BIOPLASTICS KIT

MAKING MANUAL

www.amino.bio

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BIOPLASTICS KIT™
MANUAL

Getting Ready

The Experiment

More information

Welcome! Let's get started

This user guide was created to help you get the most out of your Amino Labs experience. Even if you are familiar with making new materials, bioplastics, science or other Amino Labs™ products, please take the necessary time to read through this guide. This will ensure you practice safe science as well as store, use, and get the most out of your kit. It will also let you know what to do in case of a spill or accident.

In the first section, you will learn about your kit's components, how to store them before and during your experiment, as well as a few tips on activities to complete before you get your hands 'wet'. The second section is procedural -- these are the step by step instructions on how to run your experiment. Make sure to follow our tips to ensure your best success! The third section covers "what's next"; how to keep your creations, store or dispose of any leftover ingredients and general clean up instructions. The final section is there to help you -- a glossary, troubleshooting, and our contact information.

Amino Labs is excited to welcome you to the world of Bio-design with the Bioplastics kit! The first in a series of experiment to get you making with biology! **Following this guide will help ensure that you are getting the most out of your current and future experiences to keep on making new creations. Have fun!**

Practicing safe science - Bioplastics

Science is a safe activity when you follow simple guidelines. Read on to ensure you adopt safe practices.

The kit in your hands contains only inert ingredients - no live organisms. The ingredients are all safe to use without any special containment or training. Most of the ingredients will be familiar to you from your kitchen. Even though these are familiar, you should never eat or taste any part of this kit, or any ingredients you use for science experiments. Follow these safety guidelines for your safety and the success of your experiment(s)!

We recommend the system and kits for ages 12+, under adult supervision, and 14+ with or without supervision. The cleaning instructions must be strictly followed for safety and experiment success. Make sure to store the kit per the instructions found in this booklet.

- Do not eat or drink near your experiments. Keep your experiment at least 10 feet from food, drinks, *etc*. Under no circumstances should you eat any of the kit's content.

- Wash your hands before and after your experiment,

- Wear gloves, is recommended even when cleaning your station or handling the kit contents as part of a safe science practice. This will protect you from your experiment, and your experiment from you. Any latex, nitrile, or general purpose gloves you can find at the pharmacy will do. After you put your gloves on, be aware of what you touch. Try not to touch your face or scratch itches with your gloved hands!

- Clean up your work station, spills and work surface before and after use. Use a 10% solution of chlorinated bleach generously sprayed onto a paper towel and rub onto any contaminated surfaces. (Careful! This can discolor your clothes). A chlorinated spray cleaner also works.

- With the Bioplastics kits, all the discarded materials can go in the garbage. Do not pour bioplastic mixture down the sink. Find a container to hold the inactivation bag where you will discard used items. If you are familiar with other Amino Labs kit, you can use a discard container at your station to hold all your materials, just like when doing your genetic engineering experiments.

- **Eye-wear is not provided but can be worn.**

While these also include rules specific to experiments with live organisms, you can still download a biosafety poster for your space from www.amino.bio/biosafetyinaction and complete a short safety quiz at www.amino.bio/biosafety-quiz

If you would like to do a short Online lab safety course for your edification, we recommend a Government of Canada course: www.amino.bio/biosafety

The Big Plastic Problem & bio-based plastics

Plastics are a well-known material that is used to make our modern world go round. There is a high probability that you have many types of plastics around you right now. Can you spot them? You may see an appliance, electrical cord, clothing, shoes, pet food or water dish, or a garbage bin. You'll likely notice many types of plastics with many different properties. But did you know that all plastics have a similar basic chemical structure?

What are plastics?

Plastics are chains of repeating molecules linked together. These "strings" of repeating molecules are called polymers. Since these chains are called polymers, plastics are also often called polymers. You might have noticed that many plastic names start with "poly ." Now you know why! Different plastic properties are a result of the types of atoms and molecules that make up the polymer chains. These repeating molecules can either all be the same (also called a monomer) or by a few different molecules that repeat in a pattern (a true polymer).

Bakelite buttons. Source: Science History Institute, CC BY-SA 3.0, via Wikimedia Commons

The process of creating plastics is called polymerization. During polymerization, the smaller molecules, called monomers or building blocks, are chemically combined to create the polymers. The first "modern" plastic was Bakelite, a material that could not be melted again when formed. Bakelite was introduced in 1907 by chemist Leo Hendrik Baekland. He also coined the term 'plastics' to describe this new category of material. Since the introduction of plastics, most polymers have been produced with petrol-based compounds because petroleum-based compounds are cheap, easy to obtain and process, and very durable.

Just what is the Big Plastic Problem?

You are probably aware that many scientists, innovators, and students like yourself are now looking to biology to solve the big plastic problem. But why? Traditional plastics have three significant impacts on the environment: they use fossil fuels, have a large carbon footprint, and stay in the environment for hundreds of years. One major problem with plastic that needs to be addressed right away is that it is too durable! Most petroleum-based plastics are not biodegradable; instead, they degrade so slowly that they stay in the environment, landfills, beaches, and oceans for decades up to hundreds of years. While several factors impact the timeline to degrade plastics, like the polymer it is made from and the environment it is discarded in, on average, single-use plastic bottles will take about 450 years to degrade, while a plastic bag can take anywhere from 10 to 1,000 years to degrade in a landfill!

Plastic debris disintegrates over time into smaller particles of various sizes and shapes under natural processes like UV radiation and mechanical abrasion. These are referred to as secondary microplastics and form more than 80% of microplastics found in the environment. Source: Oof.cc, CC BY-SA 4.0, via Wikimedia Commons

Unfortunately, the problem doesn't end here. As we continue to learn about the presence of plastic in the environment, science indicates that some of these petroleum-based plastic actually do not entirely go away. If they are not recycled into new plastic products, petroleum-based plastics like PVC will break down into smaller pieces that stay in our environment, water systems, and soil. These are sometimes referred to as "forever plastics" or microplastics, and they've been found in all our oceans, marine life, drinking water, and even in human bodies! While the impacts of these microplastics on living organisms, plants, animals, and human health are still being studied, there is just no escaping them. Scientists have seen microplastics in most places they have looked: In deep oceans, in Arctic snow and Antarctic ice, in shellfish, table salt, drinking water, human blood, and even drifting in the air or falling with rain over mountains and cities.

This is why more and more people are looking at ways to degrade existing petroleum-based plastics. Others are working to create new types of plastics made with biological materials that can degrade quickly, do not rely


Gelatin-based bioplastics dyed with algae extract.

on fossil fuel as a source of the monomers, and have a smaller carbon footprint.

What are bioplastics?

These new types of plastics made with biological-based ingredients are called bioplastics. Early research indicates that they could be a significant improvement over petroleum-based plastics. To clarify, bioplastics are plastics made at least in part with renewable biological matter (like vegetable fats and oils, corn starch, woodchips, sawdust, and food waste), or they are plastics that can degrade in a reasonable time.

It might surprise you to learn that bioplastic production is much older than the petrol-based plastics we use today! In 1500 BC, people in Egypt were already using materials made of gelatin and casein (a protein found in milk) for furniture constructions. And it's not all ancient history either; in the 1940s, industrial pioneer Henry Ford experimented with soya bean plastics. Cool! The history and potential of bioplastics are rich and inspiring.

Today, bioplastics are being developed and tested as sustainable replacements for single-use plastics for packaging, utensils, food containers, 3D printing, fashion, and even medical implants. But since bioplastics have different properties than their petroleum-based counterparts because they are mostly made from biodegradable ingredients, it can require an innovative mindset to use them industrially. For example, since some bioplastics use ingredients that can dissolve in water, the plastics themselves can also dissolve in water! This property can be an advantage in some situations, but it can also limit its use. For example, bioplastic utensils and bowls that dissolve in contact with liquid will not help bring hot leftovers home and eat them. Like any technology, bioplastics will have their ideal use cases, current limitations, and potential for improvements.

And it is now your turn to join bioplastics innovators! Let's learn how to make bioplastics. Further, you'll learn about some typical bioplastic ingredients and how bioplastic properties compare to the traditional plastics you already know.

Discover the Bioplastics Kit™

The Bioplastics kit lets you combine different biological ingredients to create a range of fun materials including collagen-based plastics using gelatin, starch plastics, biocomposite plastics using food waste as fillers, and a red-algae plastic using agar. Creating these bioplastic will help you learn some of the basics skills of making materials.

By following the experiment instructions on the next pages, you will:

- discover the typical ingredients used in bioplastic by academia, industry and artisanally,
- gain or better your skills like weighing, mixing, measuring, dyeing, texturing, casting and extruding,
- learn how to add renewable food, agricultural or industrial waste to your bioplastic to change their properties. These additive, called bio-fillers, are often used in industry to bring additional properties to the final material.
- create a usable bioplastic product.

In a second (optional) part of the experiment, you will be able to explore material analysis skills and concepts. Keep your bioplastics once they are dry if you are interested in completing this part. You can find the material analysis instructions in the Bioplastics analysis instruction manual (www.amino.bio/instructions).

Once you have learned these fundamental plastic recipes and techniques, you will be able to start experimenting on your own. Super! This kit can be used alone or in a small group (with parent supervision if you are 14 or under). In the next pages, you will find descriptions of the kit's content.

Before you start: collecting some food waste to use as a "bio-filler"

For one of your biocomposite recipe on Day 4, you will need to add dry food wastes from your own kitchen or cafeteria. We recommend using fruit peels like those of an orange, egg shells or even used coffee grounds and letting them dry before you get to the Day 4 experiment. You will need ~5 grams of dried and powdered filler for this experiment.

