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 does it mean?
   Filterable agent
   Raw sewage
   Filter
   Filtrate:
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

   ![Diagram of titration process]

   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.