9

NUTRITION IN PLANTS – MINERAL NUTRITION

Sometimes you may observe that a potted plant kept in sunlight and provided with sufficient water does not grow. Its leaves look pale and weak. Plant may not even flower properly. Such a situation is an indication, that the plant may not be getting all that is required for normal growth and development.

In most of such situations one or more minerals required may be lacking in the soil. You might have seen farmers adding some extra manure (khad) to the soil. In this lesson you will learn the importance of mineral nutrition in plants.

OBJECTIVES

After completing this lesson, you will be able to :

- *define the terms mineral nutrition, macro and micro nutrients;*
- *explain the functions of minerals with reference to the techniques of hydroponics and aeroponics;*
- *list the role of macro and micro nutrients;*
- *mention the deficiency symptoms of macro and micro nutrients;*
- *differentiate between autotrophic and heterotrophic nutrition in plant;*
- *describe the saprophytic and parasitic modes of nutrition in plant.*

9.1 WHAT IS PLANT NUTRITION

As you know that all living organisms require food to survive, grow and reproduce so every organism takes in food and utilizes the food constituents for its requirements of growth. A series of processes are involved in the synthesis of food by plants, breaking down the food into simpler substances and utilization of these simpler substances for life processes. **Nutrition** in plants may thus be defined as a process of synthesis of food, its breakdown and utilisation for various functions in the body.

The chemical substances in food are called nutrients e.g. CO_2, water, minerals, carbohydrate, protein, fats etc. Green plants can make their own organic food from simple substances like water and carbon dioxide through the process of photosynthesis and are called autotrophs (auto : self; trophos : feeding). But the non-green plants and other organisms which cannot prepare their own food and obtain nutrition from green plants are called **heterotrophs** (heteros : different).

9.2 MINERAL NUTRITION

Now we will discuss how plants get the nutrients. You already know that carbohydrates are synthesised by the process of photosynthesis. What are the elements present in these carbohydrates?

Carbon, hydrogen and oxygen are the main elements in carbohydrates, fats and proteins. In addition to these three elements, plants need a variety of elements for their survival. These are generally referred to as mineral elements. They are absorbed by the root system of plants in the form of their salts.

The study of how plants get mineral elements and utilize them for their growth and development is called **mineral nutrition.**

If the minerals are not available to plants, specific symptoms appear due to the deficiency of a particular element. There are methods to determine the requirement of minerals by plants. Some such methods are given below.

9.3 METHODS TO DETERMINE THE REQUIREMENT OF MINERALS FOR PLANT

Minerals are absorbed by plants in **solution form.** So it is possible to grow plants in water containing the desired amount of mineral salts taking care that the aerial parts are exposed to air and light.

> This technique of growing plants in a nutrient solution in complete absence of soil is known as **Hydroponics/water culture.**
>
> It was demonstrated for the first time by a German Botanist Julius Von Sachs in the year 1980.

In water culture experiments, seedlings are made to grow in water containing the known nutrients in a particular proportion. Vigorous bubbling of the air is routinely done to provide sufficient oxygen to the root system. The culture solutions may contain all essential nutrients except the one whose importance is to be identified. Then the plant growing in it is compared to the one growing with all essential nutrients (**control experiment**).

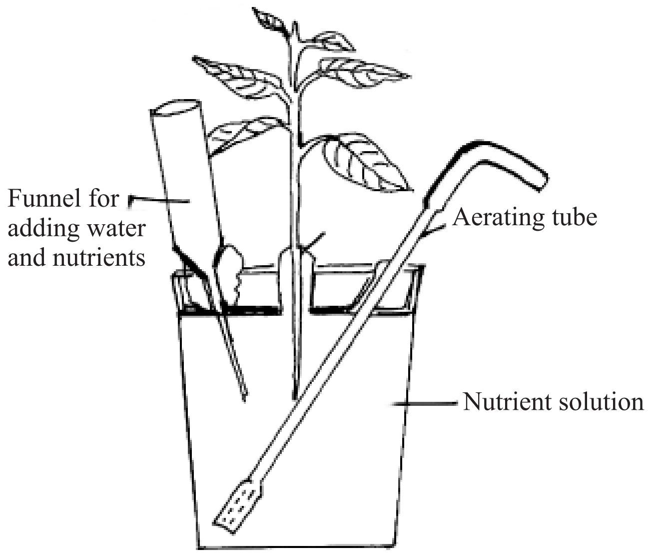


Fig. 9.1 Experimental set up for nutrient solution culture of plants.

Notes

Water culture experiments help us to understand :

(i) which element is essential for normal growth of the plant.

(ii) which element is not essential and is absorbed along with other nutrients.

(iii) how much quantity of each mineral is essential.

Hydroponics has been successfully employed for the commercial production of seedless cucumber, tomato and lettuce.

Aeroponics : Like hydroponics, aeroponics is another technique of growing plants in an air/mist environment without the use of soil.

Aeroponics is a technique of growing plants with their roots supplied with moisture present in the air. Rooted plants are placed in a special type of box. The shoots of the rooted plants are exposed to air and the roots are inside the box having computer controlled humid atmosphere. The roots are sprayed/misted for short durations with a hydro-atomized pure water/nutrient solution. This method has been developed recently. Since plants cultured by this technique get a very good growth of root hairs, it is very useful method for research purposes. Citrus plants and olives have been successfully grown through aeroponics.

INTEXT QUESTIONS 9.1

1. What are nutrients ?

   ....

2. Define aeroponics.

   ....

3. Why is it necessary to aerate nutrient solution in water culture?

   ....

9.4 ESSENTIAL MINERAL ELEMENTS

You know that 112 elements have been discovered until now. So you might be wondering whether plants require all 112 elements for their mineral nutrition. Most of the mineral elements present in soil are absorbed by roots of the plant. But all are not essential. Only **17 elements** are considered as essential for the plants. Let us now discuss the criteria for the essentiality of an element for normal plant growth.

