Production and Formulation of Bacterial Plant Growth Promoting Bioinoculants (PGPR)

During the second phase of programme, plant growth promoting (PGPR) bacterial strains R62 and R81, were used for optimization of fermentation medium, fermentor operating conditions, development of formulation and shelf life estimation experiments in order to develop an appropriate technology module for the scale-up process.

The modified Schlegel’s medium (SM-4) was used at 14 liter (L) and 300 L fermenter scale for the production. Maximum cell and siderophore concentrations were obtained when glycerol was used as a sole carbon source, with supplementation of succinic acid, under iron stress conditions. The siderophore molecules, present in the cell-free supernatant, were able to control contamination levels in sterile and non-sterile carriers. Therefore a fermented broth containing both cells and siderophores was produced. To analyze the effect of temperature on lag time, pH stability, cell growth, and siderophore production, the R62 strain was cultivated at different temperatures (4°C-40°C). The optimal temperature was 28°C. Based on the observations from four runs in a 14 L fermenter, the operating parameters were fixed for a problem-free operation in the 300 L fermenter. Both strains were successfully produced in the 300 L fermenter. Sterile formulations of R62 and R81 strains were prepared using talcum powder and aluminum silicate as a carrier. Both formulations were stored at 28°C. The cell viability, pH stability of talc powder-based fluorescent pseudomonad formulations remained constant over a period of one year. In contrast, cell viability, at the end of four month, decreased and pH increased in case of aluminum silicate based formulations. Since a cold chain of delivery for bio-inoculants does not exist in India, studies were done to test the shelf life of the product formulations under harsh conditions with appropriate packaging. Survival rate in unpacked formulations, under harsh condition (70°C, 1h), was poor. The packed formulations (with insulation) gave significantly better results.

Different formulations of PGPR strains R62 and R81, based on inorganic carriers, were tested in pot and field trials in collaboration with GBPUAT at Pantnagar. The experiments were divided into two sets. In the first set, talcum and aluminum silicate carrier-based formulations were used. Vigna mungo was selected as a test crop in pot trials and wheat for field trials. Both formulations worked significantly well as compared to the untreated control in pot and field trials. There was a significant increase in dry root and dry shoot weight of Vigna –mungo plants in pot trial. In field trials, wheat seeds treated with talc powder based formulation of strain R62 and/or strain R81, showed a higher number of plant tillers/m², grain yield and straw yield as compared to those treated with aluminum silicate based formulations. In the second set, vermiculite and talcum were selected as carriers and 1% sucrose was added as an adjuvant in fermented broth of the PGPR strains R62 & R81. Various combinations were tested in pot trial experiments on wheat and Vigna mungo. Both formulations showed good results as compared to the untreated control in terms of different growth parameters (root and shoot weight, flowering time). In conclusion, it appears that talc powder and vermiculite were better carriers than aluminum silicate. Combination of strains.
R62 and R81 gave better result in comparison to individual formulations on *Vigna mungo* and wheat.

Further, we can conclude that there is no significant difference with respect to plant growth parameters, when the talc powder based formulation was prepared from broth containing sucrose or glycerol as adjuvant. The choice between vermiculite and talc powder as carrier substrate should be based on their local regional availability. Overall, a talc powder based formulation appeared as best option with regard to plant growth response as well as with regard to long shelf life characteristics.