To dry the filler: To air dry your bio-filler, leave the peel/grounds/shells to dry at room temperature (near a hot vent works well) until they are fully dry and easy to crumble. This can take up to 48 hours. To oven dry your bio-filler Set your oven to 250 degrees Fahrenheit and lay the filler on a clean cookie sheet or similar (do not grease the cookie sheet Let your filler dry in the over for a total of 1.5 hours. After 45 minutes, flip the filler so that it dries evenly. If you are using egg shells, you should place the shells in hot, boiled water for 10 minutes to clean and sterilize them before you dry them. Take out the shells after 10 minutes and pour the water down the sink.

To grind/powder the filler: If you have a mortar and pestle or spice grinder, use it to turn your filler into powder. If you don't, take a plastic bag, add your filler and close it tight. Take a bottle, rolling pin, or cylinder object and roll over the bag back and forth until you get a power. Make sure the filler is dry before you do this, otherwise it will be hard to powderize!

Kit components

Measuring/mixing beaker(s): Beaker(s) help you measure and heat the water you need to make your bioplastics, and to mix your ingredients.

Syringes : To help you measure and dispense water, glycerol and other liquid ingredients. The two syringes with the plastic caps will also be used to extrude strings of bioplastics on Day 3

Tube of oil (~10ml): This tube of vegetable oil will help you lubricate your mold and containers to easily take out your bioplastics once dry. If you make bioplastics outside of this kit, you can use household oils, like olive, canola or coconut as mold lubricant (also called mold release).

Blank labels: These blank sticky labels will help you keep track of which containers have which bioplastics as they dry. If you make more bioplastics in the future, you can use masking tape to replace the blank labels.

Silicone tray: This tray will be an extrusion and drying surface for a few of the bioplastics you will make. The tray can be washed with soap and water if needed

Paintbrush: You will use a paintbrush/cotton swab to create your bioplastic product and to oil your molds.

Petri dishes bag: You will use these petri dishes to mix and/or dry your different bioplastic mixtures.

Red, blue and yellow dyes, 3 plastic pipettes: 3 tubes of dyes and 3 pipettes to help you dispense them in your bioplastic mixture. Each pipette is assigned a color to prevent contaminating your different dye colors. With yellow, blue and red dyes you will be able to create all colors.

Molds: An array of molds including a star-shaped mold, a 5-rectangles mold, a “dog-bone” mold and 2 rectangular molds to shape some of your bioplastics.

Bottle of glycerol: Glycerol (sometimes also called glycerin) is a colorless, viscous liquid that helps softens and reduces shrinkage in bioplastics. This is because glycerol functions as a plasticizer that bonds with the polymer in your mixture. A plasticizer is something that is added to another material (usually a plastic base) to make that material softer or more pliable.

Bottle of 5% Acetic acid solution: Acetic acid is a clear, colorless, organic liquid with a pungent odor similar to household vinegar. Indeed, acetic acid is the main component of vinegar which is around ~4% acetic acid. Adding acetic acid breaks up some of the polymer chains, making the plastic less brittle.

Tube of liquid soap: A soap solution to texture a bioplastic (may be found in the weigh bag for some kits)

Weigh boats: These diamond-shaped containers are ideal for weighing powders and pouring them into your mixture. While disposable, they can be washed and reused many times when making bioplastics.

Scoopula: A small spoon-like spatula to easily transfer powders to the weigh boats. Rinse and reuse!

Pipette: While disposable, this plastic pipette can be easily rinsed and reused to texture your bioplastics.

Toothpicks: While disposable, these toothpicks can be easily rinsed and reused to mix your bioplastics and help remove them from their molds.

<u>Your powder ingredients:</u>

Gelatin: Gelatin is a colorless powder derived from collagen, which acts as a binding material and a hardening component in bioplastics. It often functions as the polymer in your mixture.

Pectin: Pectin is colorless powder obtained from fruits and plants. It is a polysaccharide starch that functions as the polymer in your mixture. It is also what gives jelliness to jams!

Agar: Agar is a yellowish powder that comes from red algae. It is sometimes used a vegetable replacement for gelatin. Like it, it acts as a binding material and a hardening component in bioplastics. It often functions as the polymer in your mixture. Agar is more flexible than gelatin.

Vegetable starch: Starch is a white flour-like powder obtained from corn, potatoes, and other vegetables. Starch itself is a polymer made by plants to store energy. It can be used as the polymer in your mixture or an additive to another polymer mix.

Chitin/Chitosan: Chitin can be found in a big range of living organisms like sea animals, insects and microorganisms, but is mostly extracted from crab and shrimp shells. It is a natural, extremely abundant polymer and the main component in the cell walls of fungi and gives the structure to the exoskeletons of insects and sea creatures.

Chitosan is one of the most important subproducts of chitin. It is created when chitin goes through a process called deacetylation. This enables the new product, chitosan, to become soluble in acidic solutions while retaining similar structural properties of chitin. This is helpful as chitin itself is insoluble in most readily available solvents.

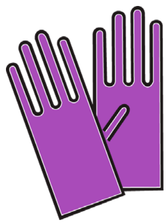
Baking Soda: Sodium bicarbonate, commonly known as baking soda or bicarbonate of soda, is a salt composed of a sodium cation and a bicarbonate anion. You'll most often find it in a white powder form. In bioplastics, in can be used to give opacity to the material, and to create textures.

Unpacking and storing kits

For a better shelf life and successful experiments, place your kit at **room temperature,** away from the light.

Items you will need

- **Access to a microwave and a timer**
- **Marker or pen**
- **Paper towels for clean up**
- **A mixing utensil** like a metal or plastic spoon
- **Small scale** to weigh ingredients from 1.0 g to 9.0 g
- **Distilled or tap water in a cup, glass or bowl** (~520 mL total for the 5 days of experiment)
- **~5g of food waste to use as a bio-filler** like dried fruit peel, egg shells or used coffee grounds. Place them in a bag to crush them into a powder once dry. We recommend using only one filler at a time.



Necessary safety supplies

- **Optional: Latex/nitrile gloves** like those found at a pharmacy. 1 pair/person if you reused them, or 5 pairs/person.
- **Soap and/or spray cleaner** bottle for cleanup

Timeline

The Bioplastics Kit™ includes 5 days of "Making" activity, and an optional 4 days of "Analyzing" activities. Each activity takes between 30 and 50 minutes, with additional drying time of the bioplastics. The drying time is about ~72 hours, depending on where you are. By placing the drying plastics in a hot, dry location, like near a heat vent, you can speed up drying. If you are keen, it is possible to complete all the "Making" experiments in one day! To complete the optional analyzing activities, you will find the instructions in a second manual *Analyzing Bioplastics at* www.amino.bio/instructions.

	Monday	Tuesday	Wednesday	Thursday	Friday
Week 1	**Making: Day 1** Bioplastic bases: Gelatin/Glycerol plastics (40min)	**Making: Day 2** Bio-foams, dyes and textures (50 min)	**Making: Day 3** Extruding strings, ribbons + strength testers (50 min)	**Making: Day 4** Bio-composites: Chitin/Pectin (30 min)	**Making: Day 5** Algea sheet (20-30 min)
Week 2	**Making: Day 5 (cont.)** Algea wallet (20 min) ***Analyzing Bioplastics Day 1:** Material properties of gelatin/glycerol plastics	***Analyzing Bioplastics Day 2:** transparency test (lux) on foams and dyes	***Analyzing Bioplastics Day 3:** Strenght test (advanced) / Walk the plank test (simple) on starch bioplastics	***Analyzing Bioplastics Day 4+:** Biodegradability tests	

*Can be done on the same day, and can be completed as soon as the bioplastics are dry using the *Analyzing Bioplastics* manual.

Day 1: Discover: Bioplastic recipes (Gelatin Bioplastic).
Goal: Test ingredients' properties and learn the basics of bioplastics.
Day 1, ~40 minutes + dry time ~72 hrs

Day 2: Transform: Bio-foams, dyes & textures
Goal: Color, mold, melt and foam your plastics.
Day 2, ~50 minutes + dry time ~72 hrs

Day 3: Extrude: Strings and ribbons (Starch Bioplastic)
Goal: Extrude your bioplastic to make rubbery strings.
Day 3, ~50 minutes + dry time ~72 hrs

Day 4: Additives: Biocomposites (Reinforced Pectin Bioplastic)
Goal: Reinforce and reuse waste to create biocomposite plastics.
Day 4, 30 minutes + dry time ~72 hrs

Day 5: Usability: Algae sheet wallet
Goal: Use all of your new skills to make a product for the everyday.
Day 5, 20 minutes + dry time 24-48 hrs

Day 6, 15-60 minutes

Analyzing Bioplastic manual at www.amino.bio/instructions

A key pitfall to avoid!

In the next pages are detailed, step-by-step instructions to complete the experiment. **It is helpful to read through all of the steps for each part of the activity before starting that day's activity.**

While all the steps outlined in the experiment are important and should be followed as described, the MOST IMPORTANT consideration for success is:

Always make sure that the water is boiling before adding any powder or liquid to make sure the ingredients will fully dissolve. **<u>You have to see the water bubbling</u>!** Be careful, the bottle or bowl will be hot!

Experiment Protocol

Prepare your space

Goal Set yourself up for success.

Materials from the kit

Materials not in your kit
Spray cleaner
Paper towels
Pair of gloves

Make sure you have the necessary materials as explained on page 12, including gloves, access to a microwave, and water before you start. Also make sure that you have read and understood the safety guidelines.