9.4.1 Criteria for Essentiality of Elements

The nutrients or elements which are essential for the healthy growth of the plant are called **essential nutrients or essential elements.** The roots absorb about 60 elements from the soil. To determine which one is an essential element, the following criteria are used :

(i) An essential element is absolutely **necessary for normal growth** and **reproduction** of the plant, and should be a part of essential metabolite for plant growth.

(ii) The requirement of the element is very specific and it **cannot be replaced** by another element.

(iii) The element is **directly** or indirectly **involved** in the metabolism of a plant.

(iv) In the deficiency of an essential element, the plant would exhibit specific symptoms of deficiency, and the plant would recover from its symptoms, if supplied with the deficient element.

Example : Magnesium is said to be an essential element because it is essential for the formation of chlorophyll molecule. Its deficiency causes yellowing of leaves.

9.4.2 Types of Essential Elements

Essential elements may be required in small amounts or large amounts. Accordingly they have been grouped into two categories

Essential Elements

Micro elements/Micronutrients	Macro elements/Macro nutrients
Required in minute quantities like 0.1 mg per gram of dry matter or less than that. Also called as **trace elements.**	Required in relatively large quantities like one to 10 milligram per gram of dry matter
Examples : Manganese, Boron, cobatt Copper, Molybdenum, Iron, Zinc and Chlorine are required in very small quantities	**Examples :** Carbon, Hydrogen, Oxygen Phosphorous, Potassium, Calcium and magnesium, Nitrogen, Sulphur

9.4.3 Sources of Essential Elements for Plants

After studying the types of essential elements we will now discuss about their sources. Most of the essential elements are taken from soil, and some from the atmosphere. The table given below focuses on the sources of different essential elements.

Table 9.1 Sources of Essential Elements

Elements	Sources of the elements
Carbon	Taken as CO_2 from the atmosphere (air)
Oxygen	Absorbed in the molecular form from air or from water. It is also generated within a green plant during photosynthesis.
Hydrogen	Released from water during photosynthesis in the green plant
Nitrogen	Absorbed by the plants as nitrate ion (NO_3^-) or as ammonium ion (NH_4^+) from the soil. Some organisms like bacteria and cynobacteria can fix nitrogen from air directly.
Potassium, calcium iron, phosphorus, sulphur magnesium	absorbed from the soil (are actually derived from the weathering of rocks. So they are called mineral elements). They are absorbed in the ionic forms e.g. K^+, Ca^{2+}, Fe^{3+}, $H_2PO_4^-$/ HPO_4^{2-} etc.

INTEXT QUESTIONS 9.2

1. In which form do plants get oxygen?

   ....

2. Molybdenum is a micronutrient. Give reason.

   ....

3. Why are carbon, oxygen, potassium and sulphur called macronutrients?

   ....

9.5 ROLE OF MACRO AND MICRO NUTRIENTS

Essential elements perform various functions. They carry out several metabolic processes in the plant cells like the maintenance of turgidity of cell, transportation of electrons, membrane permeability and enzyme activity. Essential elements also act as important constituents of the biomolecules and co-enzymes. Various functions of the macro and micro nutrients are given in the following table.

The forms in which the elements are taken in and their functions are described in the table given below -

Table 9.2 Essential Elements and their Functions

Element	Form in which the element is taken in	Region of the plant that requires the element	Function
Nitrogen, N	NO_2^-, NO_3^- or NH_4^+ ions	All tissues, particularly in meristematic tissues	Required for the synthesis of amino acids, proteins nucleic acids, vitamins, hormones, coenzymes, ATP and chlorophyll.
Phosphorus, P	$H_2PO_4^-$ or HPO_4^{2-}	Young tissues from the older metabolically less active cells	Required for the synthesis of nucleic acids phospho-lipids, ATP, NAD and NADP. Constituent of cell membrane and some proteins.
Potassium, K	K^+	Meristematic tissues buds, leaves and root tips.	Activates enzymes, associated with K^+/Na^+ pump in active transport, anion-cation balance in the cells. Brings about opening and closing of stomata. Common in cell sap in plant cell vacuole and helps in turgidity of cells.
Calcium, Ca	Ca^{2+}	Meristematic and differentiating tissues Accumulates in older leaves	Present as calcium pectate in the middle lamella of cell walls that joins the adjacent cells together. Activates enzymes needed for the growth of root and shoot tip. Needed for normal cell wall development. Required for cell division, cell enlargement.
Magnesium, Mg	Mg^{2+}	Leaves of the plant	Forms part of the chlorophyll molecule. Activates enzymes of phosphate metabolism. Important for synthesis of DNA and RNA. Essential for binding of ribosome subunits.
Sulphur, S	SO_4^{2-}	Stem and root tips young leaves of the plant	As a constituent of amino acids cysteine and methionine and of some proteins. Present in co-enzyme A, vitamin thiamine, biotin and ferredoxin. Increases root development. Increases the nodule formation in legumes.

Iron, Fe	Fe^{3+}	Leaves and seeds	Needed for the synthesis of chlorophyll. As a constituent of ferredoxin and cytochromes. Activates the enzymes catalase.
Manganese Mn	Mn^{2+}	All tissues. Collects along the leaf veins.	Activates many enzymes of photosynthesis, respiration and N_2 metabolism. Acts as electron donor for chlorophyll b. Involved in decarboxylation reactions during respiration.
Molybdenum Mo	MoO_2^{2+}	All tissues particularly in roots	Required for nitrogen fixation. Activates the enzyme nitrate reductase.
Boron, B	BO_3^{3-} or $B_4O_7^{2-}$	Leaves and seeds	Increases the uptake of water and calcium. Essential for meristem activity and growth of pollen tube. Involved in translocation of carbohydrates
Copper, Cu	Cu^{2+}	All tissues	Component of oxidase enzymes and plastocyanin. Involved in electron transport in photosynthesis.
Zinc, Zn	Zn^{2+}	All tissues	Component of indoleacetic acid – a plant hormone. Activates dehydrogenases and carboxylases. Present in enzyme carbonic anhydrase
Chlorine, Cl	Cl^-	All tissues	Essential for oxygen evolution in photosynthesis. Anion-cation balance in cells.

