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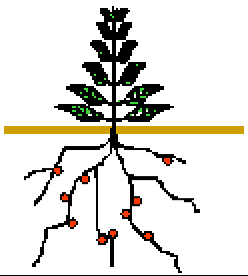
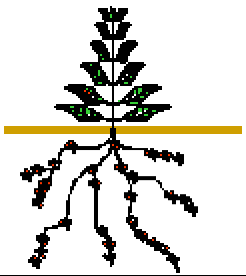
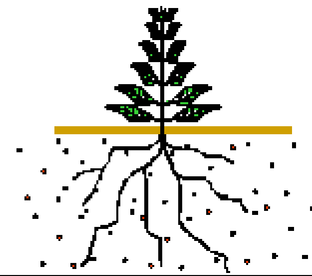
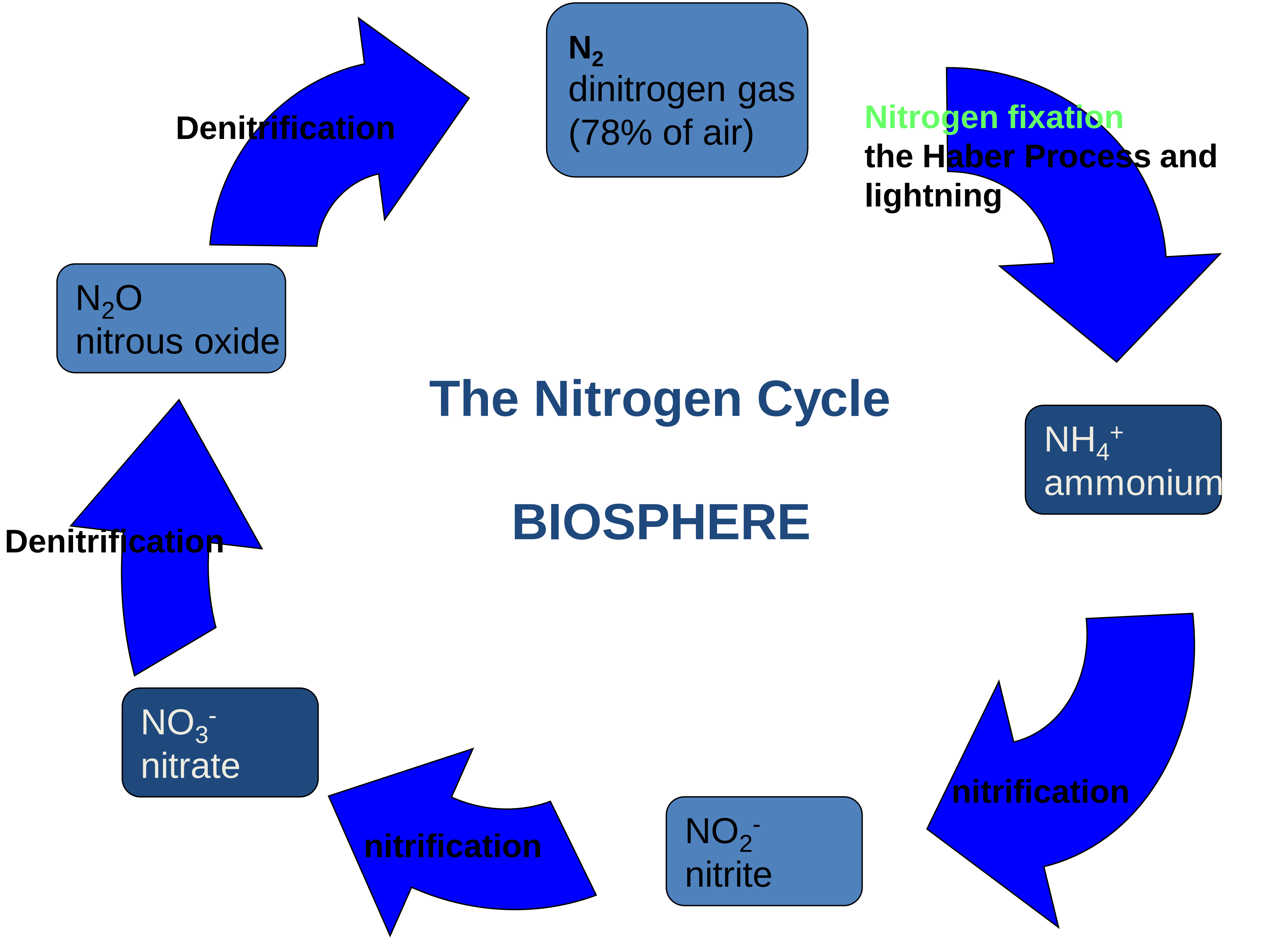
Biofertilizers

