# Dyeing to Degrade...

*A Bioplastics Experiment (Version A)*

**Introduction:**

Plastics or synthetic polymers are materials that are ubiquitous in our daily lives. Consider the carpet on the floor, the cell phone in your hand, or the container storing your lunch. The characteristics of these materials – their strength, resistance to temperature change, flexibility, and the ability to be molded into various shapes – make them indispensable to modern society. Currently, the vast majority of plastics are derived from fossil fuels which are non-renewable and are quickly being depleted. Of even more concern is the accumulation of plastics both on land and in water causing harmful effects to humans and the environment. Though some plastics can be recycled, only 9.5% of the total plastic waste generated in 2014 was recovered for recycling.[1] Additionally, most plastics are non-degradable and will last hundreds, if not thousands of years.

Paul Anastas and John Warner developed 12 green chemistry principles that focus on sustainability and provide a framework for scientists to use when designing new products or processes. Principle seven states "a raw material or feedstock should be renewable rather than depleting whenever technically and economically practical." Principle ten states "not only do we want materials and products to come from renewable resources, but we would also like them to not persist in the environment."[2]

The purpose of this experiment is to synthesize and evaluate the degradability of polymeric materials made from renewable and non-toxic components also known as bioplastics. Bioplastics are plastics created from plant materials like corn, sugarcane, or potatoes instead of oil and are often compostable. In this experiment, your bioplastics will be made of citric acid, glycerol, and tapioca starch.[3] Citric acid (Figure 1) is a natural preservative present in citrus fruits. Glycerol (Figure 2) is the backbone of all fats and oils and is a common food additive. Plant starches (Figure 3) are themselves polymeric and easily isolated from corn, potato, and tapioca root and used in many processed foods. You will use a combination of these starting materials to synthesize large molecular weight polymers (many repeating units). For more details about polymer structures and properties see the student supplemental information (at the end of this handout).

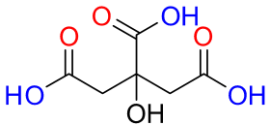



**Figure 1.** Citric acid

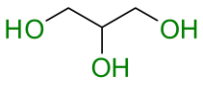



**Figure 2.** Glycerol

[1] U.S. Environmental Protection Agency, Wastes – Resource Conservation – common Wastes & Materials http://www.epa.gov/osw/conserve/materials/plastics.htm (accessed July 2017).

[2] Anastas, P. T.; Warner, J.C. Green Chemistry: Theory and Practice, Oxford University Press, 1998, p. 30.

[3] Washam, C. Plastics Go Green. *Chemmatters* **2010**, No. April, 10–12.

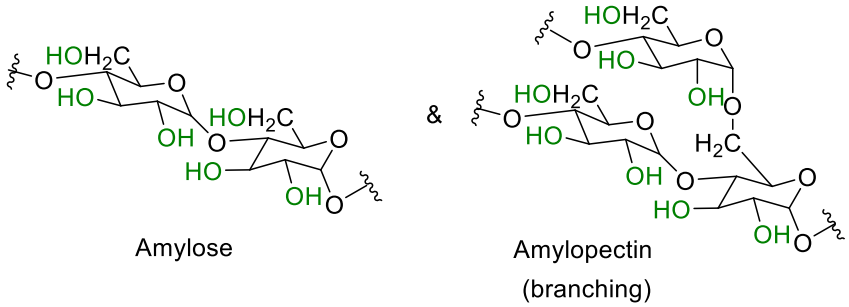
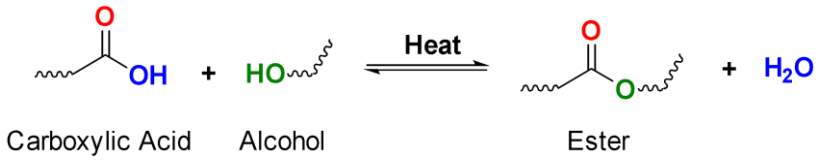



