

## Endophytic Bacteria *Bacillus amyloliquefaciens*(Pj2) Imparted Growth Promotion and Enhanced Drought Stress Tolerance of Peanut

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### Abstract

Endophytic bacteria have been described for abiotic stress mitigation and plant improvement. The present study aims to isolate the endophytic bacteria from stress-tolerant *P.juliflora* plant roots and evaluate properties of PGP *in vitro* and the effect of endophyte in non-host plants *i.e.*, peanut (*Arachis hypogaea* L.) under drought stress conditions. Among the fifteen individual endophytic bacterial strains isolated, five strains (Pj2, Pj4, Pj6, Pj10, Pj12) displayed drought tolerance. Further, one of the selected strains *Bacillus amyloliquefaciens* TG4 (Pj2), produced extracellular enzymes and plant growth-promoting activities. Plant growth-promoting activity of Pj2 in a non-host crop, the peanut, was established by infecting it, and its effect was studied in induced drought stress for ten days. The Pj2 inoculation positively affects peanut growth morphological and biochemical parameters under irrigated and drought stress conditions compared to non-inoculated plants. Our data suggest that peanut plants can be colonized with the endophytic bacteria for improved drought tolerance.

**Keywords:** Drought stress, Peanut, Endophytic bacteria, Plant Growth promotion bacteria, *Bacillus amyloliquefaciens*

### Introduction:

The steady rise in the global population

demands 50 percent more food than the current production in three decades (1). The gap between population growth and food is increasing. On the contrary, the change in the global climate and rise in the atmospheric temperature make the dry climatic zones drier and more wet climatic zones to the damper. The lack of precipitation in the dry climatic zones increases the prolonged drought stress. It was estimated that by the year 2030, water shortage due to the prolonged drought in several parts of the globe might affect 40% of the population; as a result, 700 million people, livestock, and crops will be at risk (2, 3). Several abiotic factors also severely affected plant growth and yield (4,5,6). Thus, to take up the challenge, there is imperative to develop multi-stress tolerant crops (7,8).

Advanced technologies like genetic engineering (GE) and breeding by marker-assisted selection (MAS) have enormously accelerated the generation of high-yielding stress-tolerant crop plants (9). Due to regulatory issues, several countries have not accepted the cultivation of genetically engineered crop plants (10,11). Recently, the scientific community developed a novel eco-friendly, cost-effective strategy using the microbial community to develop stress-tolerant crop plants. Research findings suggest that plant-microbe interaction boosts the plant's natural defense mechanism

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against environmental and biotic cues. This eco-friendly strategy would overcome the drawbacks of MAS and GE approaches (12,13,14).

Plants colonize with different kinds of microbial complexes. Plant-associated microbial complex includes bacteria, fungi, archaea, and protists (15,16). Microbes feed on the plant's exudates, which aids in acquiring nutrients, tolerance to biotic/abiotic stressors, and the removal of hazardous pollutants from the soil. They also solubilize phosphates, produce iron-chelating siderophores, and fix atmospheric nitrogen through biological nitrogen-fixing. Plant-associated microbes are known to produce a variety of compounds that can enhance plant growth and the ability to cope with a variety of stress conditions (17, 18, 19).

The host plant chooses the microbial species from the surrounding environment to get maximum benefits from the colonizer. Some microbes associate with exterior regions of roots and leaves called rhizosphere and phyllosphere, respectively (20). While some microbial species enter the interior regions of plants called "endophytes." According to recent research, the rhizosphere and endophytic bacteria have many variations in their genomic regions, which might explain why endophytic bacteria colonize the interiors of plants. Bacterial endophytes serve the host plant in various ways, including promotion growth, defending against pathogenic organisms, and providing environmental signals. Endophytic bacteria communicate and interact with their host plants by producing signal molecules more effectively than rhizospheric bacteria in some adverse conditions (21, 22, 23, 24, 25).

