Symbiotic relationship with plants (1)

 Nitrogen fixation Symbiotic nitrogen fixation Rhizobium, Cyanobacteria, Azolla Associative nitrogen fixation Bacterial nitrogen fixation in the root zone of rice Pseudomonas, Alcaligenes

Non-symbiotic nitrogen fixer bacteria

Proteobacteria group Cyanobacteria group Gram positive bacteria group Green sulfur bacteria group General Archaea group

Nitrogen fixation

N₂+2H⁺+8 e⁻+16ATP → 2NH₃+H₂+16ADP+16 phosphate Nitrogenase

 $NH_4^+ \rightarrow Glutamine \rightarrow Glutamic acid \rightarrow \rightarrow$ Protein, Nucleic acid Ammonia assimilation enzymes Root nodule bacteria (Rhizobium, Bradyrhizobium, Azorhizobium)

Grouped into Proteobacteria a

Symbiosis with legume plants and Ulmaceae plant, Parasponia

Legume plants are grown in 250×10^{6} ha of land in the world, and 140 kg ha⁻¹ of nitrogen are fixed.

(Total agricultural land area in the world is 1406×10^{6} ha, in Japan 5.1 $\times 10^{6}$ ha)





Cross section of root nodule.

Soy bean root Bacteroid tissue nodule Root nodules

Nitrogen fixation and the fertility of paddy field

- Cyanobacteria in the paddy water.
- Azolla
- Sesbania as green manure grown on the ridges of rice field
- Associative nitrogen fixing bacteria in the rhizosphere of rice





Sesbania

Cyanobacteria (blue green algae) Have symbiotic relationships with Lichens and mosses, **Azolla (Fern plant) Cycad family (Gymnosperm) Gunnera** (Angiosperm)





Azolla

Actinomycetes (Frankia) Belongs to Gram positive bacteria. Forms root nodules with alder and Alnus firma as well as many other angiosperm trees in the temperate and tropic region.



Alder trees in the wetland

Symbiotic relationship with plants (2)

Mycorrhizal fungi

Symbiosis with 70 % of terrestrial plants.

Fungi residing in the surface layer or inside of roots.

Help the absorption of phosphate and water.

Arbuscular mycorrhizal fungi Ectomycorrhiza

Transfer of nitrogen from N fixing plants to other plants



Mycorrhizal fungi in Timothy roots



Antagonism with disease causing germs

Abundant and heterogenous microbial flora prevents the spread of disease causing germs. *Bacillus subtilis* controls the crop diseases. *Pseudomonas* bacteria prevent the seedling diseases of tomato.

Non pathogenic *Fusarium* prevents various wilting and soft-rotting diseases of vegetables.

Microbial biomass in soil

Microbial biomass

occupies $0.3 \sim 5.0$ % of total soil carbon.

In mineral soils $2\sim 3\%$, in volcanic ash soils $0.3\sim 1.0\%$ in average.

Numbers of microbes: 10⁷~10⁹ / g soil (10 millions ~1 billion /g)

(Fungi occupy ca. 70 % in upland field, grassland, forest, and orchard field, while bacteria occupy 80 – 98 % of total microbes in paddy soils.)

Total C and N, and biomass C contents in some Japanese soils.

Kind of soils	Texture	Total C (Mg/ha)	Total N (Mg/ha)	Biomass C (kg/ha)
Immature sand dune soil	S	9.4	0.86	32
Light colored ando soil	L	33.4	3.36	114
Humic ando soil	SiL	110	8.33	234
Brown forest soil	CL	20.6	1.69	276
Dark red soil	LiC	83.8	7.49	1,155

Sakamoto and Hodono: SSPN, 46, 483-490 (2000) 土壌サイエンス入門 (Introduction to soil science, 2005) p.169 Methods of soil biomass determination

1) Direct counting method

2) Culture method

3) Biochemical method

1. Direct counting Jones-Mollison method

Count and measure the microbes in soil suspension with hemo-cytometer.

Fluorescent immuno-staining method

Staining the specific bacterium by its fluorescent antibody.

2. Culture method **Dilution Plate Method (DP) Most Probable Number Method** (MPN) **Substrate Induced Respiration Method (SIR)**

3. Biochemical method

Chloroform fumigation method (applicable to all microbes)

→fumigation-culture and fumigation-extraction methods

ATP method (applicable to all microbes) Phospho-lipid (applicable to all microbes) Muramic acid, diamino-pimelic acid (for bacteria) Ergosterol (for fungi) Microcalorimeter (applicable to all microbes)



Functions of soil microbial biomass

- 1) Decomposer of organic matter
- 2) Source and stock of soil nutrients

Growth of crops and nutrients from microbial biomass

Relative % of nutrients contained in the microbial biomass of upland field. (Anderson and Domsch, 1980)

	С	Ν	Р	K	Ca		
Bacteria	25	4.5	1.5	0.8	0.4		
Fungi	75	10.5	10.1	9.0	1.0		
Total	100	15.0	11.6	9.8	1.4		
Amounts of biomass nutrients in the field (kg/ha)							
		108	83	70	11		

Showing the balance of nutrient uptake by crops and nutrients supplied from microbial biomass.

Nutrients in microbial biomass and absorbed amounts by crops

	Biomass nutrients	Absorbed nutrients
Germany, upland	100 kg N/ha	40 kg N/ha
England, upland	17 kg P/ha	6.8 kg P/ha
England, pasture	56.8 kg P/ha	22.7 kg P/ha
Philippines,	44 – 156	40 - 100
paddy field	kg N / ha	kg N / ha

