As a key component of proteins, nitrogen is essential for all life. Nitrogen moves in a cycle from the atmosphere into the soil and plant system, through the food chain of animals and people, and back to the atmosphere. Most of the nitrogen available for cycling is in the atmosphere in a form that can only be used by certain bacteria. One group of bacteria called rhizobia can convert atmospheric nitrogen into a form that plants can use. Through a symbiotic association with members of the legume family, rhizobia convert atmospheric nitrogen to ammonia within root-nodules on the plants. This process is called biological nitrogen fixation (BNF). It is a crucial source of nitrogen for agriculture.

Nitrogen is the nutrient that is most often in short supply, limiting plant growth. For good crop production farmers usually need to add nitrogen to their soils, either as organic amendments, as inorganic fertilizer or by managing nitrogen fixation through the legume-rhizobia symbiosis. This third approach is the subject of this training manual.

**KEY CONCEPTS**

- Nitrogen is an essential element for all living things. It is a key component of proteins.
- Nitrogen moves through nature in a cyclic manner.
- Specialized bacteria can convert nitrogen from the atmosphere into a form that plants can use. This process is called biological nitrogen fixation (BNF).
- Through a symbiotic relationship with legume plants, rhizobia provide the plant with “fixed” nitrogen, which the plant uses for its growth.
- Inorganic nitrogen fertilizer is produced by chemical nitrogen fixation, an expensive process requiring nonrenewable energy inputs.
- Biological nitrogen fixation in the rhizobia-legume symbiosis is an inexpensive, valuable management option for farmers.
NITROGEN IN NATURE

All living things need nitrogen (N), because it is the key component of amino acids, the building blocks of proteins. Almost 98% of all nitrogen is bound-up in primary rocks in the earth, but this nitrogen is not available to cycle through plants, animals, soil, and air. The remaining 2% of `unbound' nitrogen cycles above and through the earth. It is distributed in the following proportions:

- In the atmosphere..........................................................99.96%
- In soil humus.................................................................0.02%
- In sea-bottom organic compounds....................................0.01%
- In living plants, animals, and microorganisms...................0.005%


The air is almost 80% nitrogen gas (N\textsubscript{2}), but plants and animals cannot use nitrogen directly from the air to make protein. Yet the amount of nitrogen present in all of the world's soil humus and living plants and animals is extremely small—far too little to support the continued productivity of natural or agricultural systems. These systems depend on atmospheric nitrogen that is converted, or fixed, into a chemical compound that plants can use.

The manufacture of nitrogen fertilizers involves chemical fixation of nitrogen. In nature, a small amount of nitrogen is converted through lightning discharges and volcanic emissions, but most nitrogen fixation is due to certain types of bacteria and other microorganisms. The process by which they convert atmospheric N\textsubscript{2} into a form that plants, and ultimately animals, can use is called biological nitrogen fixation or BNF.

Once N\textsubscript{2} from the air is fixed, it moves through nature in a series of interlocking, cyclic reactions. First it is transformed to NH\textsubscript{3} and then to protein. Protein accumulated by plants passes through the food chain to animals and people. Residues returned to the soil by plants, animals, and people also contain nitrogen as protein. These residues are decomposed by bacteria in the soil, in a process called mineralization.

The mineralization process results in the release of nitrogen as ammonium (NH\textsubscript{4}). Some ammonium is absorbed by plants through their roots, but most is converted by other soil bacteria to nitrate (NO\textsubscript{3}). This nitrate moves easily through water in the soil and is absorbed by plants. Within the plant, the nitrate (or ammonium) is converted into protein. Nitrate can also be changed back into nitrogen gas (N\textsubscript{2}), permitting its return to the atmosphere. This conversion, called denitrification, is done by other bacteria found in soil and water.
In summary, atmospheric $N_2$ is converted by specialized bacteria working in symbiosis with plants into $NH_3$—a process called biological nitrogen fixation. The plants then convert the $NH_3$ into proteins. Eventually these proteins enter the soil as residues. Other specialized bacteria convert the nitrogen in these residues into $NH_4$ and $NO_3$, which can be taken up by plants. Some $NO_3$ is converted back to atmospheric $N_2$. 
Nitrogen is a key component of all proteins, necessary for the growth, maintenance, and repair of body structures. The bones, muscles, skin, and other solid parts of the body are made up largely of proteins. Proteins also provide energy and help protect people against disease and malnutrition. We cannot synthesize our own proteins: We must obtain them from our food, either from animal products (cheese, eggs, meat, and milk) or from plant products (legumes, grains, nuts, and vegetables).