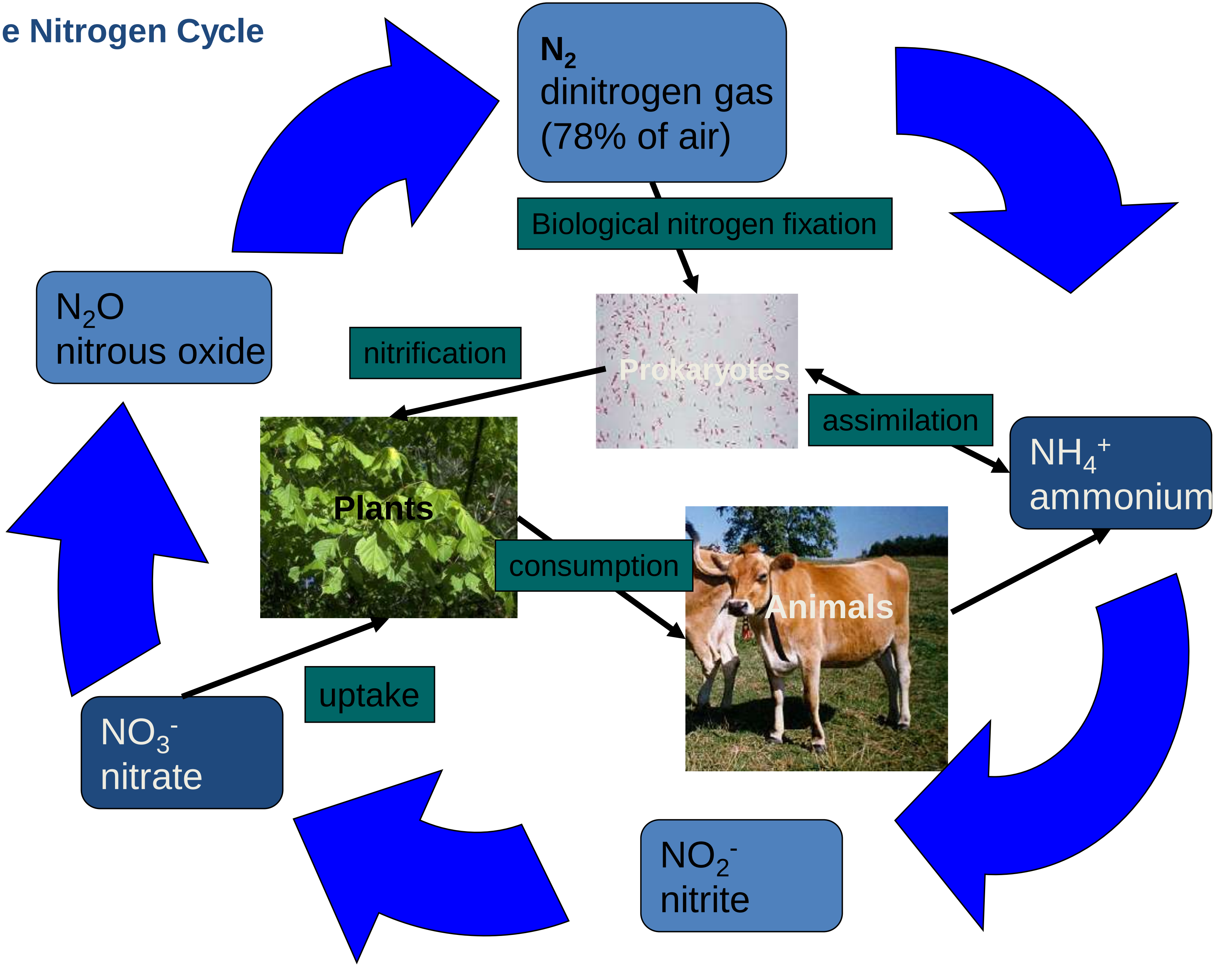
- Nitrogen Fixation

System of N_2 fixation (and microbes involved) (N_2 → NH_3)	SYMBIOSIS (e.g. *Rhizobium*) 	ASSOCIATION (e.g. *Azospirillum)* 	FREE-LIVING (e.g.*Rhodospiorillum*) 	
Energy source (Organic C)	Sucrose from the host plant	Root exudates from the host plant	Heterotroph (plant residues)	Autotroph (photo-synthesis)
Estimates of fixation rate (kg N/ha/y)	50-400	10-200	1-2	10-80



N_2
dinitrogen gas
(78% of air)
Nitrogen fixation
the Haber Process and lightning
NH_4^+
ammonium
nitrification
NO_2^-
nitrite
nitrification
NO_3^-
nitrate
Denitrification
N_2O
nitrous oxide
Denitrification
The Nitrogen Cycle
BIOSPHERE

