SAND CULTURE

- In this method, sand is used as rooting medium and nutrient solution is added to it.
- This method has following weakness:
  - The sand being highly alkaline in nature, has to be treated with acid before use.
  - The sand gets very warm during summer and very cool in winter, hence may cause injury to the root system
  - The water holding capacity of sand is low hence it requires frequent watering.

ESSENTIAL ELEMENTS

- An essential element is the one which has a specific structural or physiological role and without which plants cannot complete their life cycle.
- The important characteristics of an essential element are:
  (i) The element is involved in the nutrition of plants.
  (ii) It is required for completion of vegetative or reproductive growth of the plant.
  (iii) Element cannot be replaced by another element.
- Minerals salts dissolved in soil solution are constantly passing downwards along with percolating (gravitational) water. The phenomenon is called leaching.
- Functions of essential minerals are :
  (i) Constitute of organic molecules example C,H,O,N,P,S,Fe,Mg, Ca
  (ii) Oxidation – Reduction reaction. example Fe and Cu, with variable valency
  (iii) Catalytic effect: Component or cofactors of enzyme example Zn, Cu, K, S etc.
  (iv) Osmotic pressure: Minerals slats and their ions present in cell sap exert an osmotic pressure for maintaining turgidity of plants and absorption of water
  (v) Turgor movements: They are mainly controlled by influx and efflux of K^+ ions
  (vi) Membrane permeability: Na^+, K^+ ions are known to increase membrane permeability while Ca^{2+} and other divalent decreases the same

- Essential elements are differentiated into

1. Macro-element (macro nutrients)
   They are those essential elements which are required by plants in quantity of more than 1 milligram/ gram or 10 mmol/kg of dry matter. Essential macronutrients area nine in number – C, H, O, N, P, K, S, Mg and Ca. Silicon and sodium occur in some plants in the range of macronutrients
2. Micro-element (micronutrient)
   They are those essential elements which are required by plants in quantity of less than 1 milligram or 10m mol/kg of dry matter. Micronutrients are eight in number Fe, Mn, Zn, Cu, Mo, B, Cl and Ni. Functional elements which occur in the range of micronutrients are Cobalt, Vanadium and Aluminium.

MAJOR ELEMENTS

CARBON, HYDROGEN AND OXYGEN

- The three elements are taken up by the plant in the form of CO_2 and water.
- All the components of living organism contains C, H, O particularly carbohydrates, lipids and nucleic acid.
- Oxygen is terminal electron acceptor in respiration. Full respiratory activities can continue even when the concentration of oxygen is just 1%. The pH is finally governed by hydrogen.

NITROGEN

- Nitrogen is generally taken up by the plant in the form of nitrates (NO_3^-), nitrite (NO_2^-) and ammonium salts and rarely in molecular form.
- It is present in amino acids, which constituent for proteins, in purines and pyrimidines that form nucleic acids, coenzyme A, Cytochromes, alkaloids and vitamins (B_1, B_2 , B_6)

Deficiency symptoms

- Chlorosis (yellowing) first appearing in older leaves then in younger leaves.
- Decrease in protein synthesis.
- Decrease in cell size and cell division.
- Plants remains stunded.
- Delay or complete suppression of flowering.

PHOSPHORUS

- The plant absorbs it in the form of phosphate ion.
- The acidic soil has monovalent form ($H_2PO_4^-$) the intermediate pH soil has divalent ion (HPO_4^{-2}) and alkaline has trivalent form (PO_4^{3-}).
- At pH 6, both monovalent and divalent phosphate ion are present.
- IT is a constituent of nucleic acids, phospholipids, coenzymes I and II (NAD, NADP), coenzyme A, found in phosphoproteins.
- The main function of phosphorus is energy transfer.
- The carbohydrates becomes more reactive due to the presence of phosphorus.
- Heavy concentration of phosphorus is found in the meristems.

