Biology of Nitrogen Fixation

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What is the importance of Nitrogen to plants?

- Nitrogen is one of the most abundant element, in the form of nitrogen gas in Earth's atmosphere.
- Nitrogen is a critical limiting element for plant growth and production.
- It is the major component of chlorophyll as well as amino acids.
- It is found in the important biomolecules such as ATP and nucleic acids.
- Plants can only utilize reduced form of nitrogen.

Plants acquire nitrogen by:

- The addition of ammonia/nitrate fertilizer or manure to soil.
- The release of these compounds from organic matter decomposition.
- Conversion of atmospheric nitrogen into the compounds by natural process such as lightning.
- Biological nitrogen fixation.

Biological Nitrogen Fixation

- *Beijerinck* in 1901 by special group prokaryotes.
- These organisms utilize the enzyme *Nitrogenase* to convert the atmospheric nitrogen to ammonia (NH3).

Nitrogen fixing prokaryotes

- Aquatic organisms – **Cyanobacteria**.
- Free living soil bacteria- ***Azotobacter***.
- Associative bacteria- ***Azospirillum***.
- Symbiotic Bacteria – ***Rhizobium.***

Nitrogen Fixation by Free living microorganisms

- Many soil bacteria fix significant level of nitrogen without the direct interaction with other organism.
- Examples are ***Azotobacter, Bacillus, Clostrodium, Klebsiella*** etc.

Associative nitrogen fixation

- Some bacteria form close association with several members of *Poaceae* and fix appreciable amounts of nitrogen within the rhizosphere of the host plants.
- Examples: ***Azospirillum***

Symbiotic Nitrogen fixation

- Many microorganisms fix nitrogen symbiotically by partnering with host plant.
- The plant provide sugar by photosynthesis that are utilized by nitrogen fixing microorganisms for the energy it needs to fix nitrogen.
- In exchange of these carbon sources the mo provides fixed nitrogen to host plant for its growth .

Examples of symbiotic nitrogen fixing micro *o*rganisms are the bacteria with non-leguminous plants and the bacteria with leguminous plants producing root nodules.

- Frankia
- Rhizobium
- Bradyrhizobium

The Process

Nitrogen fixing bacteria

In biological nitrogen fixation two moles of ammonia are produced from one mole of nitrogen gas, using 16 moles of ATP and a supply of electrons and protons (hydrogen ions):

$$N_2 + 8H+ + 8e^- + 16\ ATP = 2NH_3 + H_2 + 16ADP + 16\ Pi$$

This reaction is performed exclusively by prokaryotes (the bacteria and related organisms), using an enzyme complex termed nitrogenase. This enzyme consists of two proteins - an iron protein and a molybdenum-iron protein.

