Module 15: Lytic Cycle of Viruses

OBJECTIVES:

The main objectives of this lecture are:

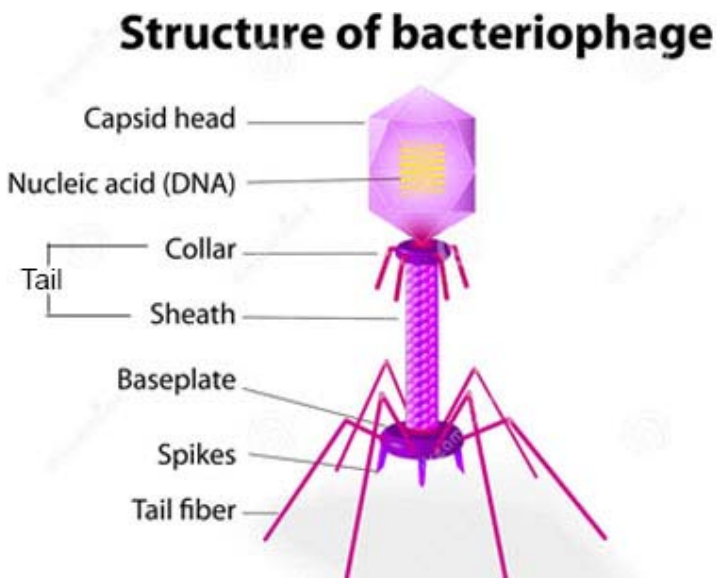
- to discuss the structure of bacteriophage
- to study the general characteristics of bacteriophage
- to discuss the various steps involved in infection of host cells
- to discuss the multiplication cycle of bacteriophage.

INTRODUCTION

Viruses are the obligate intracellular parasites. One group virus known as the bacteriophage infects the bacteria. They use bacterial cell machinery system for its own multiplication and development. Bacteriophage like all viruses also contains the nucleic acid and protein. The genetic material may be either DNA or RNA but never both. The nucleic acids of the phages often contain unusual or modified bases which protects the phage nucleic acid from nucleases that breakdown the phage nucleic acid during phage infection. Some of the examples of bacteriophages are the φX174, T-even phages such as T2, T4 and T6 that infect E.coli and Temperate phages such as λ and μ.

STRUCTURE OF BACTERIOPHAGE

Bacteriophages have different shapes and sizes. T_4 phage is one of the largest among the bacteriophages and it is approximately 200 nm long and 80-100 nm wide. But generally all phages range in size from 24-200 nm in length. Some of the phages may have icosahedral capsid while others are filamentous. Inside the capsid contains the nucleic acid and the capsid acts as the protective covering of the nucleic acid. A hollow tube like structure known as tails is also attached to the capsid which aids in injecting their genetic material inside the host during infection. In T4 phage, the tail is surrounded by a contractile sheath and basal plate like structure present at the end of tail from which the tail fibres are attached. The function of the tail fibres is it helps in attachment of the phage to bacteria while the contractile sheath helps in contraction during infection. Some of the phages do not contain tail fibres at the end.



GENERAL CHARACTERISTICS OF BACTERIOPHAGE

There are two main types of bacterial viruses –

- Lytic or virulent phage
- Temperate or avirulent phage

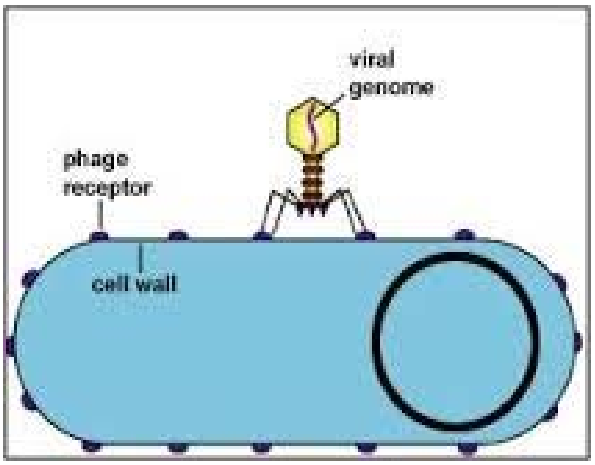
During infection by lytic or virulent phage the cells responds by producing large numbers of new viruses and at the end of incubation period the host cells bursts and release new phages to infect other host cells and hence it is termed as lytic cycle. Whereas in the temperate phage infection the viral nucleic acid is carried and replicated in the host bacterial cells from one generation to the other without the involvement of cell lysis.

