SOUTHERN AND WESTERN REGIONS
JUNE 2017

GRDC GROWNOTES™

TIPS & TACTICS

LEGUMES AND NITROGEN FIXATION

GRDC GRAINS RESEARCH & DEVELOPMENT CORPORATION

Use the four-step 'nod check' to assess your legume nodulation

For the current legume crop to gain nitrogen (N), and to increase soil N for future rotations, legumes need to be adequately nodulated by root nodule bacteria (rhizobia). Nodulation is important for lupin, lentil, field pea, faba bean, vetch and chickpea crops and for pastures (e.g. medic).

The national benefit of rhizobia-fixed nitrogen (N) for Australia's pasture and pulse legumes has been estimated at $2.4 billion a year. This is based on N fixation rates of about 110 kg N/ha/year, legume areas of 25 million ha and N fertiliser (urea) costs to the grower of $0.70/kg N (equating to $0.88/kg plant-available N in the soil).

Photo 1: Lupin roots with adequate nodulation will fix substantially more N than roots with poor nodulation. Nodules on the plant on the right have been cut to show the red/pink colour inside the nodules, which indicate N fixation.
Source: Ross Ballard, SARDI

KEY POINTS

- All legumes sown by Australian growers require specific root nodule bacteria (rhizobia) to fix nitrogen (N).
- Roots need to have adequate numbers of active nodules to ensure optimal amounts of N will be fixed by the rhizobia.
- Nodules can form from soil rhizobia or through inoculation.
- Growers should investigate the extent of nodulation in grain and pasture legumes, irrespective of whether the seed was inoculated or not.
- Conduct 'nod checks' in late winter or early spring; ideally 10–12 weeks after sowing.
- Aim for a score of 'adequate' to be confident your crops have a sufficient number of nodules to fix abundant N.

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An easy four-step 'nod check'

Has your inoculation been successful?

If you did not inoculate, should you do so next time?

To answer these questions, growers should conduct nodulation assessments in late winter or early spring (about 10–12 weeks after sowing) using this four-step process.

You'll need some basic equipment (Photo 2): buckets, a shovel and water at least.

In addition, a clean chemical drum cut in half or a white piece of plastic can help to examine the clean roots more easily.

Photo 2: Mandatory equipment needed to collect samples and check for nodulation: buckets, a shovel and water.
Source: Maarten Ryder, University of Adelaide

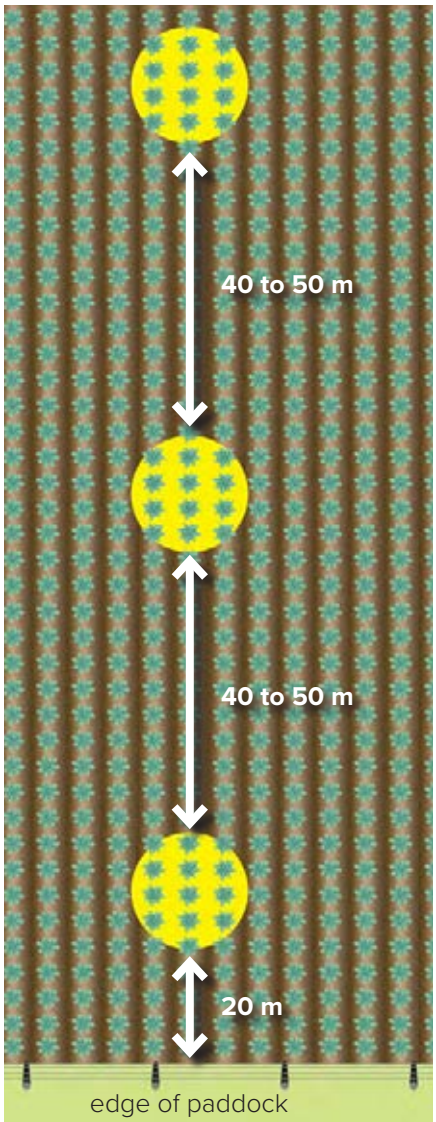


Figure 1: Sampling pattern for collecting plants. Sample at the yellow marks beginning 20 m from the edge of the paddock.
Source: Maarten Ryder

Step 1. Collect samples

Collect about 30 plants: 10 plants at 3 sample spots (Figure 1). Put each sample of 10 in a separate bucket.

