

Student Guide: How is Yogurt Made?

Yogurt contains harmless live bacteria (typically *Lactobacillus* and non-pathogen *Streptococcus* species). You can observe how these bacteria affect milk, and you can test what happens when you add antibiotics. Throughout this lab it will be very important to practice good sterile techniques to avoid adding bacteria from the environment or from your skin to the experiment.

Notes: Wear gloves, goggles and other appropriate safety gear. To kill any environmental bacteria on your gloves, sterilize gloves with alcohol by pouring some alcohol on and rubbing it around the gloves until it dries. Do not touch anything with your gloves that you do not need to touch for this experiment. If necessary, alcohol your gloves again during the experiment.

The test tubes are sterile, and you should keep them tightly capped except when you need to add something to the tubes. When you remove the test tube cap, try to hold it with your alcohol-sterilized glove instead of setting it down on a surface that might have bacteria.

The box of ultrahigh-temperature (UHT) pasteurized milk is sterile. Any bacteria in the milk were killed when it was heated to a high temperature. This means the milk can be stored at room temperature until it is opened and exposed to environmental bacteria. (Normal milk still has some bacteria, so it must be refrigerated and will still go bad within a week or so as those bacteria multiply in the milk.)

Procedure:

1. Label four test tubes with your or your group's name and one each of the following labels:

-Yogurt	-Antibiotic
+Yogurt	-Antibiotic
-Yogurt	+Antibiotic
+Yogurt	+Antibiotic
2. Carefully remove the sterile straw from its plastic on the side of the UHT milk box and insert it into the box. The straw should form a tight seal as it passes through the foil opening. Carefully use the straw to put 10 ml of milk into each of the four test tubes (measure the amount added on the tube). If the straw dribbles milk on the table, clean up the mess with paper towels and ask your teacher for help.
3. You will use one inoculation needle to transfer yogurt bacteria and another to transfer antibiotic. To avoid cross-contamination, do not get the needles confused and wipe them off thoroughly between uses. Before each use of an inoculation needle, pass its tip through a flame for a few seconds to

kill any bacteria; then let it cool for a few seconds without setting it down (it might pick up more environmental bacteria from the table or other surfaces).

4. Dip a flame-sterilized inoculation needle into a freshly opened cup of yogurt, get a tiny blob of yogurt on the tip and add that yogurt blob to one of the +Yogurt tubes. Clean and flame sterilize the inoculation needle again and use it to add a tiny yogurt blob to the other +Yogurt tube.
5. Use another flame-sterilized inoculation needle to add a small blob of antibiotic ointment to one of the +Antibiotic tubes. To get most of the antibiotic off the needle, you may need to swish the needle around in the milk in the tube and/or gently scrape the needle against the inside wall of the tube. Clean and flame the inoculation needle again and use it to add a small antibiotic blob to the other +Antibiotic tube.
6. Seal the tubes tightly, and shake and swirl them around to mix their contents thoroughly. Give the tubes to your teacher to incubate at 37° C overnight, or at room temperature for two to three days.
7. What do you predict will happen to the milk in each of the four tubes after they have been incubated? Record your thoughts below.

Tube 1: -Yogurt -Antibiotic

Tube 2: +Yogurt -Antibiotic

Tube 3: -Yogurt +Antibiotic

Tube 4: +Yogurt +Antibiotic

8. After the tubes have been incubated and you get them back from your teacher, keep the tubes sealed while closely observing the color and texture of the milk in each tube. Try slowly leaning the tubes to one side or the other. Try gently turning the tubes upside down. Record your observations for each tube:

Tube 1: -Yogurt -Antibiotic

Tube 2: +Yogurt -Antibiotic

Tube 3: -Yogurt +Antibiotic

Tube 4: +Yogurt +Antibiotic

Answer the following questions to analyze your results:

1. Did the results match your predictions for each test tube? Explain.