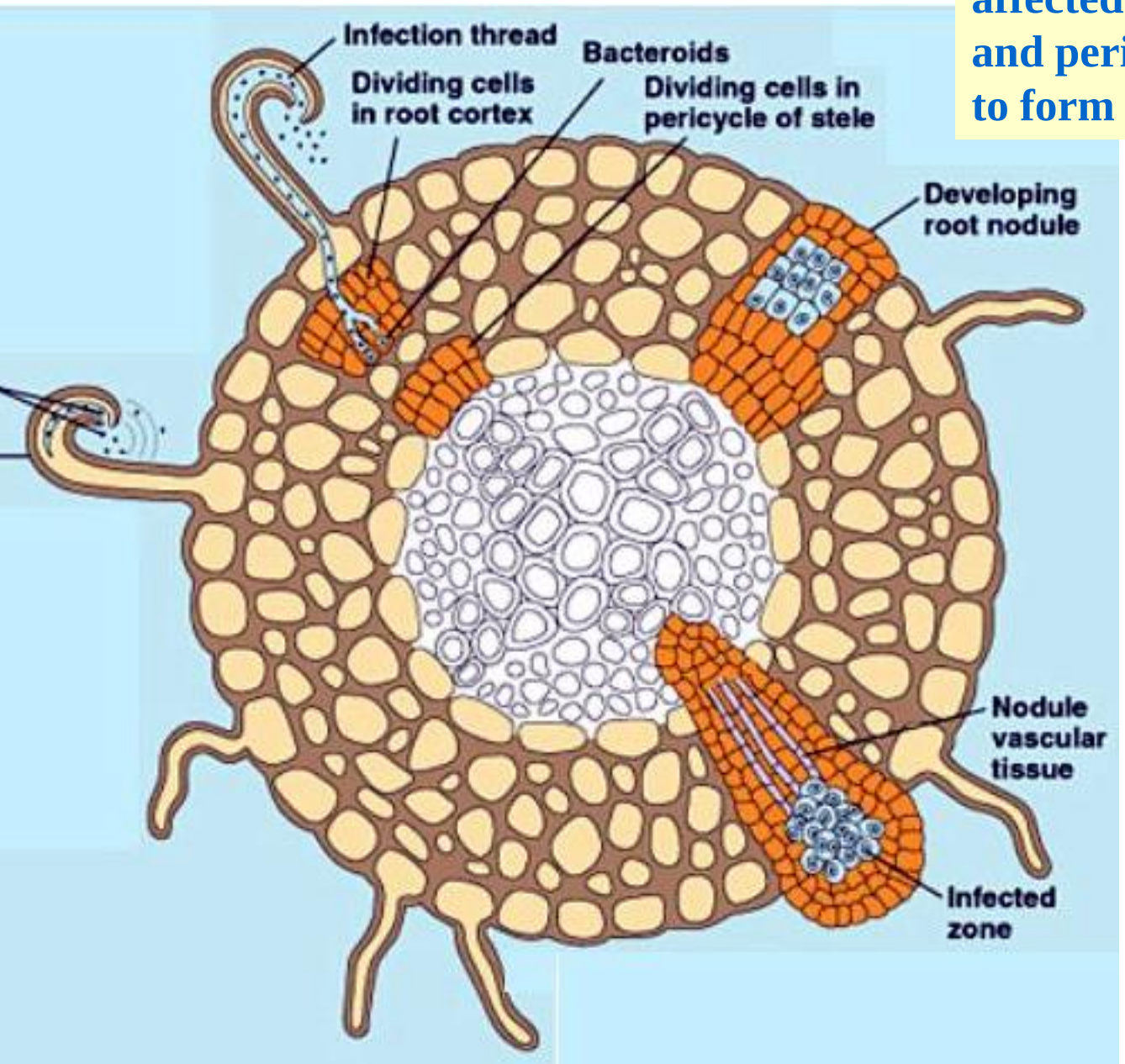
A point of special interest is that the nitrogenase enzyme complex is highly sensitive to oxygen