**Figure 3.** General structure of plant starches comprised of mixtures of amylose and amylopectin

**Understanding the Reactions:**

*Citric acid and Glycerol:*

The combination of citric acid (represented by the carboxylic acid) and glycerol (represented by the alcohol) involves the formation of an ester bond as illustrated by Scheme 1 below. In this reaction the carboxylic acid groups of citric acid react with the alcohol groups to form the ester bond and eliminate water. Strong heating and longer reaction times are required to drive off the water, therefore, these samples will be heated in an oven at 100 °C for 2-7 days. Note that this is an equilibrium reaction and Le Chatelier's Principle would apply.




**Scheme 1**. General ester formation.

*Starch and Glycerol:*

The starch/glycerol polymer sample is different from the citric acid polymer. First, vinegar is used to break down the amylopectin branching and create linear chains of the amylose and dextrin (smaller linear chains like amylose). Since there are no carboxylic acids in either the glycerol or amylose, ester bonds are not produced. Instead, a network of hydrogen bonds is created. The alcohol groups on the glycerol molecule forms hydrogen bonds with the alcohol groups associated with the starch (Figure 4) which adds flexibility to the polymers. This is called a plasticizer. This is not a chemical reaction so longer reaction times and heating are not required. Also, loss of water in the context of Le Chatelier's Principle is not relevant.

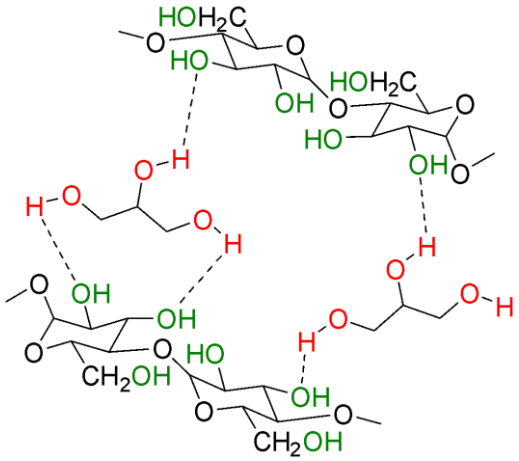


**Figure 4.** Hydrogen bonding between glycerol and starch (amylose)

*Starch and Citric Acid:*

Because the starch has alcohol functional groups and the citric acid has carboxylic acid groups, there is some possibility for formation of ester bonds formation. However, strong heating (100 °C) and longer reaction times would be needed. There is also the high potential for hydrogen bonding between the alcohol groups and carboxylic acid groups.

# Part 1: Synthesis of Bioplastic Samples

**Purpose:**

The purpose of part 1 of this experiment is to synthesize three different bioplastic samples. The samples will be stored and left to dry for part 2.

**Safety:**

- Wear safety goggles.
- Label all beakers/containers of chemicals at your workstation.
- Use caution when working with the hot plate and hot glassware.
- Consult the MSDS for more information about the compounds you are working with.

**Materials:**

*Chemicals (per group):*

- Tapioca starch (5.2 grams)
- Citric acid (2.5 grams)
- DI Water (22 mL)
- White vinegar (2 mL)
- Glycerol (3.5 grams)
- Canola oil cooking spray (small amount)
- Yellow food coloring (3 drops)

*Equipment (per group):*

- Scale (1)
- Small beakers (4)
    - Suggested: (2) - 100-150 mL beakers and (2) - 50 mL beakers
- Weigh paper (2 pieces)
- Scoopulas (2)
- 10 mL graduated cylinder (1)
- Glass stir rod (1)
- Small aluminum boats (3) – 4.4 x 1.3 cm (hold 20 mL)
- Hot plate (1)
- Tongs (1)
- Paper towel (1)
- Permanent marker for labeling

**Pre-Lab Questions:**

1. Three different polymers will be made: 1) a starch and citric acid polymer, 2) a starch and glycerol polymer, and 3) a citric acid and glycerol polymer. Based on the reactions described in the introduction, what types of bonds/interactions will be formed upon synthesis of each of these polymers.