Once colonized, microbes produce a variety of compounds and enzymes that can protect the host plants from the adverse effects of several abiotic factors and have good growth and development. Endophytes generate antioxidative enzymes like POD, SOD, GR, CAT, APX, some organic compounds like proline, glycine betaine, organic acids etc., along with

the ability to fix free nitrogen and produce phytohormones (26,27,28, 29). Endophytes also transport the heavy metals across the cell membrane, assist in depositing metals in the intra and extracellular spaces or within their cell walls, form metal complexes and metal redox reactions (30,31,32). Thus, endophytic bacteria assist the plants in alleviating the effect of abiotic stress and improve plant's growth and development. So far, several endophytic bacteria have been successfully employed to ameliorate the plant tolerance to abiotic stress (33,34,35,36). Recently, progress has been made in isolating culturable endophytic bacteria from crop plants and wild-type plants (37;38;39). Further, these strains were utilized to elucidate their role individually for drought or salt or heavy metal stress tolerance. However, crop plants encounter multiple stresses concurrently at field conditions. Hence, there is a need to identify the endophytes for broad host range multiple abiotic stress tolerance (14,40,41,42,43,44).

Recently, there has been a spark of interest in isolating endophytic bacteria from the plants grown in extreme climatic zones and their utilization to enhance stress tolerance in plants/crops (45,43,27,46). Scientists assume that several abiotic and biotic factors of the extreme environment might have shaped these bacterial communities to grow in extreme conditions. These bacteria might have the ability to ameliorate the multiple stresses and help the plants in their proper growth and development. For instance, in a study, halotolerant bacterial strains from a weed *Psoralea corylifolia* grown in salt-affected soils could enhance the salt-tolerant capacity of non-host wheat crop (47).

Similarly, endophytic bacterial strains of a drought-tolerant foxtail millet from a semi-arid region of northeastern China, improved drought tolerance in foxtail millet under laboratory conditions (48). The *Bacillus* endophytic strains from the five desert plants, *i.e.*, *Zygophyllum simplex*, *Panicum antidotale*, *Tribulus terrestris*, *Euphorbium officinarum*, and *Lasiurus scindicus* induce salt tolerance in a non-host plant

*Arabidopsis thaliana* by accelerating its growth and development(49). These studies indicate that endophytic bacteria from stress-tolerant plants from extreme environments provide greater abiotic stress tolerance in non-host plants.

*Prosopis juliflora* (Sw) DC (mesquite) is a fast-growing, moderate-sized, thorny, perennial deciduous tree that belongs to the Fabaceae family (50) that grows in a warm and dry tropical climate that reach high temperatures upto 48°C and low annual rainfall of 250–600 mm (51,52). It grows under adverse climatic conditions in different soils, including sandy, alkaline, *saline*, and rocky. The well-meshed roots penetrate deeper layers of the soil. Being drought-hardy, salt-tolerant, disease-resistant, and it does not require any special care for rehabilitating marginal lands or wastelands (53,54). Endophytic bacteria isolated from *P. juliflora* grown in the tannery industry are reported to have chromium resistance and have the ability to enhance growth and development of non-host ryegrass under heavy metal contaminated conditions (55). Thus, in the present study *P. juliflora* was considered a potential candidate plant for endophytic bacterial mining with additional beneficial properties. It was also considered one of the most widespread heavy metal hyperaccumulating plants (56, 57).

## Materials and Methods

### Isolation of endophytic bacteria:

The *P. juliflora* plant samples were collected from the Botanical garden, Yogi Vemana University, Y.S.R Kadapa, Andhra Pradesh, India (14.472°N.78.7091°E), which is a semi-arid region and comprises alkaline soil (pH 8.0-9.0). Three plants were randomly selected from different regions, uprooted completely, cleaned to remove soil adherents on the surface, washed with tap water thoroughly, and rinsed three times with deionized water. Healthy root samples were separated out and surface sterilized using 70% ethanol followed by 1% HgCl<sub>2</sub> for one min each in laminar airflow. Following the surface

sterilization, the root pieces are washed thrice with sterile double distilled water to remove traces of sterilants. To validate the efficacy of surface sterilization, tiny root pieces and filtrate of the final wash were transferred onto Petri plates with LB agar. Further, sterilized root pieces were macerated in sterile double distilled water in laminar airflow. Following the serial dilution method, 10 µl of each diluted extract was transferred onto sterile Petriplates containing SLP media supplemented with 10 mg L<sup>-1</sup> of fungicidin, spread the contents uniformly using a sterile glassrod, and incubated 28°C for 72 hours (58). Individual isolated colonies of bacteria with unique morphological and growth patterns were selected, transferred to a test tube containing 10 ml of sterile SLP media, and incubated at 28°C for 72 hours. The bacterial liquid culture was diluted using 50 % glycerol and stored at -80°C.