Deficiency symptoms

- Accumulation of anthocyanin.
- Older leaves becomes chlorotic while younger leaves remain green.
- Formation of necrotic (dead) spots on the lamina of leaves, petioles and fruits
- Decrease in protein synthesis.
- Plants becomes stunted i.e. decrease in shoot growth.
- Root growth also restricted.
- Distortion in leaf shape.
- Slow maturation of fruits.
- Premature leaf fall.

SULPHUR

- The soil has sulphur in both organic and inorganic forms.
- It is taken up by the plant as SO_4^-.
- The sulphate ions are weakly absorbed.
- With the increase of soil pH, the adsorption decrease.
- The organic sulphur becomes available to plants through biological oxidation as the microorganisms change it to sulphuric acid.
- The sulphur is present in amino acids
- It is important in FeS proteins in photosynthesis and ferrodoxin synthesis.

Deficiency symptoms

- Chlorosis (yellowing due to degradation of chlorophyll) followed by anthocyanin development; the younger leaves show chlorosis before older ones.
- Reduced growth with hard woody stem.
- Extensive growth of root system.
- Decrease in stroma lamellae and increase in grana stacking.
- Increase in starch and sucrose accumulation and decrease in reducing sugars.

POTASSIUM

- Potassium is present in the soil in fixed, exchangeable and soluble forms.
- It is absorbed by the plant in ionic form (K^-).
- It is the commonest free ion in the cell which is found in abundance in the meristems.
- It plays a significant role in closing and opening of stomata.
- It also maintains differential permeability of cytomembranes and apical dominance.
- It is needed by enzymes DNA polymerase.
- It acts as enzyme activator in peptide bond synthesis.
- It is important in the enzymatic reactions of respiration, photosynthesis, chlorophyll synthesis, etc.
- It is important in the enzymatic reactions of respiration, photosynthesis, chlorophyll synthesis etc.

Deficiency Symptoms

- Plant growth becomes stunted due to shortening of internodes
- Mottling and chlorosis first appears in older leaves
- Necrosis at the tip and margin of leaves, leaf tip curves down.
- Destruction of pith cells of tomato and increased differentiation of phloem elements.
- Decrease in carbohydrate metabolism and increase in respiration.

CALCIUM

- Calcium is the cation available in fertile soil in plenty.
- The middle lamella is composed of calcium and magnesium pectate.
- It also influences permeability character of cell membrane.

- The calcium salt of lecithin is perhaps involved in the formation of cell membranes.
- Calcium is involved in organization of mitotic spindle.
- It acts as enzyme activators.

Deficiency symptoms

- Ultimate death of meristems as found in shoot, leaf and root tips.
- Chlorosis along the margins of younger leaves.
- Distortion of leaf shape.
- Roots poorly developed.

IRON

- It's availability depends upon the pH of the soil.
- Due to increased soil pH, the plants begins to show iron deficiency symptoms.
- It is present in cytochromes and freedoxin.
- It is also component in various peroxidases and flavoprotein.
- It is also needed in chlorophyll synthesis.

Deficiency symptoms

- Extensive interveinal chlorosis in leaves; only the young leaves.
- Inhibition of chloroplast formation.
- Aerobic respiration severely affected.

MAGNESIUM

- It is present in soil in water soluble form as silicates.
- It is component of chlorophyll and an important building substances for ribosomal subunits.
- It acts as an activator for several enzymes and also needed by CO_2 fixing enzymes.

Deficiency symptoms

- Interveinnal chlorosis is older leaves.
- Development of anthocyanin and necrotic.
- Development of collenchymas and depression of internal phloem.

MINOR ELEMENTS

MANGANESE

- It exists in soil in bivalent and tetravalent forms.
- In alkaline soil, due to oxidation, this element becomes unavailable to plants
- It acts as an enzyme activator in respiration and nitrogen metabolism.

Deficiency symptoms

- Chloroplast loses chlorophyll.
- Starch grains become yellow-green, vacuolated and finally degenerate.