1. Put on your gloves, and if you have one, your lab coat or apron.
2. You can dispose of used items in the garbage. Do not pour liquid bioplastic mixture down the drain as it could solidify in the drain. When you are ready to dispose of your used bioplastics or any extra mixture, you can let it dry before scooping it out into the garbage, or you can pour it directly in a garbage bag.
3. Choose a work surface that is stable, non-porous and easy to wipe (non-porous surfaces are smooth and sealed so liquid and air cannot go into them). Wipe down your work surface with a spray cleaner before starting
4. You will need to let some of your bioplastics dry for ~72 hours in the molds provided in your kit. Once the hot bioplastics are poured into the molds, try to avoid moving the molds for about 20 minutes until the mixture solidifies. This will help avoid spills. After that time, the mixtures will be solid enough for you to move the molds to a more convenient location for longer-term drying.

Day 1: Discover

1. Bioplastic base recipe (Gelatin/Glycerol bioplastics)

Goal **Test ingredients' properties and learn the basics of bioplastics.**

Materials from your kit
Beaker(s)
1x tube of oil
1x gelatin bag
1x glycerol bottle
Mold 1 with 5 rectangular holes
1x 10 mL syringe
1x label
1x large weigh boat (a white plastic "dish")
1x scoopula (wooden "spoon")
1x paintbrush

Materials you provide
1 mixing utensil
Microwave
Paper towels
Marker/pen
100 mL distilled or tap water
Small scale

Today, you will make 5 bioplastics using water, gelatin and glycerol as the ingredients. You will add the ingredients in difference proportions before pouring the mixture into molds for it to try. This experiment will help you learn how the concentration of ingredients can affect the dried bioplastics.

Prepare

1.1. Take your mold with the five long rectangular holes in it and set it down on a level surface

1.2. Use a marker and some labels from your kit to label the rectangles 1, 2, 3, 4, or 5.

1.3. Place a paper towel on your work surface so you can set down your stirring utensil, measuring syringes and other tools.

1.4. Dip your paintbrush or finder into the oil tube and paint a thin layer on the insides of the rectangular molds and the petri dishes with oil. The oil will prevent the bioplastic from sticking to the mold as they dry, acting as a "mold release".

Make your bioplastics

Boil your water

1.5. Fill your beaker with **100 mL** of water. Use the graduated lines of the side of the beaker to help measure.

1.6. Microwave your beaker of water until you see the water boill. This can take between 60 and 90 seconds depending on your microwave. Once you see the water boil, take your beaker out. Careful! The water and beaker will be hot.

Add gelatin

1.7. Take out your scale and make sure units of the scale are set to "grams".

1.8. Take one of the large weigh boats from your bag (the weigh boats are the soft white plastic dishes in your kit). With a marker, write "GEL" on the side of the weigh boat to identify it as the one you will use for gelatin.

1.9. Place the weigh boat on the scale and press "TARE". This will resets the scale's display to zero. Using the tare function is useful when you measure your ingredients, because you don't need to worry about the weight of the weigh boat what you are measuring.

1.10. Use your scoopula (the wooden, spoon-like scoop) to measure out 8.0 g of gelatin from the gelatin bag by adding powder bit by bit into the weigh boat. If you go over 8.0 g, scoop the gelatin powder back into your gelatin bag. Note: Get the weight as close as you can to 8.0 g. If your scale is a 3 decimal point (or more) of precision, don't have to worry about the numbers passed the first decimal place. For example, if your scale reads 8.014 g, that is precise enough for bioplastic!

1.11. Pour your the gelatin powder in your beaker and place your scoopula and weigh boat on the paper towel. You will reuse them later.

Stir and dissolve

1.12. Stir the mixture in the beaker with your mixing utensil until combined–it's okay if there are some clumps.

1.13. Microwave the mixture for 5 seconds at a time until you see it foams. Watch carefully to ensure that it does not foam over the edge of your beaker. (Don't leave your mixing utensil in the beaker when you microwave!)

1.14. Stir the mixture until smooth. If clumps persist after stirring, microwave the mixture for five additional seconds and stir. Repeat until no clumps remain.

Pour the mixture into the mold section labeled "1."

1.15. Using the large syringe, draw **5 mL** of hot bioplastic mixture from the beaker.

1.16. Position the syringe over the mold rectangle labeled "1". Slowly push down on the plunger to fill this first rectangular section of the mold with your bioplastic. . You may not need the entire 5 mL of mixture to fill the first rectangle of the mold, but make sure the bioplastic reaches the top of the mold for best results.

1.17. Pour back any remaining mixture in your syringe into your beaker. Rinse your syringe with hot water.

Congratulations, you made your first bioplastic! This bioplastic is only gelatin and water. Let's now add glycerol to see how it can change the bioplastics properties once dry.

Adjust the ingredients to your mixture to create the bioplastics for "2", "3", "4", "5"

Add glycerol

1.18. For this next bioplastic, you will use the mixture in your beaker and add a some glycerol to it to see how glycerol impacts the properties of a bioplastic once dry. Find your measuring syringe and the glycerol container. Open the glycerol container and dip the tip of the syringe into the glycerol.

1.19. Pull back on the plunger of the syringe to take **4 mL** of glycerol using the markings on the syringe. Dispense it into your mixture in the beaker.

1.20. Stri with your mixing utensil until the mixture looks combined.

1.21. Microwave until you see the mixture foam.

1.22. Stir until the mixture is smooth: you can microwave until you see the mixture foam and stir a few more times if you need.

1.23. Using the syringe dispense **5 mL** of the mixture into the mold's rectangle # "2"

You now know how to add and mix basic bioplastic ingredients: water, gelatin, and glycerol. Repeat these steps to make the bioplastic mixtures to pour into rectangular mold spaces #3, #4 and #5 using the table below. The values in the table lets you know how much of each ingredient to add to the mixture in your beaker. Add the ingredients for bioplastic 3, mix and pour it into the 3rd rectangular space in the mold. Then, move onto bioplastic 4 and then 5.

Day 1: Discover: Bioplastic basic recipes - how much of each ingredient to add to your beaker

Bioplastic	Water to add in milliliter	Glycerol to add in milliliter	Gelatin to add in grams	Concentration of ingredients *	Dispense into mold
1 - gelatin-only	100 mL	-	8 g	0% glycerol 8% gelatin	5 mL in #1
2 - low glycerol	-	4 mL	0.3 g	4% glycerol 8% gelatin	5 mL in #2
3 - equal glycerol/gelatin	-	4 mL	0.4 g	8% glycerol 8% gelatin	5 mL in #3
4 - high glycerol	-	8 mL	0.6 g	15.5% glycerol 8% gelatin	5 mL in #4
5 -equal glycerol/gelatin, low water	-	-	6.4 g	15.5% glycerol 15.5% gelatin	5 mL in #5

* go to www.amino.bio/percentconcentrations to learn more about ingredient concentrations.

1.24. After pouring in the fifth rectangle of the mold, you will still have ~75 mL of mixture in your beaker. Keep this mixture in your beaker until you start Day 2: Transform.

Drying your bioplastics

Dry

1.25. Now that you made and poured five different bioplastics in your mold, you have to let them to dry. The drying time will depend on your room temperature and humidty. Typically, it will take three days (72 hours) for them to dry. You know they are dry once they are no longer gel-like to the touch. If you can, placing them in a hot room can cut down on the drying time.

Once dry, demold your bioplastics

1.26. Once your plastics are dry (no longer gel-like to the touch), remove them from their molds by runnnig on of the toothpicks from your kit between the edge of the bio-plastic and the mold. Grab a corner of the bioplastic to slowly and gently peel it up.

Learn from your experiment

1.27. What differences are there between all your bioplastics? Compare each against each other. If you are interested in doing material science analysis on the bioplastics you created, look at the Bioplastics: Analysis manual (www.amino.bio/instructions).

1.28. Once you have observed and handled your bioplastics, put them back in their mold. You can use them again later on.

Day 2: Transform

2. Bio-foams, dyes & textures

Goal **Create a bioplastics base that you will transform in the next step.**

Materials from your kit
Beaker(s)
1x tube of oil
1x large measuring syringe
1x small measuring syringe
9x labels
1x paintbrush
2x toothpicks
1x large weigh boat (a new one)
1x plastic pipette
1x baking soda bag
1x bottle of Acetic Acid
Your dye bag
1x star mold
1x small liquid soap tube
1x silicone tray

Materials you provide
1x mixing utensil
Microwave
Paper towels
Marker/pen
Distilled or tap water in a cup or bow

Today, you will learn how to transform a bioplastic's color, texture, and shape using the your remaining bioplastic from Day 1. You will use different tools and techniques that change the visual and tactile properties of the bioplastics which you'll be able to apply to all the other bioplastics recipes you will learn throughout this kit and beyond! You will also learn how to reuse bioplastic so that if you ever have leftover mixture or make a mistake in the future, you'll be able to rescue it.

After you prepare your molds and your bioplastic mixture, you will transform your bioplastic in the following order: **Casting a red bioplastic star** **Sequins** **Blown texture (part 1)** **Baking Soda Bio-foam** **Blown texture (part 2)** **Color Opacity Tests** **Soap Bio-foam.**

Prepare

Prepare your molds.

2.1. Set your star-shaped mold, petri dishes, and the red silicon tray on a flat surface. You will be using both the lids and bottoms of the petri dishes as containers.

2.2. Dip your paint brush into the vegetable oil tube and "paint" the insides of the star mold and the petri dishes with oil to make the mold release.