### Morphological tests:

Morphological traits such as cell shape (59) and gram staining (60) were conducted according to the standard laboratory protocols.

### Selection of Drought tolerant Bacteria:

Endophytic bacteria were selected for their drought tolerance by inoculating in the SLP media, supplemented with various concentrations of PEG 6000 (0%,5%,10%, 15% and 20% which is equivalent to 0, -0.05 MPa, -0.15 MPa, -0.30 MPa and -0.49 MPa respectively). Cultures were grown in an incubator shaker with 200 rpm shaking at 28°C for 24 hrs and optical density of the cultures was recorded in a spectrophotometer at 600nm.

### Molecular identification:

Endophytic bacteria genomic DNA was isolated with a modified CTAB method (61). The 16S rDNA was amplified according to the standard protocol (62) from 100ng of genomic DNA using 27F and 1492R bacterial universal primers. The amplified PCR products were electrophoresed on 1% Agarose gel, and

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the product was purified using PCR cleanup columns (Nucleospin, Macherey Nagel, Germany). The purified PCR products were sequenced at Eurofins Genomics (Bangalore, India). The obtained nucleotide sequence was submitted to BLAST N search analysis.

#### **Extracellular enzyme activities:**

The extracellular enzyme (Cellulase, pectinase, amylase, and lipase) activities of Pj2 endophytic bacteria were investigated according to the standard procedure. The cellulase activity of Pj2 was measured by growing on agar plates containing nitrogen free base media supplemented with 0.5 percent tryptone and 0.2 percent carboxymethyl cellulose (63). The Pj2 was grown on NFB media plates and incubated at 30° C for 48 hr, and congo red (1 mg ml<sup>-1</sup>) solution was added to the plates and left for 30 min followed by destaining with 1M NaCl (64). The plates were held at 4°C overnight to observe the clear zones. The pectin metabolization activity of the Pj2 was measured by growing on Petri plates with nutrient agar media containing 0,5 percent pectin (50). The presence of distinct zones around the bacterial growth suggested that pectinase activity was present. (65). The bacterial ability to solubilize starch was identified by growing them on starch agar plates and the bacterial strains that exhibit distinct, clear zones when flooded with iodine-potassium iodide solution were considered positive bacteria for starch solubilization (66). The ability of bacteria to grow on media plates supplemented with Tween-20 for a week at 30°C, and after that incubation at 4°C for 30 min, an opaque zone around the colony was positive for lipase activity (66).

#### **Bacterial traits for plant growth promotion Nitrogen fixation capacity**

The capability of the endophytic bacteria to fix free nitrogen was determined by growing them on nitrogen-free Jensen medium (67) and Ashby media (68). The turbidity of the culture media after growing them on Jensen media for 4 days and on Jensen media for 7 days was

considered as positive for nitrogen fixation (69).

#### **Ammonia Production capacity of Endophytic Bacteria**

Ammonia production was evaluated by growing the endophytic bacteria in sterile peptone water at 30° C for 48-72 hrs followed by adding Nessler's reagent, which gives yellow color in the media is positive for ammonia production (70).

#### **Indole Acetic Acid Production**

The IAA production ability of the isolated endophytes was assessed by growing in sucrose minimal media supplemented with 0.5mg/ml tryptophan and incubated for 48 hrs at 30° C. Followed by 48 hrs incubation, 2 ml of Salkowski's reagent was mixed with 1 ml of the culture and incubated in the dark for 30 min at room temperature. The amount of IAA produced is proportionate to the pink color developed in the mixture read at 535 nm and compared against IAA standard(58).

#### **Phosphate solubilization capacity**

Endophytic bacteria inoculated on NBRIP media for 72 hrs at 30° C with clear halo zones around the colonies indicates phosphate solubilization capacity of the isolates (71). The ability is measured by using the following formula

Phosphate solubilization efficiency %=(Solubilization diameter)/(Growth diameter) X 100

#### **ACC deaminase activity**

The ability of endophytic bacterial isolates to produce ACC deaminase was measured by the amount of α-ketobutyrate generated from the cleavage of ACC by reading the absorbance at 540 nm using standard α-ketobutyrate reference curve (72).