Table 1-1. Yield and protein content of certain tropical food crops.

<table>
<thead>
<tr>
<th>Crop</th>
<th>Estimated Yield$^a$ (kg/ha)</th>
<th>Protein Content$^a$ (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Legumes</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Soybean</td>
<td>2,800</td>
<td>38.0</td>
</tr>
<tr>
<td>Lima Bean</td>
<td>3,200</td>
<td>25.0</td>
</tr>
<tr>
<td>Cowpea</td>
<td>1,800</td>
<td>25.0</td>
</tr>
<tr>
<td>Peanut</td>
<td>1,600</td>
<td>26.0</td>
</tr>
<tr>
<td>Winged bean</td>
<td>1,400</td>
<td>31.0</td>
</tr>
<tr>
<td>Chickpea</td>
<td>2,500</td>
<td>20.0</td>
</tr>
<tr>
<td>Mungbean</td>
<td>900</td>
<td>24.0</td>
</tr>
<tr>
<td><strong>Root Crops</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sweet Potato</td>
<td>20,000</td>
<td>1.3</td>
</tr>
<tr>
<td>Potato</td>
<td>15,000</td>
<td>2.0</td>
</tr>
<tr>
<td>Cassava</td>
<td>20,000</td>
<td>1.2</td>
</tr>
<tr>
<td><strong>Cereals</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rice</td>
<td>5,000</td>
<td>7.5</td>
</tr>
<tr>
<td>Maize</td>
<td>4,000</td>
<td>9.5</td>
</tr>
<tr>
<td>Sorghum</td>
<td>3,500</td>
<td>10.1</td>
</tr>
</tbody>
</table>

For legumes and cereals, the yields and protein content are for harvested seed. From R.A. Luse and P.E. Okwuraie, 1975. Role of legumes in tropical nutrition, pp. 98-100. In R.A. Luse and K.O. Rachie (eds.) Proceedings of the IITA Collaborators’ Meeting on Grain Legume Improvement. Ibadan, Nigeria: International Institute of Tropical Agriculture
Legumes Are Rich in Protein

In many countries, legumes are a main source of dietary protein. The legumes are combined with cereals or grains for a complete protein balance. Although legume crops commonly have lower yields than cereal or root crops, their protein concentration is much higher, largely because of biological nitrogen fixation. Even with lower yields, farmers can obtain more protein from legumes than from cereal or root crops.

Figure 1-2. Protein: An important part of human nutrition

NITROGEN IN AGRICULTURE

In plants and soils, nitrogen occurs mainly in organic forms such as protein and soil humus. The amount of nitrogen in each form varies: Forests, for example, hold most of their nitrogen in plant biomass and litter, whereas grasslands and croplands usually hold most of their nitrogen in soil humus. In addition to these forms of organic nitrogen, ammonium and nitrate contribute a small percentage of the total nitrogen in plants and soils.

Farm Management is Important in the Nitrogen Cycle
Nitrogen is the most commonly deficient plant nutrient. Good management of crops and soils affects the amount of nitrogen available for plant growth. How much nitrogen a soil naturally supplies to plants depends on the rate of nitrogen mineralization, which in turn depends on the amount of organic nitrogen available and on conditions of temperature, water, and aeration. Fortunately, management of temperature, water and aeration to benefit crop productivity also promotes the activity of the bacteria responsible for nitrogen mineralization.

Soil nitrogen can be increased by adding organic matter taken from somewhere else, by growing green manure crops to till into the soil, or by conserving and incorporating crop residues. In the long term, soil nitrogen depends largely on plant growth, which is the source of most soil organic matter. This generalization leads to the simple rule that nitrogen fertility is maintained most effectively by residue conservation combined with agricultural practices that produce the highest yield and greatest biomass.