The Nitrogen Cycle

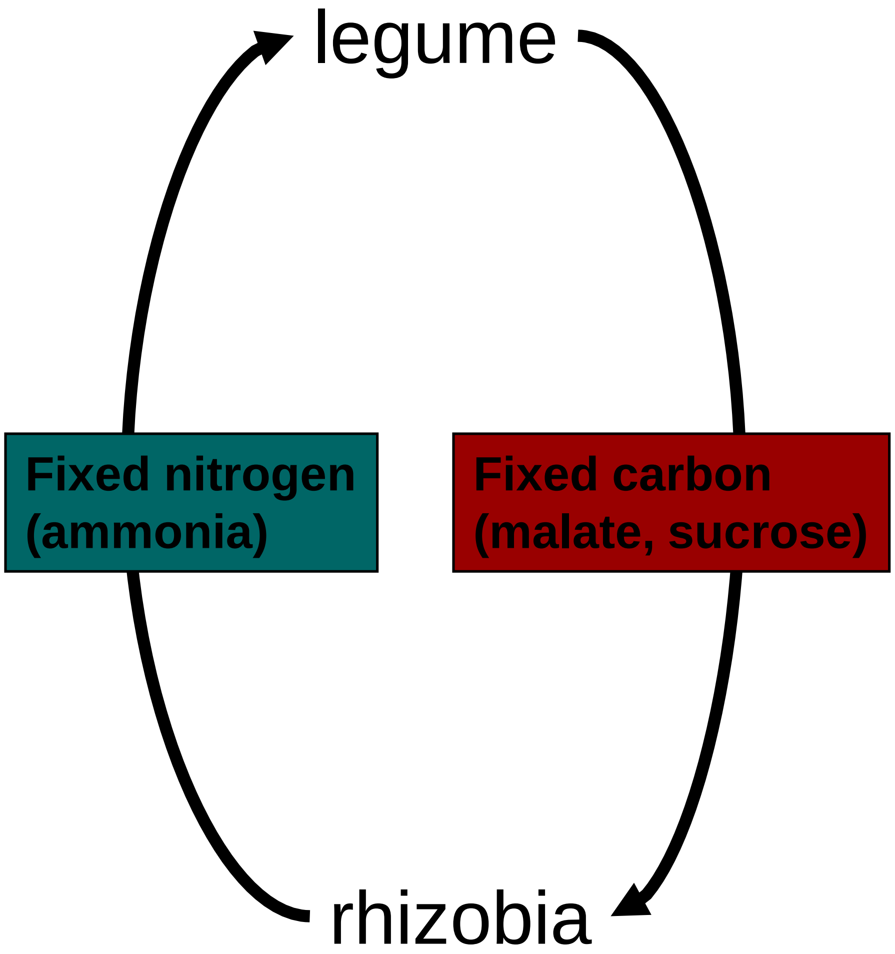


Rhizobium-legume symbioses

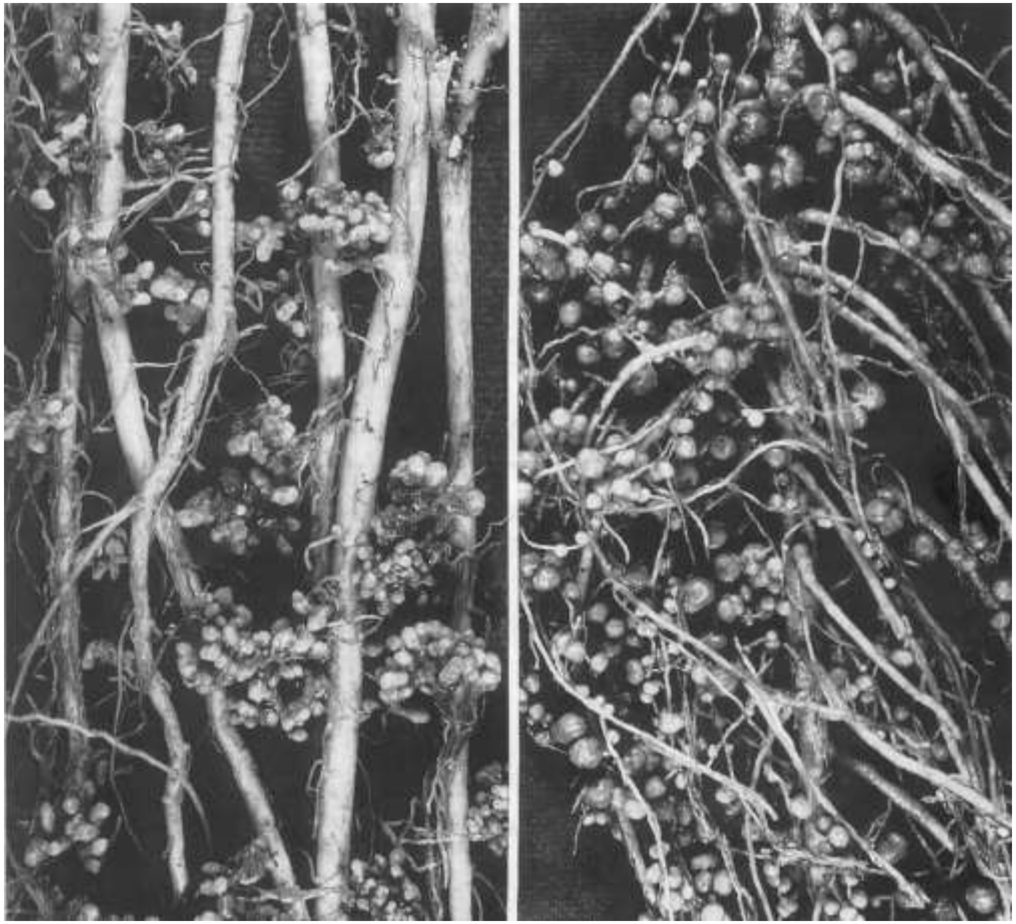
Host plant	**Bacterial symbiont**
Alfalfa	*Rhizobium meliloti*
Clover	*Rhizobium trifolii*
Soybean	*Bradyrhizobium japonicum*
Beans	*Rhizobium phaseoli*
Pea	*Rhizobium leguminosarum*
Sesbania	*Azorhizobium caulinodans*

