4. MAXIMIZING LEGUME BNF

Rhizobia and legumes have evolved together for many millions of years and have become specialized in their relationships. For instance, rhizobia that form nodules on common beans do not form nodules on soya beans, groundnuts, or field peas. Similarly rhizobia that form nodules on groundnuts do not form nodules on common beans, soya beans or field peas. Furthermore even the rhizobia that nodulate one particular legume say soya beans are not all equally good. Some strains are good N fixers while others are poor fixers. So to maximize BNF, it is recommended that farmers inoculate legume seeds with the most effective strain. Rhizobium inoculants are made using superior strains of rhizobia that are selected after rigorous testing for their ability to fix large amounts of nitrogen. So they are likely to give good results if they are well handled and applied following instructions given on the packet.

Seed dressing chemicals are toxic to rhizobia. Therefore, dressed seeds should not be coated with Rhizobium inoculant. When planting dressed legume seeds the Rhizobium inoculant should be applied directly into the planting furrows before the seeds are planted. This is best done by mixing the inoculant with water (10g of inoculant to 1 litre of water, mixed thoroughly by stirring) and applying the mixture to the planting furrows at the rate of 1 litre per 100m row.

5. UTILIZATION OF BNF

Symbiotic BNF between legumes and rhizobia and between *Azolla* and *Anabaena* can be exploited by farmers using many cropping systems:-

a) Sole cropped legumes require less N:
When sole cropped-legumes fix nitrogen, most of this N is taken up by the legume resulting in higher total yield rich in N. The legume biomass adds N to the soil.

b) Inter-cropping of legumes with non-legumes:
Some of the nitrogen fixed by the legume crop ends up in the soil when nodules rot and when old leaves are shed. This N is then taken up by the non-legume and it results in increased yield of the non-legume.

c) Legumes in crop rotations improve soil fertility:
Legumes have been used in crop rotations for a long time because they are known to add N to the soil. This because some of the nitrogen they fix remains in the soil and benefits other crops that follow the legumes in the rotation.

d) Legume green manures increase soil fertility
A legume planted in the field and ploughed into the soil when it starts podding, is referred to as green manure. Green manures add all the fixed nitrogen and organic matter to the soil with nothing being taken out of the field. This cropping system provides maximum benefit to the crop that follows the green manure.

e) Legumes as cover crops in perennial crops:
Cover crops are crops planted between rows of perennial crops like bananas, coffee, fruit trees, etc, to protect the soil from erosion. Fast growing legumes like *Calopogonium* and *Pueraria* are preferred because they fix nitrogen and add it to the soil in addition to preventing soil erosion.

f) Using pasture legumes in pastures:
Mixing legumes and grasses is known to improve pasture quality and livestock production because pasture legumes are able to fix large amounts of nitrogen, and thus produce high protein forage.

g).Agroforestry legume trees improve soil fertility:
Agroforestry legume trees like *Leucaena* and *Calliandra* fix a lot of nitrogen. They are therefore used to improve soil fertility and increase the yields of crops.

h) Using azolla to increase paddy rice yields:
The Azolla-Anabaena symbiosis also fixes a lot of nitrogen. This BNF is widely exploited in the rice growing areas to increase yields.

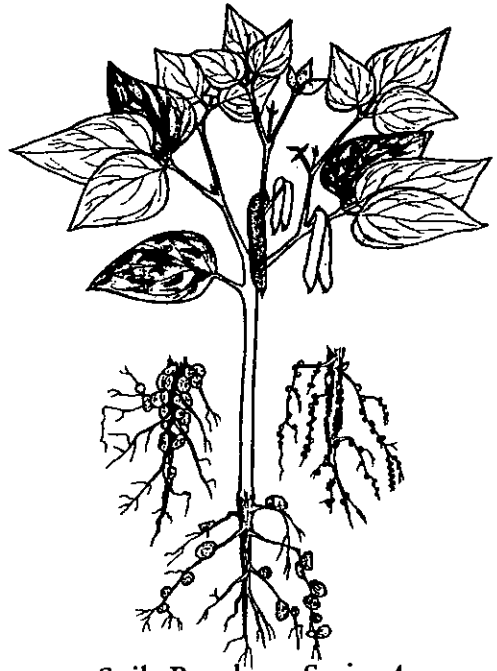
6. SOURCES OF RHIZOBIUM INOCULANT

Currently, Rhizobium inoculants for most of the important legumes in Uganda can be obtained from the Soil Science Department, Makerere University and from some agricultural input stockists around the country. An inoculant packet of 250g weight costs about Shs.1,200/= and this packet can be used to inoculate 16kg of legume seeds (enough to plant half an acre in the case of beans).

For more information, please contact:-

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BIOLOGICAL NITROGEN FIXATION (BNF)



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1. INTRODUCTION

Nitrogen (N) is a key component of all proteins and nucleic acids and hence all protoplasm, the substance from which all living things are made. It is therefore required in relatively large amounts by all living things.