INTEXT QUESTIONS 9.3

1. State any two metabolic processes for which mineral nutrition is required.

   ....

2. Which element is provided by NO_2 and NH_4 when taken up by plants?

   ....

3. State any two functions of Ca^{2+} in plants?

   ....

9.6 SYMPTOMS OF MINERAL DEFICIENCY IN PLANTS

The absence or deficiency (not present in the required amount) of any of the essential elements leads to **deficiency symptoms.** The symptoms can be studied by hydroponics. Under natural conditions, these symptoms can be taken as indicators of the mineral deficiencies in the soil.

Some common deficiency symptoms are :

- **Chlorosis** - It is the loss of chlorophyll leading to yellowing in leaves. It is caused by the deficiency of elements like K, Mg, N, S, Fe, Mn, Zn and Mo.
- **Necrosis** or death of tissues, particularly leaf tissue is caused by deficiency of K, Ca, Mg
- **Inhibition of cell division** is caused due to lack or deficiency of N, K, B, S and Mo.
- Stunted/Retarded plant growth caused by the deficiency of N, P, K, Zn, Ca
- Premature fall of leaves and buds is caused by deficiency of K, P.
- Delay in flowering is caused due to deficiency of N, S, Mo.

9.7 UPTAKE OF MINERAL ELEMENTS

Plants absorb a large number of minerals from soil. The uptake of mineral ions by the roots may be **passive** or **active**.

(a) **Passive Absorption** : It is the initial and rapid phase wherein ions are absorbed into the "outer space" of the cells, through the apoplast (Recall from lesson No. 08) pathway. It does not require use of any metabolic energy.

(b) **Active Absorption :** It is the second phase of ion uptake. The ions are taken in slowly into the 'inner space' the symplast of cells (Lesson No. 08). It needs the expenditure of metabolic energy.

The movement of ions is called **flux.** When the ions move into the cells, it is called **influx** and the outward movement of ions is called **efflux**.

The mineral ions absorbed by the root system are translocated through the xylem vessels to other parts of the plant.

INTEXT QUESTIONS 9.4

1. What is meant by 'passive absorption' of minerals by plants.

   ....

2. Name the minerals whose deficiency affects normal cell division.

   ....

3. "Deficiency of K, Ca and Mg causes necrosis of leaves". What does this statement mean ?

   ....

9.8 MODE OF NUTRITION IN PLANTS

Nutrition is plants is classified into two main categories : autotrophic and heterotrophic. Heterotrophic plants are further classified into saprophytes, parasite and insectivores.

Nutrition in Plants

- Autotrophic Nutrition
  (Auto = self; trophos = feeder)
- Heterotrophic Nutrition
  (Hetero = other; trophos = feeder
  Heterotrophic plant)
  - Saprophytic plants/
    Saprophytes
    (Sapros = rotten;
    Phyton = plant
  - Parasitic plants/Parasites
    (Para = beside; sites = food)
  - Insectivorous plants/
    Insectivores
    (eat insects)

1. Autotrophic Nutrition

It is a type of nutrition in which the living organisms manufacture their own organic food from simple inorganic raw materials. The green plants exhibit autotrophic mode of nutrition and hence called the autotrophs. The autotrophs require external energy source for the manufacture of organic substances. Green plants obtain energy from sunlight and therefore are called **photoautotrophs**. The process of synthesizing food in plant in the presence of sunlight is called **photosynthesis**. The insectivores are autotrophic but they develop specific structures to trap insects to overcome N_2 deficiency because they grow in soils having acute N_2-deficiency.

2. Heterotrophic nutrition

Certain non green organisms like fungi and many bacteria fail to synthesize their own organic nutrients from inorganic substances. These organisms are thus dependent on some other external sources for their organic nutrition. Such plants are called **heterotrophic plants** and the mode of nutrition is called **heterotrophic nutrition.**

The heterotrophic plants are broadly categorised into two main groups depending upon the source from which they get their nourishment. Saprophytes, and parasites.

(a) **Saprophytes** are those plants which grow and live on dead organic matter including animal and plant remains. Most of these plants secrete some extracellular enzymes (enzymes secreted and poured out on food) which break down the complex organic compounds into simple forms. The simple form are then absorbed by the plants. Saprophytes include mainly fungi and bacteria. Also among higher plants the Indian pipe plant *Monotropa* found in khasi hills of our country (Fig. 9.2) is a saprophyte.

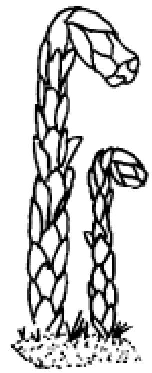


Fig. 9.2 A Monotropa, a saprophyte.

(b) **Parasitic Plants** : *Dodder* (*Cuscuta*) known locally as Amarbel/Akashbel is a parasitic plant that lacks both chlorophyll and leaves. It is a yellow colour climber that attaches itself to the host. It gives out haustoria or the suckers that get attached to the phloem of the host and derive nutrition. *Cuscuta* does not have roots in the mature condition. It produces bunches of whitish or yellowish bell shaped flowers.

Insectivorous Plants : These are plants which are autotrophic but develop adaptations to trap insects in order to **supplement the deficiency of Nitrogen in the soil**. They feed on insects. They are generally found in nitrogen deficient habitats and hence to compensate the loss, they use insects as a source of nitrogen. Some examples are given below :

