## METHODS FOR ISOLATION OF PLANT GROWTH PROMOTING RHIZO-BACTERIA (PGPR) FROM WESTERN AUSTRALIAN SOILS

## Rebecca Swift<sup>1</sup>, Jen McComb<sup>2</sup>, Giles Hardy<sup>3</sup>, Khalid El Tarabily<sup>4</sup> and Lambert Brau<sup>1</sup>

<sup>1</sup> Centre for *Rhizobium* Studies, <sup>2</sup> School of Biological Sciences and Biotechnology, and <sup>3</sup> Centre for *Phytophora* Science and Management, Murdoch University, South Street, Murdoch, Western Australia 6150, Australia

<sup>4</sup> Department of Biology, Faculty of Science, United Arab Emirates University, 17551, Al-Ain, United Arab Emirates

There is a substantial and increasing effort in industrial microbiology and biotechnology to develop microbial inoculants as a means to improve the sustainability and profitability of rural activities whilst increasing productivity. Inoculants are being developed for use as microbial biofertilizers, biocontrol agents for weed suppression, biopesticides and bioremediation agents. All of these require the addition of microorganisms to complex microbial communities. Plant Growth Promoting Rhizosphere organisms (PGPRs) are good examples of microbes that might have important roles in agriculture. PGPRs inhabit plant root rhizospheres and can affect plant growth directly by nutrient solubilisation (P, N and K) and production of plant growth regulators, and indirectly by suppression of plant pathogens by competition, release of antibiotics or siderophores.

This current study is focusing on two methods to screen potential PGPRs from a variety of rhizosphere soils in Western Australia. The first method used detects the ability of microorganisms to produce the plant growth auxin [indole-3-acetic acid (IAA)] *in vitro*. This involves separating rhizosphere soil from plant roots and plating soil dilutions on to various media. Discrete colonies are plated on to fresh media and sub-cultured several times until clean colonies are obtained. A total of 166 bacteria were isolated and 77 were successfully assayed for auxin production in the presence and absence of the IAA precursor L-tryptophan.

The second method is a much more rapid method of isolating PGPRs from rhizosphere soils. A number of PGPRs produce ACC deaminase, which can cleave the plant ethylene precursor ACC, thereby lowering the levels of ethylene in a developing or stressed plant. The method involves incubating one gram of each rhizosphere soil in minimal broth media over four days, with the final media containing ACC as the sole nitrogen source. Dilutions of the final culture are plated on to DF minimal media again with ACC as the sole nitrogen source. In effect, PGPRs that produce ACC deaminase, and can therefore use ACC as a nitrogen source, are selected. A total of 16 ACC deaminase-producing organisms were isolated using this method.

Fourteen of the highest auxin-producing organisms and 13 ACC deaminase-producing organisms were then tested for seed toxicity on clover and wheat. Six of the auxin-producing organisms progressed to field trials on peas and wheat in Kojonup in south-west Western Australia. Eight of the auxin producers and eight of the ACC deaminase producers are being tested gnotobiotically in growth pouches in the glasshouse for plant growth promotion on wheat. This will be followed with glasshouse pot trials of organisms that show significant plant growth promotion.