Step 2. Wash roots

Carefully wash off the soil in a bucket of water and rinse roots once or twice to remove remaining soil.

You may need to soak samples for up to 30 minutes for heavy soils.

Step 3. Score each plant

The best way to examine the roots is to float them in a few cm of water in a clean chemical drum cut in half or on top of a white piece of plastic.

Assess each plant as having either 'adequate' or 'poor' nodulation. Refer to photos of adequate and poor nodulation for the relevant crop or pasture legume starting from page 3.

Break open or cut a few nodules to confirm that most are a red/pink colour inside (indicating that nodules are actively fixing N) before assigning a final score.

4. Determine the overall average nodulation rating

Work out the percentage of plants adequately nodulated from each sample site. Then average this score across the three sampling locations. Use Table 1 to determine the rating for the crop in that paddock.

Table 1. Determine the overall (average) nodulation rating using the rating system: adequate, borderline, poor, none.

Rating	Percentage of plants with nodulation similar to or better than "adequate"
Adequate	70% or more
Borderline	50–70%
Poor	Less than 50%
None	No nodules present = no nitrogen fixation

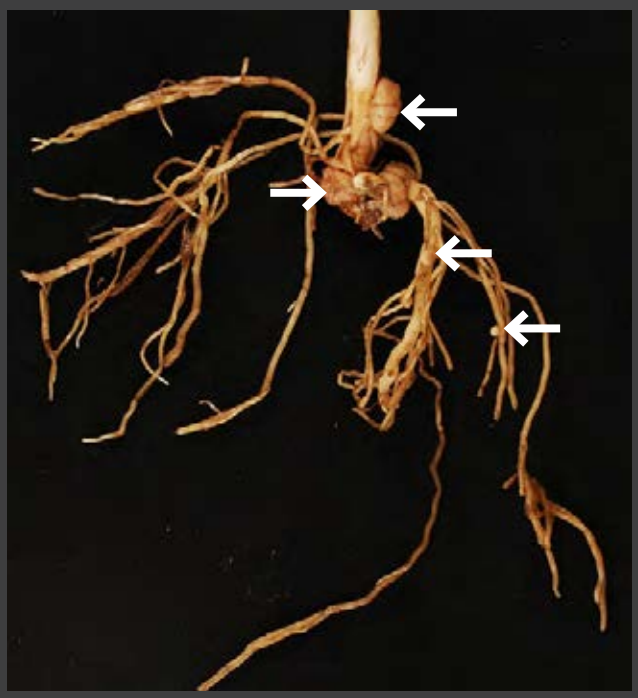
LUPIN

Adequate

Source: Ross Ballard, SARDI

Many nodules around the crown and on laterals; Plant on right: nodules have been sliced open to reveal pink interior (arrowed)

Poor



Source: Maarten Ryder, University of Adelaide

Few nodules (arrowed)

Important note: All normal lupin roots can have a pinkish layer inside the root that is unrelated to nodulation and nitrogen fixation.

FIELD PEA AND VETCH

Adequate

Source: Liz Farquharson SARDI

50 to 100 nodules

Poor

Source: Maarten Ryder, University of Adelaide

Less than 20 nodules

STRAND MEDIC seedling

Adequate

Source: Ross Ballard, SARDI

5 to 10 nodules per plant (8 in this case)

Poor

Source: Ross Ballard, SARDI

3 nodules

CHICKPEA

Adequate

Source: Andrew Heath

Adequate: 10 to 30 nodules;
(note multi-lobed nodules around crown)

Poor

Source: Maarten Ryder, University of Adelaide

FABA BEAN

Adequate

Source: Maarten Ryder, University of Adelaide

50 to 100 nodules per plant

Poor

Source: Maarten Ryder, University of Adelaide

Less than 20 nodules per plant

LENTIL

Adequate

Source: Maarten Ryder, University of Adelaide

50 to 100 nodules per plant

Poor

Source: Maarten Ryder, University of Adelaide

Less than 20 nodules

The benefits of inoculation with rhizobia

Over a decade of research (including the National Rhizobium Program and the Nitrogen Fixation Program) has confirmed that there are significant benefits from inoculating pulses and pastures with rhizobia.

Growers should consider inoculating their grain legume and pasture seed to boost the N fixation in their paddocks after assessing paddock history and soil conditions, particularly soil pH.

