

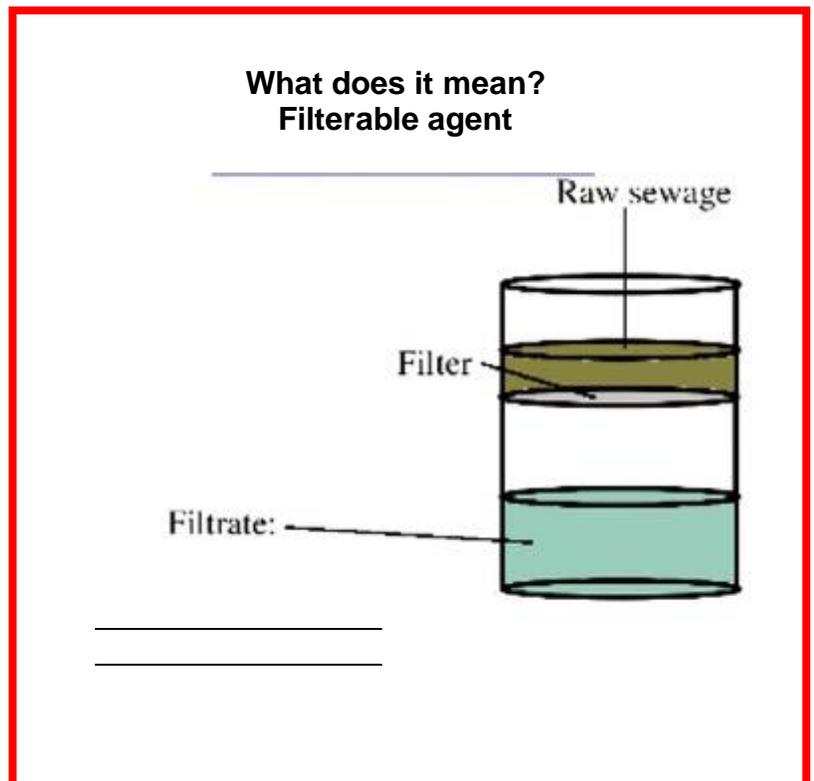
Lecture 21

I. Bacteriophages (also known as “phages”)

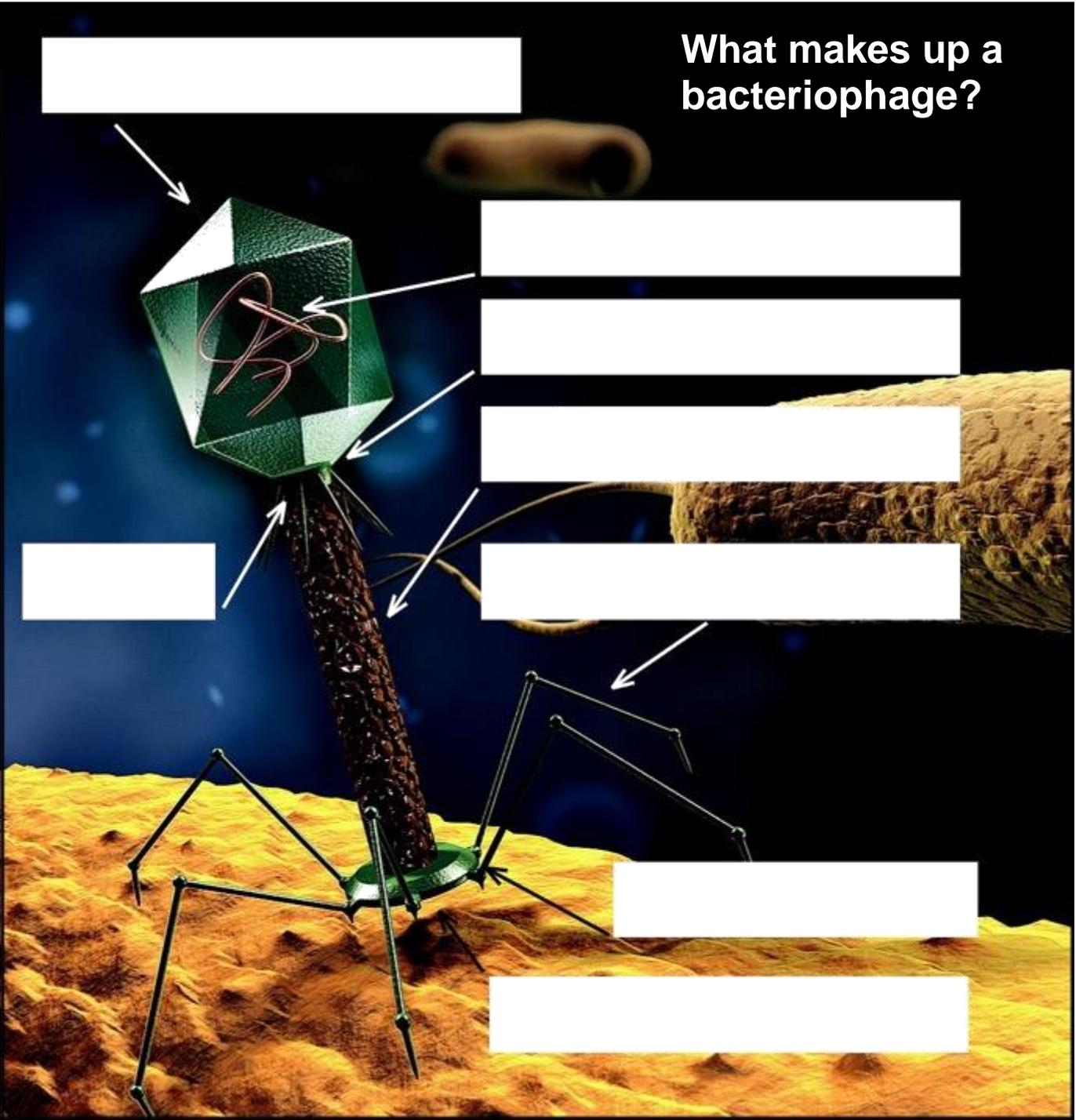
- A. A bacteriophage is a _____ that uses a _____ as its host cell.
- B. It is an _____.
- C. Phages have either an RNA or DNA genome which:
 - 1. Directs _____.
 - 2. Directs synthesis of a _____ that protects and transmits the genome between cells.
- D. Viruses are _____. (See What Does It Mean?, below)

