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B.Sc. (Hons) Fifth Semester Examination-2014
Biotechnology
LBTC-502: Plant Animal Tissue Culture
Paper Code: AU-6941

Model Answer

Section-A: Multiple question answers

Ans1. i.(b) ii. (b) iii. (a) iv. (a) v. (a) vi. (a)

vii. (a) viii. (c) ix. (a) x. (b)

Section-B: Descriptive question answers

Ans-2: Sterilization Techniques: Sterilization can be defined as any process that effectively kills or eliminates transmissible agents (such as fungi, bacteria, viruses and prions) from a surface, equipment, foods, medications, or biological culture medium.

Methods of Sterilization: The various methods of sterilization are broadly classified into two types i.e.

1. **Physical Method**
   a. Thermal (Heat) methods b. Radiation method c. Filtration method
2. **Chemical Method**
   a. Gaseous method b. Liquid method

*Explain each methods in detail

Ans-3: Clonal propagation refers to the process of asexual reproduction by multiplication of genetically identical copies of individual plants. The tissue culture methods of plant propagation, known as 'micropropagation'

(explain on the following points)

Approaches: (a) Auxiliary bud approach (b) Organogenesis (c) Embryogenesis

Stages: a) Initiation of culture b) multiple shoots formation c) rooting d) transplantation

Factors: (write few lines about each factors)

(a) genotype (b) the culture medium (c) culture environment (d) explants source

Benefits:

Ans-4a: Nutrient media

The intact plants can make their own food but the ***in vitro*** culture of plant parts or cells requires a variety of nutrients and suitable physical conditions for their growth. A typical nutrient medium consists of the following components: (write few lines on each component)

i) inorganic nutrients (both micro- and macro-elements), ii) a carbon source and energy source iii) Organic supplements, iv) Growth regulators, v) Solidifying agents, vi) pH

Ans-4b: Auxins are the phyto hormones induce cell division, cell elongation, and formation of callus in cultures. 2,4-dichlorophenoxy acetic acid is one of the most commonly added auxins in plant cell cultures.

* write different types of auxins with examples and their functions

Ans-5a: Batch cultures are initiated as single cells in 100- 250 ml flasks and are propagated by transferring regularly small aliquots of suspension to a fresh medium. Explain on the following points

Methods:

Application:

Limitations:

Ans-5b: Continuous cultures are maintained in a steady state for long period by adding fresh medium. (explain on the following heads)

Types:

a. Open continuous culture
b. closed continuous culture
    i. Turbidostat (ii)Chemosta

Methods:

Application:

Limitations:

Ans-6: HAT contains a drug Aminopterin which blocks one pathway for nucleotide synthesis making the cells to depend upon other pathway that needs HGPRT enzyme, which is absent in Myeloma cells. Therefore, Myeloma cells which do not fuse with the B cells will die, since they are HGPRT-.B cells which do not fuse with the Myeloma cells will die, because they lack tumorgenic property of immortal growth. Therefore HAT medium allows selection of Hybridoma cells which inherit HGPRT gene from B cells and tumorgenic property from Myeloma cells.

Biochemistry involved in HAT selection: (Discuss about HAT medium and how it selects hybrid cells).

Ans-7: The first subculture represents an important transition for a culture. The need to subculture implies that the primary culture has increased to occupy all of the available substrate.

From a very heterogeneous primary culture, containing many of the cell types present in the original tissue, a more homogeneous cell line emerges. Once a primary culture is subcultured (or passaged), it becomes known as a **cell line**. When a cell line is subcultured the regrowth of the cells to a point ready for the next subculture usually follows a standard pattern. A lag period after seeding is followed by a period of exponential growth, called the log phase. When the cell density (cells/cm^2 substrate) reaches a level such that all of the available substrate is occupied, or when the cell concentration (cells/ml medium) exceeds the capacity of the medium, growth ceases or is greatly reduced. **Then either the medium must be changed more frequently or the culture must be divided**.

(Explain the above statement graphically).

Criteria for Subculture: The need to subculture a monolayer is determined by the following criteria: (Explain the following criteria)

(1) **Density of Culture. (2) Exhaustion of Medium**. (3)**Time since Last Subculture**.

Stages of Subculture of Monolayer:

(Diagrammatically Explain the Subculture of Monolayer)

Ans-8: Cell fusion is a biological process in which the plasma membranes of two cells break down at the point of contact between them and the cytoplasm of the two mixes. The product of fusion was called **Homokaryon** if the two parental cells come from the same species, and **Heterokaryon** or Somatic Cell Hybrid if the fusion if both cells are not identical. Cell fusion is followed by nuclear fusion to produce uninucleate hybrid cells or **Synkaryons**. (Explain with Diagram).

Types of cell Fusion: (Explain under the following heads)

1- Spontaneous cell fusion
2- Induced cell fusion: cells can be induced to be fuse by the following methods:
    i. Viruses (ii) Electrofusion (iii)Chemicals

Selection of Hybrid Cells: (Explain HAT selection of Hybrid cells**).**