2.3. Take 9 white labels and a marker. Create the labels: 1x "Mixing", 1x "sequins", 1x "blown texture", 1x "soap bio-foam", 1x " soda bio-foam", 1x "1 drop blue", 1x "5 drops blue", 1x "10 drops blue", 1x "15 drops blue"

2.4. Stick your labels as follows:
- The "1 drop blue", "5 drops blue", "10 drops blue", and "15 drops blue" labels each go on the bottom parts of the 4 petri dishes (the bottoms are the bigger parts).
- The "blown texture", "soap bio-foam", "mixture" and "soda bio-foam" each go on one of the lid side of the petri dish.
- The "sequins" label goes on a corner of your silicon tray.

Prepare your bioplastic mixture

2.5. Add **~25 mL** of water to the beaker with the remaining bioplastic mixture from Day 1. No need to be very precise, you are only adding some of the water that evaporated

2.6. Microwave for 30 seconds and check if the mixture has liquified. If the bioplastic mixture is not fully liquid, keep microwaving it in 10 second intervals until it liquefies, You can also add some more water if needed. You should have at least 80 ml of bioplastic before you move onto the next step.

Note: **Always be careful when holding the beaker after you microwave it as the bioplastic will be hot.** You can always put your mixture back in the microwave for 5-10 seconds if it starts to harden as you are using it.

You are now ready to start transforming your bioplastic in shape, color and texture. Let's start!

Casting a red bioplastic star

Casting is a manufacturing and crafting process in which a liquid material is poured into a mold and allowed to solidify before it is taken out. The mold contains a hollow cavity of the desired shape. In this experiment, your mold has the shaped of a star. Some of your favorite objects are made this way, from jewelery to Lego blocks!

Prepare your mold

2.7. Place your star-shaped mold on a flat surface, where you will be able to leave it for at least 20 minutes before moving.

Measure, color and cast your bioplastic

2.8. Using your large syringe, measure **7 ml** of bioplastic from your beaker and dispense it into the petri dish labelled mixture. Note: If your syringe gets clogged with drying bioplastic and cannot take up any new mixture, rinse it in hot water to dissolve the dried bioplastic.

2.9. Take the dye bag and add 1 drop of red dye to your liquid bioplastic. Each pipette has been assigned a color with a dot on its label. Use the corresponding pipette with the colored dye to prevent mixing the source tubes. Keep your dye pipettes; you can place them on your paper towel for later use

2.10. Using your stirring utensil or a toothpick, stir until the color is fully combined.

Casting your bioplastic

2.11. Take your measuring syringe and draw up all the red bioplastic, then dispense it into your star mold until the liquid bioplastic reaches the top but does not overflow. Go slowly! If you have any leftover mixture in your syringe, dispense it back in the 'mixing' petri dish.

Dry

2.12. Dry the bioplastic star ~ 3 days (72 hours), or until it is no longer soft/wet to the touch. The bioplastic start to solidify after 10-20 minutes. Once it is no longer "jiggly" when you touch the mold, you can move it to another drying spot if you need.

Sequins

A sequin is a small, shiny circle usually made of metal or plastic. Sequins are used for ornamentation, especially on clothing and for fast fashion. They are quite taxing on the environment so you will learn to make some bioplastic ones today.

Measure and dye your mixture

2.13. Using the large syringe, measure **4 ml** of bioplastic from your large beaker and add it into the 'mixing' petri dish.

2.14. Add 2 drops of yellow dye to the 'mixing' petri dish and mix until fully mixed. If you want to modify the color, adding more red will give a deep orange.

Making the sequins

2.15. Use the plastic pipette to suction up some bioplastic mixture from your mixing petri dish and gently pipette out small drops of bioplastics on your silicone tray, starting in one corner of the tray. Try to create rounded drops of 5 to 10 mm, leaving at least half a centimeter between each drop.

2.16. Repeat pipetting the sequins on the tray, keeping them to one side of the tray. You can try varying the size of your drops. As they dry, they will become bioplastic sequins. You can add more dye to your bioplastics mixture as you go to change the colors of sequins. Adding more red will give you red-orange tones, adding more yellow will give you a peach color, while adding blue or green to orange will result in deeper shades of brown.

Remember that you can re-melt the bioplastic mixture if it has started to solidify. Place the petri dish in the microwave for 2 to 3 seconds at a time. Repeat if necessary.

Wash and dry

2.17. After you have made all your sequins, rinse your pipette by pipetting hot tap water up and down. Repeat until no visible residue remains.

2.18. Dry the sequins for ~2 days (48 hours), until they are no longer soft to the touch. You can move your surface to a new location after 10-20 minutes

Blown texture part 1 - Prepare your mixture for the blown texture and let sit 5-10 minutes

For the blown texture, you will create the texture on the surface of your bioplastic by infusing it with air. Part 1 is about preparing your mixture and letting it start to solidify.

Measure and dye liquid bioplastic for the blown texture petri dish.

2.19. Dispense **10 ml** of bioplastic mixture from your beaker into the 'blown texture' petri dish using the measuring syringe.

2.20. Add one drop of blue dye and two drops of yellow dye to the mixture. Mix using your toothpick until the color is fully combined.

2.21. Let this petri dish sit for about 6-10 minutes to start solidifying while you do the next bioplastic transformartion.

Baking Soda Bio-foam

For this texture, you will create a foam-like material using an old favorite: the baking soda + vinegar chemical reaction! By adding vinegar and baking soda in the bioplastic, the foam that is created during the chemical reaction will be trapped inside to give your dried sample a foamy finish

Measure and mix your bioplastic.

2.22. Dispense **8 ml** of bioplastic from the beaker into the petri dish labeled "Soda bio-foam" using the syringe. (Microwave your bioplastic to make it liquid again if needed.)

2.23. Using the large syringe, measure 3 ml of 5% acetic acid from the bottle and add it to your "Soda bio-foam" petri dish. Caution! Acetic acid has a strong smell.

2.24. Take a toothpick and mix until the vinegar is fully incorporated.

2.25. Open your bag of baking soda and pour all 3 grams of it into your mixture.

2.26. Using your toothpick, mix the powder into solution. You will see the chemical reaction happen as you mix. Keep mixing until all the powder is dissolved in the liquid.

Dry for 3 days.

2.27. Dry the baking soda bio-foam for ~3 days (72 hours), or until it is no longer soft to the touch. You will notice the bioplastic start to solidify and deflate after 20 minutes or so. This is normal. Once the bio-foam is no longer "jiggly" when you touch the mold, you can move it to another drying spot if you wish.

Blown texture part 2 - texturize!

Add texture to your bioplastic

2.28. Take your "blown texture" petri dish and the plastic pipette you just used to make sequins. Place the pipette tip inside the bioplastic so that the tip rests the bottom of the container at a slight angle. You can try different angles between 0° (straight up) and 45° to find what works best for you as you go through the next steps.

2.29. Gently press on the pipette bulb to release the air that was inside the pipette into the bioplastic mixture. You should see bubbles appearing at the surface. The bio-plastic mixture must have started to solidify for the bubbles to stay on the surface.

2.30. Take the pipette out of the mixture and release the bulb to pull in air. Place the tip back into your mixture and repeat making bubbles until the surface of the bioplastic is covered. Note: Smaller bubbles pop less easily and create more texture when the bioplastic dries

TROUBLESHOOTING: If your bubbles deflate immediately, let the bioplastic rest for an additional 1-2 minutes so that it continues to solidify before trying the bubbles again. If the mixture has solidified too much, you can always microwave it for a 2-3 seconds to bring it back to a more liquid form.

Dry

2.31. Dry the blown bioplastic for 3 days (72 hours), until it is no longer soft to the touch. For the next 10 minutes, the bioplastic will continue to solidfy, after which you can move it. You can keep an eye on it as it dries in case you want to repeat blowing air in if the bubbles pop as it dries.

Color Tests

Time to see if increasing dye in the bioplastic affects material properties. You will increase the amount of dye per dish of your 4 petri dishes, as follows:

- Dish 1: add one drop of blue dye.
- Dish 2: add four drops of blue dye for a total of 5 drops of dye in the mixture.
- Dish 3: add five drops of blue dye for a total of 10 drops of dye in the mixture.
- Dish 4: add another five drops for a total of 15 drops.

Prepare your containers

2.32. Take your 4 petri dish bottoms labeled with “1 drop blue”, “5 drops blue”, ”10 drops blue”, and “15 drops blue” and place them on a flat surface.

Color and dispense your bioplastic

2.33. Add 1 drop of blue dye directly into the remaining liquid bioplastic in your large beaker. Stir until the color is fully combined.

2.34. Measure **10 ml** of the dyed mixture using your large syringe and dispense it into the petri dish labeled “1 drop blue”.

2.35. Add 4 drops of blue dye to the liquid bioplastic in the beaker (for a total of 5 drops, counting the 1 drop you already added). Stir until fully combined and dispense **10 ml** into the petri dish labeled “5 drops blue”.

2.36. Add 5 more drops of blue dye to the beaker of bioplastic (total of 10 drops), stir until combined, and dispense **10 mL** into the “10 drops blue” petri dish

2.37. Add a final 5 drops of blue dye to the beaker (total of 15 drops), stir until the color is fully combined and dispense **10 mL** into the “15 drops blue” petri dish.

Dry

2.38. Dry the bioplastics for 3 days (72 hours), or until they are no longer soft/sticky to the touch. After 20 minutes or so, you can move them to another drying spot.

Soap Bio-foam.

For your final texture, you will use soap to create a different type of foam texture in your bioplastic.

Prepare your bioplastic mixture

2.39. You will be using the remaining bioplastic in your beaker for this step. If you want, you can add some drops of red or yellow dye to change its color.

Foam the bioplastic

2.40. Pour the entire contents of the soap tube from your kit into the large beaker and use your mixing utensil to vigorously stir in the soap, creating bubbles as you go. The more vigorous your mixing, the more bubble you will get!

