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Assessment of IAA producing microorganism in chickpea fields of Bhopal

Vineeta Ratonia and Adarsh Lalit

Mansarovar Global University, Sehore

Abstract:

There were six bacteria found potential to produce PGPR and that are *Bacillus aerius*; *Bacillus cereus*; *Pseudomonas fluorescence*; *Pseudomonas aeruginosa Bacillus subtilis* and *Azotobacterchroococcum.* A large array of bacteria including species of *Pseudomonas, Azospirillum, Azotobacter, Klebsiella, Enterobacter, Alcaligenes, Arthrobacter, Burkholderia, Bacillus and Serratia* have reported as PGPR to enhance plant growth

Keywords: Chickpea, IAA, Bacteria, HPTLC, Molecular identification

Introduction:

Chickpea (*Cicer arietinum* L.) is a member of Fabaceae family. This is a self-pollinated and diploid grain legume crop. This has high quality protein and fit for human consumption. There are five essential groups of hormones are identified i.e. auxins, gibberellins, ethylene, cytokinins, and abscisic acid. Indole-3-acetic acid (IAA) is the member of the group of phytohormones and is generally considered the most vital native Auxin (Ashrafuzzaman *et al.*, 2009). It functions as an important signal molecule in the law of plant development inclusive of organogenesis, tropic responses, cellular responses along with cell growth, division and differentiation and gene regulation (Ryu and Patten, 2008). Auxin phytohormone IAA is produced by a number of bacterial species. Specific biosynthesis pathways had been reported and redundancy for IAA biosynthesis amongst different plant-associated bacteria. Interactions among IAA generating microorganism and plant life lead to various results on the

plant aspect but varies from pathogenesis to phytostimulation. Bacterial IAA play a specific role in different microorganism-plant interactions and use phytohormone to engage with plant as a part of their colonization approach, together with phytostimulation and circumvention of basal plant defence mechanisms. Some recent reports suggest that IAA can signalling molecule in microorganism and show their direct impact on bacterial physiology (Spaepen *et al.*, 2007). A number of soil microflora are involved in synthesis of auxins in pure line culture and soil (Barazani and Friedman, 1999).

Materials and Methods:

Chickpea (*Cicer arietinum* L.) rhizospheric soil samples were collected from three different locations (Kolar, Neelbadh, Barrai) of Bhopal, Madhya Pradesh. Sidrophore production test, Phosphate solubilization and Indole production tests were performed on rhizospheric isolated bacteria. Isolates were confirmed by molecular studies. HPTLC was performed for IAA identification.

Results and Discussion:

Total ten different species of rhizobacteria were isolated from different sampling sites of Chickpea field in different year. Out of ten there were six bacteria found potential to produce PGPR and that are *Bacillus aerius*; *Bacillus cereus*; *Pseudomonas fluorescence*; *Pseudomonas aeruginosa Bacillus subtilis* and *Azotobacterchroococcum.* A large array of bacteria including species of *Pseudomonas, Azospirillum, Azotobacter, Klebsiella, Enterobacter, Alcaligenes, Arthrobacter, Burkholderia, Bacillus and Serratia* have reported as PGPR to enhance plant growth (Glick, 1995). There are variety of mechanisms by which PGPR promote plant growth like the ability to produce or change the concentration of plant growth regulators like indoleacetic acid, gibberellic acid, cytokinins and ethylene, asymbiotic N_2 fixation, antagonism against phytopathogenic microorganisms by production of siderophores, antibiotics, and cyanide, solubilization of mineral phosphates and other nutrients (Lugtenberg and Kamilova, 2009). Studies reported that chickpea rhizobacterial isolates of Bacillus, Pseudomonas and Azotobacter produced IAA whereas almost 85% of Rhizobium becomes able to produce IAA (Joseph *et al.*, 2007). Various species of bacteria like Pseudomonas, Azospirillum,

