



How Well Do Disinfectants Work?

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Experimental Procedure

Working with Biological Agents

For health and safety reasons, science fairs regulate what kinds of biological materials can be used in science fair projects. You should check with your science fair's Scientific Review Committee before starting this experiment to make sure your science fair project complies with all local rules. Many science fairs follow Intel® International Science and Engineering Fair (ISEF) regulations. For more information, visit these Science Buddies pages: [Project Involving Potentially Hazardous Biological Agents](http://www.sciencebuddies.org/science-fair-projects/competitions/biological-agents-regulations) (<http://www.sciencebuddies.org/science-fair-projects/competitions/biological-agents-regulations>) and [Scientific Review Committee](http://www.sciencebuddies.org/science-fair-projects/competitions/scientific-review-committee-src) (<http://www.sciencebuddies.org/science-fair-projects/competitions/scientific-review-committee-src>). You can also visit the webpage [ISEF Rules & Guidelines](https://www.societyforscience.org/isef/international-rules/) (<https://www.societyforscience.org/isef/international-rules/>) directly.

1. Using the permanent marker, divide the cutting board into six sections, numbering each section #1-6 like this:



2. Put on your gloves and wipe the piece of lunch meat all over the surface of the cutting board. Wipe evenly over the entire surface in circular motions. Leave out overnight, gross!
3. The next day you will cleanse each section of the cutting board with a different disinfectant and then culture the bacteria from each section on a nutrient agar plate.
4. Wear a new pair of gloves. Prepare your disinfectant solutions by numbering six small cups #1-6 using your permanent marker. Each numbered cup will match one section of your cutting board fomite.
5. Fill each cup with a different disinfectant solution and write it in a data table. You should fill the first cup with water as a negative control.
6. In your lab notebook, make a data table like Table 1, to record your results in.

Number	Type of Disinfectant	Number of Colonies				Other Observations
		A	B	C	Average	
1	Water (control)					
2						
3						
4						
5						
6						

Table 1. In your lab notebook, make a data table like this one to record your results in.

- Using the forceps or tweezers, grab a cotton ball from a NEW, unopened bag of cotton balls. Dip it into one of the solutions, and rub it on the surface of the cutting board in the matching numbered section. Be careful not to let the solution run into another section!
- After each application, throw the cotton ball into the trash and dip your forceps into an extra cup filled with full 70% ethanol or isopropyl rubbing alcohol.
- Repeat steps 7 and 8 until you have applied a different disinfectant to each square of the cutting board. Throw away your gloves. Allow the board to dry completely.
- When all of the sections of the cutting board are dry, you are ready to culture bacteria from each disinfectant treatment. Prepare three nutrient agar plates for each of the cutting board sections by numbering the bottoms of the plates "1a", "1b", "1c", "2a", "2b", "2c", etc. Arrange the plates on cookie sheets or trays lined with clean paper towels. DO NOT open the lids yet or you will contaminate your cultures! Put on a new pair of gloves.
- Use the swabs to transfer a sample from the cutting board to the agar plate. Holding the wooden end of the swab, swipe the cotton end around one section of the cutting board using a circular motion. DO NOT allow the tip of the swab to contact anything else!
- With your free hand, open the lid of the matching nutrient agar plate and swipe the cotton-tipped applicator gently across the agar surface using a zig-zag motion. Immediately replace the lid of the agar plate and secure with a few pieces of clear tape. DO NOT set the lid down while you are streaking the agar because this can contaminate the lid and change your results!
- Repeat steps 11 and 12 until you have swiped each section of the cutting board onto 3 separate nutrient agar plates. Throw away your gloves. *Tip:* Having 3 samples per cutting board section helps you tell how accurate and repeatable your results are.
- Leave the nutrient agar plates on the cookie sheets or trays in a warm place for 2-4 days, until bacterial colonies are visible.
- Count the number of colonies on each nutrient agar plate and write your results in the data table in your lab notebook, along with any other observations you have. Calculate the average, across plates "a", "b" and "c" for each cutting board section.
- Draw a picture of each plate, noting the size and color of the colonies. If you have a digital camera, you can also take a picture of each plate to put on your [Science Fair Project Display Board](http://www.sciencebuddies.org/science-fair-projects/science-fair/science-fair-project-display-boards) (<http://www.sciencebuddies.org/science-fair-projects/science-fair/science-fair-project-display-boards>).
- Graph your results and compare the activities of the different disinfectants. Which sections of the cutting board had the most colonies? Which had the least? Which disinfectants worked the best? The worst? Why do you think you got the results that you did?

Bacterial Safety

Bacteria are all around us in our daily lives and the vast majority of them are not harmful. However, for maximum safety, all bacterial cultures should always be treated as potential hazards. This means that proper handling, cleanup, and disposal are necessary. Below are a few important safety reminders.

- Keep your nose and mouth away from tubes, pipettes, or other tools that come in contact with bacterial cultures, in order to avoid ingesting or inhaling any bacteria.
- Make sure to wash your hands thoroughly after handling bacteria.
- **Proper Disposal of Bacterial Cultures**
 - Bacterial cultures, plates, and disposables that are used to manipulate the bacteria should be soaked in a 10% bleach solution (1 part bleach to 9 parts water) for 1–2 hours.
 - Use caution when handling the bleach, as it can ruin your clothes if spilled, and any disinfectant can be harmful if splashed in your eyes.
 - After bleach treatment is completed, these items can be placed in your normal household garbage.
- **Cleaning Your Work Area**
 - At the end of your experiment, use a disinfectant, such as 70% ethanol, a 10% bleach solution, or a commercial antibacterial kitchen/bath cleaning solution, to thoroughly clean any surfaces you have used.
 - Be aware of the possible hazards of disinfectants and use them carefully.

Explore More!

Interested in the science behind viral outbreaks? Check out [Coronavirus](http://www.sciencebuddies.org/science-fair-projects/Zika) (<http://www.sciencebuddies.org/science-fair-projects/Zika>).