BORON

- It is absorbed by plants as borate.
- It is involved in the transport of carbohydrates.
- It is required in pollen germination, cell differentiation etc.

Deficiency symptoms

- Death of shoot tips because boron is needed for DNA synthesis.
- Leaves develops a thick coppery texture, they curve and become brittle.
- Root growth is arrested and flowers not formed.

ZINC

- It is absorbed in ionic form.
- It availability decrease with the rise of soil pH.
- It plays a significant role in protein synthesis and also acts as enzyme activators.

Deficiency symptoms

- Interveinal chlorosis in mature leaves.
- Rosette due to shortening of internodes.
- White bud of maize.
- Mottled leaf in apple and walnut.

COPPER

- The amount of copper found in the soil solution is very low.
- It is taken in ionic form.

- It is component in phenolases, lactases and ascorbic oxidases.
- It participates in the electron transport chain in photosynthesis.

Deficiency symptoms

- Necrosis at tip of young leaves.
- Exanthemma in fruit trees.
- Wilting of entire plant occurs under acute shortage.

MOLYBDENUM

- It is present in the soil as molybdate ion.
- It is a component of nitrogenase and thus brings about fixation of gaseous nitrogen.
- It also participates in phosphorus metabolism.

Deficiency symptoms.

- Fall in the ascorbic acid content of the plant.
- Mottling and nerosis first in older leaves and then younger leaves.
- May lead to abscission of flowers.
- Whip tail of cauliflower.

CHLORINE

- Like manganese, it also needed in photolysis of water.

Deficiency symptoms

- Chlorosis of leaves followed by necrosis and bronzing of leaves.
- Stunting of root tips flower abscission, reducing fruiting.

FUNCTIONAL ELEMENTS

SODIUM

- It is also needed by blue –green algae for growth.
- In higher plants, it maintains differential permeability of cytomembranes. It also participates in nitrogen metabolism.
- C_4 plants also require sodium.

SILICON

- It is present in grass, equisetum.
- It is necessary for growth of sunflower and barley.

ALUMINIUM

- It is found to improve growth.
- Many plants are known to possess sensitivity to aluminium toxicity.

COBALT

- It is a component of vitamin B_{12}.
- A few blue-green algal and bacterium Rhizobium, a symbiotic nitrogen fixing bacteria of leguminous nodules also requires cobalt.

GALLIUM

- Fungus Aspergillus niger requires gallium for their growth.

SELENIUM

- Astragalus acts as selenium indicators.

IODINE

- Some marine algal like Laminaria accumulate iodine in huge amounts.

VANADIUM

- It is also necessary for growth of certain plants.

TOXICITY OF MICRONUTRIENTS

- Toxic concentration which retards the dry weight of tissue by about 10% is called toxic. This toxicity level differs from plant to plant.
  Example : In soybeans, manganese is toxic at concentration beyond 600 μg/g but sunflower plants shows toxicity symptoms only beyond 5300 μg/g.
- Some time due to excess of an element, uptake of another element may be reduced.
- Manganese competes with iron and magnesium for nutrient uptake. It also competes magnesium for binding with enzymes. So manganese in toxic concentration leads to deficiency of iron and magnesium.

ABSORPTION OF MINERALS

- Mineral absorption can be passive or active. Minerals usually occurs into two forms, cation and anion.
- There are two phases of mineral absorption, initial and metabolic. In the initial phase, there is rapid uptake of ions into the outer or free space of cells comprising cell walls. Ions absorbed in free space are fully exchangeable. In the metabolic phase, the ions pass into inner space comprising cytoplasm and vacuoles of cells. After entering inner space, the ions do not remain freely exchangeable with those of external medium.
- Entry of ions into outer space is passive absorption as no entry is required for it. Passage of ions into inner space requires metabolic energy. It is therefore an active absorption. Movement of ions into cells is called influx while movement of ions out of the cells is called efflux.

