Shielding Yeast From UV Radiation

In this experiment, students will explore how shielding countermeasures can help to lessen the harmful effects of UV radiation. Different sun protection factors, cloth, paper, foil, or sunglasses may be used.

Note: this activity is also suggested in Module 2. It may be adapted to include dietary countermeasures. This can be accomplished by adding vitamin C to the media 1 or 2 days prior to UV exposure. It is suggested to vary the amount of vitamin C concentration used in each yeast culture. Because vitamin C may have toxic effects in high concentrations, test a range of concentrations. For example, try different concentrations from 1 mg vitamin C per 10 ml media up to 1 g vitamin C per 10 ml media. Ensure that you have a control group that has not been exposed to vitamin C for comparison purposes.

Objectives:

- Discuss countermeasures for UV radiation.
- Describe phenotypic changes in yeast as a result of radiation damage.
- Explain why model organisms are important in countermeasures research

Research Question:

Within the context of available hat is the most effective method of preventing UV damage in yeast?

Discussion Questions:

- Why use yeast to study the effects of UV radiation?
- What are the effects of different types of sunscreen on yeast?
- How can your health be affected by exposure to ultraviolet radiation?
- Do you see any differences between areas of the Petri dish? If so, describe them.
- Did some SPF's of sunscreen protect the yeast cells better than others? Why?
- Does yeast grow less in some areas? Does it grow more than in others? Why?
- Does UV pass through plastic wrap? Plastic Petri dish covers?
- Why is it important to not expose an open yeast extract dextrose agar plate for very long? What is aseptic technique?
- What can you conclude from the results of your experiment?
- Describe another experiment you could carry out to obtain more information about the effects of UV radiation on cells.
- Why would vitamin C be used as a countermeasure?

How Do Sunscreens Work?

Sunscreens act like a very thin shield by stopping the UV radiation before it can enter the skin and cause damage. Some sunscreens contain organic molecules (such as

oxybenzone, homosalate, and PABA) that absorb UVB and/or UVA radiation. Others use inorganic pigments (such as titanium dioxide and zinc oxide or both) that absorb, scatter, and reflect both UVA and UVB light. Sunscreens are labeled with a Sun Protection Factor (SPF) rating that could also be thought of as a sunburn protection factor. For example, suppose that your skin begins to redden after 10 minutes in the sun. If you protected it with an SPF 15 sunscreen, it would take 15 times as long, or 2.5 hours, to get a comparable burn. Remember, SPF relates only to UVB protection; there is no standard measurement or rating for UVA protection in the United States.

Why Does NASA Study Yeast in Space?

Like the fruit fly, ordinary baker's yeast (*Saccharomyces cerevisiae*) also contains genes for DNA repair that are very similar to human genes with the same function. Therefore we can use yeast as a model system to explore the effects of radiation on cells. Like human cells, most yeast cells effectively repair DNA damage caused by UV radiation. However, some yeast strains have mutations that prevent them from performing certain types of DNA repair. Because they cannot repair all the damage to DNA, these cells usually die after exposure to UV radiation. In addition to sensitivity to UV radiation, yeast is also sensitive to space radiation. In a biological assessment of space radiation in low-Earth orbit, yeast inside special experiment hardware has been shown to have a decreased rate of survival following exposure to beta particles (electrons) and low-energy protons.[1] Other findings suggest there are highly coordinated gene expression responses to gamma radiation.[2] This knowledge is especially important when designing countermeasures for astronauts during long-term lunar surface operations or microgravity spacewalks.

Materials:

1. Yeast-Extract Dextrose media plates (from kit, can also be made)
2. UV-sensitive yeast suspension in liquid media and wild type yeast suspension in liquid media (this needs to be prepared from a stock sample that is purchased from a vendor). Ensure there is enough for the number of plates that will be plated (1ml of cells per plate is recommended).
3. A source of UV radiation such as direct sunlight. For this or future experiments, the radiation source could also be changed. Depending upon the size and design of the experiment, you may want to include black lights, halogen, or fluorescent light bulbs to determine if they also produce damaging radiation)
4. Several kinds of sunscreen (each with different SPF), black paper, cloth, metal foil, or other types of materials that can be used to experiment with UV shielding.
5. Sterile water, sterile pipets, and sterile toothpicks
6. Plastic wrap (to cover plates)

