

Plant Biotechnology Handbook

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Plant biotechnology is a precise process in which scientific techniques are used to develop molecular and cellular based technologies to improve plant productivity, quality and health; to improve the quality of plant products; or to prevent, reduce or eliminate constraints to plant productivity caused by diseases, pest organisms and environmental stresses. It can be defined as human intervention on plant material by means of technological instruments in order to produce permanent effects, and includes genetic engineering and gene manipulation to obtain transgenic plants. Plant genetic engineering is used to produce new inheritable combinations by introducing external DNA to plant material in an unnatural way. The results are genetically modified plants (GMPs) or transgenic plants. The key instrument used in plant biotechnology is the plant tissue culture (PTC) technique which refers to the in vitro culture of protoplasts, cells, tissues and organs. Plant biotechnology in use today relies on advanced technology, which allows plant breeders to make precise genetic changes to impart beneficial traits to plants. The application of biotechnology in agriculture has resulted in benefits to farmers, producers and consumers. Plant biotechnology has helped make both insect pest control and weed management safer and easier while safeguarding plants against disease. The worldwide demand for food, feed and modern textile fibers can only be met in the future with the help of plant biotechnology. It has the potential to open up whole new business areas that will totally redefine the current market scope and perception.

This book majorly deals with the organisms of biotechnology, herbicide resistant plants, transgenic plants with improved storage proteins, engineering for preservation of fruits, enhancing the photosynthetic efficiency, basic requirements for nitrogen fixation, animal and plant cell cultures, insecticides, cellular characteristics which influence the choice of cell, the growth of animal and plant cells immobilized within a confining matrix, virus free clones through plant tissue culture, microbial metabolism of carbon dioxide, organisms involved in the conversion of hydrogen, hydrogen utilization by aerobic hydrogen oxidizing bacteria, overproduction of microbial metabolites, regulation of metabolite synthesis etc.

The book contains measurement of plant cell growth, plant tissue culture, initiation of embryo genesis in suspension culture, micro propagation in plants, isolation of plant DNA and many more. This is very helpful book for entrepreneurs, consultants, students, institutions, researchers etc.