Azotobacter, Klebsiella, Enterobacter, Alcaligenes, Arthrobacter, Burkholderia, Bacillus and Serratia have been mentioned to enhance the plant increase (Kloepper *et al.*, 1980; Kloepper *et al.*, 1989; Okon and Labandera-Gonzalez, 1994; Glick, 1995; Joseph, 2007). Auxin phytohormone IAA is produced by a number of bacterial species. Specific biosynthesis pathways had been reported and redundancy for IAA biosynthesis amongst different plant-associated bacteria. Interactions among IAA generating microorganism and plant life lead to various results on the plant aspect but varies from pathogenesis to phytostimulation. Bacterial IAA play a specific role in different microorganism-plant interactions and use phytohormone to engage with plant as a part of their colonization approach, together with phytostimulation and circumvention of basal plant defence mechanisms. Some recent reports suggest that IAA can signalling molecule in microorganism and show their direct impact on bacterial physiology (Spaepen *et al.*, 2007). A number of soil microflora are involved in synthesis of auxins in pure line culture and soil (Barazani and Friedman, 1999). Potensial rhizobacteria which are capable to auxin biosynthesis can be used as a tool for the screening of effective PGPR culture (Khalid *et al.*, 2004). Pseudomonas bacteria, particularly *Pseudomonas fluorescens* and *Pseudomonas putida* are the most crucial forms of PGPR which produce auxin and promote the crop yield. Khakipour and his colleague in year 2008 studied the productiveness of auxins in Pseudomonas extracts through HPLC and reported a variety of auxins like indole-3-acetic acid (IAA), indole-3-pyruvic acid, indole-three-butyric acid and indole lactic acid, cytokinins and gibberellins with auxin production being quantitatively most critical (Costacurta *et al.*, 1994; Martínez-Morales *et al*, 2003; Horemans *et al.*, 1986; Cacciari *et al.*, 1989; Bottini *et al.*, 1989; Barassi *et al.*, 2007). There are some examples of Catecholate siderophores like the siderophores Enterobactin is produced by *Escherichia coli*, bacillibactin is produced by *Bacillus subtilis* and *Bacillus anthracis* and vibriobactin is produced by using *Vibrio cholerae*. The examples of siderophores with combined ligands are the siderophores azotobactin is produced with the aid of *Azotobacter vinelandii*, pyoverdine is produced by using *Pseudomonas aeruginosa* and yersiniabactin is produced by means of *Yersinia pestis* (Suttiviriya *et al.*, 2008). Species of *Pseudomonas* have the capability to make use of siderophores produced by various

other microorganism and fungi. *Pseudomonas putida* can make use of the heterologous siderophores produced by way of rhizosphere microorganisms to enhance the level of iron to be had to it in the natural habitat (Loper and Henkels, 1999). In a study conducted by Kaur and Sharma, 2013 Chickpea seeds bacterized with *Pseudomonas* sp. PGPR 2 showed a significant increase in percentage seed germination (89.7%) followed by PGPR 3 (88.9%) and LK 884 (86.4%) in desi variety PBG1. In a study, *Pseudomonas* and *Bacillus* isolates were obtained from rhizosphere of chickpea. In dual culture test, *B. subtilis* strains were shown to have more growth inhibition of pathogen on the PDA medium than *Pseudomona*s isolates. *B. subtilis* B28 isolate showed the highest inhibition percentages (51.16%) than other isolates. Also, the ability of bacterial isolates was varied in production of cyanide hydrogen, siderophore, protease and indole acetic acid (IAA) (Karimi *et al.,* 2012). Benefits provided by the inoculation of chickpea (*Cicer arietinum L.*) with selected strains have also been described, including the promotion of growth due to the increased availability of nutrients, provided by inoculation with *B. subtilis* in seeds (Canbolat *et al*., 2006). Plant growth promoting Rhizobacteria (PGPR), such as *Pseudomonas* and *Bacillus* strains, are the major root colonizers, and can elicit plant diseases, pseudomonas and bacillus strains have great potential in control the diseases of Chickpea (Singh *et al*., 2014).

Image visualization of HPTLC plate

Conclusions:

Out of that there were six bacteria found potential to produce PGPR and that are *Bacillus aerius*;

Bacillus cereus; *Pseudomonas fluorescence*; *Pseudomonas aeruginosa Bacillus subtilis* and *Azotobacterchroococcum.* Plant growth promoting Rhizobacteria (PGPR), as isolated in this study are the major root colonizers, and can elicit plant diseases, control the diseases and enhance production of Chickpea. Obtained strain of bacteria can be used for production of PGPR and can make this a commercialized strain of PGPR.

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