

# Full Length Article

# Amelioration of Drought Stress Adverse Effect and Mediating Biochemical Content of Cabbage Seedlings by Plant Growth Promoting Rhizobacteria

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#### Abstract

The goals of the study were to investigate the role of plant growth promoting rhizobacteria (PGPR) (*Bacillus megaterium* TV-6D, *Bacillus megaterium* TV-20E, *Peanibacillus polymyxa* KIN-37, and combination of Bacillus megaterium TV-6D +Pantoea agglomerans RK-92+Brevibacillus choshiensis TV-53D) in alleviating the harmfull effects of drought stress in cabbage seedlings grown under different irrigation levels. For this popuse, a pot experiment was undertaken to determine the beneficial effect of PGPR on growth and physiological and biochemical properties of cabbage seedlings grown under various irrigation levels (I1, I2, I3 and I4) which was determined considering different ratios (100, 75, 50 and 25%) of evaporated water from the reduced pan. Experimental data showed an increase in growth parameters in PGPR treated plants when compared to untreated plants under stressed conditions. TV-6D and TV-6D+RK-92+TV-53D strains were found to mitigate drought stress tolerance in cabbage plants by accumulating antioxidant enzymes, osmolytes, hormone production, and decreased electrolyte leakage in PGPR treated plants under water deficit conditions. © 2016 Friends Science Publishers

Keywords: Water deficit; PGPR; Cabbage seedling; Hormones; Antioxidants

# Introduction

Abiotic stress conditions such as drought are major limiting factors in agriculture as drought is one of the most unfavorable points for plant growing (Shao *et al.*, 2007). Water is one of the most indispensable resources for successful vegetable growing. Drought stress negatively affects yield and quality of vegetables. Vegetables need regularly water for vegetative and generative development.

Breeding of tolerant genotypes to drought stress can be one of the most important strategies. However, drought resistance has been suggested as being a 'complex trait', especially with the recent expansion of research into its genomics (Blum, 2011). This is due to complex mechanism of abiotic stress tolerance, which is controlled by minor genes. Moreover, methods used for selecting tolerant genotypes are time expendable and accordingly expensive (Athar and Ashraf, 2009). Hence, improving techniques and strategies to mitigate negative influences of drought on plant growing have received considerable attention. Recently, an alternative strategy to mitigate the deleterious effects of drought on crops using PGPR has been suggested (Forchetti et al., 2007).

PGPRs, which inhabite in soil called 'rhizosphere' influenced by plant roots can have indulgent effects on plant growth (Andreselosse et al., 2004). The PGPRs' effect on plant growth are not fully comprehended, but are considered to include asymbiotic N2 fixation and also solubilisation of mineral phosphates, and improving other plant nutrient element uptake (Cattelan et al., 1999). Some PGPRs may encourage plant growth because they improve soil structure and moisture withholding capasity, thus enhancing nutrition uptake (Kim et al., 2012). They can exert an advantageous effect on plant growth and nutrition probably due to fixation of atmospheric nitrogen as well as increasing availability of nutrients like phosphorus, iron and other microelements (Rodriguez and Fraga, 1999). Liddycoat et al. (2009) suggested that Pseudomonas spp. had positive effect on germination and early growth promotion of asparagus grown under drought stress. Figueiredo et al. (2008) determined that PGPR treatments improved plant performance and stomatal conductance of bean grown under lower irrigation levels. PGPRs have been reported to promote root formation and nutrient and water uptake under

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abiotic stress conditions (Perrig *et al.*, 2007). PGPR inoculation with positively affected plant growth of lettuce under drought stress (Sahin *et al.*, 2015).

According to our best knowledge there is no much investigation in relation to drought acclimation of cabbage seedling inoculated with PGPR. Therefore, the goal of the study was to determine the effect of the particular droughttolerant plant growth promoting rhizobacteria on some physiological and biochemical characteristics and plant growth of cabbage seedlings grown under different irrigation levels.

# **Materials and Methods**

This study was carried out under greenhouse conditions at Atatürk University, Erzurum, Turkey. Cabbage (*Brassica oleracea* var. *capitata* 'SARMA F1') seedlings were maintained under natural light conditions. Average temperature and relative humidity in greenhouse during growing period of seedling (21 May-1 July) were  $31.6\pm3.8^{\circ}$ C and  $69.4\pm4.9\%$ , respectively. Temperature and humidity were measured as daily by temperature and humidity measuring device. In addition, total evaporation measured from a reduced pan located in greenhouse during growing period of seedling was 93 mm.

