Biosynthetic pathways and Radio tracing techniques

Introduction to Biosynthetic Pathways

- Biosynthetic pathways are the natural chemical processes by which plants produce different phytochemicals.
- These pathways involve a series of enzyme-controlled reactions inside plant cells.
- In phytochemistry, biosynthetic pathways help us understand how medicinally important compounds are formed in plants.
- **Knowledge of biosynthesis is useful for:**
    - Identification of active constituents
    - Improvement of drug yield
    - Production of plant-based medicines
    - Industrial preparation of phytopharmaceuticals
    - Development of semisynthetic drugs

Importance in Herbal Industries

- Herbal industries use biosynthetic knowledge to increase the production of valuable compounds.
- Biosynthetic studies help in selecting plants with high medicinal value.
- They also help in tissue culture and metabolic engineering for large-scale production.
- Understanding pathways helps maintain quality control of herbal drugs.

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Radio Tracing Techniques

Definition

- Radio tracing techniques are methods used to study biosynthetic pathways using radioactive isotopes.
- In these techniques, radioactive atoms are introduced into precursor molecules.
- The movement of these labeled atoms is then followed inside the plant system.

Common Radioactive Isotopes Used

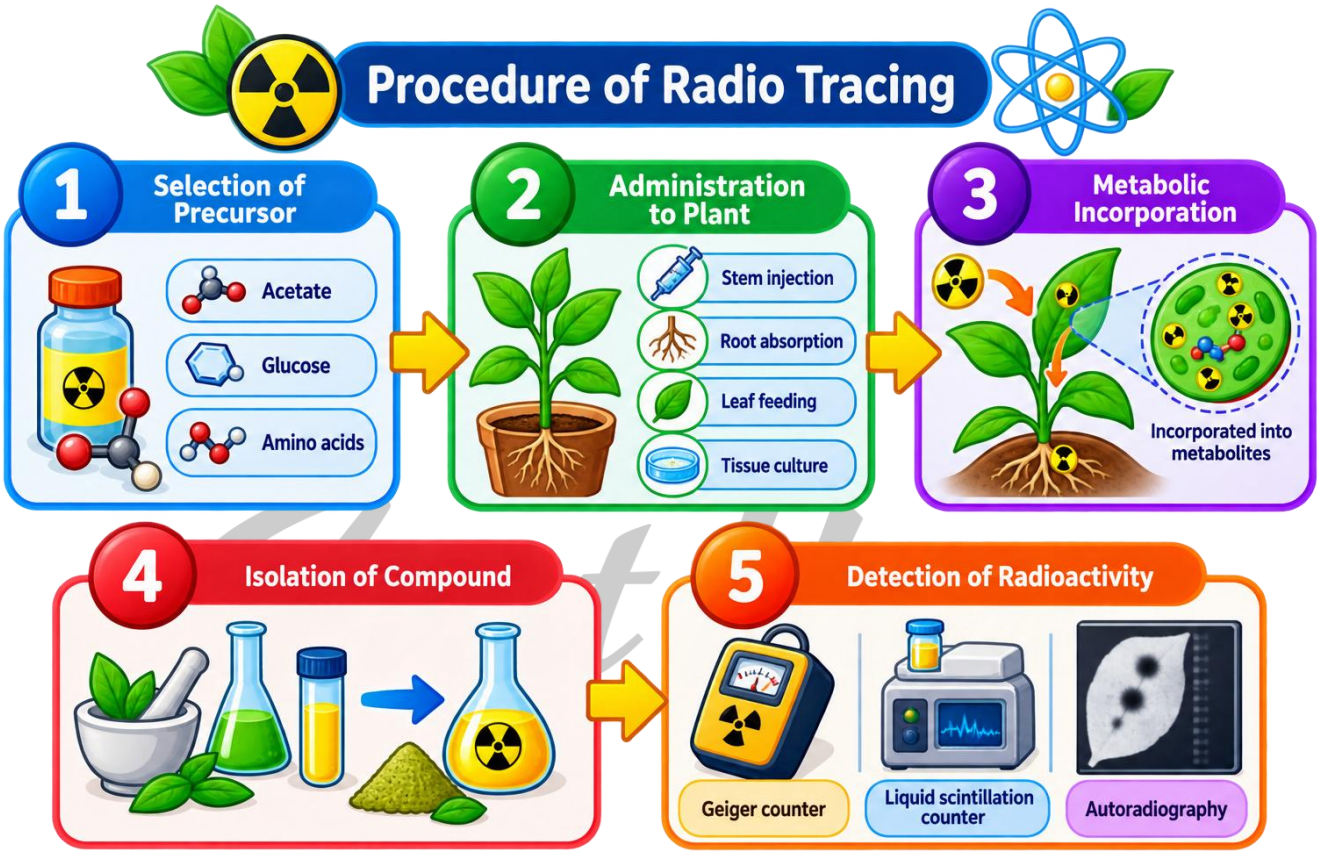
- Carbon-14 (^{14}C)
- Tritium (^{3}H)
- Phosphorus-32 (^{32}P)
- Sulfur-35 (^{35}S)