1. The organisms of biotechnology

Cells - The Basic Units

Types of Microorganism

Viruses

Prokaryotes

Eukaryotes

Algae

Protozoa

Fungi

Tissue Cultures

Animal Cells

Plant Cells

2. Transgenic plants

Herbicide Resistant Plants

1. Glyphosate Tolerant Plants

2. Sulphonylurea Tolerant Plants

3. Atrazine Tolerant Plants

4. Phosphinothricin Tolerant Plants

5. Bromoxynil Tolerant Plants

Insect Resistant Plants

1. Transgenic Plants with Bt Toxin

2. Transgenic Plants with Bt Toxin and Serine Protease

Inhibitor Gene

3. Transgenic Plants with Cowpea Trypsin Inhibitor

4. Transgenic Plants with Nicotiana glauca Proteinase Inhibitor

Virus Resistant Plants

1. Transgenic Plants with Viral Coat Protein

2. Transgenic Plants with Viral Nucleoprotein

3. Transgenic Plants with Viral SAT RNA

4. Transgenic Plants with Antisense RNA

Transgenic Plants Resistant to Fungi and Bacteria

Transgenic Plants with Improved Storage Proteins

Sweet Proteins

Enriching the Carbohydrate Contents

Improving the Quality of Oils and Fats

Male Sterility and Fertility Restoration

Changing the Flower Colours

Stress Tolerant Plants

Cold Tolerant Plants

Drought Tolerant Plants

Plant Tolerant to High Light Intensity

Engineering for Preservation of Fruits

Enhancing the Photosynthetic Efficiency

Transgenic Plants as Bioreactors

Vaccines

Interferons

Pharmaceutical Compounds

Biodegradable Plastics

3. Biological Nitrogen fixations

Non-symbiotic Nitrogen Fixation

Features Favourable for Non-symbiotic Nitrogen Fixation

1. Special separation of Nitrogen Fixing Cells

2. Protein-Nitrogenase Association

3. High Rate of Respiration

4. Time specific Nitrogenase Activity

5. Association with Rapid Oxygen Consumers

6. Presence of hydrogenase

7. Colonization

Nitrogenase

Basic requirements for Nitrogen Fixation

Mechanism of Nitrogen Reduction

Assimilation of Ammonia

Route I

Route II

Symbiotic Nitrogen Fixation

Host Specificity

Root Nodulation

Mechanism of Nitrogen Fixation

(a) Oxygen Transport by Leghaemoglobin

(b) Utilization of Oxygen by Hydrogenase

Nitrogenase

Requirement for Nitrogen reduction

Assimilation of Ammonia

4. Genetics of Nitrogen Fixation

Nif-genes of *Klebsiella Pneumoniae*

Regulation of Nif Genes

Nif-genes of *Azotobacter*

Nif-genes of *Anabaena*

Genetics of Legume-Rhizobium Nitrogen Fixation

1. Rhizobial Genes

a) Nod Genes

b) Nif Genes

c) Hup Genes

2. Legume Nodulin Genes

Leghaemoglobin Gene

Overall Regulation of Genes

Gene Transfer for Nitrogen Fixation

1. Transfer of Nif Genes to Non-Nitrogen Fixing Bacteria

2. Transfer of Nif Genes to yeasts

3. Transfer of Nif-Genes to plants

4. Transfer of Nod Genes

5. Transfer of Hup Genes

5. Mycorrhizae for Agriculture and Forestry

Mycorrhizal types and their structural and nutritional features

Ectomycorrhizae

Mechanism of ECM formation

Morphology and structure

Synthesis of mycorrhiza

Cultural study

Vesicular arbuscular Mycorrhiza

Introduction

Evolution

Taxonomy

Classification

Distribution

Lifecycle

Reproduction

Sexual reproduction

Asexual reproduction

Method of Inoculum production of VAM
Some important steps in production of VAM
Host plant/growth medium
Fertilizations/micronutrients
Chemical application
Control of fungal pathogens
Plant vesicular arbuscular mycorrhizal fungal interactions
VAM and soil biota
Control of root diseases
Endomycorrhiza fungi and tree diseases
Mechanism of disease control
6. Animal and plant cell cultures
Historical perspectives
Products and potentials
Animal cells
Immuno biologicals
1. Virus vaccines
2. Monoclonal antibodies
3. Immunoregulator materials
Insecticides
Enzymes
Hormones
Whole cells
Plant cells
Pharmaceuticals
Food additives
Agrochemicals
Perfumes
Enzymes
Speciality Chemicals
Biomass applications of plant cell cultures
Cell culture and product synthesis
The nature of animal and plant cells in culture
Cell culture initiation
Culture development
Secondary cultures
Culture replication
Industrially useful cell cultures
Substrate independent cultures
Individuality of cell lines in relation to the productivity
Culture media
Growth media
Water
Inorganic salts
Trace elements
Vitamins
Buffers
Sources of energy and carbon
Nitrogen sources
1. Defined nitrogen sources
2. Undefined nitrogen sources
Growth factors
Other ingredients

Maintenance media

Cell culture technologies

Cellular characteristics which influence the choice of cell culture technology

Mixing

Aeration

Doubling times

1. Sterilization of media

2. Sterilization of equipment

Cell stickiness

Immobilized cell systems

The growth and exploitation of cell grown on the surface of a supporting solid substratum

1. Multiple process

2. Unit process

The growth of animal and plant cells immobilized within a confining matrix

1. Gel entrapment systems

2. Applications of entrapped cells

Dynamic cell systems

Air driven systems

Impeller and air driven systems

Impeller mixed systems

7. Somaclonal variation, cell selection and genotype improvement

Somaclonal variation

Historical perspective

The manifold incidence of somaclonal variation

Range of species

Characters displaying variation

Genetic nature of somaclonal variants

Pre-existing or culture induced variation

Genetic and explant sources effects

The origin of somaclonal variation

Chromosomal abnormalities

Molecular possibilities

Gene amplification and diminution

Transposable elements

Cell selection

Disease resistance

Herbicide tolerance

Nutritional quality

Other cell selection systems

8. Virus-free clones through plant tissue culture

Distribution of viruses in plants

Techniques for eradication

Heat treatment

Chemotherapy

Meristem culture

Culture media

Factors affecting developments and rooting

Virus eradication

Major use of virus-free clones

Study effect of virus infection

Source for clonal propagation

Source for in vitro mass propagation

Concluding remarks

9. Microbial metabolism of carbon dioxide

Autotrophic carbon dioxide fixation

The calvin cycle

Molecular structure and properties of RuBP case

Phosphoribulokinase

Carboxysomes

Regulation of ribulose 1,5-biphosphate carboxydase and phosphoribulokinase synthesis