#### **Experimental Design and Bacteria Application**

The experiment was conducted as a completely randomized design with four replicates. Four levels of irrigation, I1 =100 (control), I2 = 75, I3 = 50 and I4 = 25% of evaporated water from the reduced pan, and five bacterial species treatments consisting of no bacterial inoculation (control), Bacillus megaterium TV-6D, Bacillus megaterium TV-20E, Peanibacillus polymyxa KIN-37, and combination of Bacillus megaterium TV-6D +Pantoea agglomerans RK-92+Brevibacillus choshiensis TV-53D. Bacterial strains used in this study were obtained from the culture collection unit in the Department of Plant Protection, Faculty of Agriculture at Atatürk University. We selected potential PGPR strains from a pool of 460 rhizobacterial isolates based on their 1-aminocyclopropane-1-carboxylate (ACC) deaminase-containing, auxin (IAA)-producing and, N2fixing and P-solubilizing strains. The bacterial cultures were grown on nutrient agar (NA) for routine use, and maintained in Luria Broth (LB) with 15% glycerol at -80°C for longterm storage. In previous studies, these strains were determined to have ability to grow in N-free conditions, solubilize phosphate, produce IAA, SA and GA (Kotan et al., 2014; Turan et al., 2014). The PGPR isolates also can grow and produce high amount of amino acid and hormone such as IAA, SA and GA at the drought stress of -0.73 MPa created with PEG6000 (data not shown).

Cabbage seeds were sown into strofoam trays filled with peat. Thirty days after planting (DAP) seedlings were transplanted to pots (70 and 17 cm length and width, respectively, with holes in the bottom) filled with mixture of sand, manure and loamy garden soil (v/v/v, 1:1:1). There were 4 replicates per treatment and 5 plants per replicates in different pots.

Application of the bacterial bioformulation was performed using the dipping method in which seedling roots were inoculated with the bacterial suspensions in sterile water about 20 min prior to planting. The bacterial suspension  $(1 \times 108 \text{ cfu mL}^{-1})$  was included to plastic trays containing 0.2 g of sucrose (10 mg/mL), and seedlings were soaked in this suspension. Additional applications were done at 15 days after transplanting. Bacterial suspensions (100 mL per plant) were injected into root zones of seedlings.

## **Irrigation Applications**

The plants were hand-irrigated using tap water with low electrical conductivity (0.285 dS m<sup>-1</sup>), low sodium adsorption ratio (0.47) and neutral pH (7.42). Irrigation water amounts which to be applied in each irrigation was determined considering the amounts of evaporated water from the reduced pan constructed of galvanized iron sheet (Blanco and Folegatti, 2004). Four different irrigation levels (I1, I2, I3 and I4), which were determined considering different ratios (100, 75, 50 and 25%) of evaporated water from the reduced pan, were tested. The irrigation interval was three days during growing period of seedlings. First irrigation was made together with transplanting of seedlings and all pots were fully irrigated until appears water drainage from bottom of pots. In subsequent irrigations, the depth of irrigation water used was calculated according to equation:

(1) I = Ep x IR

Where, I is the irrigation water depth (mm), Ep is the depth of water evaporated from the reduced pan with intervals of three days in the growing period of seedlings (mm) and IR is the irrigation ratio. The IR values were 1.0, 0.75, 0.50 and 0.25 for 11, I2, I3 and I4 irrigation levels, respectively. Total 13 irrigations were made during growing period of seedlings. Irrigation water amounts applied to the 11, I2, I3 and I4 treatments were 82, 61.5, 41 and 20.5 mm, respectively.

Seasonal actual evapotranspiration (ETa) of cabbage seedlings grown in pots was calculated considering the soil water balance equation (Allen *et al.*, 1998):

(2)  $ETa = I - D \pm \Delta S$ 

Where, ETa is the seasonal actual evapotranspiration (mm), I is the irrigation quantity (mm), D is the discharged water out of from pot bottom (mm), and  $\Delta$ S is the change of pot moisture content (mm). The discharge water in the I1 treatment was totally 12.9 mm during irrigation period. However, the discharge was not observed in the other irrigation treatments. Seasonal cabbage seedling actual evapotranspiration values were 76.9, 69.9, 53.6 and 39.3 mm in the I1, I2, I3 and I4 treatments, respectively.

Therefore, actual evapotranspiration values in the I2, I3 and I4 treatments were lower than the I1 treatment value by 9.1, 30.3 and 48.9%, respectively.

#### **Chlorophyll Reading Values**

Chlorophyll reading values were determined as SPAD by a portable chlorophyll meter (SPAD-502; Konica Minolta Sensing, Inc., Japan).

#### **Growth Parameters**

Forty-two days after transplanting, all the plants were harvested, and shoot fresh and root fresh weights, shoot dry and root dry weigths, stem diameter and leaf area were determined. The area of the green leaves was quantified with a leaf area meter (LI-3100, LI-COR).