#### **Plant growth promotion and drought stress tolerance**

Pot experiments were conducted in the



greenhouse facility to compare the selected bacterial growth promotion effects on peanut under drought stress conditions.

Peanut seeds (cv JL-24) were sterilized with 2% NaOCl, then washed 4-5 rinses with sterile double-distilled water. Further, seeds were also plated on LB plates to examine if surface sterilization was effective. Uniform size seeds were submerged in the bacterial suspension, and uninoculated seeds were drenched in sterile double distilled water. After incubation, 5 seeds were sown in earthen pots (27 cm x 25 cm diameter and height) containing 15 kg of autoclaved garden soil. A separate set of pots with uninoculated seeds was maintained. Pots were divided as non-inoculated irrigated and drought stress, endophyte inoculated drought stress, and irrigated. The germinated seedlings were thinned to three after two weeks of germination. Drought stress was imposed for 10 days at 55 days after germination, followed by sample collection (harvest) for further analysis.

### Physiological and biochemical analysis

The morphological and biochemical traits were measured for both irrigated and drought-stressed plants. Total root and shoot length, lateral root number, shoot root dry weight, and their ratio was measured in the harvested plants. The RWC of harvested leaves was measured according to Mayak et al. (2004) (73) for triplicates of leaves for each pot. The harvested leaves were immediately weighed to get the initial fresh weight, followed by kept in the distilled water and reweighed to get the turgid weight. Later, leaves were placed in the hot air oven at 65°C overnight and weighed to obtain dry weight. Total chlorophyll was estimated using a method of Wellburn (74), and for free proline content, Bates (75) method was used. By following the methods of Dubois et al., (76), soluble sugars were estimated in the samples.

### Statistical Analysis:

Software SPSS v17 was used to analyze the statistical significance of the data, and

ANOVA was estimated for significance ( $p \leq 0.05$ ) level.

### Results and Discussion

A total of 15 bacterial isolates with morphologically unique features were selected on SLP media. These endophytic bacterial cultures were screened for drought tolerance using PEG 6000. Most of the endophytic bacteria (Pj2, Pj4, Pj6, Pj10, Pj12) displayed drought tolerance up to 15% (-0.3 Mpa). The endophytic bacterial strain Pj2 exhibited maximum growth at all PEG stress levels, which was selected for further experiments (Fig.1).

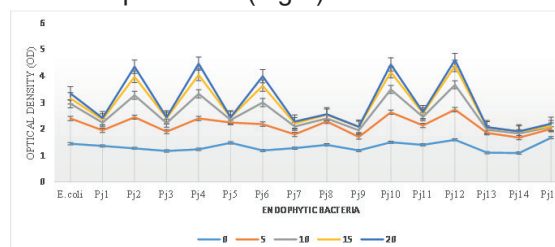


Figure 1. Effect of drought stress (0, 0.05,-0.15,-0.30,-0.49 MPa) on the growth of endophytic bacterial isolates in the SLP broth supplemented of (0,5,10,20% of PEG 6000). The values are the means of three replicates of OD (optical density) of bacterial isolates (n=3).

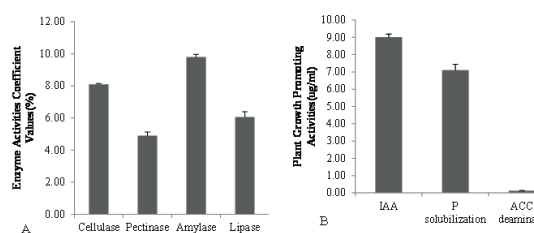


Figure 2. Plant growth-promotion activities of Pj2 endophytic bacteria. (A) Extracellular enzymes (Cellulase, Pectinase, Amylase, Lipase) (B) IAA production, Phosphate solubilization, ACC deaminase production

### Molecular identification of Pj2:

PCR amplification of endophytic bacterial genomic DNA with 16sRNA specific primers 27F and 1492R yielded an expected ~1400 bp PCR product. The single fragment of PCR

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was purified using PCR cleanup column and the purified fragment was subjected to sanger sequence analysis on both ends. The forward and reverse sequence obtained was aligned, and the contig was subjected to BLAST N analysis. The BLAST N analysis of the 16srRNA displayed 100% sequence similarity with *Bacillus amyloliquefaciens* TG4 strain. The nucleotide sequence was submitted to GenBank and obtained the accession number OL691085.