PASSIVE MINERAL ABSORPTION

- Passive mineral absorption is absorption of minerals by physical process which do not involve any direct expenditure of metabolic energy. It can occur through
  (a) Diffusion: It is passive movement of minerals along the gradient of their chemical potential across the cell membranes. Diffusion is of two types.
    (i) Passive diffusion – Membrane have pores and channel for diffusion. No energy is required for opening of channels.
    (ii) Facilitated diffusion – Cell membranes possess permeases or special protein particles that facilitate the passage of mineral ions over them without involving expenditure of energy.
  (b) Mass flow: Minerals are swept into root and passed into plant parts along the current of water caused by transpiration pull.
  (c) Donnan equilibrium: Occurrence of a non-diffusible ion in the cell interior will cause the similarly charged external ion to diffuse inwardly. The other ion will also diffuse inwardly till the multiple of free cations and inions in the interior becomes equal the multiple of free cations and anions in the external medium.
  (d) Ionic exchange: The root passes out ions in order to absorb similarly charged cations and anions.

ACTIVE MINERAL ABSORPTION

- It occurs against concentration, chemical potential gradient. Metabolic energy is utilized for this. Tare of respiration increases to meet the requirement of energy. The excess of respiration is called slat respiration. It provides extra ATP for active absorption.
- Active mineral absorption can cause accumulation of minerals. The process involves specific carriers and vesicles. A carrier is activated by energy from ATP, picks up ions from surface of membrane, froms ion-carrier complex, moves across the membrane and reach the other surface. Here, the complex breaks and ions are released. Active transport can be uniport (single ion), symport (two in the same direction) and antiport (two in opposite direction).

ION TRAFFIC INTO ROOT

- Typically, inorganic ions are taken up from the soil water via the roots to the xylem. This takes place by two pathways- apoplast and symplast.
- The apoplastic pathway, basically involves diffusion and bulk flow of water from cell to cell through spaces between cell wall polysaccharides. The ion entering the cell wall of epidermis, cell wall of pericycle and finally accumulate in xylem vessels.
- In symplastic pathway, ions entering the cytoplasm, cortex, endodermis of pericycle, through plasmodesmata and lastly to xylem vessels.

FACTORS AFFECTING SALT ABSORPTION

- Temperature – The rate of salt absorption is directly proportional to temperature within cardinal limits
- Light – The effect of light is indirect. When there is sufficient light, more photosynthesis occurs. As a result more food energy becomes available and uptake increases.
- Oxygen – The increased oxygen tension helps in increased uptake of salts. No active salt uptake is possible in absence of oxygen.
- Growth – The growth of a plant or tissue provides more surface area and binding sites for mineral ions. As a result, salt absorption is enhanced.
- pH – A plant readily taken up monovalent ions. If pH becomes alkaline, the absorption of bivalent and trivalent ion is favoured.
- Mineral interaction – The absorprtion of one type of ion is affected by other types. The absorption of K^+ is affected by Ca^{2+}, Mg^{2+} and other polyvalent ions.

However, the uptake of K^+ and Br^- becomes possible in presence of Ca^{2+} ions. There is mutual completion in the absorption of K, Rb and Cs ions

NITROGEN METABOLISM

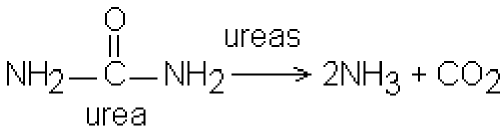
- A regular supply of nitrogen to the plants maintained through nitrogen cycle.

ABIOLOGICAL NITROGEN FIXATION

- Abiological nitrogen fixation is of two types:

Industrial nitrogen fixation

- Nitrogen and hydrogen combine to form ammonia industrially, under pressure and temperature.



