Australian Inoculants Research Group

Rhizobial Inoculation

Improved quality and productivity in Australia's variable climate: Grain Legumes

Grain legumes are valuable break crops in Australian cropping systems as they reduce production risks associated with weeds, pests and diseases as well as contribute to improvements in soil health. Some pulses are also considered valuable cash crops in their own right. Certainly, they play an essential role in nitrogen (N) supply to subsequent field crops through their mutually symbiotic association with rhizobia, a group of specialised soil bacteria.

Why is relationship between grain legumes and rhizobia beneficial?

Rhizobia infect the roots of grain legumes during seed germination, then multiply rapidly to form root nodules. They are dependent on the host for water, nutrients and energy. In exchange, they supply N in a form that can be readily taken-up by the developing plant to boost growth. This conversion of gaseous atmospheric N (N_2) into ammonia (NH_3) is called N fixation. Concentrated N residues are also left in the soil after harvest of a well nodulated legume crop, providing an enduring legacy of N for following crops of an intensive rotation system, used to optimise grain yield and protein. The value of this beneficial biological relationship is equivalent to 2.7 million tonnes of manufactured N being applied in Australian agriculture (GRDC, 2014).

There is variation across grain legumes in capability to fix N, from weak (e.g. Navy bean) to strong (e.g. Lupin). The more biomass a crop produces, the more N it fixes. Therefore, environmental factors that impact upon plant vigour, such as soil moisture or soil constraints, will also affect the rate of N fixation. Any short-fall in the crop N budget has to be supplied from soil mineralised or fertiliser N sources. Conversely, if there is an over-supply of N from these sources, rhizobia are less active and nodulation and N fixation become suppressed.

When is inoculation needed to ensure a beneficial relationship between grain legumes and rhizobia?

The purpose of inoculation is to supply selected elite rhizobial strains in large numbers to the roots of the legumes soon after germination to optimise effective nodulation, symbiotic N fixation and grain yield, while decreasing input costs. Although Australian soils have developed substantial populations of rhizobia, the suitable strain for the selected legume crop may be absent from the soil if the legume has not previously been grown or where long-term rhizobial survival may be impacted by certain soil constraints such as acidity or prolonged drying. Grain legumes must be inoculated with the correct rhizobial strain for nodulation and N fixation.

Inoculant products are specifically manufactured to contain live, unique rhizobia that are highly effective in nodulation and N fixation for a specific host group of legumes. When applied in sufficiently high numbers inoculant strains will successfully outcompete established soil rhizobia.

Inoculate when:

- the particular legume has not been grown in the paddock previously, or it has been longer than 4 years since the legume was grown
- there are newly introduced advanced strains with increased effectiveness and survival than those used in the paddock in the past
- the presence of acidic or highly alkaline soils in the paddock may limit survival of the rhizobia in the soil
- the paddock is subjected to particularly prolonged hot, dry conditions
- the legume has specific rhizobial requirements, e.g. chickpea

Latest research recommends that rates should be doubled if the paddock is being sown to a new grain legume, soil moisture is limiting but rain is forecast within 7 days and if the soil is very acidic (<5.5 pH_{Ca}).

The Australian Inoculants Research Group (AIRG) is responsible for maintaining approved rhizobial strains for commercial release to manufacturers. The Rhizobial Inoculant Strains Table is updated annually and can be accessed directly from the website. It is good practice to check the recommended rhizobial strain for the legume host group during crop planning and prior to product purchase. Each host group is identified by a dedicated letter on product packaging e.g. Group F for faba beans, broad beans & lentils.

***What* product is best for the beneficial relationship between grain legumes and rhizobia?**

Successful production and use of inoculants starts with quality and concentration of the right rhizobial strain in the product. Quality assurance is provided by the AIRG's independent Green Tick program that tests manufactured batches of inoculant products against stringent rhizobial viability and product quality criteria, as well as contaminant and moisture thresholds.

Standards for legume inoculants are based on scientific research that has defined the number of rhizobia required for adequate nodulation. Whilst testing is undertaken at the point of manufacture, the way any inoculant product is transported and stored along the supply-chain affects the viability of the living rhizobia. Site specific environmental conditions, such as climate, extreme weather events, soil moisture and soil pH have an influence on the type and rate of inoculant best used and the effectiveness of inoculation.