#### **Hormone Analysis**

Extraction and purification processes were made according to Kuraishi *et al.* (1991) and Battal and Tileklioğlu (2001). The hormones were analyzed by high performance liquid chromatography (HPLC).

# Electrolyte Leakage (EL), H<sub>2</sub>O<sub>2</sub> and Malondialdehyde (MDA) Contents

Electrolyte leakage was executed as described by Campos *et al.* (2003).  $H_2O_2$  and the malondialdehyde (MDA) were executed as described by Ozden *et al.* (2009) and Zhang *et al.* (2005).

#### **Proline Content**

Proline content was executed as described by Bates *et al.* (1973).

#### **Antioxidant Enzymes Analysis**

Catalyse activity was measured on the rate of hydrogen peroxide decomposition according the method (Tejera García *et al.*, 2007). POX activity was measured to base its capability to turn guaiacol to tetraguaiacol at 436 nm, analysis of superoxide dismutase (SOD) activity is based on the determination of inhibition in the photochemical diminution of nitroblue tetrazolium at 560 nm according to the method (Abedi and Pakniyat, 2010).

#### **Statistical Analysis**

The data were subjected to analysis of variance (ANOVA) using SPSS 18 (SPSS Inc., 2010). The experimental design was a completely randomized design with four replications. The differences between the means were compared using the least significant difference test (LSD, p < 0.05).

#### Results

#### **Plant Growth**

Irrigation levels and PGPR treatments significantly (P < 0.05) affected the growth of cabbage seedlings (Table 1, Table 2). Shoot fresh and dry weight, root fresh and dry weight, stem diameter and leaf area of cabbage seedlings were lower at I2, I3 and I4 irrigation levels as compared to I1. However, PGPR applications improved these parameters at lower irrigation levels compared to control (no PGPR inoculation). The highest head fresh and dry weight, head height and stem diameter were recorded in PGPR inoculated plants in all irrigation levels. The greatest values were obtained from TV-6D for all growth parameters. Similarly, TV-6D application provided the highest SPAD values regardless of the irrigation treatments (Table 2). The mean fresh and dry weight values in the TV-6D were 60 and 35% higher than the values of control treatment, respectively. I4 irrigated plants inoculated with TV-6D had more root fresh weight by 75% than the control ones; however, in I3 irrigated plants, this increase was 71%. The increase of 60% shoot fresh weight was recorded in I4 irrigated TV-6D inoculated plants as compared to the control. TV-6D and KIN-37 inoculation increased leaf area by 53 and 16%, respectively, over I4 irrigated as compared with the control. The plant growth improved the highest of the bacterium B. megaterium TV-6D inoculated fully irrigated plants (Table 1). An increase of 41% stem diameter was observed in TV-6D treated plants compared to the uninoculated I4 irrigated.

TV-6D, TV-20E and KIN-37 treatments in I2 irrigations were significantly (P < 0.05) not different from control treatment in I1 in terms of seedling growth. Furthermore, growth parameters of TV-6D inoculated seedlings in I3 were situatistically (P < 0.05) similar to control seedlings in I2 (Table 1, Table 2).

#### **Hormone Concentration**

IAA, GA and SA contents gave significant reduction under lower irrigation levels but ABA increased under water deficit conditions (Fig. 1). The application of combination of TV-6D+RK-92+TV-53D caused to the increase in IAA and GA production fully irrigated control treatment, which peaks (1.42 and 87.7 ng/ $\mu$ L) at control treatment and then starts declining at lower irrigation levels (Fig. 1a, 1). The production of IAA and GA in I4 was 0.79 and 76.9  $ng/\mu L$ with combination of TV-6D+RK-92+TV-53D respectively, whereas the production was 0.66 and 60 ng/µL under uninoculated conditions, respectively. In respect to the ABA and SA provision, different results were also observed in cabbage plants as affected by PGPR applications in different irrigation levels. ABA content drastically increased with decreasing irrigation levels. The greatest ABA values at low irrigation levels generally obtained from TV-6D and combination of TV-6D+RK-92+TV-53D.

