

# **METHODS OF NITROGEN FIXATION**

**Dr. KRISHNAKUMAR J**

**HOD & Assistant Professor**

**Department of Botany**

**Cardamom Planters' Association College**

**Pankajam Nagar**

**Bodinayakanur 625 582**

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CARDAMOM PLANTERS' ASSOCIATION

## METHODS OF NITROGEN FIXATION

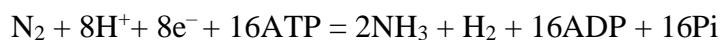
### ***Process # 1. Atmospheric Nitrogen Fixation:***

The enormous energy of lightning breaks nitrogen molecules and enables them to combine with the oxygen in the air forming nitrogen oxides. These dissolve in water forming nitrates, that are carried to the earth. Atmospheric nitrogen fixation probably contributes about 5-8% of the total nitrogen fixed.

### ***Process # 2. Biological Nitrogen Fixation:***

Biological nitrogen fixation occurs when atmospheric nitrogen (N=N) is reduced to ammonia by complex set of enzymes called nitrogenase and huge expenditure of ATP. Although the first stable product is ammonia, it is quickly incorporated into protein and other organic nitrogen compounds.

**It can be represented by the following equation:**



Nitrogenase enzyme

Two molecules of ammonia are produced from one molecule of nitrogen gas, at the expense of 16 molecules of ATP and a supply of electrons and protons (H ions), in the presence of enzyme complex nitrogenase. The enzyme is a biological catalyst found naturally only in certain microorganisms called diazotrophs.

**Micro-organisms that fix atmospheric nitrogen are:**

#### **(a) Aerobic:**

Bacteria Azotobacter, Beijerinckia, Klesbiella, Cyanobacteria e.g., Nostoc. Anabaena

#### **(b) Anaerobic:**

Bacteria Clostridium, Desulfovibria, Purple sulphur bacteria. Green sulphur bacteria.

#### **(c) Symbiotic with plants:**

##### **(i) Legumes:**

Leguminous plants of family Fabaceae (Taxa such as Clovers. Soya-beans, Alfalfa, pea-nuts etc.) contain symbiotic bacteria called Rhizobium (Bacillus radicola) within the nodules in their root system. These crops are also referred as green manure.

##### **(ii) Other Plants:**

1. Frankia (soil bacteria) forms nitrogen fixing root nodules in actinorrhizal plants e.g., Alder, Bayberry etc. (sometimes called actinorrhizae).

2. Parasponia also able to interact with Rhizobium and forms root fixing nodules. Azospirillum, Azotobacter (free living nitrogen fixing bacteria) grow in the rhizosphere of grasses and several other plants and fix atmospheric nitrogen. Spirillum lipoferum is associated with cereal grasses.
3. These are also some other symbiotic nitrogen fixing associations for e.g., Cyanobacteria (Nostoc, Anabaena) in the coralloid roots of Cycas and thallus of Anthoceros, mosquito – fern Azolla and some species of lichens such as Loberia and Peltigera etc.
4. Biological nitrogen fixation was discovered by the German agronomist Hermann Hellriegel and Dutch microbiologists M. Beijerinck.

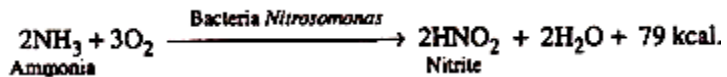
***Process # 3. Industrial Nitrogen Fixation:***

1. Under great pressure, at temperature of 600°C and with the use of catalyst, nitrogen and hydrogen (usually derived from natural gas or petroleum) can be combined to form Ammonia (NH<sub>3</sub>).
2. It can be used directly as fertilizer but most of it is further processed to form urea and ammonium nitrate, (NH<sub>4</sub>NO<sub>3</sub>).
3. Artificial fertilizer production is now the largest source of human-produced fixed nitrogen in the Earth's ecosystem.
4. The most common method is Haber process. This process requires a high pressure (200 atms) and high temperature (at-least 400°C).
5. Nitrogen fixation is essential for agriculture and manufacturing of fertilizers. It is also an important process in the manufacturing of explosives e.g., gunpowder, dynamite, TNT etc.
6. These nitrates are absorbed by the plants for the manufacture of complex nitrogenous compounds.
7. Plants in turn are eaten by animals. When the plants and animals die, certain bacteria, fungi and other microbes start acting on them and decompose the protein of dead organisms into ammonia.
8. Such bacteria are called ammonifying bacteria and the process is known as ammonification.
9. Ammonia is converted into soluble ammonium compounds which are released in the soil or water.

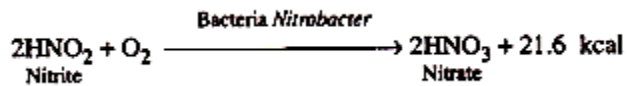
10. Enzymes involved in ammonification are GS (Gln synthetase, GOGAT (Glu 2-oxoglutarate aminotransferase), GDH (Glu dehydrogenase) etc.
11. Ammonia is converted into soluble ammonium compounds which are released in the soil or water.
12. Ammonia or the ammonium compounds are converted into nitric acid and then into nitrate by nitrifying bacteria.

**This process is known as nitrification and occurs in two steps:**

**Step I:** Ammonia is converted into nitrite.



**Step II:** Nitrite are converted into nitrates



In reverse reaction, some bacteria break of nitrates, nitrites and ammonium compounds to molecular nitrogen which is returned to atmosphere to complete the nitrogen cycle. Such bacteria are called denitrifying bacteria e.g., *Pseudomonas denitrificans*, *Thiobacillus denitrificans*, *Micrococcus*, *Bacillus licheniformis* etc. and this process is known as de-nitrification. This inorganic nitrogen is again recycled into the organic system upon absorption by the plants

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