The reductive carboxylic acid cycle

The anaerobic non-phototrophic autotrophs

Heterotrophic carbon dioxide fixation

10. Microbial metabolism of Hydrogen

Introduction

The role of Hydrogen in the biosphere

Enzyme catalysing the evolution and oxidation of Hydrogen

H₂ :+ Ferredoxin Oxidoreductase

H₂ : Ferricytochrome C3 oxidoreductase

H₂ : NAD- Oxidoreductase

H₂ : Coenzyme F420 oxidoreductase

Membrane-bound hydrogenases

Formate hydrogenlyase

Nitrogenase

Organisms involved in the conversion of hydrogen

Hydrogen-producing micro-organisms

Anaerobic conditions

1. Fermentation and fermentative bacteria

2. Anoxygenic photosynthesis and phototrophic bacteria

3. Oxygenic Phototrophic bacteria (Cyanobacteria)

4. Oxygenic green algae

Aerobic conditions : Nitrogen fixing bacteria

Hydrogen consisting organisms

Hydrogen utilization by anaerobes

1. Nitrate-reducing denitrifying bacteria

2. Sulfate reducing bacteria

3. Methanogenic bacteria

4. Acetogenic bacteria

5. Furmarate-reducing bacteria

Hydrogen utilization by phototrophs

1. Anoxygenic phototrophs

2. Cyan bacteria

3. Green algae

Hydrogen utilization by aerobic hydrogen-oxidizing bacteria

The potential use of Hydrogenases and hydrogen in biotechnology

11. Microbial growth dynamics

Microbial growth in unlimited environments

Basic growth equation from cell number increase

Basic growth equation from increment increase in the population over a small growth time.

Basic growth equations.

Microbial growth in limited environments

Growth limitation by substrate exhaustion

Variation in the observed growth yield
Influence of the growth-limiting substrate on growth rate
Deviation of the Monod equation at High substrate concentrations
Basic growth limiting substrate equation
Modelling microbial growth in limited environments
The logistic equation
The saturation model
Microbial growth in open environments
Chemostat growth kinetics
The dilution rate
The dilution rate and biomass concentration
The dilution rate and growth limiting substrate concentration
Biomass and growth-limiting substrate concentrations in the steady state
Determination of $\hat{\mu}_{max}$ from washout kinetics
Establishing and maintaining the steady state
Deviations from theoretical chemostat kinetics
Influence of variation in the observed growth yield
Microbial competition
Competition in closed environments
Competition in open environments
12. Stoichiometry of microbial growth
Growth yields and material balances
Relation between ATP production and growth yields, YATP
Influence of growth rate and maintenance energy on YATP :
anaerobic chemostat cultures
Aerobic yield studies and the influence of the efficiency of
oxidative phosphorylation on growth yields
Theoretical calculations on the ATP requirements for the formation
of microbial biomass
Influence of Cell Composition
Influence of the carbon source and complexity of the medium
Theoretical calculations on the ATP requirement for the
formation of
microbial biomass
Influence of the Nitrogen source
Influence of the carbon assimilation pathway of the growth substrate
Energy-dissipating mechanisms during growth with excess
carbon and source.
Influence of the degree of reduction of the growth substrate
Heat production
The stoichiometry of product formation
13. Ageing and death in microbes
Basic principles
Death of microbes
Ageing of microbes
Viability among microbes
Survival of populations : Cryptic growth
Injury among microbes
Stress and survival
The physiological status of the population
Overt and actual stress
Starvation
Substrate accelerated death (SAD)

Metabolic and structural injury
Thymine less death
Survival of slowly growing bacteria
Differentiation and survival
14. Effect of environment on microbial activity
Mechanisms of micro-organisms response to the environment
Primary response due to direct chemical or physicochemical effects
Enzyme inhibition and stimulation
Induction and repression of protein synthesis
Changes in cell morphology
Change in genotype
Dissolved oxygen
Cell Interactions with oxygen
Respiration
Oxygen incorporation
Oxygen as an inhibitor
Oxygen as an enzyme regulator
Measurement of dissolved oxygen
Generalized response to DOT
Diffusion limitation
Response of growing micro-organisms
Respiration rate
Change in cell constituents
Changes in metabolic products
Transient responses to changes in DOT
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Redox potential
Responses to carbon dioxide
Requirement for carbon dioxide
Inhibition by carbon dioxide
Water activity
Introduction
Halotolerance and halophily
Effects of pH
Introduction
Cellular level responses
Intracellular pH
Effects of pH membrane function
Effects of pH on uptake of substrate
Effects of pH on products of metabolism
Effects of pH on cell morphology and structure
Effects of pH on the chemical environment
Effects of pH on flocculation and adhesion
Optimum pH values for growth
Causes of pH changes in cultures
Product formation
Nutrient uptake
Oxidation/reduction reaction
Change in buffering capacity
Control of pH
By means of a buffer
By balancing metabolism
By feedback control