The extracellular enzyme activity results indicated that the endophytic bacterial strain PJ2 produced positive results for cellulase (8.0), pectinase (5.0), amylase (10.0) activity, and negative results for lipase (Fig.2A). The PJ2 strain also displayed phosphate solubilization activity. The PJ2 exhibited positive results for the production of ammonia and nitrogen fixation. The endophytic bacterial strain showed more outstanding IAA production (9.0 U) and ACC deaminase activity (0.12U) (Fig.2B). Thus, PJ2 exhibited plant growth promotion activities.

The PJ2 endophytic bacterial strain was inoculated to the JL-24 peanut genotype. Both non-inoculated and PJ2 inoculated peanut plants were subjected to drought stress; stress was imposed by withholding water for 10 days from 55 days after sowing. Drought stress has shown its effect both on inoculated and non-inoculated plants, but the extent of its effect is more on non-inoculated plants compared to inoculated plants. Inoculated plants that were exposed to drought performed better in terms of the growth rate, including shoot and fresh root weight and their dry weight ratio, root length, shoot length compared to non-inoculated plants under drought stress conditions (Fig.3). The same pattern has been observed in both treatments (inoculated and non-inoculated), even in irrigated conditions. The inoculated plants' length was 53.3% more under irrigated conditions while 54.5% under the drought stress conditions compared to non-inoculated plants. The SFW was increased by 44.4% under irrigated conditions, while it was recorded as 40% under

drought stress conditions. A similar trend was observed for SDW; the irrigated plants recorded with an increase of 58.8%; drought-stressed plants 53.8% of SDW compared to the non-inoculated plants. The PJ2 inoculated irrigated plants displayed an increase of 83.3% of RL, whereas the drought-affected plants recorded 60% more than the non-inoculated plants. The PJ2 inoculated peanut plants compared to non-inoculated plants recorded higher RFW of 43.3% under irrigated, 25% under drought stress, and 7.14%, 21.05% of RDW under irrigated, drought-stressed conditions, respectively.

As depicted in Fig.4C, the PJ2 inoculated irrigated and water-deprived peanut plants showed an enhanced leaf RWC by 33% and 5%, respectively than the non-inoculated plants. The accumulations of total soluble sugars (Fig.4B) are higher (12.5, 13.3% under irrigated, drought-stressed, respectively) in the PJ2 inoculated plants than control non-inoculated ones. The biosynthesis of free proline is also more in the PJ2 inoculated plants than in non-inoculated peanut plants under irrigated (28.5%) and drought-stressed (11.1%) conditions (Fig.4A). The total chlorophyll content was more (15% under irrigated and 40% under drought stress) in PJ2 inoculated plants than in non-inoculated (Fig.4D).

The data from the present study supported the assumption that endophytic bacteria from the plants that grow in extreme environmental conditions would attribute the drought tolerance to the inoculated plants. In this present study, 15 endophytic bacteria were isolated from the interior root region of *P.juliflora*. These endophytic bacteria were screened using PEG 6000 to assess their drought tolerance. Among the 15 endophytic bacteria, five bacterial isolates could grow up to a minimum (-0.3 Mpa) water potential. Endophytic bacteria from wheat could tolerate -0.3MPa water potential, and on inoculation, they improved drought tolerance in wheat (77). The one (PJ2) was selected among