II. Types of Bacteriophages

- A. _____
 - 1. Phages that multiply rapidly in host cells and then destroy them through lysis (the _____).
 - 2. Bacteriophage T4 is a virulent phage.
- B. _____
 - 1. Phages that can follow either the lytic or lysogenic pathway (lambda bacteriophage)
 - i. In the lysogenic pathway, phages produce a repressor protein that prevents the replication of phage DNA. Instead, this DNA is _____ into the host cell's chromosomes and the phage is then called a _____. The host cell will then replicate and produce daughter cells that contain the prophage within their DNA. These cells are called _____ bacteria.



What makes up a bacteriophage?



III. Lytic phage replication cycle (based on bacteriophage T4 of *E. coli*)

A. Step 1: _____

1. Adsorption proteins on the phage tail fibers attach to _____ on the surface of the bacterial cell.
2. As more tail fibers make contact with the bacterial cell, the _____ on the cell surface.

B. Step 2: _____

1. Conformational changes occur in the phage tail and the _____.
2. The phage genome is _____ out of the phage head, through the core and _____.

C. Step 3: _____

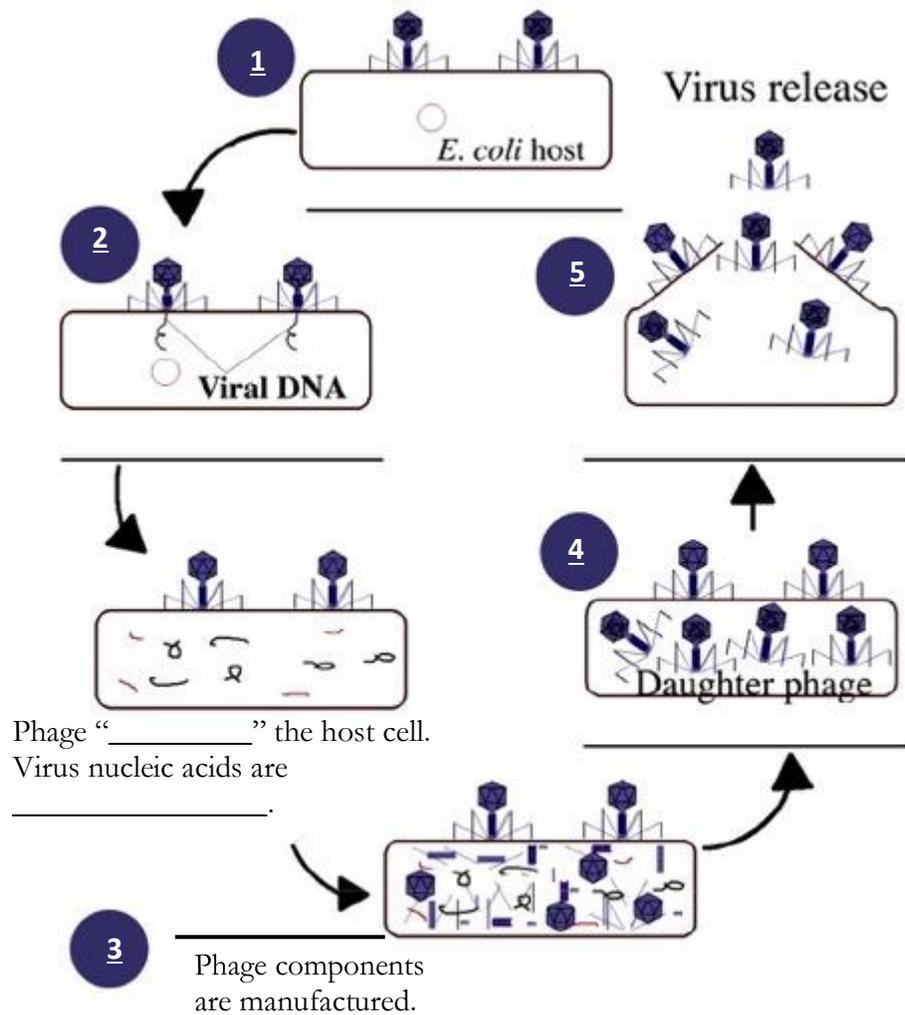
1. The phage _____ transcription and translation of the bacteria's mRNA. The host RNA polymerase starts synthesizing phage mRNA encoding for protein factors and enzymes required to _____, degrade host DNA and manufacture viral nucleic acids.
 - i. _____ of the phage genome are made.
 - ii. Many copies of the _____ are also produced.