INFECTION OF HOST CELLS

The various steps involved in infection includes –

a. Adsorption
b. Penetration
c. Synthesis of phage nucleic acid and proteins
d. Assembly and
e. Release

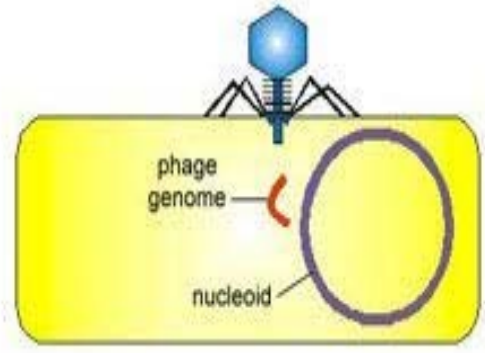
ADSORPTION - Bacteriophage attached to the specific receptor site on the host cell membrane. The nature of the receptor varies with the phage. The cell wall polysaccharides and proteins, teichoic acids, flagella and pilli serves as the receptor. In T-even phages the attachment begins when the tail fiber comes in contact with the appropriate receptor site and the baseplate settles down on the surface. This binding is carried out by electrostatic interactions and is influenced by pH and presence of ions such as Mg^{2+} and Ca^{2+}. Conformational changes occur in the baseplate and sheath when the baseplate is seated firmly on the cell membrane and the tail sheath contracts. Further, the central tube or core is pushed through the bacterial wall.



PENETRATION – the penetration of the phage into the host cell is mostly mechanical but it may be facilitated by localized digestion of certain cell surfaces structures either by phage enzymes carried on the tail of the phage or by viral activation of host degradative enzymes. Penetration in T-even phages is achieved when –

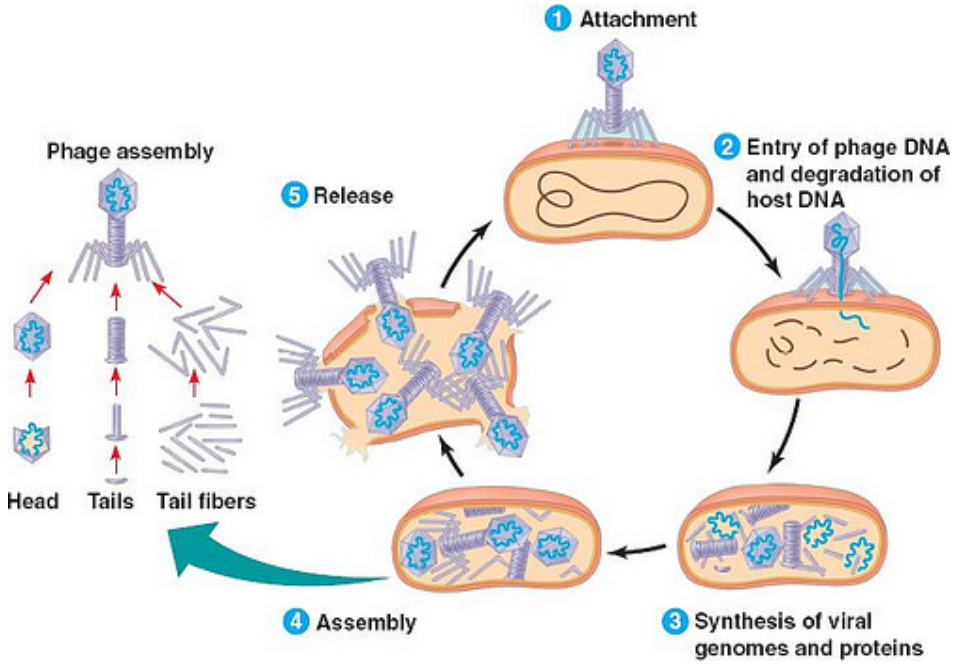
a. Tail fibers of the virus attach to the cell and hold the tail firmly against the cell wall.
b. The sheath contracts, driving the tail core into the cell through the cell wall and membrane and
c. The virus injects its DNA.

The protein coat that forms the phage head and the tail structure of the virus remain outside the cell.



