Course-B.Sc. part III

E-content : Online class 8/9/20 ; 9/9/20

Nitrogen Metabolism

Nitrogen is the most abundant element in the atmosphere having 78% concentration in the air. This is present in the molecular form of N_2, having triple bond between the two N-atoms. Due to triple bond, it cannot be taken directly by the plants. Plants take nitrogen in the forms of nitrate (NO_3^-) and ammonium ions (NH_4^+). Nitrogen is universally present in all organisms in different forms. It is component of proteins, chlorophyll, enzymes, nucleic acids, hormones. Plants take nitrogen from the soil in the forms of nitrate or ammonium salts. All these forms of nitrogen are produced as a result of fixation of N_2 and inter conversion of ammonia to nitrate and vice-versa. Conversion of different forms of N_2 to other forms takes place by exchanging compounds between atmosphere, living organisms and soil in the form of a cycle, known as nitrogen cycle. When a plant or animal or microorganisms die, their organic components reach the soil and are acted upon by the degrading bacteria, which degrade the organic compounds to their elemental level. Proteins, amino acids, nucleic acids, hormones and other nitrogen containing compounds are degraded. Therefore, four different forms of nitrogen are present in the soil viz., (i) NO_3^- (ii) NO_2^- (iii) NH_4^+ salts & (IV) organic nitrogenous compounds.

The most effective usable nitrogen source is NO_3^-. However, ammonia is rapidly absorbed by the plants and used directly. The best source of nitrogen for plants is ammonium nitrate. Soil microorganisms typically produce ammonia from organic compounds. Generally, urea is first converted to ammonia by an enzyme *urease* before it is utilized.

Urea-----→ *Urease*---------→ $2\ NH_3 + CO_2$

Plant /Animal residues---→Proteins---proteolysis--→Amino acids-------ammonification --→ NH_4^+

Nitrate Assimilation in Plants

Nitrate absorbed by the plants get converted to amino acids and amides before incorporating into proteins and other macromolecules. Reduction of nitrate into ammonia is called nitrogen assimilation. The overall summary equation of reduction to nitrate is as follows-

$NO_3^- + 8\ e^- + 10\ H^+$ ----------→ $NH_4 + 3H_2O$

This is a two step reaction, which can be explained as follows:-

In the first step, nitrate is converted to nitrite. The reaction is catalyzes by enzyme **nitrate reductase.** This requires NADH as an electron donor. In some species it is NADPH as coenzyme. It also requires FADH as a prosthetic group, cytochrome$_{557}$ as an electron carrier and molybdenum (Mo) as an activator of enzyme.

$NADH + H^+ + FAD$ ----------→ $NAD^+ + FADH_2$

$FADH_2$ + Mo(ox)----------→ Mo(Red) + $2H^+$ + FAD

Mo(Red) + $2H^+$ + NO_3----------→ Mo(Ox) + NO_2

In the second step, nitrite is reduced to ammonium ion. This reaction is catalyzed by an enzyme nitrite reductase. In the reaction ferredoxin acts as electron donor.

The overall reaction is as follows-

NO_2 + 6 e^- + $4H^+$----------→ NH_4^+ + $2H_2O$

Ammonification –

Nitrogenous compounds present in the dead remains of plants and animals, excretory materials are degraded by a group of bacteria. This is degraded by bacteria like *Bacillus, Proteus, Pseudomonas, Clostridium* and some fungi. They secrete enzyme proteinases, which degrade proteins to peptides. In the next stage peptides are broken to their units amino acids by the enzyme peptidases.

Proteins-------Proteinases----→ Peptides----------peptidases-------→ Amino acids

Amino acid degradation-

Amino acids formed by the breakdown of proteins, are further degraded by many microorganisms.

Alanine + ½ O_2 ----------Alanine deaminase-----→ Pyruvic acid + NH_3

Fate of ammonia-

Ammonia produced in the above steps, may be lost to the atmosphere. However, most of the ammonia is absorbed by the plants or it can be converted to nitrate by nitrification.

Nitrification-

Conversion of ammonia to nitrate is called as nitrification. This takes place by a group of nitrifying bacteria, *Nitrosomonas* and *Nitrobacter*. In the first step, ammonia is oxidized to nitrite by the bacteria *Nitrosomonas*. In the next step nitrite is oxidized by *Nitrobacter* , converting nitrite to nitrate.

NH_4^+ + 3/2 O_2←====*Nitrosomonas*======→ NO_2^- + H_2O + H^+ + Energy (-84K cal)

NO_2^- + ½ O_2---------- *Nitrobacter*----------→ NO_3^- + Energy (–17.8 K cal.)

Nitrogen Fixation –

Atmosphere contains molecular nitrogen (N_2), which is not accessible to plants. Plants take ammonium and nitrate ions from soil. Conversion of N_2 to NH_3 by either physical or by biological agencies is called nitrogen fixation.

Physical nitrogen fixation

Following reaction takes place in atmosphere as a physical process such as lightening, thunder storms –

$N_2 + O_2$ ---------Lightening ----→ 2NO

HNO_3 + Ca/K salts----------→ Ca/K nitrates

Biological nitrogen fixation-

This is mediated by microorganisms present in the soil. Many bacteria and blue green algae (Cyanobacteria) fix atmospheric nitrogen either in free living state or in association with plants, called as asymbiotic and symbiotic nitrogen fixation respectively. They reduce nitrogen to ammonia,the reaction catalyzed by an enzyme ***nitrogenase***.

Asymbiotic N_2 fixation

Free living soil bacteria which fix nitrogen are as following-

Aerobic bacteria- *Azotobacter Beijerinckia, Derxia*

Anaerobic photosynthetic bacteria- *Clostridium, Rhodopseudomonas,Rhodospirillum*

Chemosynthetic bacteria- *Desulfovibrio*

Free living fungi- *Pullularia, and Yeasts.*

Heterocystous Cyanobacteria- *Nostoc, Anabaena, Calothrix*

-- Continued