---

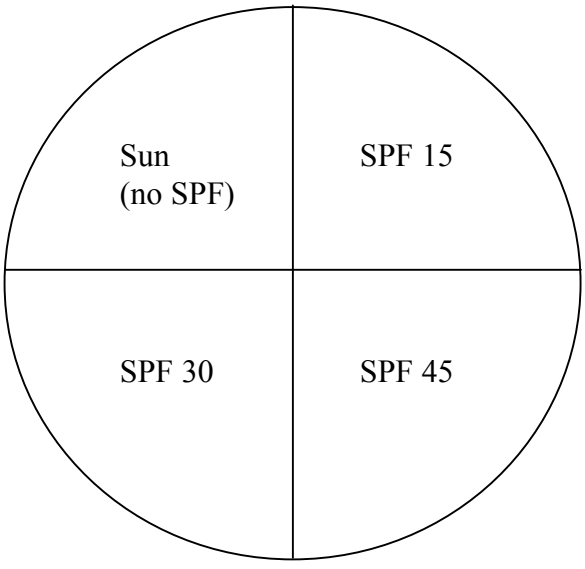
[1] http://www.spaceflight.esa.int/users/index.cfm?act=default.page&level=11&page=2120

[2] http://www.sanger.ac.uk/PostGenomics/S_pombe/docs/851.pdf

7. UV-sensitive yeast suspension in liquid media with varying concentrations of vitamin C and wild type yeast suspension in liquid media with varying concentrations of vitamin C (optional).

Directions:

1. Ensure that your hands and the work area are clean. Use soap and water and wipe your hands and your work area with alcohol and a paper towel. Good aseptic technique will ensure that the plates do not get contaminated with other organisms.
2. In this step you will plate the yeast suspension. You may want to do this for each group or allow the students to perform the task. Swirl the container of UV-sensitive yeast. Using aseptic technique and a sterile pipet, add 1 ml of the yeast cell suspension uniformly on top of the agar in the Petri dish for every plate that will be used in the experiment. Close the lid. Gently tilt and rotate the dish to spread the liquid. If there are places the liquid does not cover, reopen the dish and use the rounded end of a sterile toothpick to move the liquid over them. Sterile glass beads could also be used to spread the cells across the plate by moving the plate side to side when the beads and sample are on the agar. Let the liquid soak into the agar (remove beads if used by dumping them out). Place the Petri dish in a dark place for 10-20 minutes until the liquid soaks into the media. Note: If only few colonies are desired on the plate, do several serial dilutions of the cell suspension to reduce the initial concentration of cells plated.
3. Label the dish (see the diagram at the end of this activity) by drawing lines on the top and bottom of the dish to divide it into 4 parts (you could divide it into more parts, depending upon the number of countermeasures you are investigating). Label one area "sun" as a control, and use the other three areas to test sunscreens or other items like cloth, foil, paper, or plastic. Ensure that one area on all plates do not get UV exposure (cover it with black paper during UV exposure) or make certain that at least one entire plate per group is designated as the control, which does not get UV exposure. Label each area on both the top and the bottom of the dish and tape the 2 halves of the Petri dish together along the side so that the lid does not rotate. For one group (or the entire class), have the students remove the lid and replace it with plastic wrap (tape it on tightly). This will test any possible shielding effects of the cover.



4. Spread sunscreen on the lid of the Petri dish (or on the plastic wrap) in the places you marked; use an equal amount in each section and spread the sunscreen evenly. You can also use plastic, foil, etc. instead of sunscreen. If you labeled an area "no sun," tape a square of dark paper over it. Make sure you know exactly where each

sun screening material is used.

5. Expose the Petri dish to the sun or to a UV light. Vary the appropriate exposure times for the students from 20 minutes (in midday summer sun) to as much as 4 hours (in midmorning winter sun) per dish. If you are exposing the Petri dish to the sun, make sure that the surface of the agar is aimed directly at the sun (perpendicular to the incoming radiation). If students are careful, the lids could be removed and replaced with some clear plastic wrap during the exposure (to reduce any possible shielding effects of the lid). Consider allowing one group to remove the lid for a direct exposure.
6. After the exposure, wipe the sunscreen off the lid of the Petri dish. This will reduce the mess. Remove any other materials that were tested. If the students used the plastic wrap, just remove the wrap and replace it with the original lid. Place the Petri dish upside down in an incubator or in a dark place and let it grow for 1-2 days in an incubator at 30˚C or 3-4 days at room temperature.
7. If desired, repeat these steps with a wild type strain as a control for comparison.
8. Compare the amount of yeast that has grown in different areas of the Petri dish and draw conclusions.
9. Note: If a wild or UV sensitive yeast suspension with varying concentrations of vitamin C is used, it is suggested to refrain from using additional shielding countermeasures on at least one plate of each yeast strain. This way, if an effect is seen, you will know it is due to the vitamin C in the media, and not due to sunscreen, foil, or other countermeasures.

Suggested Resources:

1) **Carolina Biological Supply Company** http://www.carolina.com/

Catalog #17-3603, kit contains: UV-Sensitive and wild-type yeast strains, yeast medium; Petri dishes; and sterile, distilled water, sterile dilution tubes, glass beads, pipets, and instructions. $44.00 each.

Catalog #17-3603B is a 6-Station Student Kit for $79.50 each.

2) **Genetics Science Learning Center**, http://gslc.genetics.utah.edu

References:

Another more advanced experiment example can be found at:
http://www.phys.ksu.edu/gene/d3.html