Plants obtain nitrogen from the soil as nitrate (NO_3^-) and ammonium (NH_4^+) ions. However, only 0.04% of all the nitrogen on earth is found in the soil and oceans where it is mostly held up in humus and other organic materials from which it is released very slowly. It is estimated that only 1-2% of all the organic matter in the soil undergoes decomposition in a year, releasing about 22-45kg of nitrogen in a hectare of land. Yet plants take up nitrogen from the soil in relatively large amounts (Table 1). Thus out of the 16 plant nutrients taken up from the soil, nitrogen is quite often found to be the most deficient in many of the cultivated soils of the world.

Table 1: Amount of major nutrients taken up by crops.

Crop	Yield (t/ha)	nutrients taken up in kg/ha					
		N	P	K	Ca	Mg	S
Beans (Grain+Haulms)	10.0	240	73	192	120	55	28
Maize (Grain)	6.3	163	63	121	17	37	17
Cassava (Whole plant)	18.0	95	23	77	23	52	7
Bananas (Bunches)	22.5	148	30	420	154	66	14

2. NITROGEN FIXATION

The bulk (99.96%) of N on earth is found in the air (atmosphere) where it is present as an inert gas (N_2) that cannot be used by living things. It has to react with other elements like hydrogen (H) and oxygen (O) to form compounds, like ammonia (NH_3), which can be used by plants. This process of converting atmospheric N to nitrogenous compounds that can be used by plants and other living things is called "nitrogen fixation". This atmospheric nitrogen fixation is the main source of nitrogen for all living things on earth.

Nitrogen fixation is done in two ways:-

a) Artificially

Nitrogen gas (N_2) from the air is combined with hydrogen gas (H_2) from water or natural gas /methane (CH_4) to form ammonia (NH_3). This process which is used to manufacture inorganic industrial plant. The reaction occurs at a high temperature of 500°C, under high pressure (200-1000 atmospheres), in presence of a catalyst (magnesium). The process uses a lot of energy and is therefore very expensive, hence the high cost of inorganic fertilizers produced.

b) Biologically

Some micro-organisms (a few families of bacteria, algae and fungi) living freely in the soil or inside parts of plants (e.g., rhizobia living inside the nodules on the roots of legumes or the blue green algae *Anabaena*, living inside the small water plant known as *Azolla*), combine N gas from the air with hydrogen obtained from water in the soil to form ammonia (NH_3). This process, known as "Biological Nitrogen Fixation" (BNF), is carried out under normal soil conditions using an enzyme called nitrogenase found in micro-organisms that fix atmospheric N.

BNF also has a high energy demand. However, the energy used in this process comes from the microorganisms in the case of the free-living nitrogen fixers or from the plants for the microorganisms living inside some parts of plants. This makes BNF a very cheap source of nitrogen.

3. N ADDED TO SOIL THROUGH BNF.

Because of the high-energy demand of the BNF process, micro-organisms living freely in the soil are not able to fix much nitrogen— up to 25kg of N in a hectare of land annually.

However, micro-organisms living in symbiotic relationships with plants (e.g. rhizobia and *Anabaena* associations), are able to fix large amounts of nitrogen (Table 2), because they get energy in form of carbohydrates from the associated plants. It is therefore these microorganisms (rhizobia and Anabaena) which are very important as nitrogen fixers.

Table 2: N amount fixed by Legumes and Azolla

Legume	kg N fixed/ha	
a) Grain legumes:		
Groundnuts	37-247	per season
Pigeon peas	2-224	per season
Cowpeas	11-201	per season
Soybeans	159-227	per season
Field peas	48-75	per season
Common Beans	40-49	per season
Centrosema	126-395	per year
Macroptilium	46-147	per year
Alfalfa	200-250	per year
Clover	100-150	per year
c) Agroforestry trees:		
Leucaena	74-584	per year
Sesbania	542	per year
Calliandra	218	per year
Cassia	394	per year
d) Green manure/cover crops		
Calapogonium	126-182	per year
Pueraria	136-182	per year
Canavalia	142-206	per season
Crotalaria	63-188	per season
Mucuna	70-130	
e) Azolla in rice paddies	200-580	per season.

The amounts of N fixed by the legume-rhizobia and *Azolla-Anabaena* symbioses vary very much between different types of legumes and even within the same type of legume (Table 2). This is because the BNF process is affected by very many factors. Any factor that interferes with the proper growth and functioning of either the host plants or the microorganisms upsets the BNF process.

Some of the major factors affecting BNF include

a) High levels of available nitrogen in the soil suppress both nodulation and BNF.

b) The genetic potential of both the plants and the micro-organisms: some legumes e.g. soya beans can fix large amounts of nitrogen while others like common beans are genetically poor fixers; strains of rhizobia also vary widely in the amount of nitrogen they can fix.

c) Availability of other plant nutrients like phosphorus, iron, molybdenum, sulphur etc which affect the plant growth and many of which are important components of the nitrogenase, the enzyme responsible for BNF.

d) Adverse environmental and soil factors, e.g, prolonged drought, soils which are very acidic, very high soil temperatures, etc.

e) Poor management/handling of both plants and microorganisms.