(i) Pitcher plant : *Nepenthes* (ii) Sundew : *Drosera*

(iii) Venus flytrap : *Dionaea* (iv) Bladderwort : *Utricularia*

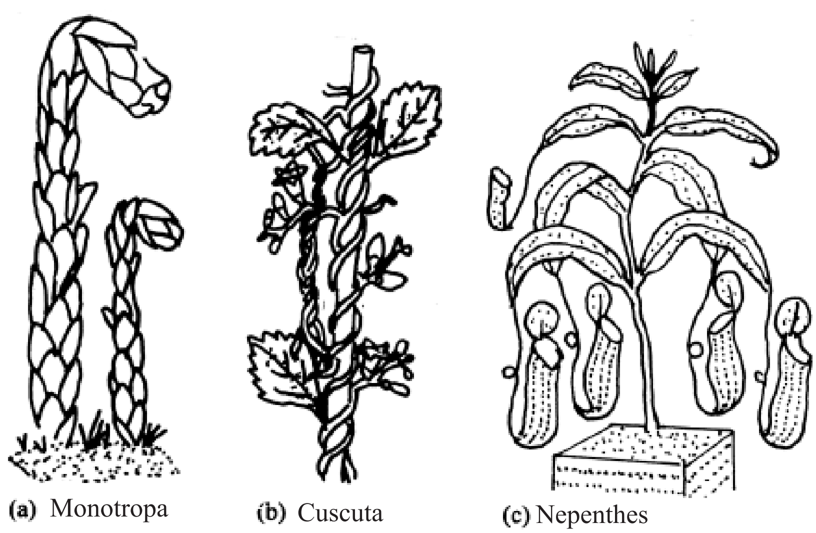


Fig. 9.3 Heterotrophic plants : (a) *Monotropa* (Indian pipe plant (b) *Cuscuta* (dodder) and (c) *Nepenthes* (pitcher plant)

Pitcher plant (Nepenthes) : It is found in north eastern India, Borneo and in many regions of North America.

These plants grow well in wet soils. The leaves are modified in the form of pitchers. The pitcher has nectar producing glands below its rim. Shiny surface of the pitcher and nectar secreted by nectar glands attract the insects. Insects once trapped can not escape due to the presence of numerous downward pointing hairs in the pitcher. The digestive glands present at the base of pitcher secrete enzymes. The insects are digested by the enzymes and the products which are mainly aminoacids are absorbed by the inner surface of leaves (pitcher).

INTEXT QUESTIONS 9.5

1. Give one point of difference between autotrophic and heterotrophic nutrition.

   ....

2. Name a plant which exhibits parasitic mode of nutrition.

   ....

3. Why does pitcher plant eat insects when it is capable of carrying out photosynthesis?

   ....

WHAT YOU HAVE LEARNT

- Plants have the nutritional requirement of various inorganic and organic raw materials for building their structure and maintaining body functions.
- Nutrition is the sum total of processes involving intake or synthesis of food and its utilisation.
- Plants generally derive their inorganic nutrients from soil, water and atmosphere.
- The absorption, distribution and metabolism of various mineral elements by plants is called mineral nutrition.
- Plants require 17 essential elements. They are C, H, O, N, P, K, S, Mg, Ca, Fe, B, Mn, Cu, Zn, Mo, Cl and Co.
- The essentiality of minerals may be determined by employing the technique of hydroponics and aeroponics.
- Inorganic nutrients are broadly classified into two categories-micronutrients and macronutrients on the basis of the amount required by plant.
- Absence of any one element may cause deficiency symptoms in plants. These symptoms include reduction in growth, delaying of flowering, chlorosis, necrosis, early leaf fall, wilting etc.

- The minerals are taken by the roots through passive or active absorption.
- Basically, there are two modes of nutritioin autotrophic and heterotrophic.
- In autotrophic nutrition the organisms (plants) manufacture their own food from inorganic raw materials by photosynthesis or chemosynthesis.
- In heterotrophic nutrition the organism is dependent on other external sources for its organic nutrition.
- Heterotrophic plants are broadly categorised into two main groups: saprophytes, and parasites.
- Insectivorous plants are special type of autotrophic plants which grow in N_2-deficient soils and develop adaptations to trap insects to overcome N_2-deficiency.

TERMINAL EXERCISES

1. Which element can be obtained from both mineral and non-mineral sources.
2. Deficiency of which essential element causes yellowing of leaves in certain plants and why?
3. Why is magnesium included among essential elements?
4. What are the criteria of essentiality of elements?
5. Differentiate between micro and macro nutrients.
6. Why do biologists grow plants by hydroponics technique?
7. Explain the uptake of mineral nutrients by the plants.
8. Give the deficiency symptoms of nitrogen, phosphorus and potassium.
9. Differentiate between the different modes of heterotrophic nutrition in plants.
10. Write notes on :
    (i) Aeroponics
    (ii) Insectivorous plants
    (iii) Active absorption of minerals by plants

ANSWERS TO INTEXT QUESTIONS

9.1 1. Nutrients are the chemical substances in food

2. A technique of growing plants with roots supplied with moisture present in the atmosphere.

3. To supply oxygen in sufficient quantity

9.2 1. Molecular form from air or water

2. Required by plant in very small quantity. 0.1 mg per gram of dry matter or less.

3. They are required in large quantities 1-10 mg per gram of dry matter.

9.3 1 Membrane permeability, turgidity of cell, transport of electrons, enzyme activity (any two)

2. Nitrogen

3. See table 9.2

9.4 1. Without expenditure of energy

2. N, K, S, Mo (any two)

3. The deficiency causes death of leaf tissues

9.5 1. Autotrophs synthesize their own food, heterotrophs depend on others for food

2. *Cuscuta* (dodder)

3. Because it grows in a nitrogen deficient habitat.

10

NITROGEN METABOLISM

All the living organisms are basically composed of carbon, hydrogen, oxygen, nitrogen and many other forms of chemical elements. These elements contribute to finally organize various biomolecules present in a cell. Nitrogen is next to carbon in importance in living organisms. In a living cell, nitrogen is an important constituent of amino acids, proteins, enzymes, vitamins, alkaloids and some growth hormones. Therefore, study of nitrogen metabolism is absolutely essential because the entire life process is dependent on these nitrogen-containing molecules. In this lesson, you will learn about various aspects of nitrogen metabolism including nitrogen fixation and nitrogen assimilation in plants.

