animal-derived fats (i.e., butter).

We will be analyzing the original vegetable oil and your partially hydrogenated product from the using capillary gas chromatography (GC). The GC analysis requires a more volatile sample than a triglyceride. To increase the volatility, you will react the original vegetable oil and your partially hydrogenated product with sodium methoxide (the sodium salt of methyl alcohol, $Na^+CH_3O^-$) to convert the triglyceride ester to three fatty acid methyl ester molecules (FAMEs) and glycerol (propane-1,2,3-triol).

Prelab Questions:

1. Draw a triglyceride with the following carbon chains 16:1 cis-9; 18:2 cis-9,12; 18:3 cis-9,12,15.
2. Draw the same triglyceride after a successful hydrogenation reaction.
3. Draw the three methyl ester molecules that would result from a transesterification reaction of the triglyceride drawn in question #1.
4. Draw glycerol (propane-1,2,3-triol).
5. Why do you think the individual methyl ester molecules will have a higher volatility than the original triglyceride?

Partial hydrogenation of a vegetable oil

- Set up a boiling water bath on a hot plate using a 250 mL beaker.
- Select a medium test tube with a smooth top (no nicks or chips in the glass). Use a spatula to add a small pinch of palladium on charcoal. Add a vegetable oil to a depth of about 1 cm.
- Fill a balloon with hydrogen gas to about the size of a grapefruit, pinch the neck of the balloon, and slip it over the top of the test tube. Place the test tube with balloon in the water bath and agitate the mixture every few minutes by shaking the test tube gently.
- After 10 minutes of heating, pinch the neck of the balloon and remove. While you hold the inflated balloon, your partner should pour a few drops of the mixture onto the bottom of an inverted beaker. Replace the hydrogen-filled balloon and continue the agitation and heating as before. Record your observations of the mixture immediately after pouring and when it cools.
- Check again every 10 minutes, watching for visible signs that a chemical reaction has occurred and recording your observations. Why does hydrogenation change the properties of the mixture?
- Save your product. Clean up using soap to wash your glassware. Why?

Transesterification (FAME preparation) of the original vegetable oil

- To a large test tube, add 3 drops of the original (unreacted – from the commercial bottle) vegetable oil sample to be analyzed. It is important to use the same oil that you used at the start of the previous lab. Add 2 mL of 0.5 mole NaOH/ L dry methanol and 1 mL dry tetrahydrofuran (THF). Place the test tube in a water bath at 40°C for 10 minutes. Remove the test tube from the water bath, allow it to cool to room temperature. Add 2 drops of glacial acetic acid and 5 mL of distilled water. Swirl the test tube to mix.
- To extract the fatty acid methyl esters from the reaction mixture, add 5 mL of heptane. Stopper the tube and gently shake the test tube to mix. After the layers separate, use a dropper to transfer the upper heptane layer into a small, dry beaker. Be careful not to collect any charcoal. Repeat the procedure with another 5 mL of heptane, combining the two heptane extractions. Add a spatula full of sodium sulfate to the heptane solution to absorb water.
- Pour the heptane solution into a labelled, clean, dry 50 mL beaker, leaving the sodium sulfate behind.
- Leave the beaker in the hood to evaporate off the heptane solvent until the next lab.

Transesterification (FAME preparation) for the hydrogenated vegetable oil product.
Use the product from the hydrogenation reaction in the ester exchange reaction following the same transesterification procedure as above.

Gas Chromatography Analysis (GC)
The GC instrument has a capillary inside an oven. The oven is warm enough to evaporate a sufficiently volatile sample into the gas phase. The individual sample molecules are carried through the capillary by a carrier gas (in this case helium) where they repeatedly interact with the surface of the chemically-modified capillary. A detector at the end of the capillary records the time required for molecules to transit the length of the capillary known as the retention time. Each type of molecule will have a specific retention time. GC is a comparative analytical technique. The retention time of an unknown is compared to a known standard that has also been analyzed on the same instrument. For this lab, you will analyze the transesterification products of both the partially-hydrogenated product and the original vegetable oil produced in Lab 15. The retention times will be compared to a FAME standard that was analyzed by the instructor.

Take a pen, notebook, and both beakers (one with the esters of the original oil and the other with the esters of the partially hydrogenated product) to the GC in SC 410. Inject 0.2 µL of the ester solution. The GC is equipped with a capillary Supelcowax 10 column, and the oven programmed to hold at 230°C for 6 minutes, then increase to 250°C. The following are the GC Instrument Parameters:

Inlet Parameters

- Split 100:1 with Helium carrier gas
- Injector temp 275 °C
- Pressure 23.3 psi
- Total flow 120.2 mL min^{-1}

Detector Parameters

- Detector temp 300 °C
- Reference flow 30 mL min^{-1}
- Helium flow 2.0 mL min^{-1}

Use the chromatograms of the FAME standards, provided by the instructor, to identify each peak in your chromatograms by the retention time of the peaks. Label each peak with the name of the fatty acid it represents and with the condensed notation (e.g., 18:2 or 20:0). Label each peak with the relative percentage composition.

1. How did the macroscopic properties of your oil change during hydrogenation reaction?

2. What change at the atomic/molecular level would account for the observable macroscopic property changes during hydrogenation? Why?

3. Attach the gas chromatographs for the GC analysis **before** and **after** the hydrogenation reaction. Complete the following table.

NAME	% Composition **Before** Hydrogenation	% Composition **After** Hydrogenation	Change in % **[(After-Before)/Before]**	**Chemical Line Notation**
Methyl palmitate (C16:0)				
Methyl stearate (C18:0)				
Methyl oleate (C18:1 cis-9)				
Methyl linoleate (C18:2 cis-9,12)				
Methyl linolenate (C18:3 cis-9,12,15)				

4. For each FAME, interpret how the change in % composition upon hydrogenation is accounted for by the chemical reactions taking place during the hydrogenation reaction.

    a. Methyl palmitate (C16:0)
    b. Methyl stearate (C18:0)
    c. Methyl oleate (C18:1 cis-9)
    d. Methyl linoleate (C18:2 cis-9,12)
    e. Methyl linolenate (C18:3 cis-9,12,15)