2. Which polymer sample(s) would you expect to be the strongest? Explain.

**Procedures:**

*Sample 1: Starch and Citric acid Polymer:*

1. Mass 3.2 g of tapioca starch directly into a 150 mL beaker.
2. Mass 0.5 g of citric acid on a piece of weigh paper and add it to the beaker.
3. Measure 10.0 mL of DI water with a graduated cylinder and add it to the beaker.
4. Measure 1.0 mL of white vinegar with a graduated cylinder and add it to the beaker.
5. Stir the contents of the beaker with a glass stir rod to mix completely.
6. Place the beaker on a hot plate set to a low setting (2-3 of 10).
7. Stir the mixture frequently until it begins to thicken (~10-15 minutes). Do not overheat or mixture will get clumpy. If it clumps form, remove from heat and stir vigorously.
8. Once thickened, remove the beaker from the hot plate.
9. Add 1 drop yellow food coloring and stir with a glass stir rod to mix completely.
10. Label the bottom of an aluminum dish as sample 1 and with your names.
11. Transfer the mixture to the aluminum dish and place back on the hot plate until the polymer is translucent throughout (~30 minutes).
12. Remove the aluminum dish from the heat and store it to dry (48-72 hours) until the second portion of the experiment.

*Sample 2: Starch and Glycerol Polymer:*

1. Mass 1.1–1.5 g of glycerol directly into a 150 mL beaker.
2. Mass 2.0 g of tapioca starch on a piece of weigh paper and add it to the beaker.
3. Measure 10.0 mL of DI water with a graduated cylinder and add it to the beaker.
4. Measure 1.0 mL of white vinegar with a graduated cylinder and add it to the beaker.
5. Stir the contents of the beaker with a glass stir rod to mix completely.
6. Place the beaker on a hot plate set to a low setting (2-3 of 10).
7. Stir the mixture frequently until it begins to thicken (~10-15 minutes). Do not overheat or mixture will get clumpy. If it clumps form, remove from heat and stir vigorously.
8. Once thickened, remove the beaker from the hot plate.
9. Add 1 drop yellow food coloring and stir with a glass stir rod to mix completely.
10. Label the bottom of an aluminum dish as sample 2 and with your names.
11. Transfer the mixture to the aluminum dish and place back on the hot plate until the polymer is translucent throughout (~40 minutes).
12. Remove the aluminum dish from the heat and store it to dry (48-72 hours) until the second portion of the experiment.

*Sample 3: Citric acid and Glycerol Polymer:*

1. Mass 2.0 g of glycerol directly into a 50 mL beaker.
2. In a separate 50 mL beaker, mass 2.0 g of citric acid directly into the beaker. Measure 2.0 mL of DI water and add to the beaker. Stir with a glass stir rod to dissolve completely.
3. Add 1 drop of yellow food coloring to the citric acid solution and stir with a glass stir rod to mix completely.
4. Add the citric acid solution to the glycerol and stir with a glass stir rod to mix completely.
5. Label the bottom of an aluminum dish as sample 3 and with your names.
6. Lightly spray a paper towel with canola oil spray and wipe in the bottom of an aluminum dish.
7. Pour the citric acid and glycerol solution into the aluminum dish.
8. Place the aluminum dish in a 100 °C oven for 2–7 days. Heating at temperatures above 110 $^{o}$C can result in the formation of bubbles, which may affect results.

**Waste Disposal:** All of the starting materials are common, non-toxic chemicals often found in foods and can be disposed of in the trash or dissolved in water and disposed of down the sink.