OBJECTIVES

After completing this lesson, you will be able to:

- *describe the modes of nitrogen fixation (both biological and abiological);*
- *explain the steps involved in nitrogen fixation by free living organisms;*
- *explain the mode of symbiotic nitrogen fixation in leguminous plants;*
- *describe the assimilation of nitrate and ammonia by plants;*
- *describe amino acid synthesis in plants.*

10.1 MOLECULAR NITROGEN

Nitrogen is primarily present in the atmosphere freely as dinitrogen or nitrogen gas. It is present in the combined form as Chile saltpetre or sodium nitrate and Chile in South America is the major source of this nitrate nitrogen.

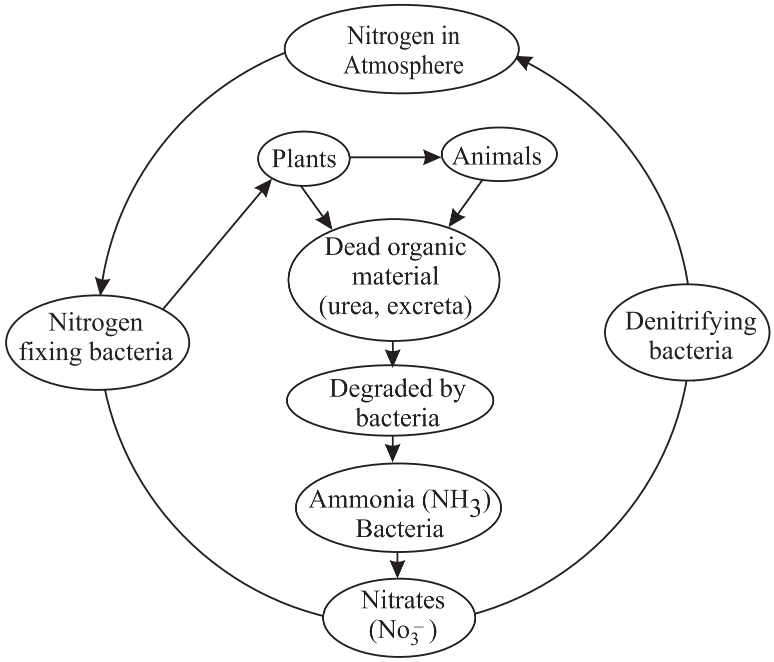
Molecular Nitrogen or diatomic nitrogen (N_2) is highly stable as it is triple bonded ($N\equiv N$). Because of this stability, molecular nitrogen as such is not very reactive in the atmosphere under normal conditions. In the atmosphere molecular nitrogen is 78.03% by volume and it has a very low boiling point (-195.8^{o}C) which is even lower than that of oxygen. Proteins present in living organisms contain about 16% nitrogen.

10.1.1 Nitrogen Cycle

Nitrogen is an essential constituent of living beings. Nitrogenous bases are part of nucleic acids and proteins are made up of amino acids of which Nitrogen is an important consituent. You already know about the importance of these two biomolecules.

Air has 78% N_2 but most of the living beings cannot utilize this atmospheric Nitrogen. Nitrogen cycle converts this nitrogen into a usable form. Lightning fixes Nitrogen to NH_3, and nitrogen fixing bacteria like *Rhizobium* (which live in roots of leguminous plants like pea, rajma, beans, pulses etc.) also convert N_2 into NH_3. Most plants absorb nitrates from soil and reduce it to NH_3 in the cells for further metabolic reactions. Dead organisms and their excreta like urea are decomposed by bacteria into NH_3 and by a different set of bacteria into nitrates. These are left in the soil for use by plants. In this way Nitrogen cycle is self regulated but human activities have caused steady loss of soil Nitrogen.

NITROGEN CYCLE



INTEXT QUESTIONS 10.1

1. What is the percent by volume of nitrogen gas in the atmosphere?

   ....

2. Name two biomolecules that contain nitrogen in plants.

   ....

3. Why nitrogen is a stable molecule?

   ....

4. What is the percentage of nitrogen in protein?

   ....

5. What is the boiling point of nitrogen?

   ....

6. Choose the correct option:

   Nitrogen fixation is the conversion of :

   (a) atmospheric Nitrogen $\xrightarrow{\text{into}}$ Nitrates

   (b) atmospheric Nitrogen $\xrightarrow{\text{into}}$ Ammonia

   (c) atmospheric Nitrogen $\xrightarrow{\text{into}}$ Amino acids

   (d) both (a) and (b)

7. Nitrogen content of biosphere remains constant because of :

   (a) Nitrogen cycle

   (b) Nitrogen fixation

   (c) Industrial pollution

   (d) Absorption of nitrogen

8. Nitrates are converted into nitrogen by microbes called ....

10.2 NITROGEN FIXATION (BIOLOGICAL AND ABIOLOGICAL)

The conversion of molecular nitrogen into compounds of nitrogen especially ammonia is called **nitrogen fixation**. Nitrogen fixation, is a reductive process i.e., nitrogen fixation will stop if there is no reducing condition or if oxygen is present. This nitrogen fixation may take place by two different methods – abiological and biological.

10.2.1 Abiological nitrogen fixation

In abiological nitrogen fixation the nitrogen is reduced to ammonia without involving any living cell. Abiological fixation can be of two types : industrial and natural. For example, in the Haber's process, synthetic ammonia is produced by passing a mixture of nitrogen and hydrogen through a bed of catalyst (iron oxides) at a very high temperature and pressure.

$$N_2 + 3H_2 \xrightarrow[\text{1000 atmosphere}]{500°\text{C}} 2NH_3$$

This is industrial fixation wherein nitrogen gets reduced to ammonia.

In natural process nitrogen can be fixed especially during electrical discharges in the atmosphere. It may occur during lightning storms when nitrogen in the atmosphere can combine with oxygen to form oxides of nitrogen

$$N_2 + O_2 \longrightarrow 2NO \xrightarrow{O_2} 2NO_2$$

These oxides of nitrogen may be hydrated and trickle down to earth as combined nitrite and nitrate.

10.2.2 Biological nitrogen fixation

Chemically, this process is same as abiological. Biological nitrogen fixation is reduction of molecular nitrogen to ammonia by a living cell in the presence of enzymes called nitrogenases.

INTEXT QUESTIONS 10.2

1. Define nitrogen fixation.

   ....