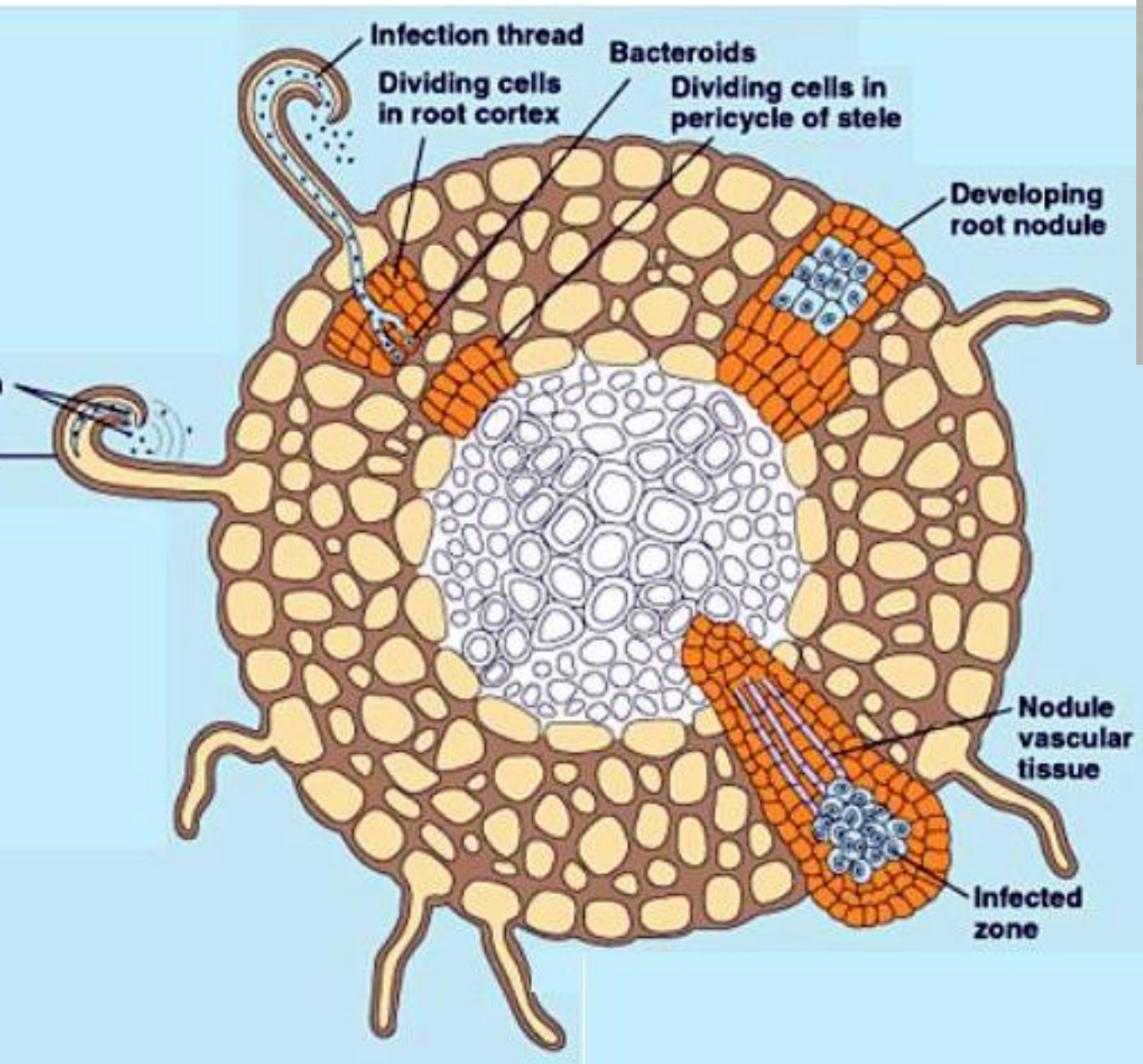
Roots emit chemical signals that attract Rhizobium bacteria. The bacteria emit signals that stimulate root hairs to elongate, and to form an infection thread by an invagination of the plasma membrane

Soya bean infection with Rhizobium

The bacteria penetrate the root cortex within the infection thread. Plant cells start dividing and vesicles containing the bacteria, bacteriods, bud into the cells from the branching infection thread

Growth continues in the affected regions of the cortex and pericycle and these fuse to form the nodule