Complete listing can be found at at:
http://cmgm.stanford.edu/~mbarnett/rhiz.htm

Both plant and bacterial factors determine specificity


legume
Fixed nitrogen
(ammonia)
Fixed carbon
(malate, sucrose)
rhizobia

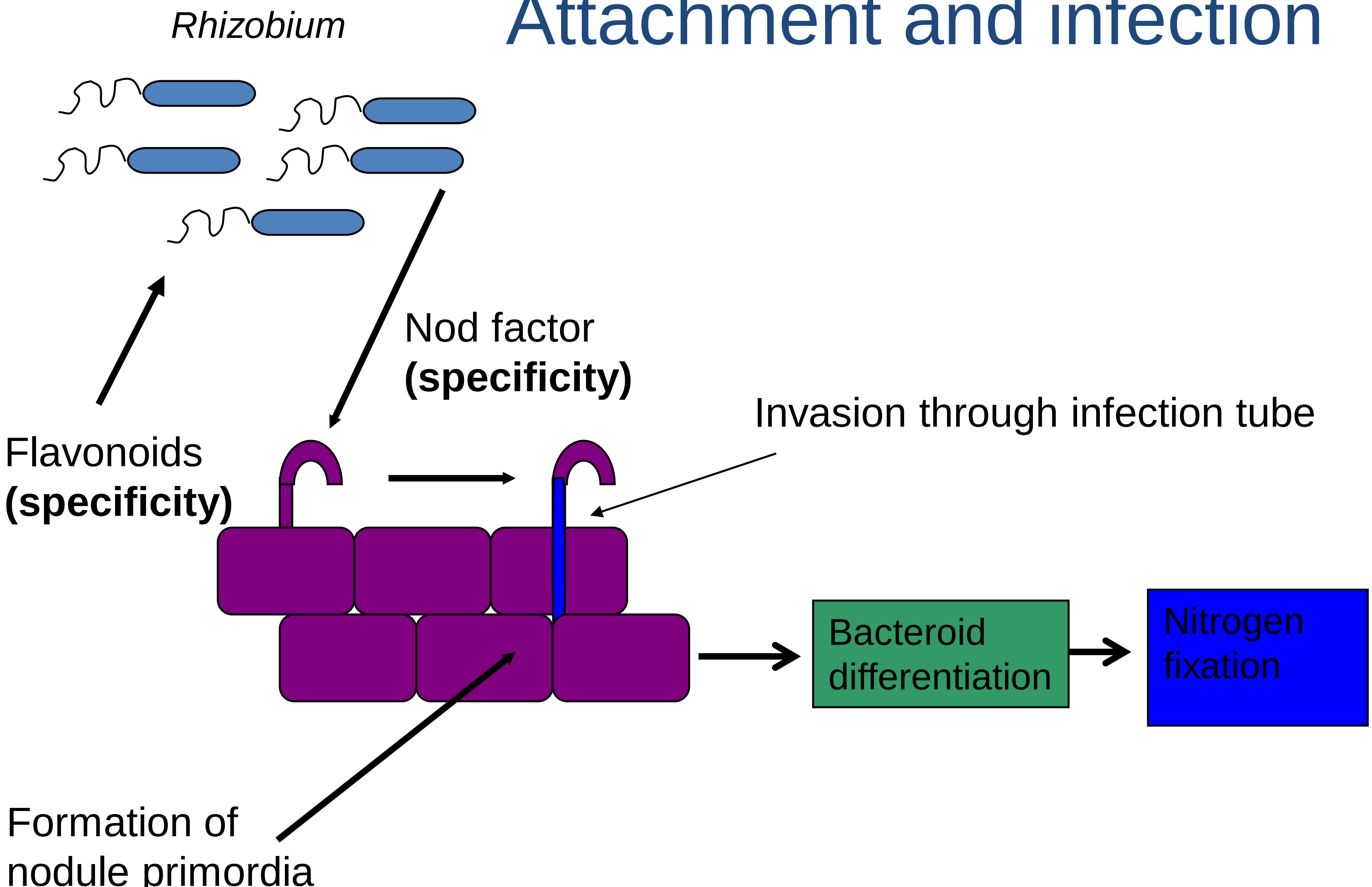
Obvious signs of nodulation by common rhizobial species



MEDICAGO
(alfalfa)