**Post-Lab Questions:**

1. Record your observations of the polymer samples before they are dry.

| | |
|---|---|
| *Sample 1:* | |
| *Sample 2:* | |
| *Sample 3:* | |

2. What properties/qualities do you think the polymer samples will have once they are dry?

# Part 2: Degradation of Bioplastic Samples

**Purpose:**

The purpose of part 2 of this experiment is to examine the degradation of the bioplastic samples that you synthesized during part 1 via spectrometry or colorimetry. You will also be qualitatively examine the degradation a PET or PLA plastic brought from home.

**Safety:**

- Wear safety goggles.
- Label all beakers/containers of chemicals at your workstation.
- Sodium hydroxide is a skin and eye irritant. Use caution and wash any exposed areas immediately.
- Consult the MSDS for more information about the compounds you are working with.

**Materials:**

*Chemicals (per group):*

- Bioplastic samples from part 1
- PLA or PET plastic sample (brought by students from home)
- 0.0160 drops/mL standard yellow dye solution (10 mL)
- 1.0 M sodium hydroxide (up to 36 mL)
- DI water (13.5 mL)

*Equipment (per group):*

- Spectrometer or colorimeter (optional if smartphone is available)
- Smartphone (optional if spectrometer or colorimeter is available)
    - Suggested: downloaded camera application (like Camera FV5-Lite)
- White paper
- Timer
- Cuvettes (8)
- 10 mL test tubes (3)
- 10 mL graduated cylinder (1)
- Pipettes or droppers (1)
- Kimwipes (several)
- Electronic balance (1)
- Glass stir rod (1)
- Scissors (1)

**Pre-Lab Questions:**

1. Record your observations of the polymer samples now that they are dry. In addition, record your observations of the PLA or PET sample brought from home.

| | |
|---|---|
| *Sample 1:* | |
| *Sample 2:* | |
| *Sample 3:* | |
| *PLA or PET Sample* | |

2. What is PET? What resource is it made from? Is it recyclable? Is it degradable?

3. What is PLA? What resource is it made from? Is it recyclable? Is it degradable?

4. Based on bonding, how do you predict the rates of degradation will compare for the three bioplastic samples?

**Procedures:**

*Degradation of PLA or PET Sample (brought from home):*

1. Cut up the plastic sample that you brought and add ~0.5 g of sample a small beakers. You may want to cut your sample into smaller pieces.
2. Add 10-15 mL of 1 M NaOH to the beaker.
3. Let the sample sit for ~1 hour while you are running your other degradations.
4. At the end of 1 hour, make note any evidence of degradation of the plastic sample.

*Degradation of Bioplastic Samples:*

**Measurement via Spectrometer:**

*Calibration Curve:*

1. Obtain 5 cuvettes and three test tubes.
2. In the first cuvette, add 3.5 mL of standard yellow solution and label this standard 4.
3. In the first test tube combine 1.0 mL of DI water and 3.0 mL of the standard yellow solution.
4. In the second test tube combine 2.0 mL of DI water and 2.0 mL of the standard yellow solution.
5. In the third test tube combine 3.0 mL of DI water and 1.0 mL of the standard yellow solution.
6. Pour 3.5 mL of each solution into the second, third, and fourth cuvettes and label these standard 3, standard 2, and standard 1, respectively.
7. In the fifth cuvette add 3.5 mL of DI water and label this blank.
8. Wipe each cuvette with a Kimwipe before placing in the spectrometer.
9. Record the absorbance of each cuvette in the data table below.

*Degradation:*

1. Mass ~ 0.1 g of polymer, by cutting the polymer with a scissors to an appropriate size (try to use only one piece). Record the exact mass of sample used.
2. Add 3.5 mL of 1 M NaOH to a cuvette.
3. Calibrate the spectrometer to 425 nm.
4. Add a polymer sample to the cuvette and start a timer considering this 0 minutes.
5. Stir the sample for 3 seconds and let sit (to allow particulates in the starch samples to settle) until the timer reaches 5 minutes.
6. Place the cuvette in the spectrometer to take a 5 minute reading.
7. Repeat steps 5 and 6 until there is no more sample visible in the cuvette.
8. Repeat for each different polymer sample.