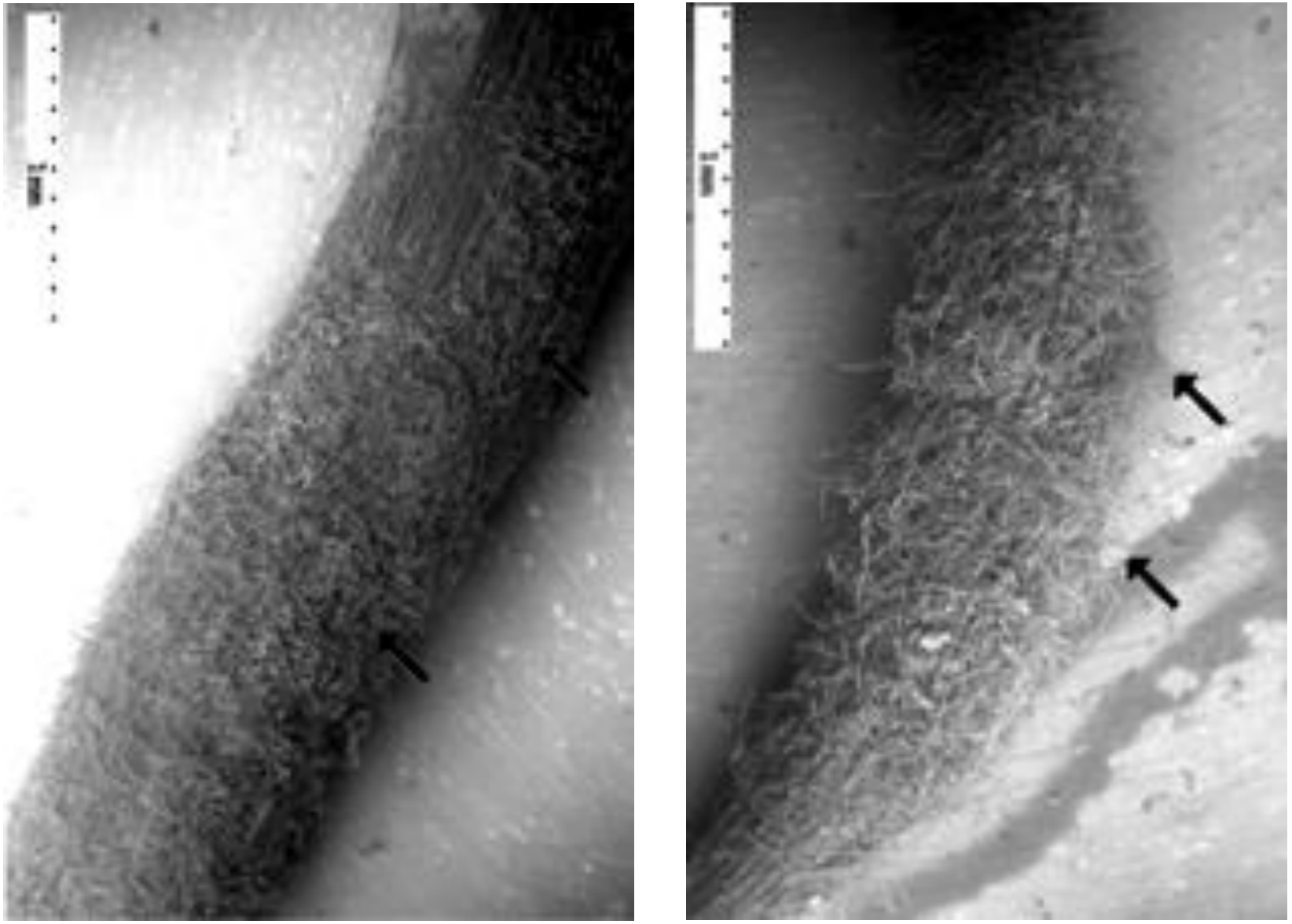


The nodule grows and vascular tissue connecting it to the plant's xylem and phloem develops

Lens culinaris (lentil) root nodulation.