SYNTHESIS OF PHAGE NUCLEIC ACIDS AND PROTEINS - After the phage DNA is injected, the synthesis of host DNA, RNA and protein is halted and the cell is forced to make the viral constituents. The RNA polymerase starts synthesizing phage mRNA. This mRNA and all the other mRNA (mRNA that are transcribed before phage DNA is made) directs the synthesis of protein factors and enzymes required to take over the host cell and manufacture viral nucleic acids. The host DNA are degraded to nucleotides by some virus specific enzymes, thereby halting the host gene expression and providing the raw material for virus DNA synthesis. The bacterial RNA polymerase is modified by the other early proteins and a phage head proteins that are injected along with the phage DNA. The polymerase than recognised the promoters on the viral DNA rather than bacterial promoter sequences so that virus genes are

transcribed. Ultimately within short period of time the virus DNA synthesis commences. For the synthesis of T4 DNA considerable preparation is required as it contains hydroxymethylcytosine(HMC) instead of cytosine. This HMC must be synthesized by two phage encoded enzymes before DNA replication can begin. After T4 DNA has been synthesized, it is glucosylated by the addition of glucose to the HMC residues. Glucosylated HMC residues protect T4 DNA from attack by restriction enzymes which would cleave the viral DNA at specific points and destroy it. In T4 DNA a base sequence is repeated at both the ends of the DNA molecule which is known as terminal redundancy. After many DNA copies are made, about 6-10 copies are joined by their terminally redundant ends with the aid of several enzymes. This long DNA strand is composed of several units linked together with the same orientation known as concatemers. During phage assembly this concatemers are cleaved in a way that the genome is slightly longer than the T4 gene set.



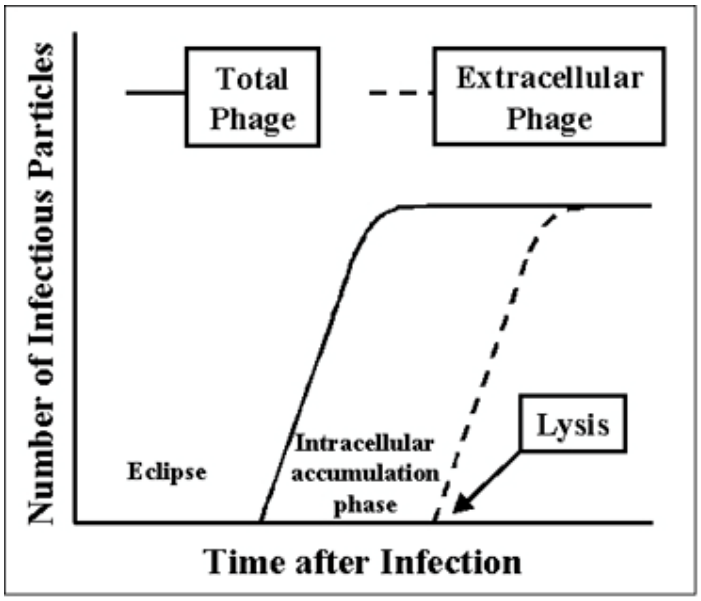
ASSEMBLY – the assembly in T4 phage is complex. The late mRNA directs the synthesis of three kinds of proteins - phage structural proteins, proteins that help with phage assembly without becoming part of the virion structure and proteins involved in cell lysis and phage release. In T4 phage late mRNA transcription begins about 9 minutes after the DNA is injected into the bacteria and simultaneously all the proteins required for phage assembly are synthesized. After the baseplate is constructed the tail tube is built on it and the sheath is assemble around the tube. The procapsid of the phage is constructed separately of more than 10 proteins and then combines with the tail assembly. The procapsid is assembled with the help of a protein known as the scaffolding proteins. Further, the tail fibres are attached to the baseplate after the head and tail have come together. Finally the DNA is drawn into the complete shell efficiently.

RELEASE – Many T4 genes are involved in the release process. One directs the synthesis of a lysozyme that attack the peptidoglycan of the bacterial cell wall. Another phage protein damages the bacterial plasma membrane and allowing lysozyme to reach the cell wall beyond it and inhibits energy metabolism reactions located in the membrane.

MULTIPLICATION CYCLE OF BACTERIOPHAGE

The sequence of events initiated by the injection of the phage nucleic acid and culminating in the release of newly synthesized virions is termed as the viral multiplication or the replication cycle. Viral multiplication involved two phases or period –

1. Eclipse period
2. Intracellular accumulation phase



ECLIPSE PERIOD - During the eclipse phase, no infectious phage particles can be found either inside or outside the bacterial cell. The phage nucleic acid takes over the host biosynthetic machinery and phage specific m-RNA's and proteins are made. There is an orderly expression of phage directed macromolecular synthesis, just as one sees in animal virus infections. Early m-RNA's code for early proteins which are needed for phage DNA synthesis and for shutting off host DNA, RNA and protein biosynthesis. In some cases the early proteins actually degrade the host chromosome. After phage DNA is made late m-RNA's and late proteins are made. The late proteins are the structural proteins that comprise the phage as well as the proteins needed for lysis of the bacterial cell.