Natural abiological nitrogen fixation

- Due to lightening and thundering of clouds, N_2 and O_2 of the air react to from nitric oxide (NO)
- The nitric oxide is further oxidized with the help of O_2 to form nitrogen peroxide (NO_2)
- NO_2 combines with H_2O to form nitrous acid (HNO_2) and nitric acid (NHO_3)
- The acid falls along with rain water.
- Now it acts with alkaline radicals to form water soluble NO_3^- (nitrates) and NO_2^- (nitrides)

  $N_2 + O_2 \rightarrow 2NO$ (in presence of lightening)

  $2NO + O_2 \rightarrow 2NO_2$

  $2NO_2 + H_2O \rightarrow HNO_2 + HNO_3$

BIOLOGICAL NITROGEN FIXATION

- Some blue green algal (Anabaena, Nostoc) symbiotic bacteria (Rhizobium) and free living bacteria (Azotobacter) pick up atmospheric nitrogen, reduce it to ammonia, combines with organic acids to form amino acids

Free living nitrogen fixing bacteria

- Azotobacter, Beijerinckai (both acrobic) and Bacillus, Clostridium (anacrobic) are saprotropjic bacteria that perform nitrogen fixation.

- Desulphovibrio is chemotrophic nitrogen fixing bacterium.
- Rhodopsludomonas, Rhodospirillum and Chromatium are nitrogen fixing anaerobic photoautrophic bacteria.

Free living nitrogen fixing cyanobacteria

- Many free living blue-green algae perform nitrogen fixation. Example, Anabaena, Nostoc, Calothirs, Aulosira, trichodesmium.
- Aulosira ferilissima is the most active nitrogen fixing rice field while cylinderospermum is active in sugarcane and maize fields.

Symbiotic nitrogen fixing cyanobacteria

- Anabaena and Nostoc species are common symbionts in lichens, Anthoceros, Azolla and cycad roots.
- Azolla pinnata (a water fern) has anabaena, azolla in its fronds. It is often inoculated in rice field for nitrogen fixation.

Symbiotic nitrogen fixing bacteria

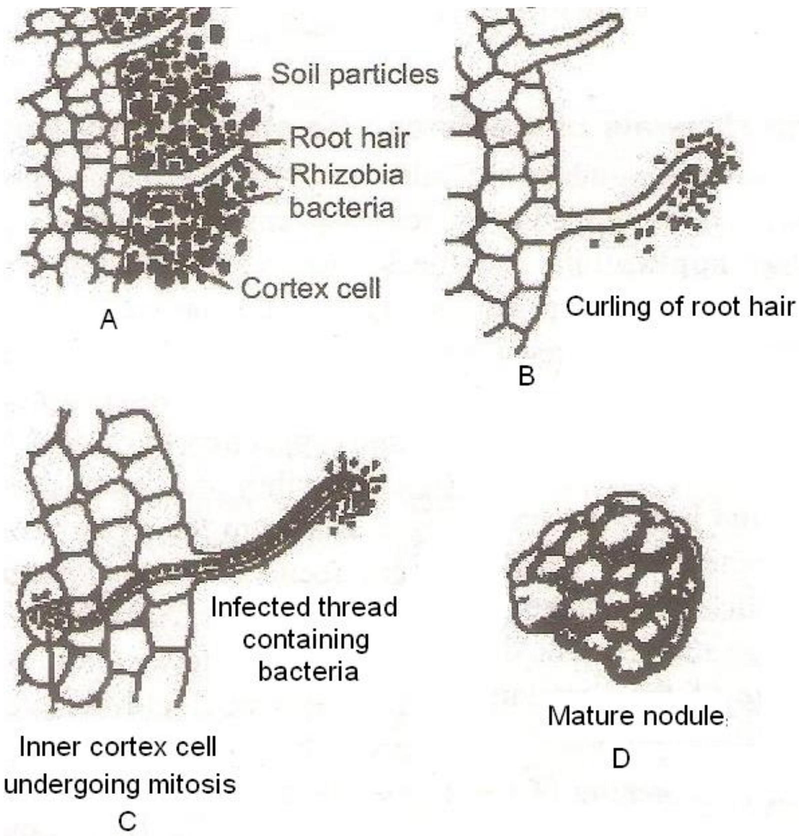
- Sesbania rostrata has Rhizobium in root nodules and Aerorhizobium in stem nodules.
- Frankia is symbiont in root nodulues of several non-legume plants like casuarinas (Australian pine), Myrica and Alnus (Alder)
- Xanthomonas and Mycobacterium form symbiotic association with the leaves of several members of rubiaceae and myrsinaceae.
- Rhizobium is the most important for crop lands because it is associated with pulses and other legumes of family fabaceae.