2.41. Pour some of your mixture, foam and all, into your petri dish lid labeled “foamed plastic” until it is full, but not overflowing. You may have leftover bioplastic foam. You can pour it into your empty ‘mixing’ petri dish, or discard of it in the garbage can.

Dry

2.42. Dry the bioplastics for 3 days (72 hours), or until they are no longer soft/sticky to the touch. After 20 minutes or so, you can move them to another drying spot.

Wash up

2.43. Wash your beaker, stirring utensil, measuring syringes and any other item that has touched wet bioplastic mixture with hot water in the sink. Rinse well with water, until you can no longer smell soap. Dry with a paper towel.

Your results

Take out your dried bioplastics

2.44. Once your plastics are dry (no longer gel-like to the touch), remove them from their containers by running a toothpick between the edge of the bioplastic and the container. Grab a corner of the bioplastic to slowly and gently peel it up.

Learn from your experiment

2.45. What differences are there between all your transformed plastics? Have a look at each sample, and compare them to your Day 1 bioplastics. If you are interested in doing material science analysis, look at the Bioplastics: Analysis manual (www.amino.bio/instructions).

2.46. Once you have observed and handled your bioplastics, put them back in their identified containers. You can use them again later on.

Day 3: Extrude

3. Extruding strings and ribbons (starch bioplastic).

Goal **Create 5 bioplastics mixtures to use for extrusion tests.**

Materials from your kit
Beaker
1x tube of oil (10 mL)
1x silicon sheet
1x three mold casting tray
2x large weighing boat
1x gelatin bag (30 g)
1x glycerol container (30 mL)
1x bag of cornstarch (6 g)
2x syringes with caps (3 mL)
1x large measuring syringe (10 mL)
1x scoopula

Materials you provide
1x mixing utensil
Microwave
Distilled or tap water
Timer
Scale

You will now learn how to extrude bioplastic strings out of a bioplastic recipe that has a bit of a rubber feel. To do this, you will use a new ingredient, vegetable starch and a new technique using syringes. You will also create “dumbbell” out of the same recipe that can be used to test the material strenght of your plastic later on.

You will make 5 different strings and 3 “dumbbell” shapes using 3 version of the same recipe: a control recipe where all the ingredients are in the same proportion, an increased glycerol recipe where there is more glycerol than starch or gelatin, and an increased gelatin recipe with more gelatin than starch or glycerol. For the last two recipes, you will also vary the amount of time you wait before you extrude the strings. Each of these different strings and dumbells will be dyed a different color so you can easily identify them. You will make and extrude bioplastic in the following order:

The control recipe: A. **Red “dumbbell” cast** B. **Red string:** no waiting time before extrusion.

The increased glycerol recipe: C. **Blue “dumbbell” cast** D. **Blue string:** no wait before extrusion. E. **Green string:** ~30 min wait before extrusion

The increased gelatin recipe: F. **Yellow “dumbbell” cast** G. **Yellow string:** no wait before extrusion. H. **Orange string** ~30 min wait time before extrusion

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Prepare

Get your space, molds and water ready.

3.1. Set down your red silicon tray and your mold with the 3 “dumbbell” shapes in it. Choose a very flat and level surface. If your sequins are mostly dry, peel them off and set them aside. You can place them in the empty petri dish bag, for example.

3.2. Fill your cup or bowl with ~75 mL of water and set down one or two paper towels near your work area.

Control bioplastic recipe

Create the bioplastic

3.3. Using your large syringe, measure **15 ml** of water from your cup and dispense it into your beaker. Microwave until the water reaches a rolling boil.

3.4. Add **1 ml** of glycerol to the beaker.

3.5. Using your GEL weigh boat, and the scale, add **1 g** of gelatin to the beaker. (Remember to TARE your scale!)

3.6. Take a new weigh boat and label it “STARCH” . Use it to add **1 g** of starch to the beaker.

3.7. Stir until roughly combined–it’s okay if there are some clumps - and microwave the mixture for 10 seconds at a time until you see it foams. Watch the mixture carefully to ensure that it does not boil or foam over the edge of your bowl.

3.8. Stir until smooth. The bioplastic should be relatively clear but may have bubbles. If clumps persists microwave for five more seconds and stir. Repeat until smooth.

3.9. Add 1 drop of red dye to the bioplastic mixture. Stir with your mixing utensil until the color is even.

A. Red "dumbbell" cast

3.10. Measure **4 mL** of liquid bioplastic using your large syringe and dispense it into one of the dumbell shape in the mold. Go SLOW as you dispense the bioplastic to avoid bubbles. If you press too hard on the plunger, the material can extrude too quickly and create bubbles which weaken your bioplastics. If some bioplastic drips on the sides of the syringe or on the table, clean it with a wet paper towel.

B. Red string

You will use the syringe to extrude long strands on the silicone tray.

Measure out the bioplastic and extrude

3.11. Draw the **10 mL** of remaining red bioplastic from the beaker into your large syringe.

3.12. Remove the air from the syringe by pointing the end of the syringe to the ceiling and gently pressing on the plunger until all the air in the syringe comes out. Go slowly to avoid mess!

3.13. Place the end of the syringe at the top of the silicon tray and extrude a string by applying gentle, even pressure on the plunger with your thumb as you drag the end of the syringe slowly on the surface of the silicone tray until you reach the bottom edge of the tray. Note: Go SLOW! If you press too hard on the plunger, the material will extrude too quickly and create a pool of liquid, rather than thin strands.

3.14. Continue to make strands of bioplastic until you have used up all your red bioplastic. Make the strings as long as you can, but keep them close together on the tray since you will need the space for 5 different strings on the same tray.

TROUBLESHOOTING: Your mixture may extrude unevenly if you press too hard or too fast on the plunger and/or if there are air bubbles in your mixture. If this happens, you can: remove the plunger from the syringe and use your mixing utensil to scoop up the mixture back into the syringe. Then, place your syringe upright into a microwave-safe container like your beaker and microwave for 5 seconds. Try extruding again!

Leave to dry and wash your tools

3.15. Your bioplastic will start to dry. Wash your beaker and mixing utensil with hot water before moving onto the next recipe.

Increased glycerol bioplastic recipe

With this bioplastic recipe using higher glycerol content, you will make one blue dumbbell shape, one blue string, and after letting the bioplastic mixture wait for ~30 minutes in a syringe, a green string. Waiting before extruding the bioplastic with the syringe will give you a different result in your string despite it being the same recipe!

Create the bioplastic

3.16. Create your bioplastic by adding **15 mL** of water to your beaker and microwaving it until it boils. Then, add **2.5 mL** of glycerol, **1 g** of gelatin and **1 g** of starch.

3.17. Stir, microwave for 5 seconds, then mix until smooth.

3.18. Add 1 drops of blue dye to the bioplastic mixture. Stir until the color is even.

A. Blue "dumbbell" cast

3.19. Measure **4 mL** of liquid bioplastic using your large syringe and dispense it into one of the dumbell shape in the mold. Remember to go slow to prevent air bubbles.

B. Blue string

Measure out the bioplastic and extrude

3.20. Draw **6 mL** of blue bioplastic from the beaker into your large syringe and remove the air from the syringe. Keep the rest of the bioplastic mixture in your beaker for now.

3.21. Extrude your blue string as you did before until your syringe is empty.

B. Green string

For this string, you will prepare your syringe but wait 30 minutes before extruding it

Prepare

3.22. Add 3 drop of yellow dye to the remaining blue bioplastic in your beaker and mix. If needed, you can microwave to liquify the bioplastic to mix the color in.

3.23. Take the small syringe with its colored cap. Remove the cap and draw up the green bioplastic mixture until the syringe is full, about 4 mL.

3.24. Remove the air from the syringe as you did before and place the colored cap back on the syringe.

3.25. Make note of the time, or start a 30 minute timer. You can continue to mix and extrude your yellow strings while you wait.

3.26. After 30 minutes, you will extrude a green string using the same method as above. The difference will be in the way the string holds its shape as you extrude it.

How do I know if I am ready to extrude after the 30 minute wait?

3.27. When ready to extrude, the mixture in the syringe should not flow like a liquid when you move the syringe, and the syringe should not be warm to the touch. If you start extruding the strings and the mixture flows like a liquid, wait longer.

3.28. To verify if you are ready start extruding the string on the silicone tray to create a string of ~1 cm. Is it jelly-like and holding its shape? Perfect. You are ready to extrude. If not, let the mixture sit longer.

If you want to speed up the experiment, you can place the syringe in a refrigerator or freezer, but be sure to check back often on your syringe as you do not want the mixture to solidify too much. If it does won't be able to extrude at all! If this happens, place the syringe in a microwave safe container and heat up 2-3 seconds at a time until the mixture is liquid again. Then, restart your timer and wait until the mixture solidifies to a jelly-like consistency.

Increased gelatin bioplastic recipe

With this bioplastic recipe using higher gelatin content, you will make one yellow dumbbell shape, one yellow string, and after letting the bioplastic mixture wait for ~30 minutes in a syringe, an orange string. Once again, waiting before extruding the last string will give you a different result in your string despite it being the same recipe!

Create the bioplastic

3.29. Create your bioplastic by adding **15 mL** of water to your beaker and microwaving it until it boils. Then, add **1 g** of glycerol, **2.5 g** of gelatin and **1 g** of starch.

3.30. Stir, microwave for 5 seconds, then mix until smooth.

3.31. Add 3 drops of yellow dye to the bioplastic mixture. Stir until the color is even.

A. Yellow "dumbbell" cast

3.32. Measure **4 mL** of liquid bioplastic using your large syringe and dispense it into one of the dumbell shape in the mold. Remember to go slow to prevent air bubbles.