**Measurement via Smartphone:**

*Calibration Curve and Degradation:*

1. Line up your calibration curve cuvettes from the spectrometer portion of the experiment in front of a white background.
2. Set up a smartphone with an app that allows you to lock white balance and focus such as Camera FV-5 Lite. Set to take pictures once every five minutes.
3. To each of three clean cuvettes add 3.5 mL of 1M NaOH and put in front of the white background to the left of the other cuvettes.
4. Mass ~ 0.1 g of each polymer, by cutting the polymer with a scissors to an appropriate size (try to use only one piece). Record the exact mass of sample used.
5. Add each polymer sample to a separate cuvette of NaOH putting sample 1 in the leftmost cuvette, then sample 2 in the next cuvette, and sample 3 in the next and start a timer, considering this 0 minutes.

6. Stir each polymer sample for 3 seconds with a glass stir rod and let sit until the timer reaches 5 minutes.
7. Take a picture.
8. Repeat steps 5 and 6 until there is no more sample visible in the cuvette.
9. View the pictures on a computer with a color picking application.
10. Choose three points in each cuvette to record the blue intensity, being careful to avoid shadows and glares.

**Waste Disposal:**
All of the degradation materials are common, non-toxic chemicals and can be disposed of in the trash or dissolved in water and disposed of down the sink.

**Data:**
Data Table 1. Absorbance of Standard Solutions (Measurement via Spectrometer):

| | Blank | Standard 1 | Standard 2 | Standard 3 | Standard 4 |
|---|---|---|---|---|---|
| Absorbance | | | | | |

Data Table 2. Exact Mass of Polymer Sample Used (Measurement via Spectrometer):

| | Sample 1: Starch and Citric acid | Sample 2: Starch and Glycerol | Sample 3: Citric acid and Glycerol |
|---|---|---|---|
| Mass | | | |

Data Table 3. Absorbance of Degrading Sample (Measurement via Spectrometer):

| | Sample 1: Starch and Citric acid | Sample 2: Starch and Glycerol | Sample 3: Citric acid and Glycerol |
|---|---|---|---|
| Time (min) | Absorbance | Absorbance | Absorbance |
| 0 | | | |
| 5 | | | |
| 10 | | | |
| 15 | | | |
| 20 | | | |
| 25 | | | |
| 30 | | | |
| 35 | | | |
| 40 | | | |
| 45 | | | |

| | | | |
|---|---|---|---|
| 50 | | | |
| 55 | | | |
| 60 | | | |

Data Table 4. Intensity (B) of Standard Solutions (Measurement via Smartphone):

| Standard Solution | $B_1$ | $B_2$ | $B_3$ |
|---|---|---|---|
| blank | | | |
| 1 | | | |
| 2 | | | |
| 3 | | | |
| 4 | | | |

Data Table 5. Exact Mass of Polymer Sample Used (Measurement via Smartphone):

| | Sample 1: Starch and Citric acid | Sample 2: Starch and Glycerol | Sample 3: Citric acid and Glycerol |
|---|---|---|---|
| Mass | | | |

Data Table 6. Intensity (B) of Degrading Sample (Measurement via Smartphone):

| | Starch and Citric acid | | | Starch and Glycerol | | | Citric acid and Glycerol | | |
|---|---|---|---|---|---|---|---|---|---|
| Time (min) | $B_1$ | $B_2$ | $B_3$ | $B_1$ | $B_2$ | $B_3$ | $B_1$ | $B_2$ | $B_3$ |
| 0 | | | | | | | | | |
| 5 | | | | | | | | | |
| 10 | | | | | | | | | |
| 15 | | | | | | | | | |
| 20 | | | | | | | | | |
| 25 | | | | | | | | | |
| 30 | | | | | | | | | |
| 35 | | | | | | | | | |
| 40 | | | | | | | | | |
| 45 | | | | | | | | | |
| 50 | | | | | | | | | |
| 55 | | | | | | | | | |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| 60 | | | | | | | | | |