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Principle

- Plants absorb radioactive precursor compounds.
- During biosynthesis, the radioactive atoms become part of newly formed phytochemicals.
- Scientists detect the radioactive compounds using special instruments.
- This helps identify intermediate compounds and biosynthetic sequences.

Procedure of Radio Tracing



Step 1: Selection of Precursor

- A suitable precursor containing a radioactive isotope is selected.
- **Example:**
  - Radioactive acetate
  - Radioactive glucose
  - Radioactive amino acids

Step 2: Administration to Plant

- **The radioactive precursor is supplied to the plant through:**
  - Stem injection
  - Root absorption
  - Leaf feeding
  - Tissue culture medium

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Step 3: Metabolic Incorporation

- The plant metabolizes the precursor during normal biosynthesis.
- The radioactive atom becomes incorporated into secondary metabolites.

Step 4: Isolation of Compound

- The synthesized phytochemical is isolated from plant material.

Step 5: Detection of Radioactivity

- **Radioactivity is measured using:**
  - Geiger counter
  - Liquid scintillation counter
  - Autoradiography

Applications in Phytochemistry

- Identification of biosynthetic pathways
- Detection of precursor-product relationships
- Study of intermediate compounds
- Understanding enzyme mechanisms
- Improvement of medicinal compound production

Importance in Herbal Drug Industry

- Helps in increasing yield of active constituents
- Assists in development of improved plant varieties
- Useful in biotechnological production of herbal drugs
- Helps standardize medicinal plant products

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Alkaloids

Alkaloids

Definition

- Alkaloids are natural nitrogen-containing organic compounds mainly found in plants.
- They show strong physiological and medicinal effects on humans and animals.
- Most alkaloids are basic in nature due to nitrogen atoms.
- They are widely used in herbal and pharmaceutical industries.

General Characteristics

A. Physical Properties

- Mostly colorless crystalline solids
- Some are amorphous substances
- A few are liquids, like nicotine
- Usually have a bitter taste

B. Chemical Properties

- Generally alkaline in nature
- Free alkaloids are insoluble in water
- Alkaloidal salts are soluble in water
- Soluble in organic solvents like chloroform, ether, and alcohol

Biosynthesis of Alkaloids

- Mainly synthesized from amino acids such as ornithine, lysine, tyrosine, tryptophan, and phenylalanine
- Important reactions include decarboxylation, methylation, oxidation, and cyclization

Methods of Isolation



A. Acid–Base Extraction

- Plant material is dried, powdered, and treated with dilute acid

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- Alkaloids form water-soluble salts
- Alkali releases free alkaloids, which are extracted using organic solvents

B. Solvent Extraction

- Organic solvents dissolve alkaloids from plant material
- Extract is concentrated to obtain crude alkaloids

C. Chromatographic Methods

- TLC, Column Chromatography, and HPLC are commonly used
- Help in separation, purification, and quality control

Methods of Characterization

A. Chemical Tests

- **Dragendorff's Test** → orange precipitate
- **Mayer's Test** → cream precipitate
- **Wagner's Test** → reddish-brown precipitate

B. Spectroscopic Methods

- **UV** → conjugated systems
- **IR** → functional groups
- **NMR** → molecular structure
- **Mass Spectroscopy** → molecular weight

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Purification Methods

- **Recrystallization** gives pure crystals
- **Column Chromatography and HPLC** provide purified alkaloids

Importance in Herbal Industries

- Important active compounds in herbal medicines
- Used in pharmaceutical and nutraceutical products
- Increase the commercial value of medicinal plants
- Many alkaloids are valuable medicinal products for export

Ephedrine

Biological Source

- Ephedrine is obtained from the dried stems of Ephedra species.
- **Important species include:**

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