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TITLE: Effectiveness of Sunscreens Against Ultra Violet Light Experiment

INTRODUCTION: With enough exposure Ultra Violet (UV) light is lethal to bacteria. Ultra violet light may also cause damage to human skin cells. It is thought this damage may destroy the ability of the skin cells to control cell division, thus leading to a type of skin cancer call melanoma. For this reason people are encouraged to protect their skin from the potentially harmful effects of UV light by using sunscreen creams. Sunscreen creams have different Sun Protective Factors (SPF). You may purchase sun block with SPF values of 15, 30, or 45. The higher SPF values in theory give greater protection from UV light. In today's laboratory we will design experiments to compare two different kinds of sunscreen (SPF 15 vs. SPF 30; or current sunscreen vs. expired, etc.) to evaluate their effectiveness to protect bacteria from the effects of UV light.

Because it is unethical to use humans to conduct scientific experiments, we will use *Escherichia coli*, a type of bacteria, as a substitute for human skin in today's experiment. This is called a model. We will test the efficacy of sunscreen to block UV light on bacteria, and then we may use that information to consider the efficacy of the same sunscreen on human skin cells. In this experiment each group will formulate its own question and hypothesis regarding the effectiveness of a variety of sunscreens to protect bacteria from cell death.

State the question that you are asking in this experiment (*do sunscreens with higher SPF values provide greater protection; do different brands of sunscreen provide greater protection; is duration of exposure for different brands a factor, etc*). This statement will become the purpose of your experiment and written in the Introduction section of your lab report.

HYPOTHESIS: State your hypothesis.

MATERIALS AND METHODS: This is a two- week experiment. During the first week you will formulate your question and hypothesis, inoculate Petri dishes with bacteria, and then cover the dishes with Saran wrap that has a

covering of sunscreen. You will then expose your samples to UV light. During the second week you will perform a colony count of those *E. coli* that survived exposure to the UV light. This is necessary to calculate the percent survival of the bacteria. The way you design and conduct your experiment may be slightly different from your classmates, however, the basic procedure will be the same for all. Be clear about the question you are asking, and the design of the experiment before you start.

Escherichia coli is not a harmful bacterium, however, you should wash your hands before and after handling *E. coli*, and you should use care not to spill any of the sample while conducting your experiments. Contact your instructor should you spill anything.

Three important concepts that you must understand for this experiment:

Control: A scientific control is that component of the experiment that permits comparison of results. Covering half of the Petri dish with aluminum foil will block the UV light and not allow it to reach the growing cells. This will permit maximum cell growth and represents your control.

Experimental: Covering the other half of the Petri dish with sunscreen may only reduce the passage of UV light reaching the cells causing some cell death and reduced growth. This is your experimental group. In this experiment you will have two experimental groups where you compare one type of sunscreen with another. Because in all cases half of each dish will be covered with foil, we are allowing each Petri dish to serve as its own control.

Sample Size: One Petri dish represents a sample size of only one ($N=1$). This gives you no reliable basis to judge your experiment. You need to use at least three Petri dishes ($N=3$) for each test group to obtain reliable results. Your experiment will require a total of six Petri dishes: three for one sunscreen and three for the other. In each case each dish will serve as its own control.

DAY 1

Experimental setup:

-Each group will have six fresh Petri dishes at their bench. Three will be used to evaluate the growth effects of one sunscreen and three for the other sunscreen. These plates contain the growth media tryptic soy agar (TSA). This agar contains all the nutrients necessary for bacterial growth.

- Do not remove the lid of the Petri dishes. Label the bottom of your six Petri dishes with your group name and the date, and then divide the plate in half with a line. Label one side of the line 'C' for control, the other side with 'E' for experiment. Label three of the dishes with the first type of sunscreen you are testing and then the remaining three dishes with the other sunscreen you are testing. These will represent your two experimental groups. Remember, half of each plate will be covered with foil and serve as its own control.

- Glove up. While holding the lid to avoid contamination, pipette 0.1ml of bacteria from the sample into each Petri dish. Spread the sample evenly with a hockey stick throughout the entire dish. This is called a bacterial lawn. Quickly put the lid back in place.

- Cut pieces of Saran wrap large enough to cover the top of each Petri dish. Remove the lid and cover the top of the Petri dish with the Saran wrap. Snuggly fold the edges beneath the bottom of the petri dish. Secure the edges of the Saran wrap with masking tape. Keep the Petri dish lids for later use.

- Weigh out (*unless of course you are testing the efficacy of different amounts of the same sunscreen*) 0.5 gram of sunscreen for each petri dish. Apply the sunscreen evenly to the Saran wrap using the same technique for each.

-Cut and fold in place pieces of aluminum foil large enough to cover the control half of each Petri dish.

Method:

-Locate the UV lamp you will be using for the experiment. Note the wavelength in nanometers, and note the time of exposure necessary to kill the bacteria.

-Expose your Petri dishes to the UV light using the same technique (duration, position, and distance from light source) for each.

-Once your samples have been exposed to the UV light, remove the aluminum foil and the Saran wrap. Put the Petri dish lids back on.

-Stack your plates into groups of three and tape them together with the masking tape. Make sure your group name is clearly written on the masking tape. Place them, inverted (*explain the problem with condensation*), into the carriers provided.

DAY 2

RESULTS: After 24 hours of bacterial growth at (37° C/ 98.6° F), the Petri dishes will be removed from the incubator by the TMCC staff and stored in the refrigerator at 4°C. This will arrest the growth until you can complete your experiment. You will be able to answer your experimental question as to which sunscreen offers better protection by counting the number of bacterial colonies (Colony Count) present on the control and experimental sides of each Petri dish. The number of colonies represents the bacteria that survived the UV light and reflects the amount of protection the sunscreen offered.

Accurately do a colony count on the control sides of the three Petri dishes for your first sunscreen. These numbers represent your raw data. Add these numbers together and divide by three. This number represents your averaged data for that control. Repeat the control colony counts on the three plates containing the second sunscreen tested. Add these numbers together and divide by three to obtain an average. Now perform colony counts on the experimental sides of the three plates from each of your two experimental groups. Record the raw data and average for each group.

Calculate the % survival for each experimental group:

Percent survival= $\frac{\text{Average colony count with sunscreen (experimental)}}{\text{Average colony count with aluminum foil (control)}}$

- 1) Prepare a table showing your raw data and averaged data for both the experimental groups and their respective controls.
- 2) Make one bar graph comparing the percent survival of your two experimental groups.

Write a few sentences summarizing your results from the table and graph. Do not interpret your results here.

DISCUSSION: This is where you will explain your results from the data and the graphs you have made. Was your question answered? Was your hypothesis supported or not? Compare results in control and experimental groups. Do you have confidence in your results? State your conclusions. Can we use bacteria as a model to explain the effects of UV light on human skin cells? Why or why not? Suggest further experiments that may help clarify your results.