D. Step 4: _____

1. Capsid head and the tail proteins are _____ into mature phage particles and the DNA is _____ within the phage head. The newly assembled phages are called _____.

E. Step 5: _____

1. Daughter phages lyse the host cell and are released to infect other bacteria.



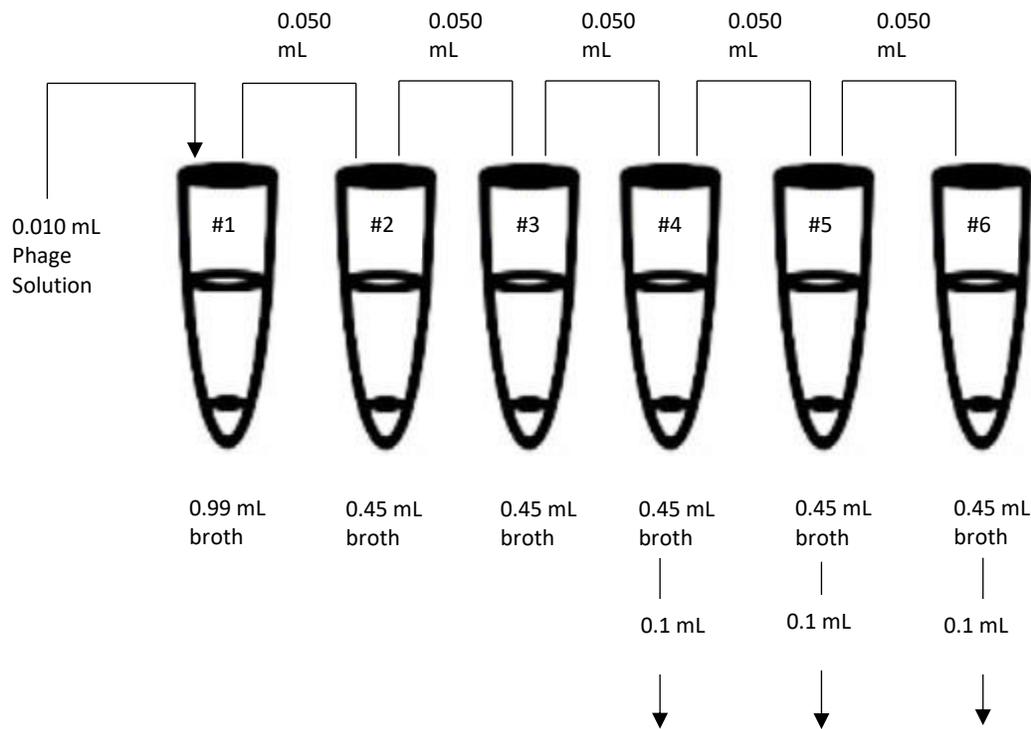
IV. How to isolate viruses

- A. First, the host (bacteria, mammalian cells, etc.) has to be _____ with the virus. The virus is then able to replicate and is _____.
 - B. Next, the host cell must be _____. This allows the viruses to _____ (liquid in which the cells were grown).
 - C. The supernatant is then _____. The filter allows the viruses to pass through with the fluid but does not allow pieces of the lysed cells to pass.
 - D. The end product is a fluid that should _____. During the last lab period, we spotted this fluid onto three quadrants of a TSA plate inoculated with *E. coli*. Today, we should see a lawn of *E. coli* with some small _____ where phages have infected and lysed the bacteria. These clear zones are called _____.
- Remember that, theoretically, _____.

V. Determining bacteriophage titer

- A. Viruses are too small to be seen using a light microscope, so we look at plaques to determine their titer.

1. Titration of bacteriophage



Placed into soft agar with host cells and plated

- B. _____ traps the *E. coli* and the diluted virus between the two layers of agar leading to _____.

Notes for Experiment 23:

- Make sure everything is ready before you go to the water baths so the soft agar _____ prematurely.
- Next time, we will count the plaques and determine the virus titer.