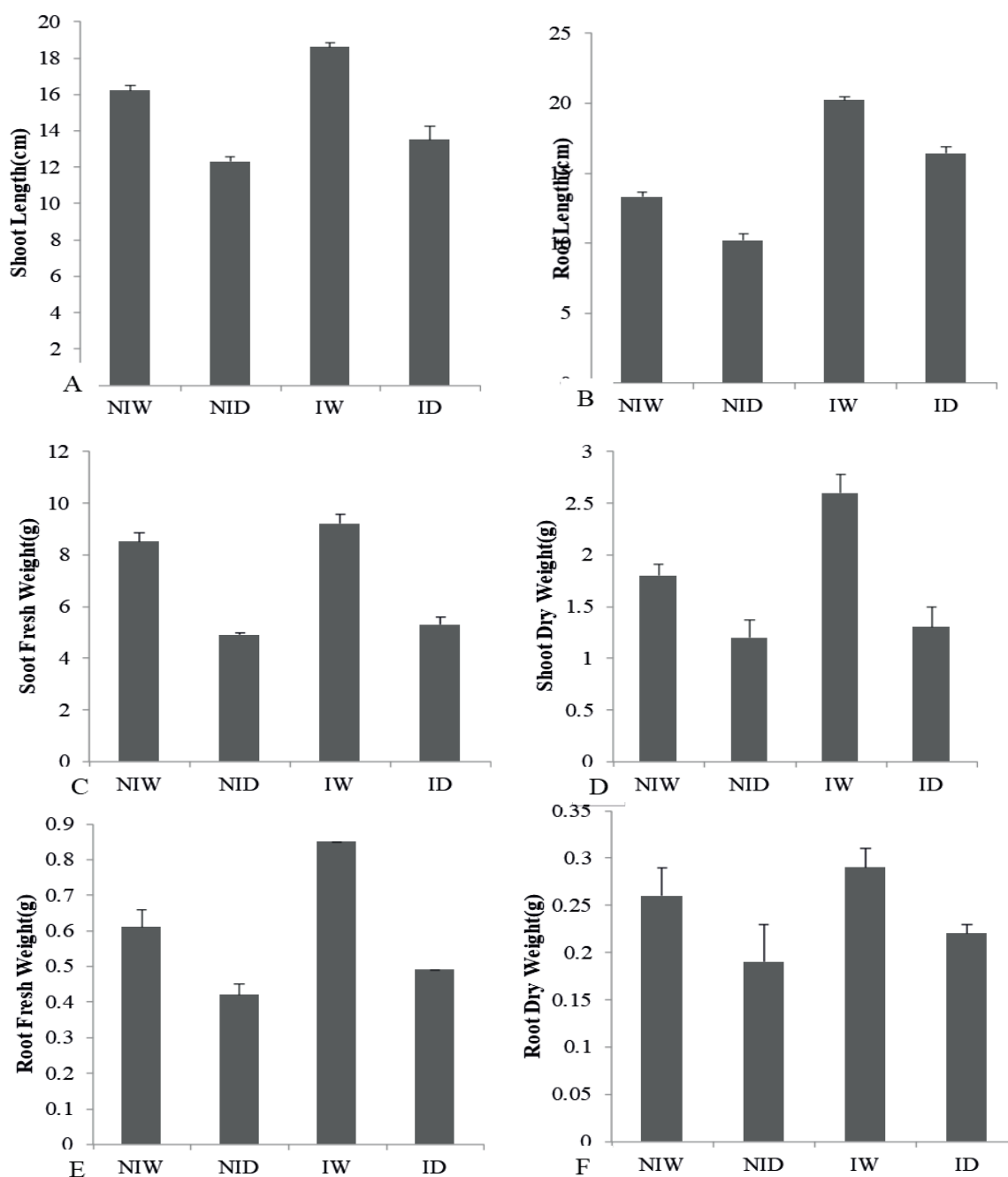


Figure.3. Effect of Inoculation of drought-tolerant bacterial endophyte on morphological parameters of peanut grown under irrigated and drought stress condition. A) Shoot length B) Root length C) Shoot fresh weight D) Shoot dry weight, E) Root fresh weight, F) Root dry weight  
 NIW: Non- inoculated Irrigated; NID: Non- inoculated drought stress  
 IW: Inoculated Irrigated; ID: Inoculated drought stress. Data represents mean value $\pm$ S.E

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the drought-tolerant endophytic bacteria, and 16S rRNA sequence analysis revealed it as *Bacillus amyloliquefaciens* TG4 strain. Several studies documented the abiotic stress tolerance potentiality of *Bacillus amyloliquefaciens* strains in several plant species (78,79,80,81,82).

Further, the extracellular enzyme production capacity of the endophytic bacteria was tested. The Pj2 bacteria exhibited positive results for cellulase, pectinase, and amylase activity. Cellulolytic enzymes are essential for endophytic bacteria, and they help for the penetration of endophytic bacteria into the host cell and the establishment of symbiotic relationships. The extracellular enzymes also provide pathogen resistance and promote plant growth (83,84). In a similar study, endophytic bacteria from the Egypt desert plants *Fagoniamollis Delile* and *Achillea fragrantissima* produced extracellular enzymes; application of these bacteria to the Zea mays showed higher nutrient uptake rates and improved the growth (27).