Where a legume has been grown in the paddock previously and where soil conditions are conducive to the survival of rhizobia (e.g. neutral to alkaline pH), inoculation may not be needed. However, in acidic soils (except for lupins) or where the legume has not been grown before, inoculation is strongly recommended as a positive response to inoculation can be expected (GRDC, 2012).

A recent trial showed that inoculated faba beans in western Victoria yielded 2.7 t/ha. This was a yield of 1 t/ha more than the uninoculated crop. The inoculated crop also added 180 kg/ha of extra fixed N to the soil, emphasising the extra value of grain legumes to the farming system (Denton et al., 2013).

Responses are not always obvious and even when the impact on legume yield is small, inoculating legumes with rhizobia can deliver substantial improvements in fixed N inputs to the farming system more generally. In addition, the cost of inoculation is small, relative to the potential benefits.

The price of carbon-based fossil fuels, used to produce nitrogenous fertilisers, is expected to increase substantially in the future. As this occurs, the value of fixed nitrogen from legumes to Australian growers will increase.

What to do next if the nodulation score is poor

1. Sample elsewhere in the paddock to see if it is a localised problem or not.
2. Complete the Troubleshooting questions (box to the right).
3. Look for further information. Refer to 'Useful resources' on page 8.

Poor nodulation can be caused by a number of factors including low rhizobia numbers, problems with incorrect use of inoculant or crop stress.

TROUBLESHOOTING QUESTIONS: POSSIBLE REASONS FOR POOR NODULATION

- Was the legume dry sown into a paddock without a background of suitable rhizobia?
- Was the legume sown into acidic soil (pH less than 5.5 in $CaCl_2$)? The exception is lupin, which can nodulate in acidic soils.
- Should you have inoculated because rhizobia numbers were low in the soil?
- Was the soil waterlogged for an extended period during the growing season? Or were there other factors that caused crop stress such as poor nutrition or root disease?
- Is there evidence of herbicide damage? For example, Group B herbicide (e.g. sulfonylurea) residues in alkaline soils can dramatically inhibit nodulation of herbicide-tolerant legumes in following years, as these herbicides are slow to break down in these soils.
- Was the correct inoculant group used? Always check the labels to make sure.
- Was the inoculant mixed with low quality water (e.g. saline or chlorinated)?
- Was the inoculant mixed with other amendments that reduced its survival? For example, peat slurry or freeze-dried inoculant can be affected by mixing with certain types of seed dressings.
- Was the inoculant combined with fertilisers, pesticides, trace elements or organic amendments either in the spray tank or during application to the soil? Many of these are toxic to rhizobia.
- Was the inoculant stored correctly? It should have been stored in cool conditions before use.
- Did you leave inoculated seed unsown for too long instead of following recommended timeframes for sowing after inoculation?

Note: If it is the first time growing a particular legume crop in a paddock, or the soil is very acidic, the rate of inoculant application can be doubled. When doubling the inoculant rate for seed coating, a test run is recommended, to ensure the coated seed does not cause blockage of seeders and augers.

Can I inoculate my crops after sowing?

By the time poor nodulation is detected it is usually difficult to remedy. Poorly nodulated pulse crops will use more soil N than adequately nodulated crops, and will fix less N from the air.

By assessing nodulation, growers can see how successful inoculation has been, and, for uninoculated legumes, whether inoculation is likely to be beneficial next time the crop is grown in that paddock.

Inoculant is always best applied at sowing; however, where a problem with the inoculation process is known to have occurred (e.g. wrong inoculant used) then possible remedies, if applied immediately, include:

- adding slurry or liquid inoculant to the irrigation water for flood or sprinkler-irrigated fields only
- drilling in a granular inoculant close to the original sowing furrow or oversowing with a low rate of inoculated seed, before germination.

Responses to remedial inoculation will decline within days or weeks of plant emergence, as mature roots are less likely to form nodules. After this time, the only option to increase N availability for the crop is to apply N fertiliser where it is cost effective.

FAQs

Will N fixation be different if the nodules formed from existing soil rhizobia or though inoculation?