**Do Soils Have Enough Nitrogen for Crops?**

Even with high soil organic matter, nitrogen mineralization is rarely fast enough to meet the nitrogen demands of vigorously growing crops. Farmers therefore supplement the soil's ammonium or nitrate, either directly by adding inorganic nitrogen fertilizers or indirectly by adding organic materials or promoting biological nitrogen fixation.

![Energy requirements of synthesized N fertilizer](image)

**Figure 1.3. Energy requirements of synthesized N fertilizer**
Inorganic nitrogen fertilizer was first synthesized in 1921. It is produced industrially by chemically fixing the $\text{N}_2$ gas in the air to form ammonium. This requires energy-expensive components and processes, which accounts for most of the fertilizer's cost. The use of synthetic nitrogen fertilizer has grown tremendously since its introduction, representing a truly significant alteration of the nitrogen cycle. It is estimated that the annual amount of $\text{N}_2$ now fixed industrially—about 60 to 80 million tons—equals the amount fixed by biological nitrogen fixation.

**BIOLOGICAL NITROGEN FIXATION (BNF)**

Bacteria are the only organisms capable of biological nitrogen fixation (BNF). There are bacteria that carry out this process on their own, and others that fix nitrogen while housed within the root system of plants (symbiotic nitrogen fixation). While symbiotic nitrogen fixation is associated with several plant families, the fixation associated with legumes has the most benefits for agriculture. Symbiotic nitrogen fixation benefits both the bacteria and the plant, and can be managed to benefit entire agricultural systems. More specific information about symbiotic BNF is given in Module 4.

**Many Familiar Crops are Legumes That Support BNF**

The nitrogen-fixing symbiosis most beneficial to agriculture occurs between a type of soil bacteria called rhizobia and the legume plant family (Leguminosae). The legume family is large: It includes many familiar plants such as peanuts (groundnuts), soybeans, green beans, mungbeans, faba beans, cowpeas, chickpeas, lima beans and others, plus many pasture and tree crops. When these legumes meet the right rhizobia, they produce small growths on their roots, called nodules, where the rhizobia grow and fix nitrogen. The plant feeds the rhizobia the energy that they need for growth, and in return the rhizobia give up their fixed nitrogen to the plant, which converts it to protein. If other growth conditions are adequate, this process of biological nitrogen fixation can supply the legume with most or all of the nitrogen it needs for optimal yield, without addition of nitrogen fertilizer.

**Different Rhizobia for Different Legumes**

Rhizobia and legumes have evolved together over many millennia to become fairly specialized. For example, the rhizobia that cause nodules and fix nitrogen with faba beans cannot do so with soybeans, and vice versa. Rhizobia may be present in the soil but may fail to fix enough nitrogen for a legume plant, either because there are too few rhizobia or because they cannot fix nitrogen with that particular legume species. The appropriate rhizobia can be added to the seed before planting or to the seed bed. The process of adding rhizobia is called inoculation. Rhizobial inoculants are produced commercially and packaged with a carrier material such as peat. They are commonly available in countries where legumes are an important part of the agricultural production system. Module 3 discusses the specific rhizobia required by different legumes, and Module 5 explains the use of inoculants.
Figure 1-4. Root nodules of soybean (*Glycine max*) nitrogen fixing bacteria (rhizobia) are located within the nodules.

**How Do Rhizobia and Legumes Get Together?**

Rhizobia present in the soil or introduced by inoculation begin to multiply on the surface of the young roots of an emerging legume plant. They enter the roots through root hairs. After about two weeks, small bumps appear on the root. These bumps eventually become larger and mature into fully functional nodules. The nodules produce nitrogen for the legume throughout the growing season, with peak activity usually at about the time of flowering. The number, size, and activity of the nodules depend on several factors—most importantly on the number and effectiveness of the rhizobia present at planting plus all the environmental and management factors that influence the general growth and vigor of the plant. When the plant is harvested or dies, the nodules wither and fall off the roots. As they decay in the soil, some of the rhizobia are released. If soil fertility, temperature, and moisture conditions are favorable, the released rhizobia may survive in sufficient numbers to inoculate new plants of the same legume species the following season.
THE NEED FOR BNF IN AGRICULTURAL SYSTEMS

Since nitrogen is commonly the most important nutrient limiting crop-production systems, it must be managed properly to obtain good yields. This can be achieved by adding nitrogen fertilizer or manure, or through BNF in the legume-rhizobia symbiosis. Fertilizers are expensive, both in terms of the energy used to produce them and the cost to farmers. Manure may not be available in adequate amounts, and application can be difficult and labor intensive. BNF through inoculation of legumes offers an alternative that is fairly simple and inexpensive and over the years provides a large return on the time and money invested. The introduction and maintenance of effective rhizobial populations in a farmer’s field are valuable techniques for the production of nitrogen for legume crops.