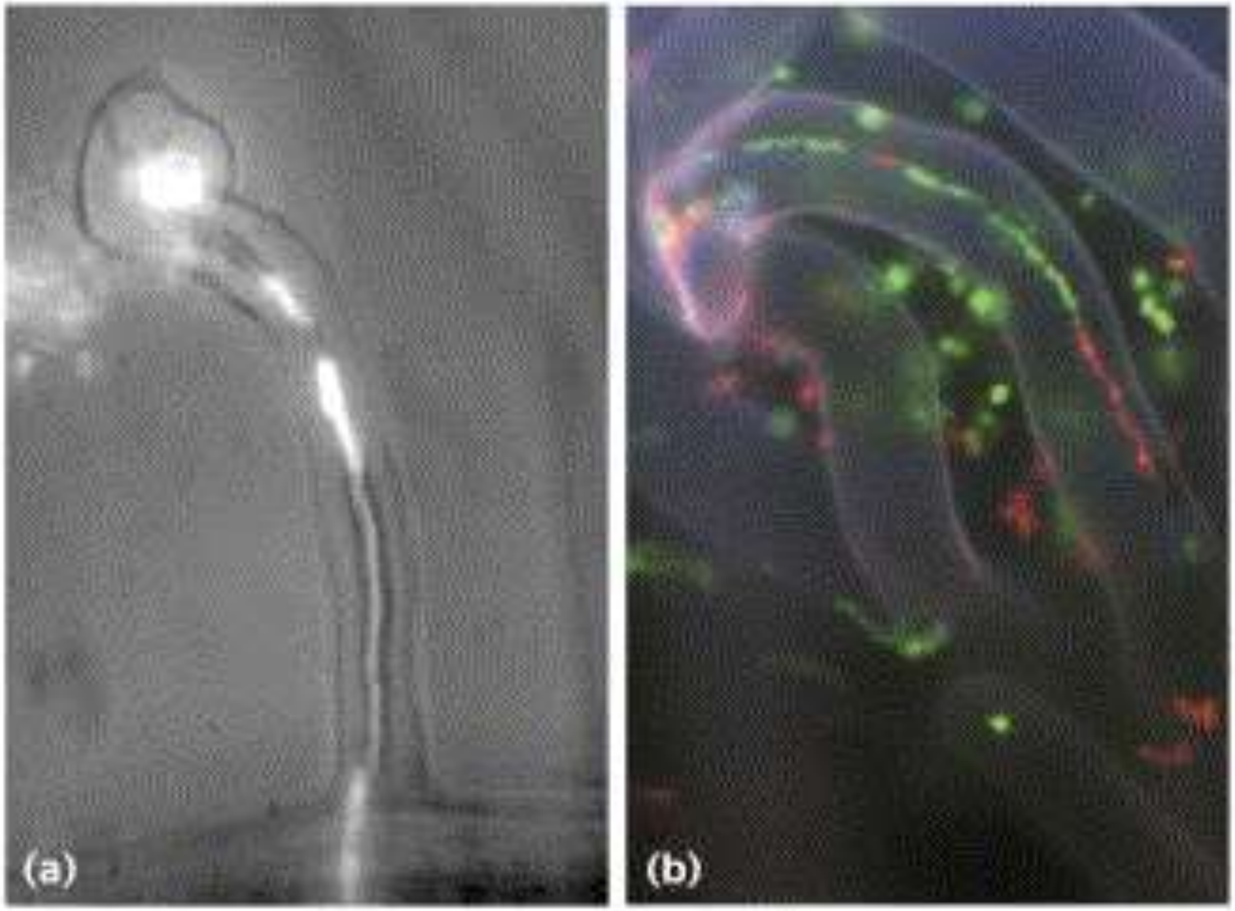
LOTUS
(birdsfoot trefoil)

Attachment and infection



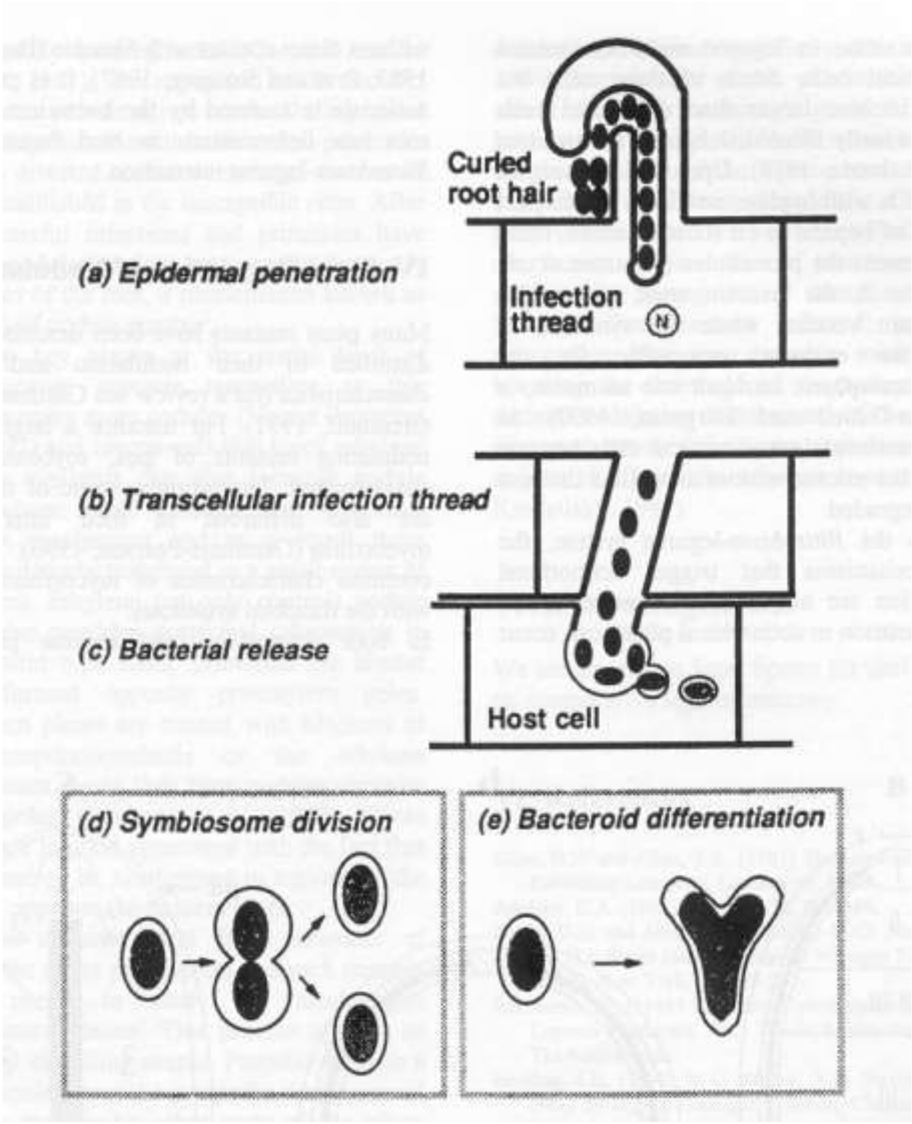
From Hirsch, 1992.
New Phyto. 122, 211-237