There can be differences in the location of nodules on the root system. Where growers have inoculated the seed of a first-time crop, they may see nodules restricted to the crown of the plant (where root meets shoot) and the upper taproot. These nodules will boost the early growth of seedlings. Where there is a background of correct soil rhizobia, legumes will have nodules spread more widely over the root system on crown, taproots and laterals, because the rhizobia are distributed through the soil. Commercial inoculant rhizobia are generally more effective in fixing nitrogen than background soil rhizobia, so there may be added N fixation benefits from the nodules produced by fresh inoculation.

How can I be sure that the inoculant I'm considering is effective?

Inoculants that carry the 'Green Tick' logo (meaning they are approved by the Australian Inoculants Research Group) meet minimum quality standards (purity and number of rhizobia per gram of product). The use of inoculants displaying the 'Green Tick' logo is strongly recommended.

I forgot to inoculate my seeds. Can I spray inoculant onto the top of the soil or directly onto the legume crop or pasture?

Unless there is access to irrigation water to enable the rapid movement of inoculant into the soil, this approach does not have a high chance of success. Drilling in a low rate of inoculated seed or the recommended rate of granular inoculant may be helpful prior to germination (see 'Can I inoculate my crops after sowing').

Apart from inoculating, what else can I do to improve nodulation?

The most important factors are soil nutrition (particularly soil N levels) and herbicide residues. If soil N levels are very high, nodule formation will be decreased; the plant will opt to take up the available N from the soil. Residues from some herbicides, e.g. sulfonylurea, are known to retard root growth and development and the ability of roots to form nodules and then fix N. This is more often seen on alkaline soils. A larger legume biomass will fix more N. Managing time of sowing, crop nutrition and disease and weed pressures to optimise dry matter production of the pulse crop will generally encourage more N fixation. (Read more about N fixation by field pea in Farquharson *et al*, 2016.)

How much should I pay attention to the colour of the nodules?

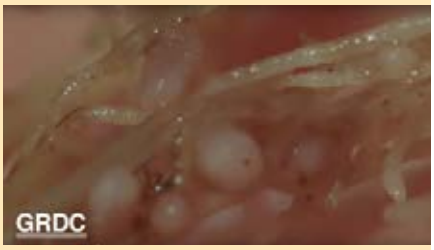
A red/pink colour inside the nodule indicates it is fixing N so it is important to look for this. However, growers need to note that lupin roots have a pink-coloured layer inside and this layer is not a nitrogen-fixing nodule.

VIDEO SERIES: TOP TIPS FOR SAMPLING AND ASSESSING NODULATION

Maarten Ryder explains why nodulation is important, how to sample plants and the steps to assess nodulation for the paddock.

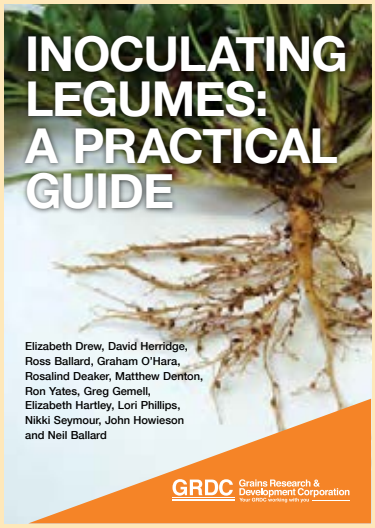
Sampling plants
https://grdc.com.au/archive/video/2015/09/episode-17-september-2015/bfnbsem64t0

Washing and preparing plants
https://grdc.com.au/archive/video/2015/09/episode-17-september-2015/0vl7cly-k9w



Scoring nodulation
https://grdc.com.au/archive/video/2015/09/episode-17-september-2015/nd303sfltdk

USEFUL RESOURCES



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GRDC (2015) GroundCover TV Episode 17, September 2015, https://grdc.com.au/Media-Centre/GroundCover-TV/2015/09/Episode-17-September-2015

GRDC (2013) Inoculating Legumes: The Back Pocket Guide, September 2013, https://grdc.com.au/resources-and-publications/all-publications/publications/2013/09/grdc-bpg-inoculatinglegumes

MORE INFORMATION

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E Farquharson, N Charman and R Ballard (2016) Optimising nitrogen fixation in southern farming systems. Grains Research and Development Corporation, 9 February 2016 https://grdc.com.au/Research-and-Development/GRDC-Update-Papers/2016/02/Optimising-nitrogen-fixation-in-southern-farming-systems

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