**Data Analysis and Calculations:**

*Calibration Curve (Beer's Law Plot) from Spectrometer Measurements*

1. Calculate the concentration of each standard solution and record in the table below.

| Blank | Standard 1 | Standard 2 | Standard 3 | Standard 4 |
|---|---|---|---|---|
| | | | | 0.0160 drops/mL |

2. Construct a concentration vs. absorbance plot for the standard solutions.

3. Add a linear trendline to this plot and record the equation for the line below. Note the correlation coefficient ($R^2$) value. If this value is not sufficiently close to 1, you may consider remaking your standard solutions.

*Calibration Curve (Beer's Law Plot) from Smartphone Measurements*

1. Calculate the concentration of each standard solution and record in the table below.

| Blank | Standard 1 | Standard 2 | Standard 3 | Standard 4 |
|---|---|---|---|---|
| | | | | 0.0160 drops/mL |

2. Calculate the average blue intensity ($B_{AVG}$) for each standard solution and record in the table below.

| | Blank | Standard 1 | Standard 2 | Standard 3 | Standard 4 |
|---|---|---|---|---|---|
| $B_{AVG}$ | | | | | |

3. Convert the average intensity to an absorbance value using the equation: $A = -\log(I_{sample}/I_{blank})$ where A is the absorbance of the sample and I is the intensity of the sample. Record the values in the table below.

| | Blank | Standard 1 | Standard 2 | Standard 3 | Standard 4 |
|---|---|---|---|---|---|
| Absorbance | | | | | |

4. Construct a concentration vs. absorbance plot for the standard solutions.

5. Add a linear trendline to this plot and record the equation for the line below. Note the correlation coefficient ($R^2$) value. If this value is not sufficiently close to 1, you may consider remaking your standard solutions.

*Comparing the Rates of the Degradation (From Spectrometer and Smartphone Measurements):*

1. Using the equation for the trendline, convert absorbance to concentration for absorbance measurement.

2. Construct a plot of concentration vs. time.

3. Repeat this for each different sample.

4. Compare the time it took for each sample to degrade and list in order of fastest to slowest.

5. Compare the slopes of the concentration vs time plots and list of order of fastest rate to slowest rate.

6. Do the orders in question 4 and 5 agree? If not, hypothesize why absorbance may deviate from true values for some of the samples.

**Post-Lab Questions:**

1. Based on the bonding, did the samples degrade at the relative rates expected? Explain.

2. How did the degradation of the bioplastic samples compare to the degradation of the PLA or PET sample brought from home?

# Supplemental Information for Students
# Polymer Primer

**Polymer Basics:**

A polymer is a large molecule made of small repeating units called monomers. The number of repeating units is designated by "*n*" and can vary in number from 50 to 100,000. See a representation of a simple polymer made from one monomer unit in Figure 1a. A well-known example of a synthetic polymer would be polystyrene. Styrofoam cups and containers are composed of polystyrene. Polymers made by combining two or more different monomers are called copolymers and can be represented as illustrated in Figure 1b. A well-known example a synthetic copolymer is styrene-butadiene-styrene (SBS) rubber. Tires and the soles of shoes are composed of SBS rubber. Additionally, polymer chains can be covalently bonded to each other through a process called cross-linking (Figure 1c). The physical and mechanical properties of a polymer depends on the structure of the monomer(s) used, the size of the polymer chain ("*n*" value), the composition and positioning of each repeating monomer chain, extent of cross-linking, and degree of entanglement of the polymer chains (Figure 1d).