Bacteria divide as they traverse infection thread

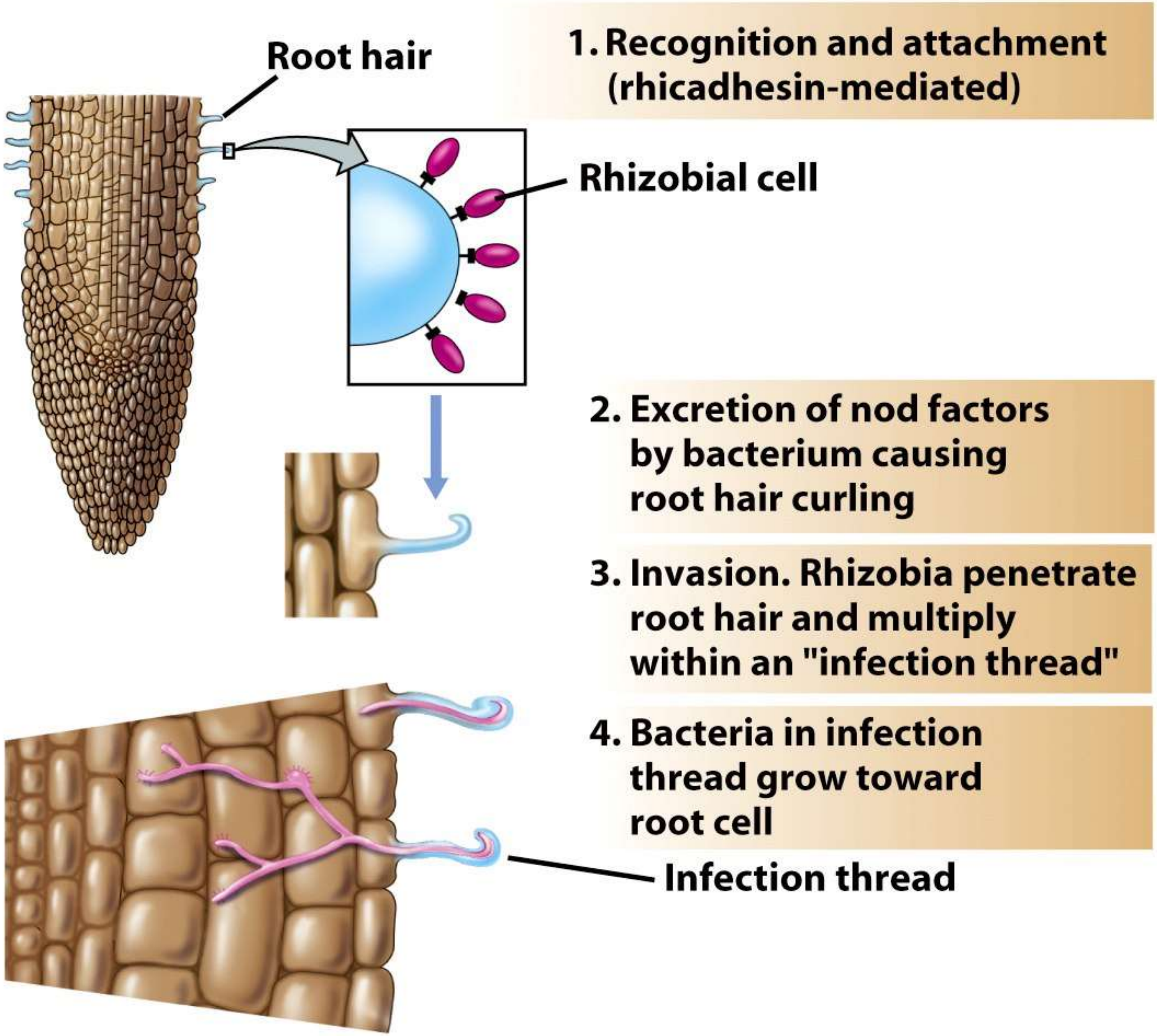


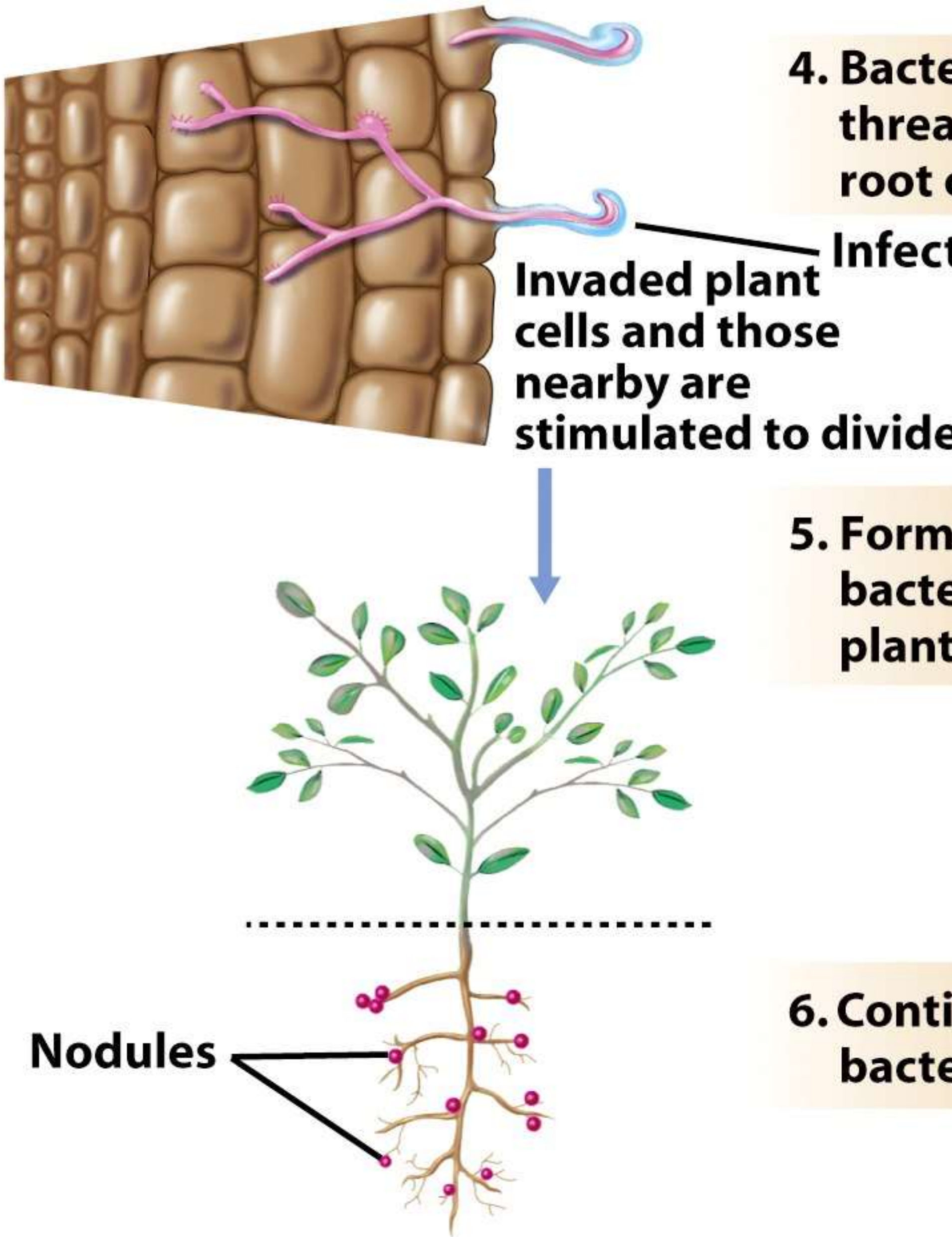
Current Opinion in Microbiology

Nodule development



Enlargement of the nodule, nitrogen fixation and exchange of nutrients

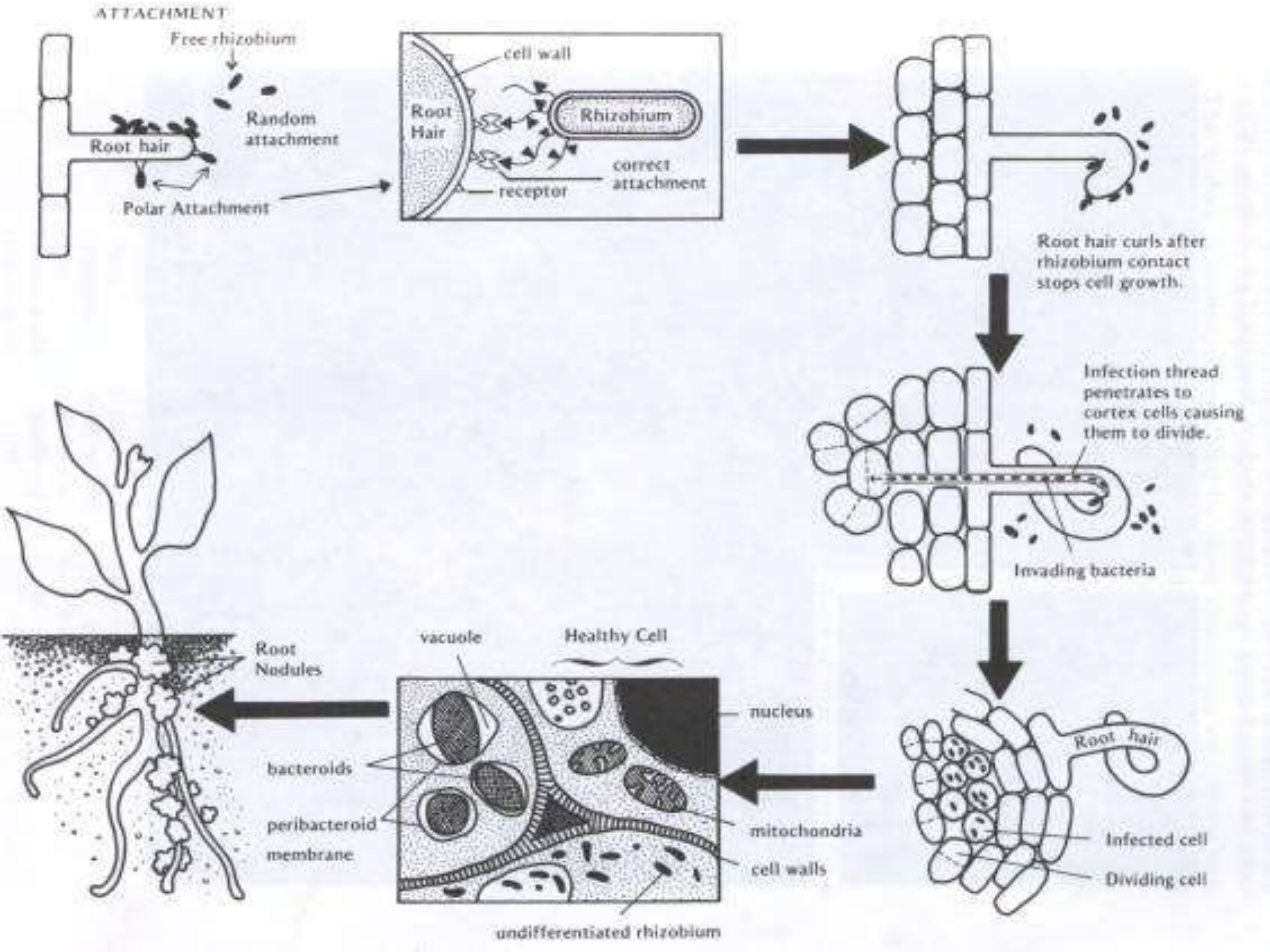

Root hair
1. Recognition and attachment (rhicadhesin-mediated)
Rhizobial cell
2. Excretion of nod factors by bacterium causing root hair curling
3. Invasion. Rhizobia penetrate root hair and multiply within an "infection thread"
4. Bacteria in infection thread grow toward root cell
Infection thread



4. Bacteria in infection thread grow toward root cell

5. Formation of bacteroid state within plant cell

6. Continued plant and bacterial cell division



ATTACHMENT
