Soybean Nodulation Process

- Nitrogen (N) fixation is a symbiotic process between soybean plants and rhizobia soil bacteria where atmospheric N is converted to a form that is available to the plants.
- Nitrogen fixation occurs in soybean root nodules and begins when the plant is between the V2 and V3 growth stages and reaches its maximum level when the plant is at about R5.5 stage.
- Understanding the factors that lead to nodulation failure can help avoid N deficiency and potential yield reduction in soybean.

As soybean planting season approaches, farmers may be thinking of planting a field that is new to a soybean crop or has been out of soybean production for several years. These situations may cause concerns in achieving enough root nodulation for adequate N fixation throughout the growing season, which may reduce yield potential.

How are Nodules Formed?

For successful atmospheric N fixation to occur, adequate populations of N-fixing bacteria (*Bradyrhizobia japonicum,* in the genus Rhizobium) should either be available in the soil or applied to soybean seed to help nodules form on the roots.

Shortly after seedlings emerge, the first nodules are formed and become visible as they increase in size. The initial step in nodulation is a successful penetration of the bacteria into the root hair of a soybean seedling and the formation of an infection thread. The thread forms and grows to the base of the root hair. Root nodules may result from multiple infection threads or double infections from a single thread. Swelling develops near the tip of the infection thread.

Around the V2 to V3 stage, the N fixation process begins in the nodule.[1] Rhizobium bacteria convert atmospheric N to ammonium (NH_4), which is a form of N available to the plant, and in turn, the plant provides carbohydrates to the bacteria to survive. A successful nodulation by the V3 to V4 growth stage should produce 8 to 10 healthy nodules per plant. The number of nodules per healthy plant (several hundred) and the amount of N fixed is maximized around the R5.5 stage.[1, 2]

Nitrogen Fertilization

The application of N fertilizer to a soybean crop is not recommended, as it generally does not increase yield potential.[1] The greater the supply of N in the soil, the less N fixation occurs. As the amounts of applied N increase, the number of nodules decreases and the bacteria become less active.[2] Soybean planted in fields with excessive residual nitrate should be closely monitored. If nodulation has been severely inhibited and N deficiency symptoms appear (Figure 1), additional N during pod fill may be helpful.[5] Research has shown conflicting results regarding late-season N applications, with the greatest success occurring when N was applied via irrigation system in high-yield situations.[5]

Figure 1. Yellow leaves due to temporary N deficiency caused by wet soil and reduced nodulation.

Nodule Viability Assessment

After the second or third trifoliate has emerged, dig up a few (at least 10) soybean plants, without pulling to avoid stripping nodules off the roots. After digging, soak plants in a bucket of water to loosen the soil so that you don't remove too many nodules. If nodules are present, they should be large and active. If there are less than five nodules per plant, resample the same field one week later. Just prior to flowering, there should be 8 to 20 large (2 to 4 mm) and active nodules per plant (Figure 2). Nodules found on the tap roots are probably the result of the current season's inoculation. Nodules developed on the lateral roots may be from existing rhizobium bacteria, depending on the bacteria's movement through the soil or if soil-applied inoculants were used. New nodules are formed throughout much of the season, ending during pod-filling.

If the internal tissue of a dissected nodule is pink to red in color, the nodule is active and N fixation is occurring, but this is not a measure of the efficiency of the nodule. Some nodules are pink to red but are not very effective at

Figure 2. Comparison between a well nodulated plant (left) and a poorly nodulated plant (right). Photo courtesy of Stu Duncan, Kansas State University.

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producing ammonium. Nodules that are green, brown, or mushy are not fixing N, while immature nodules are small, white, and have yet to fix N.

Rhizobium can be added as a liquid, as granular peat inoculants, or as a peat-based powder if soils do not contain high populations. They can be seed-applied or used in-furrow.

When are Inoculants Most Important?

Inoculants can have a positive impact in many growing situations including the following:

Figure 3. Photo courtesy of Stu Duncan, Kansas State University.

- Field has no previous history of soybean production (Figure 3).
- Soil pH is below 6.0. Greater response from inoculants can be expected in fields with a pH below 6.0.
- Fields that have been flooded for several days can create anaerobic conditions for the rhizobia.
- Compaction and cool soil temperatures, due to no-till practices, can reduce nodulation.
- Fields with sandy soils and low organic matter (less than 1%) need to be inoculated every year.[2] These soils generally have very low populations of rhizobia bacteria.

Unsuccessful Nodulation

The following conditions are most likely to cause poor nodulation and reduce N fixation:

- Fields with low soil rhizobia bacteria populations and/or fields with a high residual of soil N from a previous forage legume, such as alfalfa, clover, or other crop, or due to manure application.

  Fields new to soybean should always be double inoculated with one application preferably to the soil. Applying inoculants to both the seed and soil can potentially increase nodulation.
- Poor quality inoculants due to improper storage time and conditions.

  Follow the inoculant expiration date and proper storage conditions to preserve the inoculant's viability. Additionally, avoid exposing inoculant to direct sunlight or excessive heat.
- Dry conditions, excessive moisture, or flooding for several days. Due to anaerobic conditions, nodules rot, turn brown, and die if soils are saturated for at least three days.
- Hail damage, root diseases, or iron deficiency chlorosis (IDC) symptoms early in the season.
- Soil pH levels below 6.0 or above 8.0.[3]
- Soil compaction can limit rooting and the development of root hairs that are hosts for rhizobia to colonize and develop root nodules.
- Symptoms of inadequate nodulation include yellowing and stunting of soybean plants.

Summary

- The symbiotic relationship of a soybean plant with soil rhizobia bacteria converts atmospheric N into usable ammonia (NH_4) within plant root nodules that grow on the root system.
- Between 50 to 75% of the soybean total N requirement is obtained through the N fixation process.
- Provided adequate nodulation and N fixation, soybean plants can grow well in low N soils.
- Healthy and active dissected nodules are pink to red in color.

Sources:
[1] Pedersen, P. 2004. When do we need to inoculate our soybean seeds? Integrated Crop Management. Iowa State University. www.ipm.iastate.edu.
[2] Pedersen, P. 2003. Soybean seed inoculation. Integrated Crop Management. Iowa State University. www.ipm.iastate.edu.
[3] Staton, M. 2014. Identifying and responding to soybean inoculation failures. Michigan State University. http://msue.anr.msu.edu.
[4] Kandel, H. 2012. Soybean nodulation. Crop and Pest Report. North Dakota State University. www.ag.ndsu.edu.
[5] Larson, K., Rice, C., and Roozeboom, K. 2012. Successful soybean nodulation without rhizobia. 344 Agronomy e-Updates, Kansas State University Extension.
We would like to thank Dr. Stu Duncan, Kansas State University, for granting permission for the reprint of soybean images.
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Web sources verified 3/30/2015.

For additional agronomic information, please contact your local seed representative.