The Pj2 endophyte has positive PGP characters, such as ammonia, IAA, ACC deaminase, solubilization of phosphate, and nitrogen fixation capability. The PGP traits directly or indirectly improve plant's growth and development. Endophytic bacteria produce ammonia and carbon dioxide by hydrolyzing the urea. Ammonia can be used as a nitrogen source by plants (69,85). Thus, ammonia-producing bacteria are responsible for the good growth and development of the host plant. Soil-bound rock phosphates are solubilized by the organic acids produced by these endophytic bacteria that can be easily observed by the plants and can increase their growth and development (86,87).

In the present study, Pj2 demonstrated phosphate solubilization activity. The phosphate-solubilizing endophytic bacteria from wild poplar improve the phosphate uptake capacity (88). The earlier studies noted improved plant growth and development after applying phosphate

solubilizing bacteria to many crop plants in vegetable crops, including tomato and pepper (89); Chinese fir seedlings (90).

Some endophytic bacteria comprise genes for biological nitrogen fixation (BNF) that can convert free atmospheric nitrogen into ammonia and nitrate within the host plant, directly supplying it to the host as a nitrogen source. Endophytic bacteria are more efficient in BNF than the rhizospheric and rhizoplane microbes (91,92). In this study, Pj2 displayed a positive result for nitrogen fixation. Several endophytic bacteria which have the nitrogen fixation capability also provided drought tolerance. The endophytic bacteria from seeds of invasive *Lactucaserriola* with BNF capability also improved the growth and development in host plant *Arabidopsis* under drought stress (93).

IAA is an important phytohormone, controlling various physio-growth and developmental processes (94,95,96). It is present in plants, but some plant-associated microorganisms also produce IAA (97,98). Though the production of IAA concentration varies in different microorganisms, when the microorganisms were inoculated to the plants, synergistic effects of both microorganism and native plant IAA certainly influence the growth and development of the host plant. The application of IAA-produced endophytic bacteria improved the drought tolerance in maize (99); wheat (100). In the present study Pj2 also produced the IAA. The ethylene biosynthesis precursor ACC is breakdown into amines and ketobutyric acid by ACC deaminase, preventing the production of ethylene under stress conditions. Ethylene is known to cause a deleterious effect on the plants, and the bacteria with ACC deaminase producing capacity can decrease the effect of ethylene on plants by stopping its synthesis (101). The external application of ACC deaminase has improved tomato and pepper's drought tolerance (73), and the same pattern has been observed in maize (102).