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Cellular-level Responses
Temperature ranges for growth
Response of growth rate to temperature
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Effects of temperature on cellular components
1. Membranes
2. DNA
3. RNA
4. Proteins
Cultural effects of temperature
Response to temperature shifts
Effects on substrate utilization
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Generation of shear
Effects of shear on filamentous fungi
Effects of shear on protozoa and animal cells in culture
Effects of products on shear rate
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Relevance and importance of lipids
Lipid composition of micro-organism
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Yeasts
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Oleaginous micro-organism
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Factors influencing lipid biosynthesis
Growth rate
Substrate
Temperature
Growth substrate
Oxygen
pH and salinity
Other factors
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Acetyl- CoA carboxylase
Fatty acid synthetase
Origin of acetyl - CoA
Bacteria
Eukaryotic micro-organism
Biosynthesis of unsaturated fatty acids
Biosynthesis of other fatty acids
Biosynthesis of lipids from fatty acids
Triacylglycerols
Phospholipids
Waxes
Poly β -hydroxybutyrate
Microbial metabolism of alkanes and fatty acids

Alkane-utilizing organisms

Uptake of alkanes

Mechanisms of alkane oxidation

Oxidation of primary alcohols to fatty acids

Metabolism of fatty acids derived from alkanes

ÅŸ-oxidation

a-oxidation

Microbial products derived from alkanes

Fatty alcohols and aldehydes

Fatty acids

Surfactants

16. Microbial metabolism of aromatic compounds

Fission of the Benzene nucleus

Pereparation of nucleus for aerobic fission

Reactions which follow ring fission

Pathways of degradation

Meta fission pathways

Degradation of 4-hydroxyphenylacetic, homopropionic, homoglutamic

Homogentisic and gentisic acids

Proocatechuate 4,5 dioxygenase

Degradation of 3-O-Methylglucic acid: Biological formation of methanol

Ortho fission pathway

Separation of pathways used for aromatic catabolism by bacteria

Catabolism of aromatic compounds in *Trichosporon cutaneum*

Degradation of aromatic industrial pollutants and pesticides

Complete mineralization

Catabolic plasmids

Release of halogen substrates from benzene nucleus

Incomplete degradation of aromatics

17. Bacterial respiration

The generation of the proton motive force

Bacterial respiratory chains

Respiration linked proton translocation

The proton motive force

The utilization of the proton motive force

ATP synthesis

Active transport of solutes

Biotechnological aspects of bacterial respiration

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18. Mechanisms of enzyme catalysis

The events in an enzyme catalysed reaction

Enzyme mechanisms

Enzyme kinetics

Binding of the substrate to the enzyme

Conformational changes

Covalent bond making and breaking

Glucose isomerase

19. Enzyme evolution

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Feedback regulation
Limiting accumulation of end products
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Amino-acid production by genetically engineered strains of E-Coli and related organisms
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20. Microbial photosynthesis
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General characteristics of microbial photosynthesis
Structure and synthesis of photosynthetic pigments
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The eubacterial photosynthetic microbes
Introduction
The anoxygenic phototrophic bacteria
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The Cyano bacteria: oxygenic photosynthesis in a diverse prokaryotic group
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Alternative export mechanisms; post translocational secretion
Aspects of enzyme secretion in fungi
Regulation of Extracellular enzyme synthesis
Regulation of protein synthesis
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Catabolite repression
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22. Overproduction of microbial metabolites
Effects of nutrient limitation
Effects of pH and uncouplers of oxidative phosphorylation
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23. Regulation of metabolite synthesis
A phospholactase system in Klebsiella
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Regulatory mutations
Modular pathways
Evolution of an aliphatic amidase in pseudomonas
Evolution of a new β -Galactosidase in E-Coli
Properties of the wild-type proteins
Evolution of lactose utilization
Evolution of new activities for ebg enzymes
Evolution of the ebg repressor
Decryptifying Existing Genes

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