7 days after infection

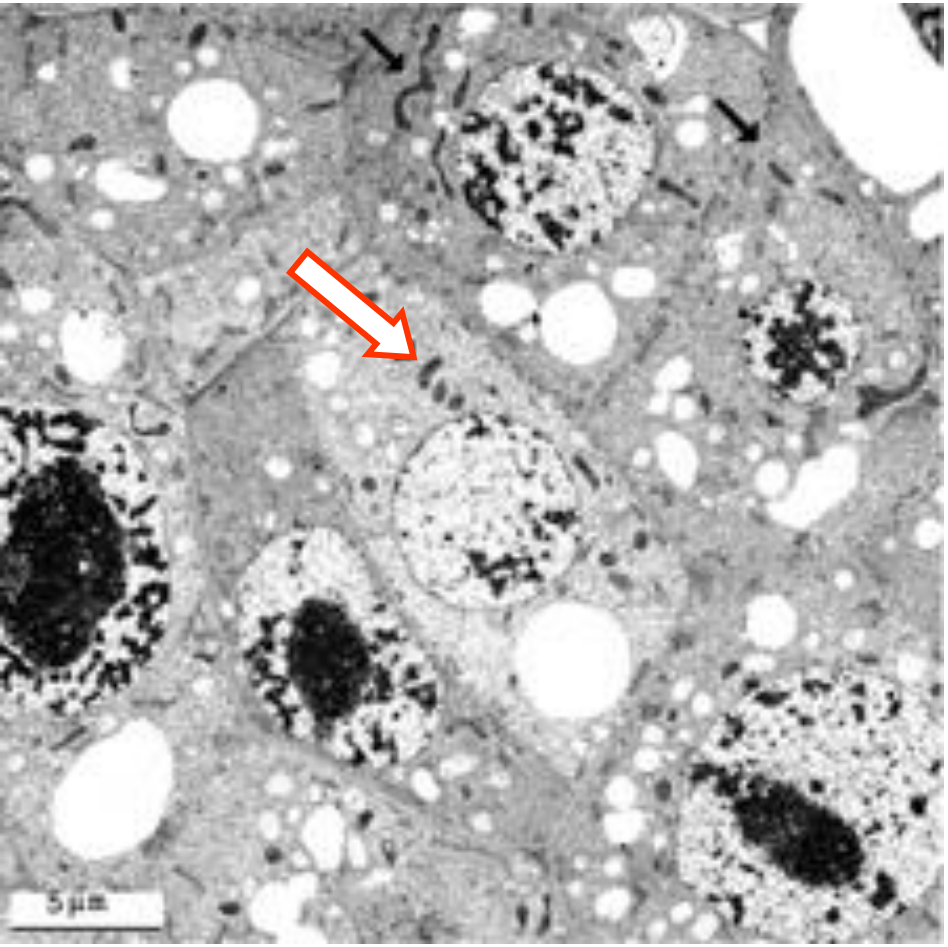
12 days after infection



http://www.sunderland.ac.uk/~es0man/tem2.htm

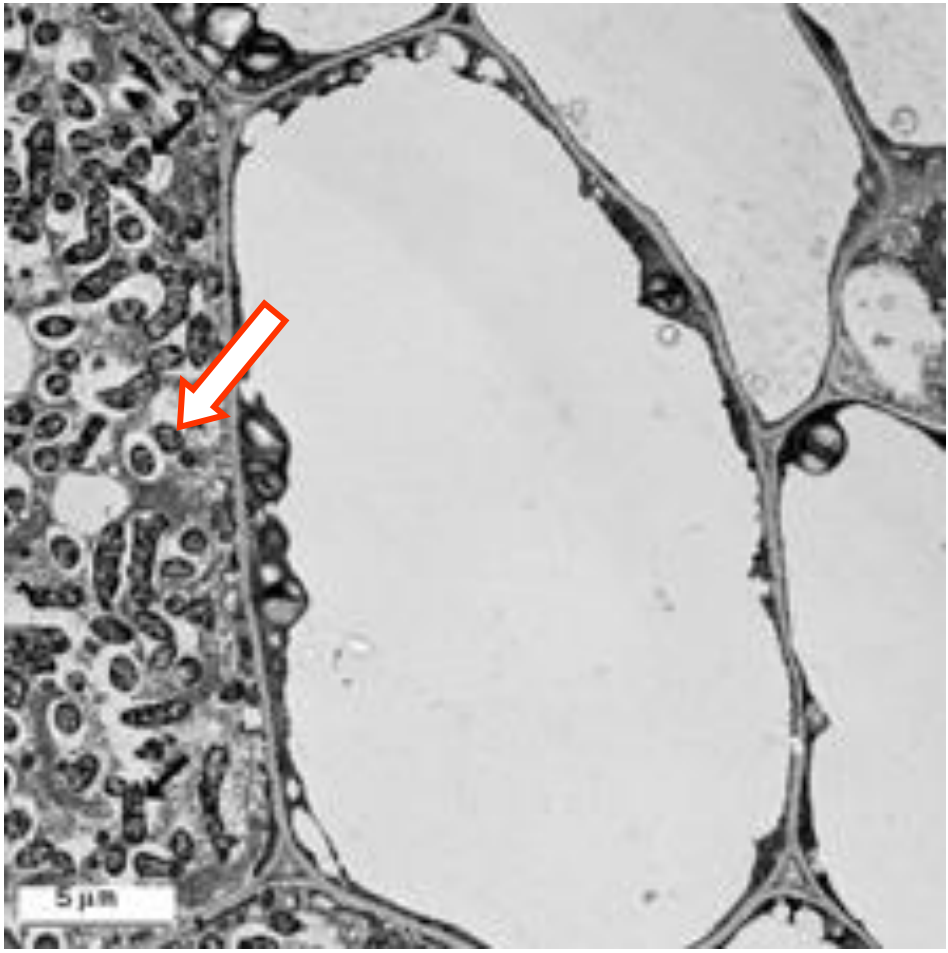
7 days

Small numbers of bacteria



12 days

Large numbers of bacteria

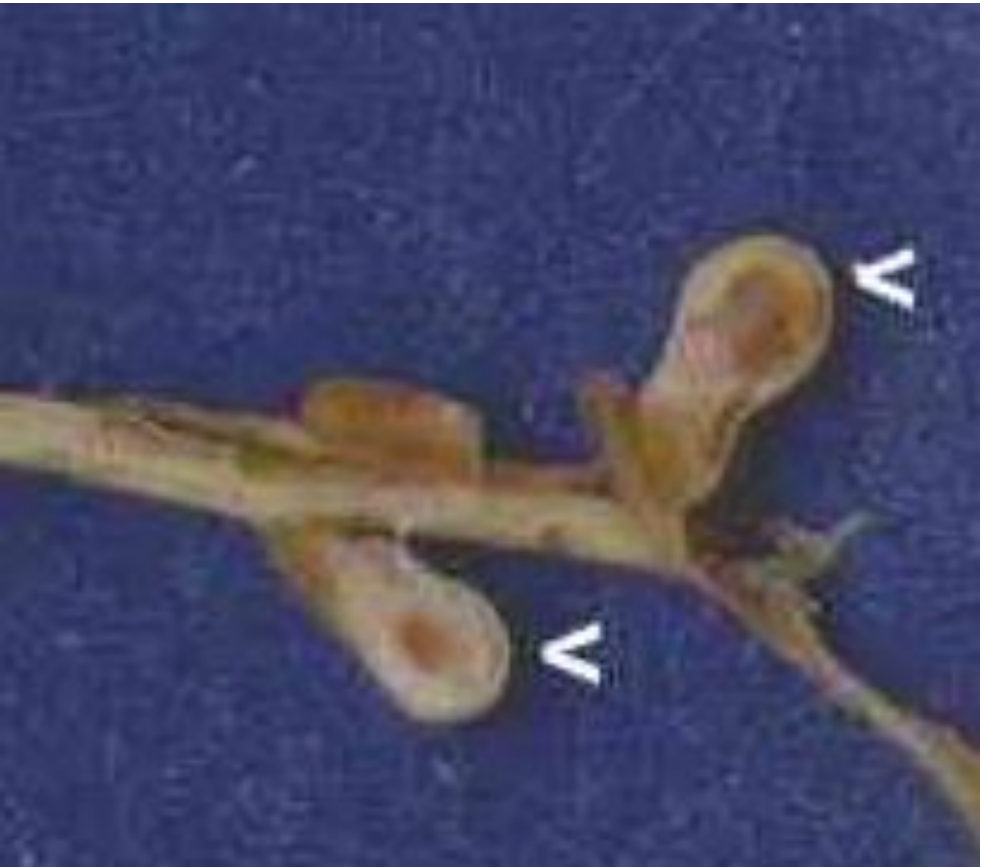


http://www.sunderland.ac.uk/~es0man/tem2.htm

Part of a clover root system bearing naturally occurring nodules of *Rhizobium*. Each nodule is about 2-3 mm long.

http://helios.bto.ed.ac.uk/bto/microbes/nitrogen.htm

In symbiotic nitrogen-fixing organisms such as *Rhizobium*, root nodules can contain oxygen-scavenging molecules such as leghaemoglobin, which shows as a pink colour when the active nitrogen-fixing nodules of legume roots are cut open. Leghaemoglobin may regulate the supply of oxygen to the nodule tissues in the same way as haemoglobin regulates the supply of oxygen to mammalian tissues



Clover root nodules.

Leghaemoglobin is found only in the nodules and is not produced by either the bacterium or the plant when grown alone.

Some legumes are better at fixing nitrogen than others. Common beans are poor fixers (less than 50 lbs per acre) and fix less than their nitrogen needs. Maximum economic yield for beans in New Mexico requires an additional 30-50 lbs of fertilizer nitrogen per acre. However, if beans are not nodulated, yields often remain low, regardless of the amount of nitrogen applied.

Other grain legumes such as peanuts, cowpeas, soybeans, and faba beans are good nitrogen fixers, and will fix all of their nitrogen needs. These legumes may fix up to 250 lbs of nitrogen per acre and are not usually fertilized. If large amounts of nitrogen are applied, the plant literally slows or shuts down the nitrogen fixation process.

There are many research programs attempting genetic improvement of nitrogen fixation, e.g., alfalfa

SUMMARY

- Nitrogen is an essential nutrient for plant growth and development but is unavailable in its most prevalent form as atmospheric nitrogen.
- Plants instead depend upon combined or fixed form of nitrogen such as ammonia and nitrate.
- Much of these nitrogen is provided in the form of industrially produced nitrogen fertilizers to crops.

- Use of these fertilizers lead to world wide ecological problems such as formation of coastal dead zone.
- On the other hand Biological Nitrogen Fixation offers a natural means of providing nitrogen for plants.
- Conventional agriculture has depended upon this process to produce the commercial fertilizer for plant growth.
- There is need to promote its use.

THANKS