Pulse Advisor

SASKATCHEWAN pulse Growers

Nodulation and Nitrogen Fixation Field Assessment Guide

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Accurate field measurements of nitrogen fixation responses to inoculation with *Rhizobium* are often difficult, undependable, and expensive. However, nitrogen fixation can be estimated through an assessment of nodulation and plant growth characteristics.

This guide will help growers and agronomists learn how to assess nodulation and nitrogen fixing potential in the field.

Nodule Assessment Timing

Nodulation assessments should be done during early flowering. Nodule formation begins approximately 14 days after crop emergence, but under certain conditions formation may take three to four weeks.

Nodule numbers and nitrogen fixation rates are generally at a maximum during early- to mid-flowering. After flowering, nodule efficiency is reduced and they begin to shut down.

Figure 1. Faba bean flowering.
Source: Saskatchewan Ministry of Agriculture

Assessment Procedure

To assess the nodulation and nitrogen fixation potential of a pulse crop, select five areas that are typical of that field at early flowering. Follow the steps listed below in each of the five areas:

1. Evaluate plant growth and vigour of the area according to the assessment codes shown in the following column.
2. With a shovel, carefully dig up a minimum of two plants per area. Do not pull plants out of soil as nodules are delicately attached to roots and can be easily lost.
3. Carefully examine plant roots to assess the nodules. Depending on the soil type and condition, this may require gently agitating the roots in water.
4. Assess the overall nodulation by comparing the calculated scores to those provided for the three categories in the assessment guide.

Assessment Codes

1. **Plant Growth and Vigour**
   Assess colour and overall health of the plant

• Plants are green and vigorous	5
• Plants are green and relatively small	3
• Plants slightly chlorotic (yellow)	2
• Plants very chlorotic	1

Poor nitrogen fixation can cause nitrogen deficiency symptoms such as yellowing of the leaves at the base of the plant prior to flowering and poor plant development.

Figure 2. Good (left) and poor (right) nodulation in a pulse crop.
Source: Saskatchewan Ministry of Agriculture

2. **Colour and Abundance**

• Greater than five clusters of pink pigmented nodules	5
• Three to five cluster groups of mostly pink nodules	3
• Less than three clusters of nodules OR white or green nodules	1
• No nodules OR white or green nodules	0

Nitrogen fixation efficiency can be estimated with nodule color and the number of nodule clusters present. Carefully slice open the nodules. The strong pink color of the nodules is caused by the presence of leghaemoglobin, which must be present for active nitrogen fixation. If a nodule is brown, white, or green it is considered non-effective.

Figure 3. Assess colour and abundance.
Source: Saskatchewan Ministry of Agriculture

3. **Nodule Position**

- Both crown and lateral nodulation 3
- Mostly crown nodulation only 2
- Mostly lateral nodulation only 1

Predominantly crown nodulation is observed when seed is inoculated. Lateral nodulation is prevalent when native rhizobia species exist in the soil or when granular inoculants are used. The crown region of a plant is generally the area of soil surrounding the seed. The approximate size of this region varies according to the crop.

Figure 4. Nodule position.
Source: Saskatchewan Ministry of Agriculture

4. **Total Score**

11—13 **Effective Nodulation**
Numerous nodules that have good nitrogen fixing potential.

7—10 **Nodulation Less Effective**
Nodules present with limited nitrogen fixing potential.

1—6 **Poor Nodulation**
Few nodules present with very little to no nitrogen fixation potential.

In the field, a healthy plant does not always reflect effective nodulation and active nitrogen fixation. Localized soil environments, particularly with variations in soil nitrogen, may stimulate vigorous growth of the plant. Such situations are only apparent when the plants are excavated and examined for the presence of active nitrogen-fixing nodules.