Can Legumes Provide Nitrogen to Other Crops?

Nitrogen can be added to cropping systems by growing legumes as rotational crops, rotational pastures, green manures, intercrops, or range plants. The benefits that accrue from productive legume crops can extend to subsequent non-legume crops. The nitrogen in residues from the legumes becomes available to other crops through the process of nitrogen mineralization. The amount of nitrogen provided depends on several factors: These will be discussed in more detail in Module 4. Nitrogen fixation stops if there is already too much nitrogen in the soil or if plants are growing poorly due to some other factor. Nitrogen fixation by legumes is most effective in soils with low organic matter when legumes grow vigorously and are well nodulated.

Figure 1-5. BNF meets a legume’s need for nitrogen
BNF Has Other Advantages Compared to Fertilizer Nitrogen

It is easy to add too much inorganic nitrogen fertilizer to a cropping system. Crops usually use only one-half or less of the fertilizer nitrogen applied. Nitrogen fertilizer that is not used by plants can cause water pollution. Rain and irrigation water carry the unused fertilizer into streams or lakes, where the nitrogen compounds stimulate growth of algae and other water plants. Excess nitrate (NO$_3$) can leach into drinking water supplies, and high levels are toxic to humans.

Legumes can receive their nitrogen from BNF, without the problems associated with nitrogen fertilizer use. In addition, the cost of nitrogen fertilizer can be too high for many farmers—rhizobial inoculant costs much less.

REVIEW AND DISCUSSION

- What are the common legume crops grown and eaten by farmers in your area?
- Have you observed nodules on these legumes?
- Are these legumes commonly inoculated with rhizobia?
- How will the market price of oil/gas influence the cost of nitrogen fertilizer? Will these changes influence cropping systems?
- Can you define a typical cropping system in your region and develop a management strategy that improves the nitrogen balance of the system? Include realistic inputs and outputs of nitrogen in the system.
SUGGESTED LESSON PLAN FOR MODULE 1

TIME: One Hour +

This lesson can be taught in a very short time, especially if participants are trained in agriculture or a related field.

OBJECTIVES:

Understanding the important role of nitrogen in nature and in human nutrition. Knowing that BNF is an important part of nitrogen cycling in the environment

MATERIALS:

Demonstrations D1/1 and D1/2

Training Aids for Module 1

STEPS:

1. Assemble materials for demonstrations. Display key concepts and other visual materials.

2. Using the Module as a basis, organize your talk according to the background and skill level of your audience.

For example, begin with the broad theme of nitrogen in the environment, and move toward the more specific themes of nitrogen in agriculture, Biological Nitrogen Fixation, and the role of nitrogen in human nutrition.

3. Be sure to begin by asking questions to get people involved in the learning process and to become familiar with their knowledge level. This step is important in every module, as well as asking questions at the end of the presentation to check learning, because each module presents information that builds knowledge necessary to understand the following modules. The case studies at the end of each module should help with this.
KEY CONCEPTS

Nitrogen is an essential element for all living organisms. It is a key component of proteins.

Nitrogen moves through nature in a cyclic manner.

Bacteria are able to convert atmospheric N to ammonia in a process called biological nitrogen fixation (BNF).

In the rhizobia-legume symbiosis, rhizobia provide the plant with fixed N, which the plant uses for its growth. Inorganic N fertilizer is produced by chemical nitrogen fixation, requiring nonrenewable energy inputs.

Biological Nitrogen Fixation in the Rhizobia-legume symbiosis is an inexpensive, valuable resource option for a farmer.
Protein: An Important Part of Human Nutrition

Concentrated protein is available to people through animal products or legumes.
BNF Meets A Legume's Need for Nitrogen