2. Which industrial process is utilized for converting nitrogen to ammonia?

   ....

3. Distinguish between biological and abiological nitrogen fixation.

   ....

4. Name the enzyme that helps in nitrogen fixation in living cells.

   ....

5. Which gas prevents nitrogen fixation?

   ....

10.3 NITROGEN FIXATION BY FREE LIVING ORGANISMS AND SYMBIOTIC NITROGEN FIXATION

Nitrogen fixation is a distinctive property possessed by a select group of organisms, because of the presence of the enzyme nitrogenase in them.

The process of nitrogen fixation is primarily confined to microbial cells like bacteria and cyanobacteria. These microorganisms may be independent and free living (Table 10.1).

Table 10.1 : Some free living microbes which fix nitrogen

Organisms	Status
Clostridium	Anaerobic bacteria (Non-photosynthetic)
Klebsiella	Facultative bacteria (Non-photosynthetic)
Azotobacter	Aerobic bacteria (Non-photosynthetic)
Rhodospirillum	Purple, non-sulphur bacteria (Photosynthetic)
Anabaena	Cyanobacteria (Photosynthetic)

Some microbes may become associated with other oragnisms and fix nitrogen. The host organism may be a lower plant or higher plant. The host organism and the nitrogen fixing microbes establish a special relationship called **symbiosis** and this results in symbiotic nitrogen fixation (Table 10.2).

Table 10.2 : Some symbiotic nitrogen fixing organisms

System	Symbionts
Lichens	Cyanobacteria and Fungus.
Bryophyte	Cyanobacteria and *Anthoceros*.
Pteridophyte	Cyanobacteria and *Azolla*.
Gymnosperm	Cyanobacteria and *Cycas*.
Angiosperms	Legumes and *Rhizobium*.
Angiosperms	Non leguminous plants and actinomycete (Such as *Alnus*, *Myrica*, *Purshia*).
Angiosperm	Brazilian grass (*Digitaria*), Corn and *Azospirillum*.

10.3.1 Mechanism of Biological Fixation of Nitrogen

Nitrogen fixation requires

(i) the molecular nitrogen

(ii) a strong reducing power to reduce nitrogen like reduced FAD (Flavin adenine dinucleotide) and reduced NAD (Nicotinamide Adenine Dinucleotide)

(iii) a source of energy (ATP) to transfer hydrogen atoms from $NADH_2$ or $FADH_2$ to dinitrogen and

(iv) enzyme nitrogenase

(v) compound for trapping the ammonia formed since it is toxic to cells.

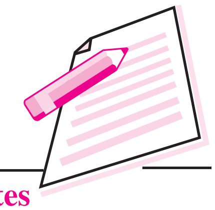
The reducing agent ($NADH_2$ and $FADH_2$) and ATP are provided by photosynthesis and respiration.

The overall **biochemical process** involves stepwise reduction of nitrogen to ammonia. The enzyme nitrogenase is a Mo-Fe containing protein and binds with molecule of nitrogen (N_2) at its binding site. This molecule of nitrogen is then acted upon by hydrogen (from the reduced coenzymes) and reduced in a stepwise manner. It first produces diamide (N_2H_2) then hydrazime (N_2H_4) and finally ammonia ($2NH_3$).

NH_3 is not liberated by the nitrogen fixers. It is toxic to the cells and therefore these fixers combine NH_3 with organic acids in the cell and form amino acids.

The general equation for nitrogen fixation may be described as follows:

$$N_2 + 16ATP + 8H^+ + 8e^- \longrightarrow 2NH_3 + 16ADP + 16Pi +$$



Molecular nitrogen is a very stable molecule. Therefore, sufficient amount of cell energy in the form of ATP is required for stepwise reduction of nitrogen to ammonia.

In legumes, nitrogen fixation occurs in specialized bodies called **root nodules**. The nodules develop due to interaction between the bacteria *Rhizobium* and the legume roots (see diagram 6.4c). The biochemical steps for nitrogen fixation are same. However, legume nodules possess special protein called LEGHEMOGLOBIN. The synthesis of leghemoglobin is the result of symbiosis because neither bacteria alone nor legume plant alone possess the protein. Recently it has been shown that a number of host genes are involved to achieve this. In addition to leghemoglobin, a group of proteins called **nodulins** are also synthesized which help in establishing symbiosis and maintaining nodule functioning.

Leghemoglobin is produced as a result of interaction between the bacterium and legume roots. Apparently, Rhizobium gene codes for Heme part and legume root cell gene codes for Globin moiety. Both the coded products together consitute the final protein leghemoglobin. During N_2-fixaion, function of Leghemoglobin is to act as Oxygen-scavenger so that the enzymes, Nitrogenages then, convert N_2 to NH_3 under anaerobic condition.