SYMBIOTIC NITROGEN FIXATION

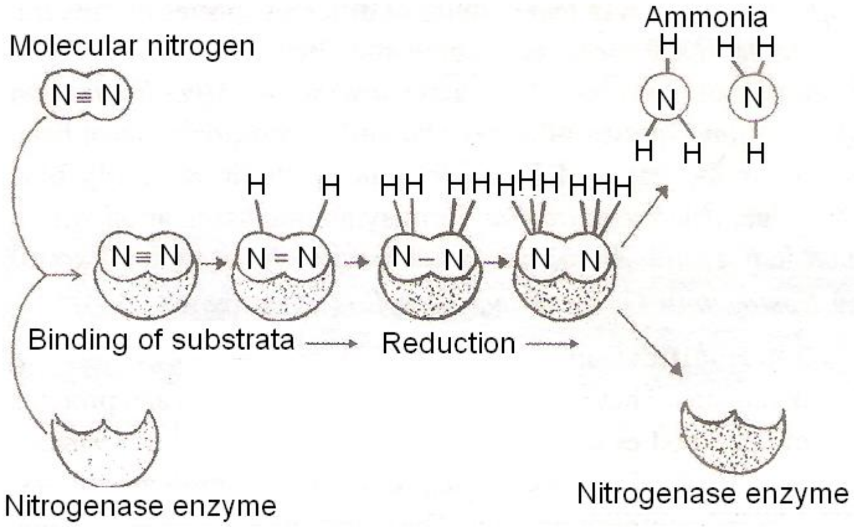
- Symbiotically bacteria are found in the root nodules of the members of family leguminosae. The best known nitrogen fixing symbiotic bacterium is Rhizobium leguminosaeum.
- Members of the family leguminosae such as beans, peas , grarams, soyabeans, etc on primary roots bears small nodules like swelling. Rhizobium penetrates to the cortex of root through infection thread. Simulated to divide more vigourously to form nodules on the root.
- Roots of the legume secrete chemical attractants. Bacteria collect over the root hairs, release nod factor that cause curling of root hairs around the bacteria. Infection thread grows alongwith multiplication of bacteria. It

branches and its end comes to lie opposite protoxylem points of vascular strand. The infected produces swellings or nodules. Nodule formation is stimulated by auxin produced by cortical cells and cytokinin liberated by invading bacteria. The infected cells and cytokinin liberated by invading bacteria. The infected cells enlarge. Bacteria stop dividing and form irregular polyhedral structures called bacteroids. However, some bacteria retain normal structures, divide and invade new areas.

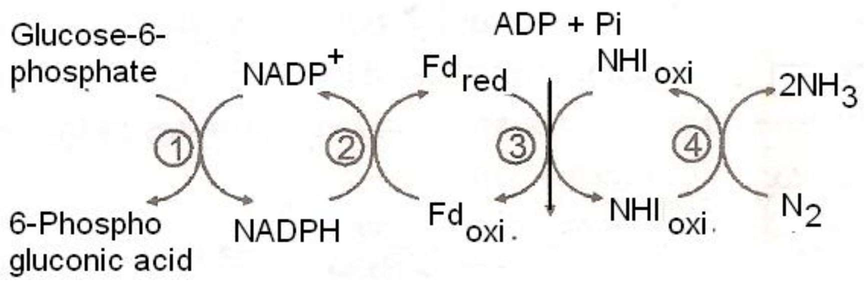
- If an infected cell bacteruiods occur in groups surrounded by host membrane. When a section a root nodules is observed, the presence of a pigment, leghaemoglobin is seen to impart pinkish colour to it. This pigment is closely realted to hemoglobin and helpful in creating optimal condition for nitrogen fixation. Like haemoglobin, leghaemoglobin is an oxygen scavenger. Fixation of nitrogen is done with the help of enzyme nitrogenase, which functions under anaerobic conditions. Leghaemoglobin combines with oxygen and protects nitrogenase.