INTRACELLULAR ACCUMULATION PHASE – At the end of eclipse period mature phages begins to accumulate intracellularly until they are release by cell lysis. In this phase the nucleic acid and structural proteins that have been made are assembled and infectious phage particles accumulate within the cell. No newly released extracellular phages can be seen until lysis begins. Hence, the time interval from infection until lysis is termed as the latent period. At the end of the multiplication cycle the phage number continue to increase until it reach the constant titre and this time interval is termed as the rise period.

CONCLUSION

Lytic cycle of virulent bacteriophages is a life cycle that ends with host cell lysis and virion release. The life cycle of T4 phage composed of several phases. In adsorption phase the phage attaches to the specific receptor sites followed by the penetration of the cell wall and insertion

of viral nucleic acid into the cell. Finally the complete virions are assembled immediately after the separate components have been constructed and the phages are released upon lysis.

Transcript

INTRODUCTION

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CONCLUSION

So that was about the lytic cycle of viruses and I'll wind up by adding that lytic cycle of virulent bacteriophages is a life cycle that ends with host cell lysis and virion release. The life cycle of T4 phage composed of several phases. In adsorption phase the phage attaches to the specific receptor sites followed by the penetration of the cell wall and insertion of viral nucleic acid into the cell. Finally the complete virions are assembled immediately after the separate components have been constructed and the phages are released upon lysis.

GLOSSARY

Flagella – A whip- like structure that helps the cell in movement

mRNA – Messenger RNA

Lysozyme – An enzyme that catalyzes the destruction of cell wall of certain bacteria

Peptidoglycan – A polymer consisting of sugar and amino acids that forms a mesh – like structure outer to the plasma membrane of certain bacteria

Intracellular – Inside the cell

Virion – A complete viral particle

Metabolism – Total chemical reaction inside the cell

Pilli – A hair – like appendage that are found on the surface of bacteria which plays a role in bacterial conjugation

Nucleases – An enzyme that catalyzes the hydrolysis of nucleic acid

FAQS

1. What are bacteriophage? Give some examples.

   Answer: Bacteriophage are the viruses that infects bacteria. Some of the bacteriophages are the lambda phage, T_4 phages etc.

2. Name some of the T- even phages.

   Answer: T- even phages are the T_2 , T_4, T_6 etc.

3. What are the function of tail fibres and contractile sheath in bacteriophage?

   Answer: The function of the tail fibres is it helps in attachment of the phage to bacteria while the contractile sheath helps in contraction during infection.

4. What are the steps involved in the phage infection to the host cell?

   Answer: The various steps involved in phage infection to the host cells are adsorption, penetration, synthesis of phage nucleic acid and proteins, assembly and release.

5. Differentiate between lytic phage and temperate phage.

Answer: In lytic or virulent phage, the cells responds by producing large numbers of new viruses during infection and at the end of incubation period the host cells bursts and release new phages to infect other host cells whereas in the temperate phage infection the viral nucleic acid is carried and replicated in the host bacterial cells from one generation to the other without the involvement of cell lysis.

6. How does penetration in T – even phages is carried out?

Answer: Penetration in T-even phages is achieved when –

g. Tail fibers of the virus attach to the cell and hold the tail firmly against the cell wall.
h. The sheath contracts, driving the tail core into the cell through the cell wall and membrane and
i. The virus injects its DNA.

7. What is viral multiplication?

Answer: The sequence of events initiated by the injection of the phage nucleic acid and culminating in the release of newly synthesized virions is termed as the viral multiplication.

8. What are the two phases involved in viral multiplication?

Answer: the two phases involved in viral multiplication are the eclipse period and the intracellular accumulation phase.

9. Explain in brief the eclipse phase during virus multiplication.

Answer: During the eclipse phase, no infectious phage particles can be found either inside or outside the bacterial cell. The phage nucleic acid takes over the host biosynthetic machinery and phage specific m-RNA's and proteins are made. Early m-RNA's code for early proteins which are needed for phage DNA synthesis and for shutting off host DNA, RNA and protein biosynthesis. In some cases the early proteins actually degrade the host chromosome. After phage DNA is made late m-RNA's and late proteins are made. The late proteins are the structural proteins that comprise the phage as well as the proteins needed for lysis of the bacterial cell.

10. What is intracellular accumulation phase during virus multiplication?

Answer: The phase during which the nucleic acid and structural proteins that have been made are assembled and infectious phage particles accumulate within the cell are called the intracellular accumulation phase.

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