B. Yellow string

Measure out the bioplastic and extrude

3.33. Draw **6 mL** of yellow bioplastic from the beaker into your large syringe and remove the air. Keep the rest of the bioplastic mixture in your beaker for now.

3.34. Extrude your yellow string as you did before until your syringe is empty.

B. **Orange string**

For this string, you will prepare your syringe but wait 30 minutes before extruding it

Prepare

3.35. Add 1 drop of red dye to the remaining yellowbioplastic in your beaker and mix. If needed, you can microwave to liquify the bioplastic to mix the color in.

3.36. Take the second small syringe with its colored cap. Remove the cap and draw up the orange bioplastic mixture until the syringe is full, about 4 mL.

3.37. Remove the air from the syringe as you did before and place the colored cap back on the syringe.

3.38. Make note of the time, or start a 30 minute timer. After 30 minutes have passed, come back to extrude your orange string. While you wait, you can wash your tools and clean up your area.

3.39. After 30 minutes, you will extrude the orange string using the same method as above. If you want to speed up the process, you can also place this syringe in a refrigerator or freezer, but be sure to check often so that it does not solidify too much!

How do I know if I am ready to extrude after the 30 minute wait time?

3.40. Just like with the other string, to verify if you are ready start extruding the string on the silicone tray to create a string of ~1 cm. Is it holding its shape? Perfect. You are ready to extrude. If not, let the mixture sit longer. Remember, just like with the green string, when ready, the mixture in the syringe should not flow when you move it, and the syringe should not be warm to the touch. If you start extruding the strings and the mixture flows like a liquid, wait longer. When you extrude, the mixture should be jelly-like and hold its cylindrical shape.

Drying and viewing all your results

Dry and peel your strings and shapes

3.41. Dry all your strings and dumbbell shapes at room temperature for ~3 days (72 hours), until they are no longer gel-like/sticky to the touch. If you can, placing them in a hot room can cut down on the drying time.

3.42. Once your bioplastics are dry (no longer gel-like to the touch), un-stick them from the extrusion surface, by slowly and gently peeling the strands from the surface.

3.43. Take out the dumbbell shapes like you did before by running a toothpick along the edges and gently peeling up the shape.

Learn from your experiment

3.44. What differences are there between all your transformed plastics? Have a look at each sample, and compare them to each other, and your previous plastics. If you are interested in doing material science analysis, look at the Bioplastics: Analysis manual. You will find various ways to analyze your samples.

Day 4: Additives

4. Biocomposites (Reinforced Bioplastics)

Goal Reinforce and reuse waste to create biocomposite plastics.

Materials from your kit
1x tube of oil (10 mL)
1x dye bag
Beaker (s)
3x large weigh boats
1x gelatin bag (30 g)
1x 60ml bottle of 5% acetic acid
1x bag of pectin powder (2.5g)
1x bag of chitosan (3g)
2x rectangular molds
1x 5g bio-filler

Materials you provide
1x Mixing utensil
Microwave, Paper towels, Marker/pen
Bio-filler: ~5 g of dried and ground fruit peel, used coffee grounds or egg shells

Today, you will explore biocomposites. Biocomposites are materials made from a mix of bioplastic and renewable fillers like food and agricultural waste. Biocomposite plastics are a good alternative to traditional petroleum products since they can gain the properties of both the bioplastic ingredients and the filler.

For your biocomposite experiment, you will create 2 recipes. Both will use their own recipes to which you will add the fillers. You will add the chitosan powder from your kit to one. Chitosan comes from chitin, which is a food waste of shellfish. To the other recipe, you'll add the food waste you collected to a pectin based plastic. We recommend using dried fruit peels, egg shells or even used coffee grounds for this since they provide the best results, but feel free to experiment with what you have.

Prepare your molds

4.1. Find the mold with the two rectangular space and oil it to prevent the bioplastic from sticking. Notice one is identfied as "chitosan" and one as "pectin."

Make the Chitosan bioplastic

Make your bioplastic mixture

4.2. Using your beaker, measure **50 ml** of water and microwave it for 60 seconds, or until it reaches a rolling boil.

4.3. Use your large measuring syringe to add **5 mL** of the 5% acetic acid to the beaker. Remember that acetic acid smells very strong! Keep it away from your eyes/face. Tip: Hold the bottle of acetic acid on its side so the syringe can reach the liquid. Ask a classmate or friend to hold the bottle while measure if needed.

4.4. Swirl the beaker to mix the two liquids.

4.5. Pour the entire contents of the chitosan (3 g) bag into the beaker.

4.6. Add **1.5 mL** of glycerol to the beaker

4.7. Add **1 g** of gelatin into the beaker.

4.8. Stir until roughly combined and microwave the solution for 10 seconds, making sure it does not boil over. Caution! The acetic acid will smell even stronger when heated.

4.9. Stir the mixture, until smooth. Repeat microwaving and stirring until your mixture is smooth. Note: This recipe clumps easily. If some clumps of powder remain after multiple attempts at microwaving and stirring, that's OK! Repeat the process up to four times then move on.

Cast your biocomposite

4.10. Pour into the "Chitosan" side of the mold. For this bioplastic, you want to pour enough mixture into the mold so that it does overflow a little. The bioplastic mixture should stay in its shape even as you have mixture over the lip of the mold because it is a biocomposite with some structure. Use a paper towel to clean any that spills.

4.11. You will have some biocomposite mixture remaining in your beaker. Have a look at your samples in the Petri dishes. Are any petri dishes empty, or can any samples be put aside? If so, pour the excess solution into the petri dish(es). If not, you can dispose of the excess mixture in the garbage.

Why make extra? You'll have noticed that this recipe was particularly hard to mix. The bio-fillers, in this case the chitosan, cause the mixture to form a paste as you mix. We've found that creating a smaller volume of bioplastic with this recipe makes it so much harder to dissolve all of the ingredients into a smooth, well-mixed bioplastic. If you do not want to discard of the excess bioplastic but do not have an empty petri dish, you can try to find a container in your class or home that is waterproof and can be washed after use. You've just made yourself a mold! If you have not yet collected and dried food waste filler for your next recipe, you can pour this bioplastic into both sides of the mold and skip the next recipe - the pection bioplastic

Leave to dry and wash your tools

4.12. Dry the chitosan bioplastic for ~48 hours at room temperature, or until no longer sticky to the touch.

4.13. Using your mixing utensil, scrape any solidified bioplastic from the large beaker into the garbage and wash the beaker, mixing utensil and measuring syringe with hot water. Dry with a paper towel. Note: Large amounts of bioplastic will plug your sink. Scrape as much bioplastic out as possible before rinsing the container.

Make the food waste-filler and pectin bioplastic

Prepare your bio-filler

4.14. If your have not yet, take your dried food waster and turn it into a powder using either a food processor, grinder, mortar and pestle, or by placing it into a plastic bag and useinga rolling pin or water bottle to crush it into a powder. The finer the powder, the better it will mix into your material. This powder is your bio-filler.

4.15. Once your bio-filler is powdered, weigh 5 g of it using your scale and a weigh boat (new or cleaned used one). It is okay if you have a bit less than 5 g of bio-filler.

Combine your ingredients

4.16. Pour **50 mL** of 5% acetic acid into your beaker and microwave for 45 seconds, or until it reaches a rolling boil. Careful with the odor as it will be stronger when hot!

4.17. Add all your pectin powder (**2.5g**) to the hot acetic acid.

4.18. Add **1 g** of gelatin and add it into the beaker.

4.19. Stir until roughly combined–it's okay if there are some clumps- and microwave the mixture for 10 seconds, making sure it does not boil over.

4.20. Stir the mixture until smooth. You can repeat microwaving and stirring until your mixture is smooth.

Add your bio-filler and cast

4.21. Take your ~5g of bio-filler powder and stir it into the mixture until smooth.

4.22. Pour into the "Pectin" side of the mold. You will also want to pour enough mixture into the mold so that it rises above the lip of the mold.

4.23. Just like with the chitosan biocomposite, you will have some biocomposite mixture remaining. Can you empty a mold, petri dish, or bowl to pour into? Great! Oil it first, then pour your extra mixture. If not, dispose of it in the garbage. Why make extra? You'll have noticed that with this recipe, achieving a smooth, well-mixed result was difficult once again. The recipe is larger than necessary for the same reasons as with the chitosan biocomposite.

Wash your tools

4.24. Scrape any solidified bioplastic from the large beaker into the garbage and wash the beaker, mixing utensil and measuring syringe with hot water. Dry with a paper towel. Remember that large amounts of bioplastic will plug your sink. Scrape as much bioplastic out as possible before rinsing the container.

Dry your bioplastics and learn from your experiment

4.25. Dry the bioplastics for ~48 hours at room temperature, or until no longer sticky to the touch. Once dry, remove them from the mold gently as they can be fragile.

4.26. Curious how other bio-fillers would influence your bioplastic? You can explore using with different materials like corn husks, sawdust, straw, and different sizes of bio-filler like fine and coarse powder, granular, or even 1 cm size pieces in your mix. To repeat at home, you can use your vinegar and store bought pectin.

4.27. Look to the Bioplastic: Analysis manual to evaluate the material properties test of your biocomposites and head to the Glossary at the end of the manual to learn more pectin, acetic acids, and bio-fillers.

Day 5: Usability

5. Algae sheet wallet

Goal Use all of your new skills to make a product for the everyday

Materials from your kit
1x Paint brush
1x Silicone sheet
Beaker (s)
1x Large weighing boat
1x Bag of agar (9 g)
1x Glycerol container (30 mL)
1x Small syringe

Materials you provide
Mixing utensil
Microwave
Paper towels

Time to create a usable object out of bioplastic - a wallet or small bag - using a new ingredient, agar, and the skills you have learned so far. You will first create an agar bioplastic sheet. Then, once dry, you will turn it into a bag or wallet and use your plastics and skills from previous days to decorate your bag/wallet. Time to get creative!