Free rhizobium
Random attachment
Root hair
Polar Attachment
cell wall
Root Hair
Rhizobium
correct attachment
receptor
Root hair curls after rhizobium contact stops cell growth.
Infection thread penetrates to cortex cells causing them to divide.
Invading bacteria
Root hair
Infected cell
Dividing cell
vacuole
Healthy Cell
nucleus
bacteroids
peribacteroid membrane
mitochondria
cell walls
undifferentiated rhizobium
Root Nodules

The nodulation process

1. Chemical recognition of root and *Rhizobium*
2. Root hairs curl
3. Formation of infection threads
4. Invasion of the roots by *Rhizobia*
5. Nodule tissue forms
6. Bacteria convert to bacteriods and begin to form nitrogenase enzyme
7. Legume provides *Rhizobia* with carbon. *Rhizobia* provide the legume with fixed N

The Nodulation Process

- Chemical recognition of roots and *Rhizobium*
- Root hair curling
- Formation of infection thread
- Invasion of roots by *Rhizobia*
- Cortical cell divisions and formation of nodule tissue
- Bacteria fix nitrogen which is transferred to plant cells in exchange for fixed carbon

Non-symbiotic nitrogen fixation

Aquatic:

Cyanobacteria
Anabaena
Nostoc

Terrestrial and rhizosphere-associated:

Azospirillum
Azotobacter
Acetobacter
Klebsiella
Clostridium

- These bacteria take nitrogen from the air (which plants cannot be used) and convert it into a form of nitrogen called ammonium (NH_4^+) which can be used by plants.
- The enzyme named nitrogenase controlls the process called nitrogen fixation and these bacteria are called nitrogen fixers.