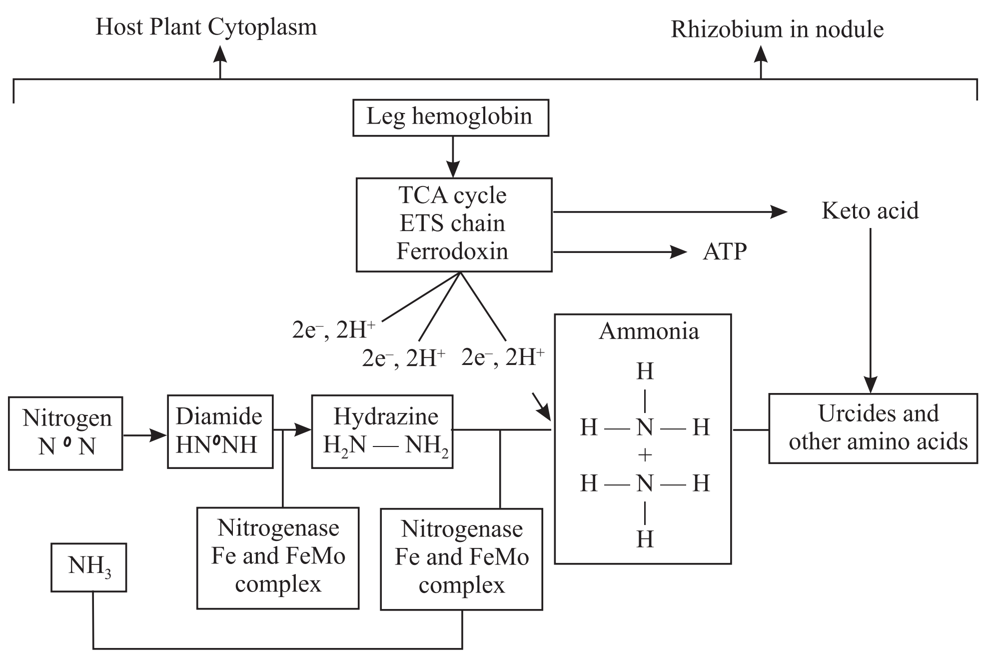


Fig. 10.1 Simplified flowsheet of biochemical steps for nitrogen fixation

Leghemoglobin is considered to lower down the partial pressure of oxygen and helps in nitrogen fixation. However, this function is specific for legumes only because free living microbes do not possess nitrogen fixing leghemoglobin. Moreover, it has also not been found in cyanobacterial symbiosis with other plants, which fix N_2 under aerobic condition.

INTEXT QUESTIONS 10.3

1. Match the following:

	A		B
(i)	*Azotobacter*	(a)	anaerobic nitrogen fixer.
(ii)	*Clostridium*	(b)	aerobic nitrogen fixer
(iii)	*Lichens*	(c)	aerobic nitrogen fixing cyanobacterium
(iv)	*Anabaena*	(d)	symbiotic nitrogen fixer.

2. Which Gymnospermous plant fixes nitrogen?

....

3. Is there any other gas evovled during nitrogen fixation? If yes, name the gas evolved.

....

4. How many ATP molecules are required to reduce a single molecule of nitrogen?

....

5. What is the major source of electrons for reduction of nitrogen?

....

6. Match the following:

	A		B
(i)	Leghemoglobin	(a)	cyanobacterium
(ii)	*Anabaena*	(b)	Legumes
(iii)	Reductive process	(c)	nitrogen fixation

7. Name the proteins that help in establishing symbiosis and maintain root nodule functioning in legumes.

....

10.4 NITRATE AND AMMONIA ASSIMILATION BY PLANTS

As pointed out in the previous section, nitrogen fixation is confined to selected microbes and plants. But all plants require nitrogen because it has a role to play in the general metabolism. Therefore, plants which do not fix nitrogen, use other combined nitrogen sources such as nitrate and ammonia for carrying on metabolic activity.

Nitrate is absorbed by most plants and reduced to ammonia with the help of two different enzymes. The first step conversion of nitrate to nitrite is catalyzed by an enzyme called nitrate reductase. This enzyme has several other important constituents including FAD, cytochrome, NADPH or NADH and molybdenum.

$$NO_3^- + NADH + H^+ \xrightarrow{\text{Nitrate reductase}} NO_2^- + NAD^+ + H_2O$$

The overall process of nitrate reduction take place in the cytosol and is an energy dependent reaction.

The enzyme nitrate reductase has been studied in many plants and it is observed that the enzyme is continuously synthesized and degraded. The enzyme nitrate reductase is inducible. This means that increase in nitrate concentration in the cytosol induces more of nitrate redutase to be synthesized. However, when excess NH_4^+ is produced then it has a negative effect on the synthesis of nitrate reductase. In plants, it has also been observed that light also increases nitrate reductase when nitrate is available.

In the second step the nitrite so formed is further reduced to ammonia and this is catalyzed by the enzyme nitrite reductase. Nitrite present in the cytosol is transported into chloroplast or plastids where it is reduced to ammonia.

$$NO_2^- + 3NADPH + 3H^+ \xrightarrow{\text{Nitrite reductase}} NH_3 + 3NADP^+$$

The enzyme nitrite reductase is able to accept electrons from sources such as NADH, NADPH or $FADH_2$. Besides, reduced ferredoxin has also been shown to provide electrons to nitrite reductase for reducing nitrite to ammonia. Ammonia so formed has to be utilized quickly by plants because accumulation of ammonia has a toxic effect. Some plants including algae leach out excess ammonia which can further be oxidized to nitrite and nitrate by microorganisms in the soil or water.

INTEXT QUESTIONS 10.4

1. Which is the most reduced form of inorganic nitrogen?

   ....

2. Match the following:

A		B
(*i*) Nitrate reductase		(a) nitrogen fixation
(*ii*) Nitrite reductase		b) nitrate reduction
(*iii*) Nitrogenase		c) nitrite reduction

3. In which part of the cell, reduction of nitrate to nitrite occurs?

   ....

4 Which is the most oxidized form of inorganic nitrogen?

   ....

5 In which plant organelle reduction of nitrite to ammonia is catlyzed by the enzyme nitrite reductase?

   ....

10.5 AMINO ACID SYNTHESIS BY PLANTS

As you have noticed that ammonia formation is achieved by plants either by (i.) nitrogen fixation or (ii) by reduction of nitrate to nitrite. Ammonium (NH_4^+) is the most reduced form of inorganic combined nitrogen. This ammonium now becomes the major source for the production of amino acids, which are the building blocks of enzymes and proteins. Amino acids have two important chemical groups. (i) amino group (NH_2) and (ii) carboxy1 group (-COOH).

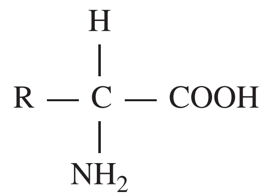


Fig. 10.2 A typical amino acid with functional groups. R represents alkyl group.

