

Welcome Students,

I am Dr. Janet Mascarenhas and I will be presenting a module on Bacterial Growth Curve. This module is from unit 2 Methods in Microbiology, from the paper Microbiology and Plant Pathology. The module number is 11.

Outline of this module are:

Stages of binary fission

Exponential increase in the cell number of bacteria

Phases of bacterial growth curve

Significance of bacterial growth curve

Learning outcome

At the end of this module, students will be able to describe the stages of binary fission, explain the growth of organisms in a batch culture, explain exponential growth of bacteria and generation time and describe the phases of the bacterial growth curve.

Now let us begin with the topic Bacterial Growth.

Now, as we all know bacteria are prokaryotic organisms. In these organisms the growth involves an increase in the number of organisms and not in the cell size. The replication in bacteria takes place by means of a asexual process which is called as binary fission.

Now, let us study the various steps in binary fission. Initially the cell shows a tightly coiled DNA inside the cell. In the second step the DNA replicates, similarly, there is synthesis of cellular material taking place inside the cell. In the next step, the cell increases in cell size and there is separation of the replicated DNA. The next step is the beginning of cell division. The cell divides by the constriction or the invagination of the cell membrane. Completion of the cell division takes place by the formation of two genetically identical daughter cells.

Growth of microorganisms in the laboratory

This can be carried out either on solid media, through the development of colonies or in liquid media by the formation of a turbid suspension.

Now, let us study the procedure for the growth of organisms in a batch culture. Batch culture means wherein no additional nutrients are added to the medium. First, we inoculate bacteria into a fixed volume of liquid culture medium. This is incubated under ideal conditions of growth. Then we need to take cell counts at fixed intervals after inoculation. Finally, we plot a graph of the cell count versus incubation time. This plot, what we see, is a 'Sigmoid Growth Curve'.

Coming to the phases of growth. There are four phases recognized in a typical sigmoid growth curve. First is the lag phase, second exponential phase, third stationary phase and the fourth is the death phase.

Now, this is a typical sigmoid S-shaped growth curve. On the x-axis time is plotted while the y-axis represents the log of number of bacteria. Now, the first stage is the lag phase, where we see that there is no increase in the number of bacteria. In the next phase, that is, the exponential phase the number of bacteria increases exponentially. This is followed by the stationary phase, where again we see that there is no increase in the number of bacteria, and the last stage is the death phase where the number of bacteria starts to decline.

Now, this broken line represents theoretical growth, wherein, if the growth is not limited, maybe due to the depletion of the nutrients, doubling will continue producing a straight line.

Now, let us study about each phase. First is the lag phase. In a freshly inoculated medium the cells take time to adjust to the new environment and prepare to divide. In this phase there is no increase in the cell number, but the cells grow in volume or mass and show an increase in the metabolic activity. Like, there might be synthesis of certain enzymes, proteins, RNA, etc. This gap of time is called the lag phase. Now, there are many factors which affect the length of the lag phase, like the type of bacterial species, the size of the inoculum, culture medium and various other environmental factors.

The next phase is the exponential phase. Now, in this phase the cells start dividing by binary fission. Now we have already studied the various steps of binary fission, where one cell divides to give rise to two genetically identical daughter cells. The cell number increases proportionally with time, therefore, this phase is termed as the exponential or the log phase. The cells divide at a constant rate as long as nutrients and the environmental conditions are ideal for growth. The bacterial population doubles at a constant interval which is called the generation time or the doubling time.

Now this slide represents the exponential increase in cell number. Now, here we see that the parental cell will divide by binary fission to give rise to two daughter cells. Now, each of these daughter cells will again divide into two cells, thereby giving a total of four cells in the second generation. Similarly, these four cells will divide again to give rise to a total number of eight cells in the third generation. So each division results in doubling the number of bacterial cells or you can say each division doubles the number of cells.

Now, what is bacterial generation or doubling time?

It is the time taken for a population to double through one round of binary fission and this is denoted as 'g', which is equal to time per number of generations

Now let us see some examples of generation time in bacteria. Now, the generation time varies according to the organism and the environmental conditions. Under ideal conditions *E. coli*, i.e., the common colon bacterium, takes 20 minutes or the generation time is 20 minutes for it to double its cell number. In *Mycobacterium tuberculosis*, this is the bacterium which causes tuberculosis, the generation time is 18 hours. While, *Mycobacterium leprae*, i.e., the organism which causes leprosy, it is a very slow growing organism and the generation time is 14 days.

Now the third phase is the stationary phase. This is also called as the plateau phase. The exponential growth it cannot continue forever in a batch culture. Batch culture refers to a closed system, wherein, no nutrients are added and the waste or the dead cells are not removed. The number of cells reaches a maximum and does not increase further. The population growth is limited due to the depletion of available nutrients, limited oxygen availability, accumulation of inhibitory by-products, etc. So, due to all these limiting factors the growth of the organism is limited in the stationary phase. Now, some bacteria produce secondary metabolites such as antibiotics in this stationary phase.

The next phase is the death phase. This is also called as the phase of decline. It is the reverse of growth during the log phase. The number of viable cells decreases geometrically or exponentially in this phase and there is irreversible loss of ability to reproduce. Death occurs due to various adverse conditions in the culture environment which could be due to the depletion of the nutrients, limitation of the space, accumulation of waste products, etc., but in some cases, some resistant bacteria might show extended survival even after the stationary phase.

Next are the factors regulating growth. So, what are the factors which regulate the growth in the medium? First is the availability of nutrients, second various environmental conditions such as temperature, oxygen, pH and osmotic pressure and the last factor is generation time.

Coming to the significance of the bacterial growth curve. The first significance is to understand population dynamics and population control in the course of infectious diseases and in food preservation. Second is to inoculate known numbers of a bacterial isolate to increase biodegradation of a toxic compound or produce antibiotics or other compounds.

Okay, let us summarize what we have studied. We have seen that the bacterial cells divide by binary fission. The growth curve represents the number of viable cells in a population over time. Then we saw that the growth curve comprises of four growth phases which is as follows: first one lag phase where the cells are metabolically active but not dividing. Second was the exponential phase which shows initial slow multiplication of the cells but later exponential growth of the cells takes place. Third was the stationary phase in which the growth rate reaches a plateau and the last stage was the death phase where there is an exponential decrease in the number of living cells.

Now these are the books or the references which I have used for this module.

These are some additional references which you can use to find more information on the internet.

Thank you.