**Figure 1a. Two representations of a polymer molecule from one monomer**

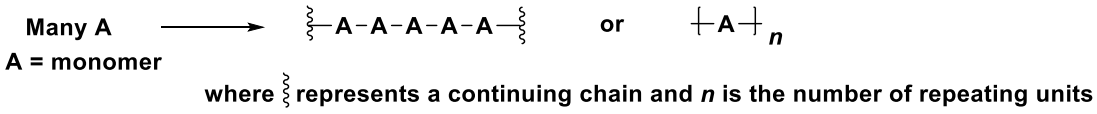



**Figure 1b. Two representations of a polymer made from two different monomers**

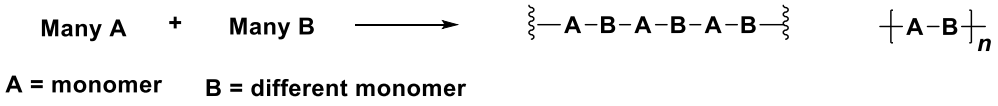



**Figure 1c. Example of cross-linking of polymer chains**

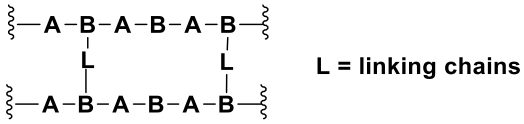
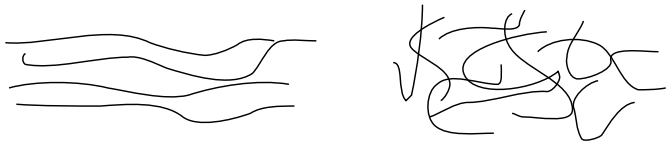
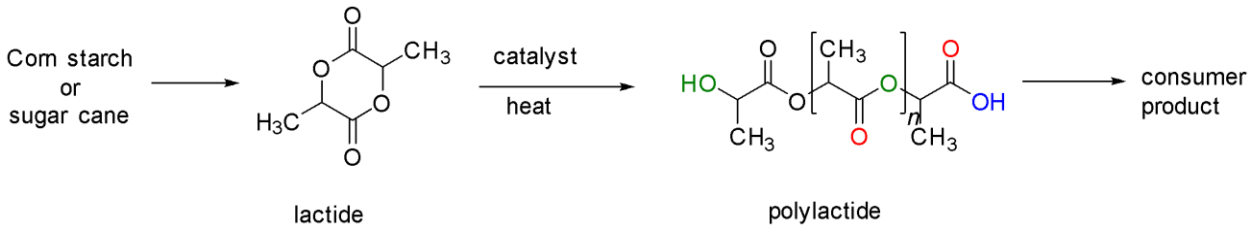



**Figure 1d. Linear packing polymer chains versus entangled polymer chains**



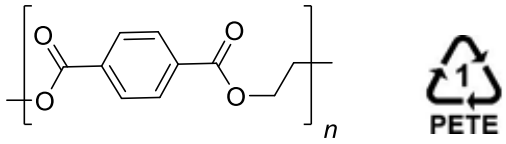

**Biodegradable Polymers/Plastics:**

A biodegradable polymer is one that can be broken down by naturally occurring microorganisms such as bacteria or algae. An example of a renewable and degradable polymer that can be found on campus in dining services is called PLA or poly(lactide). You can recognize this polymer by the word "compostable," or trade names of "Natureworks," "Greenware," or "Ingeo" on the bottom of these products. The starting material is derived from cornstarch or sugarcane and its products degrade under compostable conditions over several months. Its structure, illustrated in Scheme 1, also contains ester bond linkages.




**Scheme 1.** Generic synthesis of poly(lactide), PLA, used in compostable bioplastics

A common non-renewable polymer used for soda pop bottles is polyethylene terephthalate, PETE, which though recyclable, will not biodegrade for an estimated 450-1000 years depending on conditions. Most consumers recognize this polymer by the recycle code of “1” (Figure 2).




polyethylene terephthalate

**Figure 2.** Structure and recycle code for polyethylene terephthalate

**For Instructor’s guide, contact jwiss@umn.edu**