Ammonium so produced is the major source of amino group. However, the carboxyl group has to be provided by other organic molecule synthesized by the plants. There are two major reactions for amino acid biosynthesis in plants:

10.5.1 Reductive amination reaction:

In this reaction, ammonia combines with a keto acid. The most important keto acid is the alpha ketoglutaric acid produced during the operation of Krebs cycle (see lesson 12 Plant Respiration). The keto acid then undergoes enzymatic reductive amination to produce an amino acid.

$$\underset{\text{(keto acid)}}{\alpha\text{-ketoglutaric acid}} + NH_3 \xrightarrow{\text{glutamate dehydrogenase}} \underset{\text{(amino acid)}}{\text{Glutamic}}$$

Similarly another amino acid called aspartic acid is produced by reductive amination of oxaloacetic acid.

> It has been noted that reductive amination respresents the major 'port of entry' for ammonia into the metabolic stream in plants. This initiates synthesis of glutamic acid followed by other amino acids.

10.5.2 Transamination reaction

This is another very important reaction for amino acid biosynthesis. The reaction involves transfer of amino group, from already synthesized amino acid, to the keto acid.

$$\underset{\text{(Keto acid)}}{\alpha\text{-Ketoglutaric acid}} + \underset{\text{(Amino acid)}}{\text{Aspartic acid}} \xrightarrow{\text{Transaminase}} \underset{\text{(Amino acid)}}{\text{Glutamic acid}} + \underset{\text{(Keto acid)}}{\text{Oxaloacetic acid}}$$

In the above reaction, aspartic acid has transferred its amino group (NH_2) to the α-ketoglutaric acid to synthesize glutamic acid and release keto acid. The reaction is catalyzed by enzymes called **transaminases**. A large number of amino acids are synthesized by this transamination reaction. Amino acids are organic molecules containing nitorgen. The incorporation of amino group, from ammonium, into keto acids represents the major step for synthesis of nitrogenous organic biomolecules.

INTEXT QUESTIONS 10.5

1. Match the following:

	A	B
(i)	Amino acid	(a) keto acid
(ii)	Glutamic acid	(b) amino group and carboxyl group
(iii)	α-ketoglutaric acid	(c) amino acid

2. Name two biochemical reactions for biosynthesis of amino acids in plants.

   ....

3. Which group of enzymes catalyzes transamination reaction?

   ....

4. What is the source of amino group for amino acid synthesis in reductive amination reaction?

   ....

5. Which keto acid is the source for synthesis of glutamic acid?

   ....

WHAT YOU HAVE LEARNT

- Nitrogen is an important constituent of several biomolecules such as amino acids, proteins and enzymes.
- Molecules such as vitamins, alkaloids, nucleic acids, pigments and some growth hormones also contain nitrogen.
- Molecular nitrogen is triple bonded and stable.
- Nitrogen fixation is the reduction of nitrogen to ammonia.
- Abiological nitrogen fixation is an industrial process (Haber's process)
- Biological nitrogen fixation takes place in a living cell.
- The enzyme that catalyzes nitrogen fixation is Nitrogenase.
- Nitrogen fixation may take place in free living organisms or in symbiotic systems.
- There are many symbiotic nitrogen fixation systems such as Lichens, Pteridophytes, Bryophytes, Gymnosperms and Legumes.
- Cyanobacteria is the symbiotic component in Lichens, Bryophytes, Pteridophytes and Gymnosperms.
- In Legumes, the symbiont is a species of bacterium *Rhizobium*.
- Source of electrons and energy for nitrogen fixation is generally pyruvic acid after it enters Krebs' cycle during cell-respiration.
- Hydrogen gas evolution may also accompany nitrogen fixation process.

- Nitrate is the most oxidized form and ammonium is the most reduced form of nitrogen.
- Nitrate is reduced to nitrite by an enzyme nitrate reductase.
- Amino acids have two functional groups, namely, amino group and carboxyl group.
- Amino acids may be produced by reductive amination of keto acids.
- Amino acids may be produced by transamination reaction.
- Reductive amination reactions are catalyzed by dehydrogenases.
- Transamination reactions are catalyzed by transaminases.

TERMINAL EXERCISES

1. Define nitrogen fixation.
2. Which form of combined nitrogen may be formed during lightening storms?
3. Name three biomolecules other than enzymes and proteins, which contain nitrogen.
4. Name one aerobic and one anaerobic bacterium, which fixes nitrogen.
5. Which amino acid is synthesized due to reductive amination of α-ketoglutaric acid?
6. Differentiate between biological and abiological nitrogen fixation.
7. What is required for biological nitrogen fixation?
8. How does human hemoglobin differ from leghemoglobin?
9. What is the function of leghemoglobin?
10. What are the functional differences between nitrate reductase and nitrite reductase?
11. What is the difference between nitrogen fixation and nitrogen assimilation? Describe in brief the process of abiological nitrogen fixation.
12. Describe in brief various steps involved in biological nitrogen fixation.
13. Enumerate various free living and symbiotic nitrogen fixing systems with suitable examples.
14. What are the major differences between free living and leguminous nitrogen fixing organisms?
15. Describe in brief nitrate and nitrite reduction in plants..
17. Describe in brief the reductive amination reactions for synthesis of amino acids in plants.
18. Describe the transamination reaction for synthesis of amino acids in plants. How does this differ from reductive amination?

ANSWERS TO INTEXT QUESTIONS

10.1 1. 78.03 percent 2. proteins and enzymes.
3. Because it is triple bonded. 4. 16 percent.
5. –195.8°C. 6. (b)
7. (a) 8. Denitrifying Bacteria

10.2 1. Conversion of molecular nitrogen to ammonia.
2. Haber's process.
3. Biological nitrogen fixation takes place in a living cell and abiological fixation without a living cell.
4. Nitrogenase. 5. Oxygen.

10.3 1. (i) b (ii) a (iii) d (iv) c
2. Cycas. 3. Yes, Hydrogen gas.
4. 16 ATP 5. Reduced coenzymes such as Ferrodoxin
6. (i) b (ii) a (iii) c 7. Nodulins.

10.4 1. NH_4
2. (i) b (ii) c (iii) a
3. Cytosol. 4. Nitrate. 5. Chloroplast.

10.5 1. (i) b (ii) c (iii) a
2. Reductive amination and transamination.
3. Transaminases. 4. Ammonia.
5. Alpha ketoglutaric acid.