Pasteur's Experiment — Student Guide

INTRODUCTION / BACKGROUND INFORMATION

Historical Background

The French chemist Louis Pasteur (1822-1895) is considered by many to have established the field of microbiology. As a professor of chemistry and, later, Dean of the Lille Faculty of Science, Pasteur devoted a great amount of time and energy to the study of fermentation. Through careful, repeated experimentation, Pasteur showed that yeast is a particular type of microorganism, or germ, that converts sugar into alcohol rather than, according to the conventional scientific wisdom of his day, a general chemical catalyst. He also disproved the idea that "spontaneous generation" of germs occurs, showing by experimentation that all microorganisms arise from other microorganisms. Pasteur demonstrated that the spoilage of perishable substances could be delayed through a process of heating substances in order to destroy harmful microorganisms. This process is now termed pasteurization.

Furthermore, Pasteur suggested that just as a particular type of microorganism caused each type of fermentation, many diseases might also be caused by specific microorganisms. This laid the foundation of 'germ theory', a major, transformative step in the development of modern medicine.

Pasteur also made profound contributions to the field of immunology, discovering the bacteria that cause anthrax, and campaigning for hygiene, sanitation, and sterilization to halt the spread of disease-causing bacteria. Pasteur later showed that rabies was caused by an organism that was too small to be seen with a microscope, thus opening the door to the study of viruses. In 1885, he succeeded in developing a vaccine for rabies, and, in 1888, Pasteur became the first director of the newly established Pasteur Institute, a post he held until his death.

Theory

Biogenesis — the concept that every living thing comes from other living things— seems commonsensical or obvious to us today. However, prior to the seventeenth century, it was a generally held belief that living things could arise from non-living things, in a process called "spontaneous generation" or "abiogenesis". Thus, when people observed the appearance of maggots on a piece of rotting meat, they believed that the meat had produced, or "spontaneously generated", the maggots. Similarly, some scientists suspected that spontaneous generation was occurring when they observed that bacteria had grown in a flask of broth.

Around 1765, Lazzaro Spallanzani showed that no microorganisms would grow in broth after he boiled the broth in a flask, and then sealed the flask. Critics of Spallanzani's experiment countered that air was needed for spontaneous generation to occur. Other experimenters heated or filtered the air that entered the flasks of boiled broth. The critics then contended that these treatments changed the air somehow, destroying the "life force" in it.

In order to disprove the theory of abiogenesis, the French chemist Louis Pasteur undertook a special experiment. He believed that microorganisms, which were transported in the air on dust particles, were finding their way into the broth through the open necks of the flasks holding the broth. Pasteur designed a flask with a special S-shaped neck, like the one shown in **Figure 1**, at

allowed untreated air to enter the flask while trapping dust particles from the air in the curved S-shaped neck of the flask.

Pasteur's experiment used two flasks, each containing broth that had been sterilized. One flask was left open, and one flask had its top heated and formed into an S-shaped tube. Pasteur hypothesized that if living things were spontaneously generated, the broth in both flasks would turn cloudy due to microbial growth; if living things were not spontaneously generated, then only the broth in the open flask would turn cloudy, as a result of microbes traveling on air and dust particles and entering the flask.

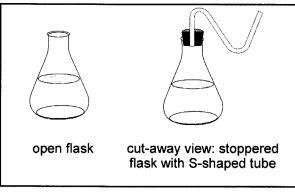
right. In contrast to a conventional flask with a straight neck, Pasteur's innovation



Figure 1

LAB OBJECTIVE

In the following lab activity, you will analyze experimental data and use your knowledge of the scientific method to determine whether or not Pasteur's experiment was valid. Your teacher will direct you to assemble a two flask set-up (**Figure 2**) which is similar to Pasteur's original experiment, or a three flask set-up (**Figure 3**) that illustrates Spallanzani's contribution as well.



open stoppered flask (Spallanzani's method) cut-away view: stoppered flask with S-shaped tube

Figure 2

Figure 3

MATERIALS NEEDED PER GROUP

Two flask set-up 200 mL of beef broth

200 mL of beef broth
Hot plate
Safety goggles
(2) 125-mL Erlenmeyer flasks
Wax pencil
(1) one-hole rubber stopper with S-tube
Lab instructions (page S2)
Student Worksheet (pages S3-S4)

Three flask set-up

300 mL of beef broth
Hot plate
Safety goggles
(3) 125-mL Erlenmeyer flasks
Wax pencil
(1) one-hole rubber stopper with S-tube
(1) solid #5 rubber stopper
Lab instructions (page S2)

Student Worksheet (pages S3-S4)

SAFETY

- Wear goggles at all times when setting up this experiment.
- Use heatproof gloves or tongs to handle hot objects.
- Use hot plates under the supervision of a teacher.
- Do not touch the hot plate or flasks while hot.
- Do not heat cracked or damaged glassware.

PROCEDURE

- Obtain two (or three, if directed by your teacher) 125-mL Erlenmeyer flasks. Inspect them for cracks. Use the
 wax pencil to write your initials (or your group's name) on each flask. Pour 100 mL of beef broth into each
 flask. Insert the S-tube stopper into one flask. Leave the other flask(s) open.
- 2. Put on your safety goggles. Place the flasks on the hot plate. Set the hot plate to the highest setting. Be careful not to touch the top of the hot plate.
- 3. Once the broth has come to a boil, let it continue to boil for 30 minutes. Turn off the hot plate. Note: The hot plate will be extremely hot for several minutes after it has been turned off; take care not to touch the hot plate.
- 4. **Be careful not to touch the hot flasks with your bare hands.** Use tongs or heatproof gloves to remove each flask from the hot plate. If you prepared and boiled two open flasks, seal one of them immediately with a solid rubber stopper. Leave the other flask open.
- 5. Observe the appearance of the liquid in each flask. Record your observations in the proper spaces of the data table on page S3.
- 6. Place the flasks in a location designated by your instructor, preferably a location in which the flasks will not be disturbed.
- 7. Observe the appearance of the liquid in each flask after one week, and, again, after two weeks. Each time, record your observations in the proper spaces of the data table.
 - Note: If the solution becomes cloudy, you are probably seeing evidence of bacterial growth. If the solution contains thin threadlike structures, you are seeing evidence of mold growth.
- 8. At your teacher's direction, answer the analysis questions on the worksheet.

Pasteur's Experiment — Student Worksheet

Name:	
Date:	
Class/Period: _	

DATA TABLE

In the proper spaces below, record what you observed in each flask on the day of the experiment, after one week, and after two weeks.

Flask	Description on Day 1	Description after one week	Description after two weeks
Open			
S-shaped neck			
Solid stopper			

ANALYSIS QUESTIONS

- 1. State the problem that the scientists of Pasteur's time were trying to address.
- 2. What was Pasteur's hypothesis?
- 3. What was the purpose of boiling the broth?
- 4. How did Pasteur's S-tube flask help to end the controversy over spontaneous generation?

5.	What are the two parts of a controlled experiment?
6.	Do you consider this recreation of Pasteur's experiment to be a controlled experiment?
7.	Was Pasteur's hypothesis correct? Explain your answer. Use the observations you recorded in your data table to support your answer.
8.	What would happen if the flask with the stopper and S-tube were disturbed, causing some of the broth to enter into the curve of the S-tube and return back into the flask? Explain your answer.
9.	Besides answering the question of spontaneous generation, what practical application could result from the information gathered from this experiment? In other words, how can we use the results of this experiment in our lives today?