- It is believed that during the process of nitrogen fixation, the free, atmospheric nitrogen is first bound to the enzyme surface and is not released until it is completed reduced to ammonia. Nitrogen fixation requires reducing power like NADPH, source of energy like ATP, enzyme nitrogenase and compounds for trapping ammonia formed by reduction of dinitrogen enzyme nitrogenase has iron and molybdenum. Both of them take part in attachment of a molecule of nitrogen. Bonds between the two atoms of nitrogen become weakened by their attachment to the metallic components. The weakened molecule of nitrogen is acted upon by hydrogen from a reduced coenzyme. It produces diamide (N_2H_2), hydrazine (N_2H_4) and then ammonia ($2NH_3$) Ammonia is not liberated. It is toxic in even small quantities. The nitrogen fixers protect themselves from it by providing organic acids

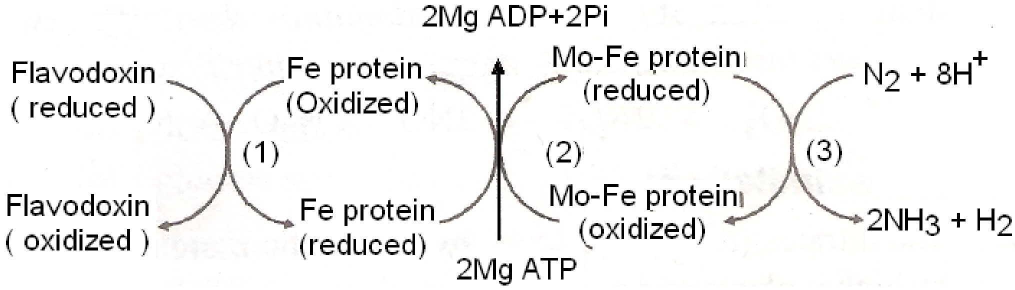


- The reduction of nitrogen into ammonia by nitrogenase in bacteroids depend upon availability of ATP and reduced substrate capable of donating hydrogen atoms to nitrogen. ATP is generated in bacteroid respiratory chain system and reduced substratory chain system and reduced substrate is obtained from host cells. Glucose-6-phosphate is considered to be reduced substrate for the process and reduced NADP together with ferredoxin function as electron carriers. The ATP interacts with non-heme iron (NH1) protein component of nitrogenase and bring about conformation change to convert it to a powerful reluctant. This powerful reductant becomes capable of transferring electrons to reduce N_2 into NH_3.



- The in vitro studies on the process reveled that at least four molecules of ATP are hydrolysed for each pair of electrons transferred to nitrogen. Thus, the reduction of one molecule of nitrogen into molecules of ammonia requires twelve molecules of ATP because six electrons are required per molecule of nitrogen reduced.

$N_2 + 6H^+ + 6e^- + 12ATP \rightarrow 2NH_3 + 12ADP + 12\ Pi$



- Ammonification – It is carried out by decay causing organisms. They act upon nitrogenous excretions and proteins of dead bodies of living organisms. Example, Bacillus ramosus, B. Vulgaris, B. mesentericus, Actinomyces. Proteins are first broken up into amino acids organic acids released in the process are used by micro-organisms from their own metabolism.

$$\text{Proteins} \xrightarrow{+H_2O} \text{R-}NH_2 \xrightarrow{+H_2O} \underset{\text{organic acid}}{ROH} + \underset{\text{ammonia}}{NH_3}$$