Prepare

5.1. If you have sequins, strings and labels remaining on your silicone tray from the previous days of experiments, they should be dry enough to move now. Place them on another surface like sheet of paper towel. Since your strings are color-coded, you do not need to worry about labels.

Make the algae sheet

Make the bioplastic mixture and cast the sheet

5.2. Measure **300 ml** of water in your beaker and microwave it until it reaches a rolling boil, about 2 minutes.

5.3. Using the large syringe, dispense **8 mL** of glycerol into the beaker.

5.4. Add the full content of the agar bag **(9 g)** into the beaker and stir until roughly combined.

5.5. Microwave the mixture for 25 seconds, or until it foams and stir until smooth. If clumps persist after stirring, microwave the mixture for 5 additional seconds and stir. Repeat until no clumps remain. You can add more water if the clumps won't dissapear otherwise.

5.6. If you would like a colored algae bioplastic, add a few drops of dye and stir it in with your mixing utensil. Repeat until you are happy with the color.

5.7. Pour all of your mixture into your silicone tray and tilt the tray to ensure that the mixture evenly covers the bottom. Make sure you are on the flatest surface you can find.

1.8. Wash your beaker, syringe, and stirring utensil with hot water in the sink, and dry with a paper towel.

Leave to dry for ~48 hours

1.9. Dry at room temperature for 48 hours, or until it is dry/no longer sticky to the touch.

Once dry, make your wallet or bag

Peel your sheet from the silicone tray

1.10. Once dry, peel the bioplastic from its tray by running your hands or a toothpick along the edge of the tray and gently taking one corner of the bioplastic to slowly peel up the entire sheet of bioplastic. Congratulations! You made an algae bioplastic sheet!

Heat water.

1.11. Fill your beaker with **~50 mL** of water and microwave until it reaches a rolling boil, about 30 seconds.

Create the bag

1.12. Position your sheet in front of you on a clean, dry surface. You can use the silicone tray or a do this directly on a table.

1.13. Use the hot water as a glue to seal the edges of your bag: take your paintbrush and dip it in the hot water, then paint the hot water onto the right and left edges of your sheet, from the top to the bottom. Note: the paintbrush should stay wet. Dip back into the hot water as many times as you need! Reheat the water if it gets too cool.

1.14. Fold the sheet in half, and press down on the right and left edges to seal them. If you have a heavy object like a large book, place it on top of your folded bioplastic to help the seal.

1.15. Leave your folded sheet to dry so that the wet edges fuse together. Depending on the amount of water used this could take a few minutes to half an hour. If sections of the edges don't glue together, repeat the steps above with the paintbrush to seal them.

Decorate your bag/wallet: get creative!

Note: Visit the Bioplastics Kit Analysis manual if you are interested in doing the analysis before continuing since you need specific bioplastics for analysis and shouldn't use them for decorating yet. Those bioplastics are are: the first day's bioplastic, the color test bioplastic with their differing numbers of blue dye, the dumbells of day 3 and the bio-composite of day 4. Once you have completed the analysis, you are free to use any bioplastic as you like!

1.16. Once you have a base for your wallet/bag, it is time to get creative! Use your string, sequins, star, and any of your previous bioplastics to personalize and create a closing mechanism for your bag or wallet. Not sure where to start? Here are some ideas:

- Create different pockets by sealing more sections together using the hot water technique to glue.
- Glue the sequins with the hot water paintbrush technique or sew them on by piercing a small hole in their center with a sewing needle. Then, use the sewing thread to secure them to the bag surface.
- Remember that you can melt any dried gelatin bioplastic back into its liquid form by adding boiling water to them, stirring and microwaving them until they liquidfy. Have a look back at the recipes for the plastics you want to melt down to figure out how much water to add. Use the same amount of water as in the original recipe. Note: Strings are much harder to meltdown because of the starch. Be careful if you try.
- Once your bioplastics are back into liquid form, you can dye them, extrude them or pour them into a new mold of your choice.
- To create more textures, you can place a textured material at the bottom of a mold before pouring your mixture on top. Don't forget to oil the textured material first! For example, you could take a aluminum foil, crumple it in the bottom of your mold and oil the surface. Then, you can pour your mixture on top and let it dry.
- To create polka dot patterns inside the bioplastic itself, you can drop sequins into freshly made clear/colored bioplastic once the bioplastic mixture has cooled a bit, but not yet solidified.
- You can even try to embed strings, twists, braids and weave into bioplastic sheets using the same technique: make a gelatin-glycerol mixture, pour it into an oiled mold, let it cool for 5 minutes and drop in your items! Leave to dry.
- To create ombre or tye-dye looks, let your bioplastic cool a few minutes before adding in different drops of dye and mixing gently until you get swirly patterns or gradient patterns. If you over mix, you can melt the plastic back up and try again with different (darker) colors.

Congratulations! You are officially "making" with bioplastics! Have fun! Don't forget to wash your tools and beaker with hot water when you are done.

CONGRATULATIONS

You have now joined the global community of bio-designers and material innovators! Happy with your material sample library and algae bioplastic product? There are many opportunities to share it online and in your community. You can also share your results with friends and our growing community: find us on Instagram, Twitter, TikTok and Facebook @aminobiolab

Don't forget, you can use these recipes again and again using ingredients you may already have at home, or that you can find at the grocery store, or at Amino Labs. For now, let's make sure you dispose of and store your remaining material correctly.

Storage, Disposal, Clean Up

After you see your results and end the experiment, it is time to clean up, dispose of and store away your materials. Disposing of experiment materials is an integral part of experimental protocols.

A. **Preserving your bioplastic sample library:** If you want to preserve them, you can place your samples with their identifying labels in a container. Or, you can attach them in a binder along with any experiment notes and material property analysis so that you can reference them in the future.

B. **Disposing of your bioplastics:** If you would like to dispose of all or some of your bioplastics samples, you can place these in the garbage, melt them down, or place them in the compost pile. Unfortunately, at this point, the bioplastics you've made cannot go into the traditional recycling stream as the recycling industry is not ready to deal with these.

C. **Disposing of your other kit material:** All tubes, bottles and plastic packaging can be disposed of in a recycling bin or garbage. Of course, they can also be washed and kept for reuse along the molds, silicone tray, paintbrush, syringes, etc. Wash with soap and water and kept for reuse around the house/classroom.

D. **Unused ingredients/items:** If you did not use all of your ingredients, you can keep them in a sealed bag for reuse later. If the ingredients are in powder form, make sure the lid of the container/tube is tight to prevent the powder from humidity. Do not eat or cook/bake with any of your science kit ingredients!

E. **Clean your workspace:** Use a spray cleaner, wipes, or hot, soapy water to clear up your work area and equipment. If you are using a bleach cleaner, always wear gloves and remember that the bleach cleaner can discolor clothes and fabrics.

Glossary

Acetic acid: You'll have noticed that acetic acid is a clear, colorless, organic liquid with a pungent odor similar to household vinegar. Indeed, acetic acid is the main component of vinegar apart from water and trace elements. Regular vinegar is around ~4% acetic acid.

Acetic acid has a variety of uses, including as raw material and solvent in the production of other chemical products, oil and gas production, and in food and pharmaceutical industries.

The acetic acid in the recipe helps the starch fully dissolve in the mixture because starch dissolves better in the presence of electrically charged ions which the acetic acid provides.

Large cellulose molecules, like starch, are long-chain polymers. In this experiment, two ingredients change the properties of the polymer plastic. By adding a small amount of acetic acid, you break up some polymer chains, making the plastic less brittle.

Algae/Agar Bioplastic: Agar is a yellowish powder that comes from algae. It acts as a binding material and a hardening component in bioplastics. In addition, it functions as a polymer in bioplastic mixtures.

Agar is a polymer created from the agarose and agaropectin found in the cell walls of red algae. It is often used as a vegetable replacement for gelatin. It is used as a thickening or gelling agent in cooking. However, agar is more flexible than gelatin. You can combine gelatin powder with agar powder to make a more flexible/hybrid material.

Agar bioplastics are a suitable replacement for disposable single-use packaging, including snack packaging and dry food packaging. And, if you've done any biotechnology or microbiology, you'll recognize agar from the substrate that microorganisms grow on. It's the same ingredient, used differently. A fun fact about agar: it affects touch capacitive screens! You might be able to make an agar bioplastic stylus for your tablet!

For agar to work successfully, it first needs to absorb water which it does by being brought to a boil (100°C) It becomes a gel at room temperature since it solidifies at 32–40 °C. Solid agar gels will start melting at 85 °C.

Agar does not hydrate (absorb water) well in acidic liquids, making gelling difficult. To use agar alongside an acidic bio-plastic ingredient, first hydrate the agar in water, allowing it to dissolve, before adding it to your acidic ingredient.

Agar shrinks a lot in size and thickness over time, and if left in a mold where it's connected to wooden edges, it will form cracks in the center. So make sure to cut the agar free from the edges of the mold after the first 24 hours of setting.

Agar bioplastic should decompose for two months in summer temperatures, taking approximately four months in cooler climates.

Biocomposites bioplastic: Biocomposites are made from a matrix of plastic and renewable fillers. Biocomposites can be made entirely of bio-based ingredients (biocomposite bioplastics) or with bio-fillers to replace some of the petroleum-based content of traditional plastics. In adding bio-fillers to traditional plastics, non-renewable ingredients can be drastically reduced.

