RAMA UNIVERSITY

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FACULTY OF ENGINEERING & TECHNOLOGY

Course: B. Tech Biotechnology
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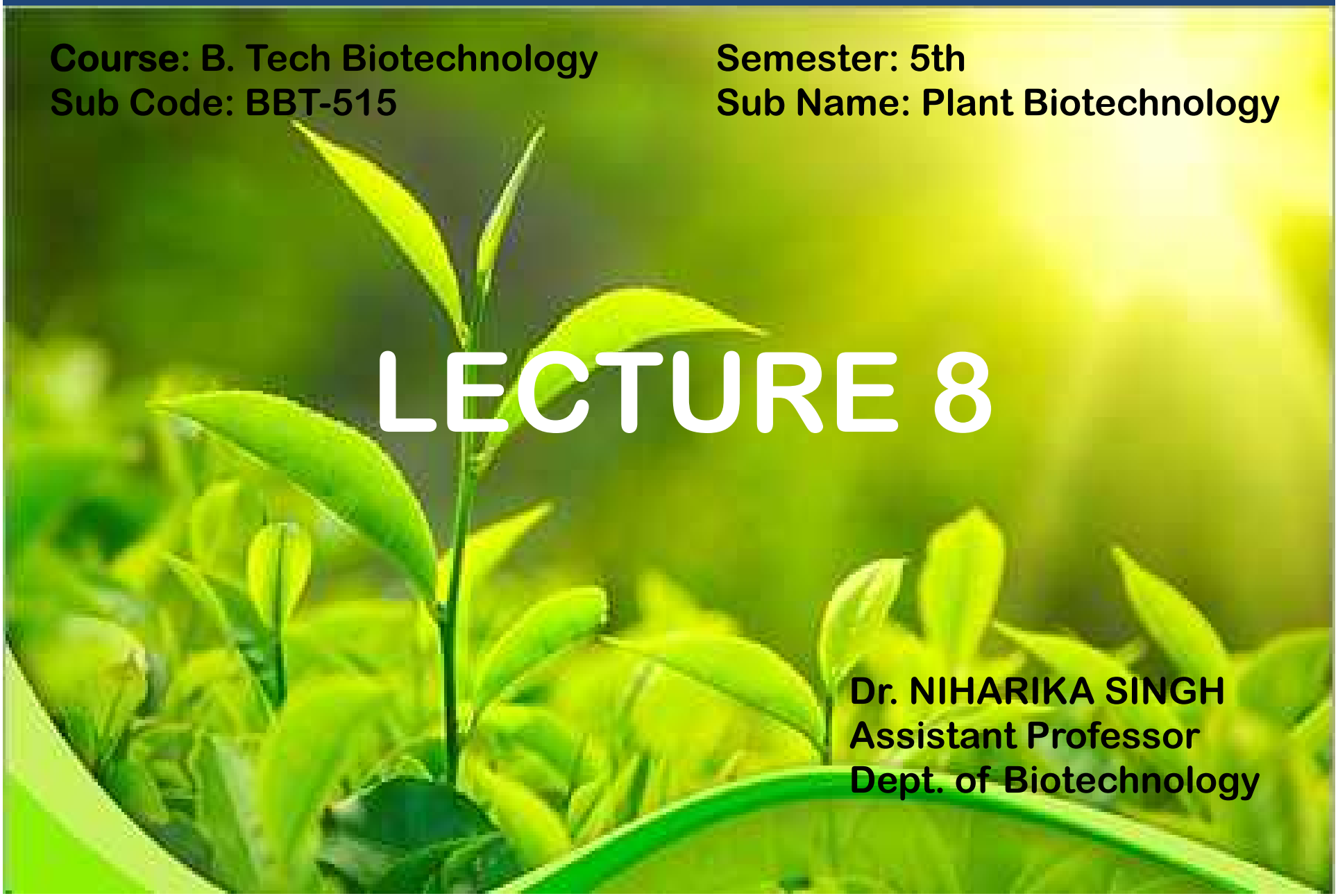

LECTURE 8

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MAINTENANACE

After sufficient time of callus growth on the same medium following changes will occur :

- ✓ Depletion of nutrient in the medium
- ✓ Gradual loss of water
- ✓ Accumulation of metabolic toxins

Hence for maintenance of growth in callus it is necessary to subculture the callus.
Subculture should be repeated after 4-5 weeks

CELL SUSPENSION CULTURE

- A friable callus is often used to initiate a liquid cell suspension culture .
- Consists of cell aggregates dispersed and growing in moving liquid media.
- It involves active proliferation of callus as undivided unit suspended or submerged in a liquid medium.
- The nutrient medium in this case is in constant agitation so as to prevent the cells from settling or aggregating in to clumps.
- Suspension cultures are normally initiated by transferring pieces of undifferentiated callus to a liquid medium which is agitated during incubation.

SUB CULTURING

- A subculture is a new cell made by transferring some or all cells from a previous culture to fresh growth medium. This action is called sub-culturing the cells.
- Sub-culture is process in which the plant tissue or explants is first subdivided and transferred into fresh culture medium.
- If tissue cultures are kept in the same culture vessel, they die in due course of time. Therefore, cells/tissues are regularly transferred into new culture vessels containing fresh media.
- It is important to note that during subculture; only a part of the culture from a vessel is transferred into the new culture vessel.
- The callus and suspension cultures may be used to achieve cell biomass production, regeneration of plantlets, production of transgenic plants and isolation of protoplasts.

METHODS OF SUBCULTURING

Basically there are three types of suspension cultures: batch culture, continuous culture and immobilized culture.

1. Batch Suspension Culture

- It means 'Culture in a fixed volume of culture medium'.
- In general, a nutrient medium and cellular inoculum are mixed, aerated and allowed to grow.
- In Batch cultures, as the cells grow, the medium is depleted of nutrients and metabolic byproducts from the cells accumulate.
- Batch cultures are characterized by - continuous changes in the medium - continuous internal changes in cellular composition - accumulation of metabolic products.
- The system is "closed" with respect to additions or removal of culture, except for circulation of air.

2. Semi-continuous Suspension Culture

- Here, the system is ‘open’.
- There is periodic removal of culture and the addition of fresh medium, due to which growth of the culture is continuously maintained.

3. Continuous Suspension Culture

- The system is ‘open’ here also.
- In this systems, volume of culture remains constant and fresh medium is added continuously to a fixed volume of growing culture and withdrawn.

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