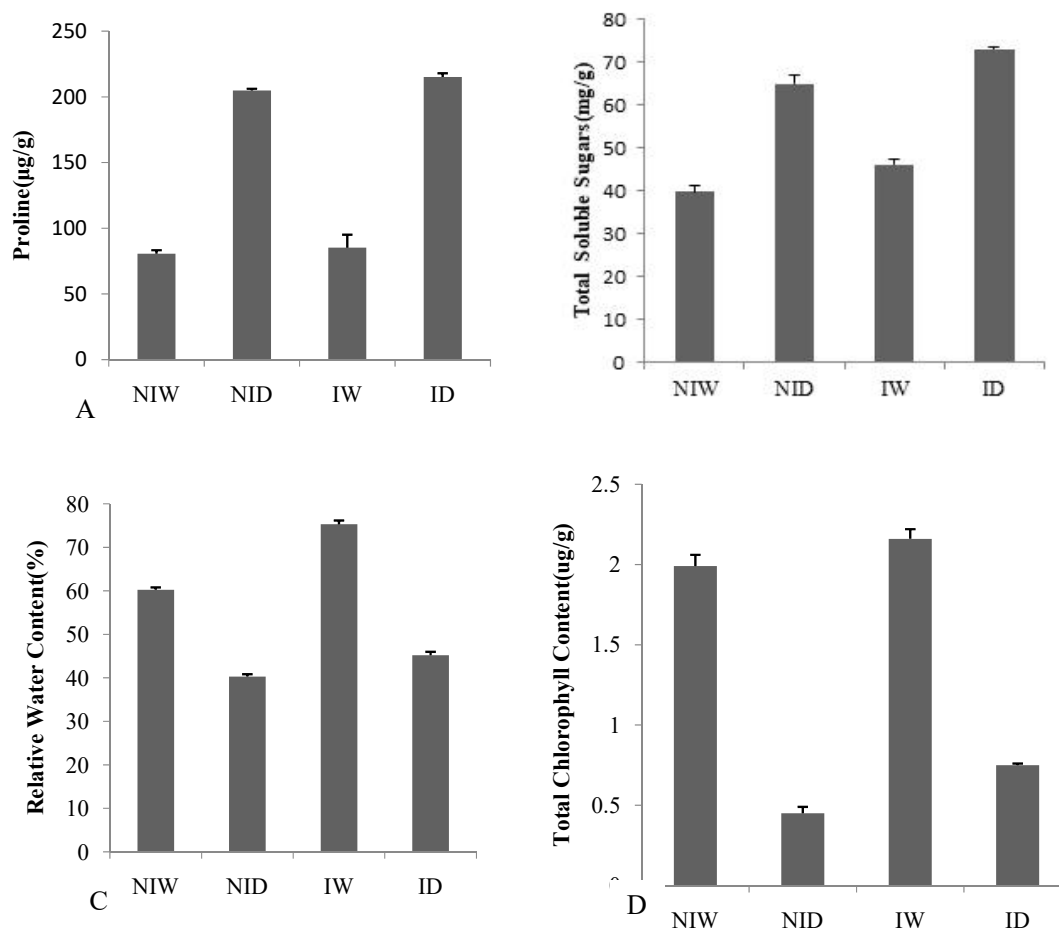


Figure.4. Effect of Inoculation of drought-tolerant bacterial endophyte on Physiological and Biochemical parameters of peanut grown under irrigated and drought stress condition. A) Free proline B) Total soluble sugars C) Relative water content D)

NIW: Non- inoculated Irrigated; NID: Non- inoculated drought stress

IW: Inoculated Irrigated; ID: Inoculated drought stress. Data represents mean value $\pm$ S.E

In the current report, the application of *Pj2* endophyte to the peanut improved the morphological, biochemical traits compared to the un-inoculated plants under irrigated well as in drought stress conditions. These results are inconsistent with the previous studies on the application of PGPR bacteria for drought tolerance in plants (103,104,105,106,107,108,77,109,110,111,93,29).

As indicated by the results, the *Pj2* inoculated peanut plants exhibited better shoot

and root biomass under irrigated and drought stress conditions. The increase in the biomass could be due to the ability of the endophyte to increase the water content, production of more IAA, and enhancement of phosphate mineral acquisition. In general, drought stress decreases the root and shoot biomass. The bacterial inoculated plants overcome the negative effect of drought stress due to enhanced root growth triggered by IAA production. The increase in the root biomass enhances the water and mineral uptake. In a previous study, inoculation *B. subtilis*

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strain B26 to the Timothy plants enhanced the root and shoot biomass ratios due to production IAA and P solubilization (110).

Plants to maintain osmotic turgor under abiotic stress conditions trigger the production of various osmolites like soluble sugars and proline (112,113). Free proline acts as an osmoprotectant, ROS scavenger, and repository of organic nitrogen, which helps the plants recover from environmental stress's adverse effects (114,115,116). Soluble sugars function as structural components of the cell. They also act as ROS scavengers and maintain osmotic balance (117,118,119,120,121). Several studies revealed that enhanced levels of free proline and soluble sugar contents alleviated abiotic stress-induced damage in several plant species. The *Bacillus amyloliquefaciens* strain NBRI-SN13 increased the rice cv Sarayu-52 proline and total soluble sugars contents under drought, salt, ABA, SA, JA, ethephon treatments (78). Similarly, Pj2 inoculated peanut plant exhibited elevated free proline and soluble sugars in the present study. The research data from the present study established the crucial role of Pj2 endophytic bacteria in promoting the growth and development of non-host plants and alleviating the impact of drought stress.

#### **Conclusion and future perspectives:**

The present study revealed that an endophytic bacteria Pj2 can successfully infect the non-host plant peanut and improve the growth and development under irrigated, drought stress conditions by improving the biomass, osmotic adjustment, and maintaining the relative water content. We have also identified several other drought-tolerant endophytic bacteria from *Prosopis juliflora*. These bacteria should be investigated individually or in the form of consortia for their drought and other abiotic stress tolerance in non-host plants. Further, molecular genetics studies are required to get more insights into the plant-bacterial interaction for imparting abiotic stress tolerance.

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