- Nitrification – it is a phenomenon of conversion of ammonium nitrogen to nitrate oxygen. It is performed in two steps – nitrite formation and nitrate formation. Both the steps can be carried out by Aspergillus flavus. In the first step, ammonium ions are oxidized to nitrites by Nitrosococcus, Nitrosomonas. Nitrites are changed to nitrates in the second step by nitrocystis, Nitrobacter.

$$2NH_3 + 3O_2 \xrightarrow[\text{Aspergillus flavus}]{\text{nitroscoccus, nitrosomonas}} 2NO_2^- + 2H^+ + H_2O + \text{energy}$$

$$2NO_2^- + O_2 \xrightarrow[\text{Aspergillus flavus}]{\text{Nitrobacter, Nitrocystis}} 2NO_3^- + \text{energy}$$

- Denitrification – under anaerobic conditions, some microorganism use nitrate and other oxidized ions as source of oxygen. In the process, nitrates are reduced to gaseous compounds of nitrogen. The latter escape from the soil. Common bacteria causing denitrification of soil are Pseudomonas denitrificans, Thiobacillus denitrificans, micrococcus denitrificans.
$2NO_3^- \rightarrow 2NO_2^- \rightarrow 2NO \rightarrow N_2O \rightarrow N_2$

NITRATE ASSIMILATION

- The nitrates (NO_3^-), produced by nitrification, are absorbed by higher plants and assimilated by the process called nitrate assimilation.
- The nitrates absorbed by plant roots get converted into amino acids and amides before incorporating acids proteins and other macromolecules.
- Reduction of nitrate into ammonia is called nitrogen assimilation.
- The overall summary equation of reduction of nitrate to ammonia is as follows NO_3^- + 8 electrons + $10H^+ \rightarrow NH_4^- + 3H_2O$

This process consists of the following two distinct enzymatic steps:

Reduction of nitrate to nitrite

- The reaction is catalyzed by enzyme nitrate reductase and occurs in the cytosol outside any organelle and require NADH as an electron donor, FAD as a prosthetic group, cytochrome b_{557} as an electron carrier and molybdenum (Mo) as an activator of enzyme

$$NO_3 NAD(P)H + H^+ \xrightarrow[\text{FADIFMN}]{\text{Nitrate reductase}} NO_2^- + H_2O + NADP^+$$

Reduction of nitrite

- The reaction is catalysed by enzyme nitrite reductase and the most probable electron donor in the reaction appears to be reduced ferredoxin
- Nitrite reductase requires power and the product of nitrite reduction is ammonia

$$2NO_2^- + 7NAD(P)H + 7H^+ \xrightarrow[\text{ferredoxin}]{\text{nitrite reductase}} 2NH_3 + 4H_2O + 7NAD(P)^+$$

SYNTHESIS OF AMINO ACIDS

- Reductive ammination – In this process, ammonia reacts with α-ketoglutaric acid and forms glutamic acid

$$\alpha\text{-ketoglutaric acid} + NH_4^+ + NAD(P)H \longrightarrow \text{Glutamate} + H_2O + NAD(P)$$

- Catalytic amidation – In the presence of enzyme synthestase and ATP, ammonia combines with amino acid glutamate to form amide glutamine. Glutamine reacts with α-ketoglutarate in the presence of reduced coenzyme to form two molecules of glutamate.

$$\text{Glutamate} + NH_4^+ + \text{ATP} \xrightarrow[\text{synthetase}]{\text{Glutamine}} \text{Glutamine} + \text{ADP} + \text{Pi}$$

$$\text{Glutamine} + \alpha\text{-ketogluarate} + \text{NAD(P) H} \xrightarrow[\text{synthetase}]{\text{glutamine}} \text{2 Glutamine} + \text{NAD(P)}$$

- Transamination – It involves the transfer of amino group from one amino acid to the keto group of organic acid. Glutamic acid is the main amino acid from which other 17 amino acids are formed through transamination. The enzyme responsible for such reaction is known as transaminase.