Bio-fillers: are organic materials that are ideally waste by-products of other industrial processes like agriculture or paper processing and combined into plastic recipes to provide different properties or limit the number of other ingredients. They are sometimes referred to as biomass. Fillers can help the material keep its shape and reduce the natural shrinkage in most bioplastic.

You can use different bio-fillers in biocomposite to obtain different materials properties. Popular bio-fillers are dried peels, eggshells, dried plant leaves, shredded paper waste, seafood wastes like shells, starch, or wood dust. You can add natural colorant by choosing naturally colorful bio-fillers.

You can also try to mix in your bio-filler to a gelatin-glycerol bioplastic recipe instead of a pectin recipe to obtain different properties.

Cornstarch: see starch

Drying: when a material dries, it can shrink and deform as the water leaves the mixture. You can remedy this by placing heavy objects on your drying plastic to compress it into the desired shape. You should always dry your bioplastics indoors and away from direct sunlight and pets, insects, and younger siblings. While many ingredients in bioplastic can be found in foods, you should never eat any of your experiments.

Gelatin: is a colorless powder derived from collagen. It acts as a binding material for bioplastic mixture and a hardening component. It also provides transparency to the piece. Gelatin functions as a polymer in bioplastics.

Gelatin bioplastics are clear, strong, oxygen permeable, and hygroscopic. The properties of gelatin bioplastics are influenced by drying temperature. When Gelatin bioplastics are dry, especially when exposed to the sun, they often get quite brittle. Even when adding more glycerol to the recipe, it is still one of the stiffer bioplastics to work with. Gelatin bioplastics can also be heated up or whetted to shape them over forms/molds and melted back down for reuse.

Gelatin is primarily used as a gelling agent in food, pharmaceutical, and cosmetic industries. When it comes to gelatin bioplastics, they can be used for alternative packaging, to create a folder/pocket, to replace stiff plastic materials you might use in your daily life.

Gelatin is often an animal-based ingredient. Some might find it problematic to use resources that require killing an animal because of religious or animal welfare beliefs. Arguments are also made that as long as there’s a meat industry, it is better to use products from the entire animal, including skin and bones. Some might consider gelatin a product that comes from a waste stream, but this is considered controversial by others. To avoid using animal-based gelatin, use agar or vegetarian gelatin.

Glycerol: is a colorless viscous liquid that helps give shape, softens, and reduces shrinkage in bioplastics. Glycerol functions as a plasticizer that bonds with the polymer in your mixture. Sometimes also called glycerin, although glycerol is a pure form, while glycerin is a mixture that contains 95% glycerol.

Glycerol is a sugar alcohol derived from animal products, plants, or petroleum (as a by-product of bio-fuel). It can also be obtained from micro algae oils, and it can be recovered from used cooking oil.

Glycerol has a low melting point (18°C) and high boiling point (290°C), which makes it suitable as a plasticizer in bio-materials. As a plasticizer, glycerol "lubricates" the plastic. If you want the plastic more pliable, add more glycerol. If you want the plastic to be stiff, add less glycerol. However, higher glycerol concentration makes the bioplastic weaker (low tensile strength), more fragile, and easier to degrade under wet and dry soil. But the flexibility of your glycerol bioplastic will make it easier to peel them out of their drying containers.

In biomaterials, glycerin can be used as a plasticizer for more flexible bioplastics, as moisturizer or softener in tanning (e.g., bacterial cellulose), as an additive for bubbling textures/mixes, and as a solvent for pigment extraction. It has antimicrobial properties.

Material sample: in general, a sample is a limited quantity of something intended to show what the whole is like. A material sample of bioplastic is then a small piece of bioplastics that you can use to understand how a larger quantity of the material would behave and what it could be useful for.

Material samples library: is a resource for material research, exploration, and experimentation consisting of an expanding collection of material samples, analysis, and notes.

Mold: a mold is a hollowed-out shape filled with a liquid or pliable material like plastic, glass, metal, or ceramic. The liquid hardens or sets inside the mold, adopting its shape. Molds are sometimes called forms or negative forms and are often used to replicate a "master" item. The mold you select will affect both the shape of your material as well as the drying process. For example, thicker materials will take longer to dry.

Pectin Bioplastic: You may have heard of pectin before and might even have made some while making jams. Pectin is a colorless powder typically obtained from fruit. It is a polysaccharide starch found in the cell walls of fruits, vegetables, and plants - it is the second most abundant component of the cell wall of all land plants. Pectin functions as the polymer in bioplastic mixtures and is what makes jams jelly!

Pectin is often used as a gelling agent in desserts, but it also has wide applications in various fields due to its use as a gelling, emulsifying or stabilizing agent and its non-toxic, bio-compatible, and biodegradable nature.

In terms of biomaterials, pectin can have different applications, including food packaging, pharmaceutics, nutraceuticals, drug delivery, tissue engineering, and cosmetics.

The properties of pectin can be improved and modified with bio-fillers or in a mixture of other gelling ingredients (polymers). For example, adding a bio-filter like peels can help keep your bioplastic shape and reduce shrinkage.

Petri dish: A petri dish is a small plastic container. It is usually used to culture (grow) microorganisms in a controlled environment but can also be used as molds for material science experiments like you just did. Petri dishes come in several sizes, including 6 cm and 10 cm. They are sometimes called "plates."

Polymers: Plastics are chains of repeating molecules linked together called polymers. Whether natural or artificial, polymers are long molecules made by linking up smaller repeating molecules. Wool, cotton, and silk are natural polymer-based materials. Cellulose, the main component of wood and paper, is also a natural polymer, as are the starch molecules made by plants. Learn more about polymers here: https://www.sciencenewsforstudents.org/article/explainer-what-are-polymers

Shrinking: See drying.

Starch: is a white flour-like powder typically obtained from corn or potatoes. Starch itself is a polymer made by plants to store energy. It can be used as the polymer in your mixture or an additive to another polymer mix.

Troubleshooting

Here are some possible common issues:

My bioplastic does not solidify/gel.
When done correctly, the bioplastic will harden into a sheet. If it is not:

1. You likely did not heat (boil) the water before or after adding the ingredients

2. You might not have added all the powder from the tube, resulting in too much water vs. ingredient(s).

3. You may not have fully dissolved the powder, meaning it cannot turn into a gel and will look cloudy. You can practice by making Jell-O! Next time heat and swirl longer to ensure the powder is fully dissolved.

My bioplastic breaks:
Don't worry; some of the bioplastics recipes give thin, brittle plastic sheets. If your sheet or sample breaks, you can:

1. Melt it back down! Go back to the recipe and add the same amount of water to your mixing bowl! Add your broken plastic sheet and microwave until boiling. Stir and pour into an oiled mold. Note that bioplastics are hard to melt back down. Try the method below to repair them instead. Note that trying to melt the biocomposites might not work depending on the filler you used.

2. You can repair most bioplastics by dipping or coating the two edges of the break with boiled water and then pressing the edges together again. Leave to dry.

If anything else causes you issues, please contact us : help@amino.bio

More Information

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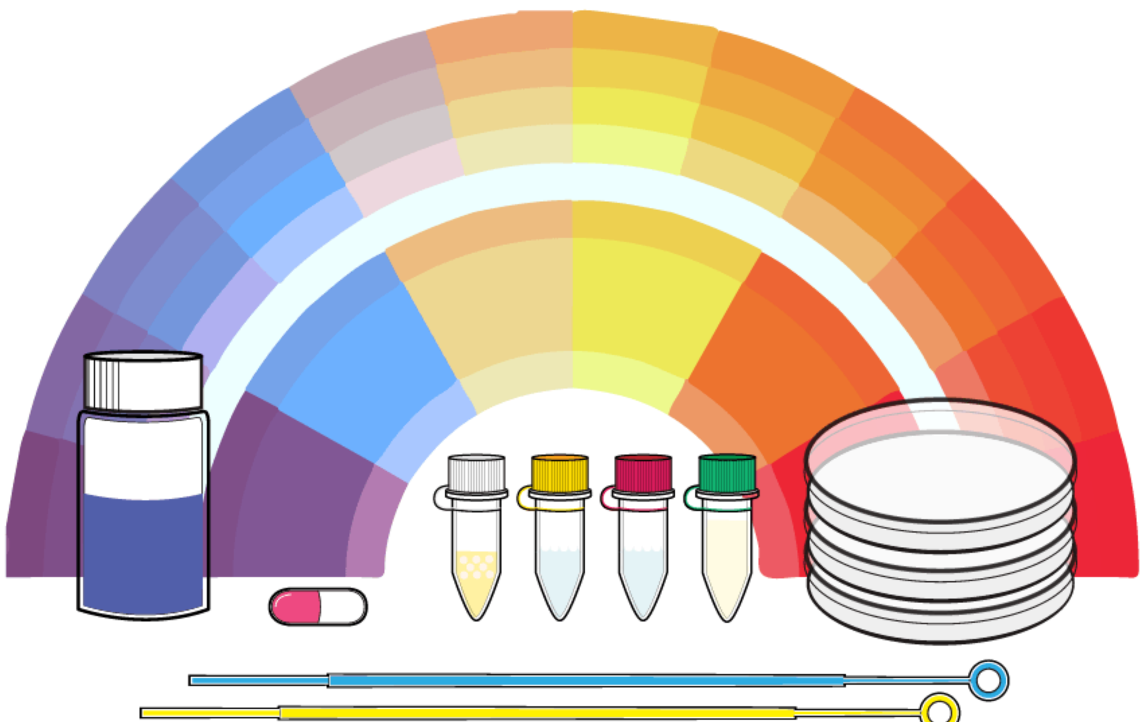

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