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**Reflection 2**

Due Tuesday, October 4, 2016

By end of lab on Tuesday, September 27, you will have a novel phage that you are working to purify and characterize. The phage will either be one that you have isolated from an environmental sample or one that you have received from the Teaching Assistants that needs to be purified and characterized.

Use the information about your phage and the laboratory manual as a reference to address the questions below. You will also need to review the Ted Talk by Uri Alon: “Why truly innovative science demands a leap into the unknown” (found here: <https://www.ted.com/talks/uri_alon_why_truly_innovative_science_demands_a_leap_into_the_unknown?language=en> ).

1. How big is a typical phage? How does this size compare to the filter we use during the Capture protocol?
   1. A phage has a diameter of about 6.5 nanometers (6.5\*10^-9 m). The filter has a pore size of 0.22 micrometers (2.2\*10^-7 m), which is about 34 times larger than the size of an average phage.
2. What is the average diameter of the plaques from your phage?
   1. Currently, there are two types of plaques found on the plates, some smaller, about 1 mm across, and others larger, about ¼ inch across.
3. How many phage might exist in a single plaque?
   1. Assuming the entire area of a plaque is covered in phage, up to 150,000 to 200,000 phage might exist in a single plaque.
4. If an isolated plaque is formed by an original single phage particle, how many infection events must have happened to form that plaque. State all your assumptions. Use the phagesdb.org to look up terms and information such as a typical burst size.
   1. A typical burst size can be anywhere from 50-200 bacteriophage/burst (assuming the midpoint, 75 phage/burst).
   2. Assuming 175000 phage live in a typical plaque.
   3. Assuming the infection rate is perfect (i.e. every phage infects a unique bacterial cell)
   4. 175000 = 75^(# of events)
   5. After 3 rounds of infection, there are 421,875 phage in a plaque
   6. 2.79632 rounds of infection creates 175,000 phage in a plaque
5. What did you accomplish in the previous week? Your answer should include a summary of your findings.
   1. Reflect on the meaning and/or implications of your findings for the work that you have done in lab thus far. Refer to the data or evidence that you have to support any claims that you make.
   2. This past week, my partner and I made webbed plates, based on the titer we calculated last week. The titer was 6.4\*10^6, which was too small to flood immediately. On Thursday, the webbed plates were inspected, and two different sized plaques were found on the plates, meaning the phage sample we have been working with was not purified completely. Streaked plates were created to isolate the phage from each plaque and see if the plaques are made by the same type of phage. I
6. What issues or challenges did you face?
   1. One challenge we faced was calculating the titer of the lysate, as many of the plaques ran together, and it was difficult to decide where unique plaques began and ended. Once the titer was calculated and the webbed plates were created, another challenge faced was the fact that two different types of plaques were found on the plates, which could mean that the phage sample we have been working with has not been purified completely.
7. Where are you at compared to the overall goal for the project? Are you ahead or behind schedule?
   1. I believe we are on track to finish the project on schedule, allowing some buffer time for future issues or challenges with the project, as happened this past week. We have finished a lot of the project’s more tedious steps, such as finding phage and isolating them on plates, but we still have some of the more complicated steps to go, such as obtaining DNA from the phage sample.
8. As discussed in the Ted Talk by Dr. Uri Alon failure is a part of the process of science and an opportunity to explore the boundaries between what is known and unknown.
   1. How many times have you tried to capture a phage by direct plating?
      1. We found phage in our sixth sample that we plated directly.
   2. Have you successfully captured a phage from your environmental samples or did you collaborate with another group?
      1. We successfully captured a phage from the environment outside the DLRC, after the four original environmental samples, plus one outside the DLRC, failed to capture phage.
   3. Reflect on your experience with failure as a part of authentic research and consider the following as you reflect:
      1. What have you learned from your failure?
         1. From my failure, I have learned that it is okay to ask for help and/or start over completely in order to do the best job possible with my responsibilities and projects.
      2. Do you think it is important to fail sometimes? Why do you think you failed? How did you change in order to be successful?
         1. I think it is important to fail sometimes because that is the best way to learn. If everything goes perfectly all the time, one will never have to problem-solve, or work success.
         2. I think I failed because I was working with a new technique and new project. I was not as careful with the aseptic technique at times as I should have been, as I did not realize how small mistakes, such as touching the outside of a bottle with a pipette tip, could affect the entire outcome of my project.
         3. To change, I became much more careful with how I conducted myself when using aseptic technique. I became almost paranoid about contamination, and used a new tip or started over whenever I believed I had made a mistake with the technique.
      3. Overall, what do you think could be the best strategy to overcome failure and be successful?
         1. The best strategy to overcome failure and be successful is to learn from every failure. Figure out what caused the failure, and how to avoid making the same mistake in the future, as I did with becoming more aware of possible causes of contamination while using aseptic technique.
9. Read the following article posted in Blackboard: “All the World’s a Phage,” a Science News article that discusses phages and their impact on the world.
   1. Based upon this article, what are the potential impacts of your semester project?
      1. My semester project will help researchers discover more and more species of bacteriophage that may aid in finding new ways to fight bacterial diseases, as these viruses kill bacteria so well. The project may help find bacteria with similar molecules to humans, which may find the cause or the cure to diseases and disorders, such as that of lupus tuberculosis, or leprosy.
   2. State 2 questions that you have from the article.
      1. Felix d’Herelle believed that bacteriophage could treat infectious diseases caused by bacteria, and now biotech firms are trying to make this happen again. Is there a way to prevent bacteriophage from infecting bacteria that do not cause disease in the body, such as those in the intestines?
      2. How long will it take the researchers to find out what the isolated genes (that number currently about 800) whose purpose is unknown do?