Integration of Biofertilizers (Mycorrhiza and PGPR) in a Green Manure Based Wheat-pulse Rotation

In this project, bio-inoculants like arbuscular mycorrhizal fungi (AMF) and plant growth promoting rhizo-bacteria (PGPR) were integrated as bio-fertilizers in wheat (Triticum aestivum L.)-pulse (leguminous plants; here Vigna mungo [L.] Hepper = urdbean) and wheat-rice (Oryza sativa L.) crop rotations at multi-locations in India. In parallel, one field trial in Switzerland on a P-deficient (phosphorus) soil was conducted to assess the effects of different AMF inoculation techniques (i.e. seed coating and row inoculation) in soybean (Glycine max (L.) Merr.). Molecular tools were developed to monitor the inoculated AMF and PGPR strains in pots and in the field in Switzerland. The overall aim of the project was to develop bio-fertilizers to increase productivity of low-input systems in marginal areas.

The Energy and Resources Institute, New Delhi (TERI), the G.B. Pant University of Agriculture &Technology, Pantnagar (GBPUAT) and the Barkatullah University Bhopal (BU) conducted field trials in India with bio-fertilizers from 2005 to 2007. These bio-fertilizers, based on bacterial (PGPR) and fungal strains (AMF), had been isolated and selected from wheat roots during the first programme phase. They were tested at nine different experimental sites over two years. The AMF or PGPR were evaluated alone and in combination each at two fertilizer levels. Since the project was targeted to low-input agriculture in marginal areas, the fertilizer input was restricted. The study sites were extending from the Himalayan feet to the Indo-Gangetic Plain. Additional trials were performed in Madhya Pradesh. One AMF inoculum used consisted of a carrier based natural consortium (Mnat), which was multiplied via host plants and comprised several AMF strains. A commercial single strain AMF (Glomus intraradices) was additionally used (Mss2), multiplied on root organ cultures (ROC). In the second experimental year, Mss1, a further AMF single strain inoculant was used, which was isolated from the Mnat consortium. The PGPR inocula constituted of two fluorescent Pseudomonas (Pf) strains (R62 and R81), applied always in combination.

FiBL was substantially involved in the planning and set up of these field trials in India, aiming at establishing a consistent data basis to evaluate the effects of bio-fertilizers in marginal areas under farmers’ practice conditions. The conceptual work and the implementation were done in close collaboration with the partners in India and an external scientific advisor. All sample analyses of these field trials were performed by the Indian partners and the results presented here relay on these analyses. For data assemblage, FiBL installed an electronic platform to which all members of SA bio-fertilizer have access. Data validation, evaluation and uniform statistical analysis was done by FiBL. A manuscript including yield as well as crop and soil quality data is in preparation.

The most exciting results of the multi-location field trial were achieved in wheat where AMF inoculation significantly increased AMF root colonisation. Over two cropping seasons (2006 and 2007) and calculated across both fertilizer levels and six sites, wheat grain yield was increased by 41% after dual inoculation with Mnat and Pf as compared to the non inoculated control, corresponding to an increase from 2.36 t ha⁻¹ to 3.35 t ha⁻¹ grain dry matter yield (mean of 11 wheat
crops, $p < 0.05$). Inoculation with AMF (Mnat or Mss2) or Pf alone augmented wheat grain yield by 29\% to 31\%, respectively. In 2007, dual inoculation by Mss1+Pf increased wheat grain yield from 2.37 t ha$^{-1}$ to 3.17 t ha$^{-1}$ by 34\%, application of Mss1 alone resulted in 14\% grain increase (2.69 t ha$^{-1}$). There was no clear trend towards a better or worse performance of the bio-fertilizers at the zero and farmers’ practice fertilizer level, respectively. However, yield increase due to bio-inoculant application compared to non-inoculated controls was highest at marginal sites with low yields.

The kharif (‘summer’) crops rice and urdbean (Vigna mungo) showed no significant responses to bio-fertilizer application in terms of grain yield in the first year of application. In the second year however, dual inoculation with Mnat+Pf increased rice grain yield by 26\% calculated over two sites. This corresponds to a yield increase from 3.35 t ha$^{-1}$ to 4.21 t ha$^{-1}$. Urdbean grain yield was increased from 0.266 t ha$^{-1}$ to 0.353 t ha$^{-1}$ by 33\% due to inoculation with Mnat.

The wheat-rice rotations were further improved by the introduction of a green manure plant, grown between wheat harvest and rice sowing. Sesbania as a leguminous green manure substantially increased yields of rice and wheat by 25 to 27\%, respectively.

Phosphorus and trace elements (Fe, Zn, Cu) of wheat were significantly higher after inoculation of AMF and Pf. This is most relevant with respect to the diet of the Indian population, because wheat is a staple crop there.

In a field trial with soybean in Embrach (Switzerland) on a P deficient soil, FiBL investigated the effects of inoculation with the ISCB strain 13 from the Botanical Institute of the University Basle (BIB), selected for its plant growth stimulation during the first programme phase. A seed coating technique with gelatine, row-inoculation and a non inoculated control were compared. All seeds were additionally supplied with the nodule forming Bradyrhizobium japonicum. Both AMF inoculation techniques significantly increased the plant density which led to increased straw yield, slightly lower numbers of pods per plant and in the tendency to a decreased grain yield in the inoculated plots. It is concluded that sowing density may be adapted when bio-fertilizers are applied. FiBL was also mass multiplying several AMF strains for inoculum production and maintaining pot cultures of Indian AMF and PGPR strains for monitoring purposes. In a new approach, the Institute of Botany, Univ. Basel (BIB) developed two different molecular tools for tracking Glomus intraradices strains within a mixed indigenous AMF community, i.e. based on specific microsatellite length or using the large subunit of mitochondrial rDNA. For monitoring of strain PGPR R62 in the field, the Laboratory of Microbiology of the University Neuchâtel (LAMUN) developed a method to detect this specific bacterial strain on the basis of differences in the ITS1 region.

The experiments have shown that bio-fertilizers improve productivity and nutrient use efficiency in marginal areas, rendering these agri-systems less dependent on external inputs such as fertilizers and energy. By this, bio-fertilizers mitigate the effects of climate change and help to guarantee food security in a world of increasing food demand and scarcer resources.

In future it will be interesting to test mid-term effects of the bio-inoculants on yield, soil and food quality. A comparison of a natural AMF consortium with a synthetic ROC AMF consortium will fill important knowledge gaps in applied soil ecology. In these experiments, the newly developed monitoring tools will be finalized, transferred to the Indian partner laboratories and evaluated under Indian field conditions. Moreover mass-multiplication, formulation, application techniques for AMF and Pf need optimisation.