Final Project Report to Protein Research Trust

Inoculation of Lupin in South Africa

January 2000 – August 2001

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Executive Summary

Widespread cultivation of lupin has resulted in the establishment of effective populations of *Bradyrhizobium* sp. (*Lupinus*) in the winter rainfall region of the Western Cape. Despite the availability of an effective commercial inoculant strain VK 10, many farmers prefer to rely on naturally-occurring soil populations of rhizobia for nodulation of their lupin crops. The aim of this study was to determine whether inoculation benefited yields in soils containing established populations of lupin rhizobia.

Field trials were carried out at sites in the Swartland (Hopefield, Langewens, Eendekuil) and Rûens (Roodebloem, Tygerhoek) regions. In addition to inoculated and uninoculated treatments, molybdenum and nitrogen fertilizer treatments were included in the experimental design.

The size and nitrogen-fixing effectiveness of rhizobial soil populations, together with the effectiveness of individual isolates, was determined in greenhouse experiments. Plants were sampled for nodule occupancy four and/or six weeks after planting, and harvested at the end of the season. Nodule occupancy was determined serologically by ELISA. Rep-PCR was used to compare genomic diversity of the soil populations.

Four soils were moderately acid (pH 5.5-5.8) and contained large populations of effective rhizobia (>5000 cells g^{-1}). In contrast, Roodebloem soil was moderately alkaline (pH 7.6) and contained a relatively small population of rhizobia (380 cells g^{-1}). Comparison with VK 10 showed that some soil isolates were equally effective while others were significantly less effective. The average isolate: VK 10 effectiveness ratio was less than 0.9 for each soil suggesting that there is potential for increasing nitrogen fixation. The isolates from Langewens had appreciably lower nitrogen-fixing effectiveness and efficiency than those from the other soils. The highly effective populations may be a useful source from which to select inoculant strains adapted to local conditions.

Nodule occupancy was determined at the Roodebloem site after 6 weeks but the site was later abandoned because of poor plant growth and survival. Following harvest, analysis of seed dry mass and protein content showed that neither inoculation, molybdenum or nitrogen fertilization treatments increased yields at the other sites. The nitrogen application reduced yields at Tygerhoek due to stimulation of grass weeds which choked lupin growth.

Inoculants were previously applied in the Swartland soils and serological analysis showed that strain VK 10 was present in 23-55% of the nodules formed on uninoculated plants. Inoculation with VK 10 failed to significantly increase nodule occupancy by this strain. Although neither of the Rûens sites were previously inoculated, a small proportion (<6%) of uninoculated plant nodules contained VK 10. Inoculation in the high population soil at Tygerhoek failed to increase VK 10 nodule occupancy. In the low population soil at Roodebloem, inoculation increased nodule occupancy by this strain to 98%.

Rep-PCR analysis of isolates revealed considerable genomic diversity in all four soils with acid pH. In contrast, isolates from the high pH soil at Roodebloem were more homogeneous. Further study of the effect of environmental conditions such as soil pH on genomic diversity is required.