# M Sc. Biotechnology (First Semester) Examination, 2013 LBTM :102 (Microbial genetics and physiology)

Answer 1. (MCQ)

 $(i) d \quad (ii) d \quad (iii) c \quad (iv) d \quad (v) d \quad (vi) a \quad (vii) f \quad (viii) b \qquad (ix) d \qquad (x) c$ 

## 2. (i) Describe about the growth kinetics of unicellular microorganism in closed system.



Growth is defined as an increase in cellular constituents and may result in an increase in a microorganism's size, population number, or both. When microorganisms are grown in a closed system, population growth remains exponential for only a few generations and then enters a stationary phase due to factors such as nutrient limitation and waste accumulation. Therefore the culture flask having four important phases like

1. Lag Phase (there is no net increase in mass, the cell is synthesizing new components)

**2.** Log/ Exponential Phase (rate of growth is constant during the exponential phase, the microorganisms are dividing and doubling in number at regular intervals).

**3. Stationary Phase** (the total number of viable microorganisms remains constant, the population may simply cease to divide though remaining metabolically active).

**4. Death Phase** (detrimental environmental changes like nutrient deprivation and the buildup of toxic wastes lead to the decline in the number of viable cells).

(Explain metabolic and physiological behavior)

## (ii) Who established the pure culture technique and what were the postulates?

German physician Robert Koch established the pure culture technique.

Koch's postulates and can be summarized as follows:

- 1. The microorganism must be present in every case of the disease but absent from healthy organisms.
- 2. The suspected microorganism must be isolated and grown in a pure culture.
- 3. The same disease must result when the isolated microorganism is inoculated into a healthy host.
- 4. The same microorganism must be isolated again from the diseased host.

(Describe about pure culture technique in brief and the points of postulates)

# (iii) What is generation time? Calculate the number of generation (n) if No bacteria inoculated and after n generation it becomes N microbial cells.

During the exponential phase each microorganism is dividing at constant intervals. Thus the population will double in number during a specific length of time called the **generation time** or **doubling time**. This situation can be illustrated with a simple example. Suppose that a culture tube is inoculated with one cell that divides every 20 minutes. The population will be 2 cells after 20 minutes, 4 cells after 40 minutes, and so forth. Because the population is doubling every generation, the increase in population is always 2n where n is the number of generations. The resulting population increase is exponential or logarithmic.

These observations can be expressed as equations for the generation time.

Let  $N_0$  = the initial population number

 $N_t$  = the population at time t

n = the number of generations in time t

Then inspection of the results in table 6.1 will show that

$$N_t = N_0 \times 2^n$$
.

Solving for n, the number of generations, where all logarithms are to the base 10,

$$\log N_t = \log N_0 + n \cdot \log 2, \text{ and}$$
$$n = \frac{\log N_t - \log N_0}{\log 2} = \frac{\log N_t - \log N_0}{0.301}$$

(solve the equation)

#### (iv) Describe about major nutritional types of Microorganisms.

In addition to the need for carbon, hydrogen, and oxygen, all organisms require sources of energy and electrons for growth to take place. Microorganisms can be grouped into nutritional classes based on how they satisfy all these requirements.

Major Nutritional Types <sup>a</sup>	Sources of Energy, Hydrogen/Electrons, and Carbon	Representative Microorganisms
Photolithotrophic autotrophy (Photolithoautotrophy)	Light energy Inorganic hydrogen/electron (H/e <sup>-</sup> ) donor CO <sub>2</sub> carbon source	Algae Purple and green sulfur bacteria Cyanobacteria
Photoorganotrophic heterotrophy (Photoorganoheterotrophy)	Light energy Organic H/e <sup>-</sup> donor Organic carbon source (CO <sub>2</sub> may also be used)	Purple nonsulfur bacteria Green nonsulfur bacteria
Chemolithotrophic autotrophy (Chemolithoautotrophy)	Chemical energy source (inorganic) Inorganic H/e <sup>-</sup> donor CO <sub>2</sub> carbon source	Sulfur-oxidizing bacteria Hydrogen bacteria Nitrifying bacteria Iron-oxidizing bacteria
Chemoorganotrophic heterotrophy (Chemoorganoheterotrophy)	Chemical energy source (organic) Organic H/e <sup>-</sup> donor Organic carbon source	Protozoa Fungi Most nonphotosynthetic bacteria (including most pathogens)

 Table 5.2
 Major Nutritional Types of Microorganisms

## (v) What is transposon and describe about mechanism of transposition.

The chromosomes of bacteria, viruses, and eucaryotic cells contain pieces of DNA that move around the genome. Such movement is called **transposition**. DNA segments that carry the genes required for this process and consequently move about chromosomes are **transposable elements** or **transposons**. They doesn't require homology between transposon and target sequence to integrate. According to their features three types of transposons mainly simple, composite and Tn3 class of transposons.

There are two important mechanism of transposition

### 1. Conservative



2. Replicative transposition.



(Explain)

#### (vi) Describe about transduction and sexduction.

**Transduction** is the transfer of bacterial genes by viruses. Bacterial genes are incorporated into a phage capsid because of errors made during the virus life cycle. The virus containing these genes then injects them into another bacterium, completing the transfer. Transduction may be the most common mechanism for gene exchange and recombination in bacteria. There are two very different kinds of transduction: **generalized** and **specialized**.





**Specialized Transduction** 

Bacterial genes on the F' plasmid are transferred with it and need not be incorporated into the recipient chromosome to be expressed. The recipient becomes F' and is a partially diploid merozygote since it has two sets of the genes carried by the plasmid. In this way specific bacterial genes may spread rapidly throughout a bacterial population. Such transfer of bacterial genes is often called **sexduction**.

(Explain)

# (vii) Describe about the direct measurement methods of microbial growth.

Microbial populations can be counted directly as follows

- 1. Measurement of Cell Numbers by Petroff-Hausser counting chambers.
- 2. Membrane filters are used to count bacteria directly.
- 3. By using spectrophotometer.

4. By using special colony counter to know cfu. In this way the spread-plate and pour-plate techniques may be used to find the number of microorganisms in a sample.

5. By using fluorescence microscopy.

(Explain)