Rhizobia inoculants are available in several formulations and each product type has advantages in certain situations. Peat products, the most common inoculant formulation, deliver the most reliable level of nodulation across a range of sowing environments and are the most cost effective. Granules are advantageous when sowing into dry situations. Liquid and freeze-dried inoculants are easily compatible with liquid injection systems.

Currently, the Green Tick is issued by the AIRG for **moist peat** products that are applied as slurry to the seed or diluted/filtered slurry injected into the seed furrow. **Liquid** inoculants, a suspension of rhizobia in a protective liquid formulation, is sprayed under low pressure into the soil in the seeding furrow. **Freeze-dried powders** require reconstitution prior to application and can be directly injected into the seedling furrow or used to coat seed. Lastly, **clay/peat granules** are applied directly into the seed furrow.

Only inoculants meeting the standards of the AIRG's independent quality testing carry the Green Tick logo. The Green Tick guarantees the presence of live rhizobia population at the point of manufacture required to successfully nodulate. This is 1 billion rhizobia per gram of moist peat product. The recommendation for rhizobial numbers on seed at sowing, when inoculated by a peat slurry inoculant, is 100,000 rhizobia per large seed (chickpeas, lupins) and 10,000 for smaller seeds (mungbeans, lentils) (GRDC 2014).

Rhizobia concentrations vary greatly across granule products and contain fewer per gram than peat-based inoculants.

***How* is the beneficial relationship between grain legumes and rhizobia best managed?**

There are factors that may compromise the tested high numbers and effectiveness of rhizobia along the supply-chain, effecting grain legume nodulation and therefore field efficacy. While the quality tests ensure that inoculants contain high numbers of effective rhizobia at the time of testing, the quality of the inoculant can be affected by the way it is treated along the supply chain and how it is applied.

Rhizobia inoculant products should be treated with consideration for the following:

- *Rhizobia are living organisms susceptible to high temperatures:* It is important that inoculants are transported and stored away from direct sunlight and in temperatures less than 25 ^{o}C for short periods but best refrigerated at 4 ^{o}C.
- *Read the expiry date before use:* Only use a product that is within date. A well stored peat or granule product will have a shelf-life of 12 months or 6 months respectively for most strains.
- *Avoid crushing or cracking:* Granular formulations can easily be turned to powder that will clog application machinery.
- *Fresh is best:* Sow seed into the ground as soon as possible after it is inoculated to maximise the potential for nodulation. Sow peat-inoculated seed within 24 hours; prepare liquid and freeze dried inoculants within 6 hours, into moist soils.
- *If sowing into dry soils:* A peat inoculant, applied at a double rate where rainfall is forecast within 7 days, is an option. As the window increases past 7 days, the risk of nodulation failure increases based upon soil type, weather, whether a fungicide has been applied to the seed or whether the soil is acidic (GRDC Webinar, 2020).
- *Rhizobia are sensitive to chemical residues:* Use clean equipment and potable (non-chlorinated) water for mixing and application.
- *Trace elements, fertilisers and pesticides are toxic to rhizobia:* Do not mix inoculant directly with these. Where application of both pesticide and inoculant are critical to crop establishment, the use of direct soil inoculation techniques should be considered.
- *Rhizobia survival on seed is reduced by fungicides:* Apply rhizobia last and limit exposure time between inoculation and sowing to <5 hours.
- *Opened inoculant product can be stored for short periods:* Air needs to be immediately expelled, the packet sealed and refrigerated at 4^{o}C. Use as soon as possible and never beyond the use by date.

The careful application of high-quality inoculants to grain legume crops increases the chances that nodulation, nitrogen fixation and yield will be optimised.

***Review* the success of the beneficial relationship between grain legumes and rhizobia**

Nodulation should not be assumed. Assess the effectiveness of the rhizobial inoculant, and the practices used, by checking the degree of nodulation from 8 to 10 weeks. In general, numerous pink nodules near the top of the root system indicate that effective nodulation has occurred.

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