Figure 5. Example assessment.
Source: Saskatchewan Ministry of Agriculture

Example Assessment Effective Nodulation *(Figure 5)*

Left Plant
Plant Growth/Vigour—5
Nodule Colour/Number—3
Nodule Position—3

Total Score—11

Right Plant
Plant Growth/Vigour—1
Nodule Colour/Number—1
Nodule Position—2

Total Score—4

Nodulating Rhizobia

Recently researchers found that inoculation rates higher than 105 cells/seed were usually required for high nodulation, nitrogen fixation, and grain yields. Therefore, Canadian standards, which require that 105 nodulating rhizobia be delivered per seed for large-seed legumes like peas, may need to be increased. They also found that coated seeds only provided benefits in acid soils, and provided little economic benefit compared to regular inoculant. Overall, population of rhizobia per seed is a critical factor. (Source: Rice et al, Evaluation of coated seeds as a *Rhizobium* delivery system for field pea)

Figure 6. Soybean roots and nodules.
Source: Saskatchewan Ministry of Agriculture

Faba Bean Inoculation

In a recent study researchers found that, in faba bean, un-inoculated and inoculated plants nodulated equally well, suggesting the presence of adequate populations of effective indigenous *Rhizobium leguminosarum bv. viciae* for nodulation of untreated plants. The indigenous rhizobia could have originated from previous field pea (*Pisum sativum L.*) crops or leguminous native plants/weeds. (Source: Ken J. Lopetinsky, et al, Contrasting Rhizobium inoculation requirements of zero-tannin faba bean and narrow-leafed lupin in Western Canada)

Meanwhile, other studies have shown beneficial results by inoculating faba beans. So, by comparing the cost of nitrogen to inoculant, it pays to inoculate. Further evaluation is giving us a better understanding of faba bean's response to granular and peat inoculants in various locations across Saskatchewan.

Figure 7. Faba bean soil vs. fresh inoculant.
Source: Garry Hnatowich

Nodulation Failure

If nodulation does not occur, or is poor, it is possible to salvage the pulse crop. According to researchers from Manitoba who studied nodulation failure on soybeans, it is best to wait to the early pod fill stage and wait for early rainfall to get nitrogen into the rooting zone. In total, soybeans will generally remove 150-200 pounds per acre (lb/ac) of nitrogen to produce 30 bushels per acre (bu/ac) of crop. According to the Canadian Fertilizer Institute, on average field peas remove 105-129 lb/ac of nitrogen to produce a 50 bu/ac crop. Lentils remove 55-67 lb/ac of nitrogen to produce a 30 bu/ac crop. Faba beans remove 154-188 lb/ac of nitrogen to produce a 50 bu/ac crop. Actual uptake and removal will vary with crop yield, crop variety, soil fertility, and from year to year. Accurate removal values can only be determined by laboratory analysis.

In the soybean trial, early application just appeared as vegetative growth. Adding nitrogen at pod fill, and also in conjunction with a rain event provides the best value of nitrogen. The field had 50 lb nitrogen residual in the soil. Higher rates of nitrogen may only keep the crop in a vegetative state longer and will not guarantee higher yields. In a salvage operation, the idea is to salvage enough yield to break even or better.

With peas and lentils a scenario was posed to pulse experts Dr. Bert Vandenberg with Crop Development Centre at the University of Saskatchewan, and Dr. Yantai Gan with Agriculture and Agri-Food Canada at Swift Current regarding what to do if inoculant was not put down at seeding time or inoculant failure has occurred. Both suggest that growers can apply 50 to 60 lb/ac of actual nitrogen as early as possible if nodulation failure has occurred or is expected, such as when inoculant application is absent or reduced. If peas or lentils have been grown in the field previously there may be enough background rhizobia to induce nodulation. Under these conditions one can wait to see if nodulation occurs at early flowering. If no nodulation is apparent, then the grower should fertilize according to recommendations as soon as possible. More studies are required to better understand the response lentils, field peas, and faba beans might have to variable time of application.

Table 1. Yield and Quality Components for Soybeans With Application of Nitrogen Fertilizer

Treatment	Yield (bu/ac)	Protein %	Nitrogen in Seed (lb N/ac)	Oil %	Seeds/lb
Check	31.4	32.8	99	23.6	2,996
50 Nitrogen at Flowering	33.7	33.8	109	23.2	2,989
50 Nitrogen at Pod Fill	39.0	34.7	130	23.0	2,915
100 Nitrogen at Flowering	33.6	36.5	118	22.2	2,792
100 Nitrogen at Pod Fill	41.4	36.0	147	22.0	2,709
100 Nitrogen at Emergence	31.5				

Source: Heard, Lange, Peters