		Shoot fresh wt (g plant <sup>-1</sup> )	Shoot dry wt (g plant <sup>-1</sup> )	Root fresh wt (g plant <sup>-1</sup> )	Root dry wt (g plant <sup>-1</sup> )
PGPR	Irrigations levels	~~ · · ·	• • • •		• • • •
	I1	100,11d	11.18cd	8,13bc	0.90cde
Control	I2	62,81gh	8.56def	5,45def	0.64gh
	13	34,99k	4.50gh	3,09fgh	0.49ijk
	I4	7,71	1.55i	2,35hi	0.37jkl
	Mean	51,40D**	6.45B**	4.76C*	0.60B*
	I1	174,08a	15.21a	13,44a	1.17b
TV-6D	12	88,93de	10.45cd	7,27bc	0.81def
	I3	53,50hij	6.93f	5,28d-g	0.65f-i
	I4	12,37 1	2.41h	4,106d-i	0.61f-i
	Mean	82.22A	8.75A	7.52A	0.82A
	I1	128,47c	15.41a	12,81a	1.43a
TV-20E	I2	92,65de	11.32c	7,99bc	0.96bcd
	13	38,30jk	4.64gh	2,93ghi	0.47ijk
	I4	7,551	1.27i	1,92i	0.36jkl
	Mean	66.74BC	8.16A	6.41AB	0.80Å
	I1	157,09b	14.38ab	12,33a	1.12bc
KIN-37	12	94,44de	10.35cde	4,78d-h	0.72efg
	I3	34,15k	4.97g	4,26d-i	0.56h-j
	I4	10,921	1.84hi	3,37e-i	0.52h-k
	Mean	74.15AB	7.89AB	6.18AB	0.73AB
	I1	126,50c	12.07bc	8,40b	0.80d-g
6D+92+53D	I2	73,86 fg	8.36ef	5,91bcd	0.68e-i
	13	42,89ijk	4.97g	5,89cde	0.66f-i
	I4	7,311	1.46i	1,92i	0.231
	Mean	62.64CD	6.71B	5.53 BC	0.59B
	Irrigation	**	**	**	**
	Irrigation x PGPR	**	**	*	**

Table 1: Cabbage seedling growth in response to PGPR treatments under different irrigation levels <sup>z</sup>

<sup>2</sup>Means with the same letters within columns are not significantly different at p < 0.05 using LSD Test. ns: P > 0.05

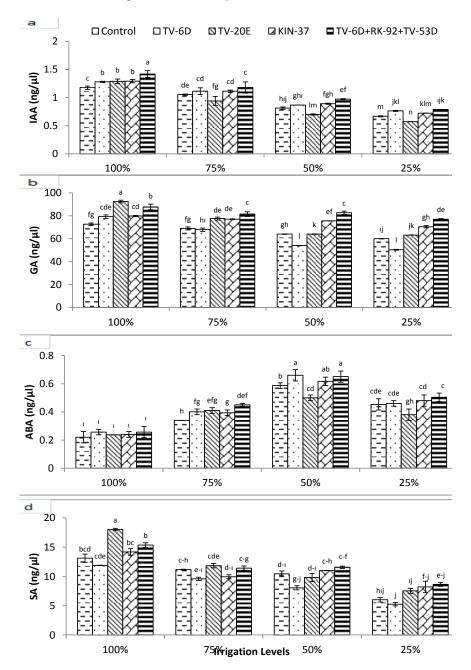
Table 2: Cabbage s	eedling sten	1 diameter. I	leaf area and i	in response to PGP	R treatments under	different irrigation levels <sup>z</sup>

		Stem Diam. (mm)	Leaf area (cm <sup>2</sup> )	Chlorophyll (SPAD)
PGPR	Irrigations levels			
	I1	10,34bcd	89,40d	56.00c-f
Control	I2	9,45cde	58,39fg	55.56def
	13	8,89ef	53,00gh	55.62def
	I4	5,12h	16,90j	54.36efg
	Mean	8.45B**	54.42C*	55.39C**
	I1	12,44a	117,62a	63.00a
ГV-6D	12	10,65b	81,65de	62.91b
	I3	10,14d	64,73ef	61.86ab
	I4	7,23g	25,82i	56.83cde
	Mean	10.11A	72.46A	61.15A
	I1	11,00b	111,21ab	56.56c-f
TV-20E	12	10,86b	92,89cd	55.03efg
	13	8,12fg	44,03h	54.65efg
	I4	4,91h	13,59j	56.05c-f
	Mean	8.72B	65.43AB	55.57C
	I1	13,46a	117,27ab	55.18efg
KIN-37	I2	9,63cde	89,58d	55.12efg
	13	7,96fg	43,81h	53.23fg
	I4	6,91g	19,56j	52.00g
	Mean	9.49A	67.56A	53.88C
	I1	9,91b-e	103,86bc	59.50abc
6D+92+53D	I2	8,55fg	69,01ef	59.01bcd
	13	8,02ef	54,88gh	56.24c-f
	I4	5,59h	12,83j	55.00efg
	Mean	8.02B	60.15BC	57.44B
	Irrigation	**	**	*
	Irrigation x PGPR	**	*	ns

<sup>2</sup>Means with the same letters within columns are not significantly different at p < 0.05 using LSD Test. ns: P > 0.05

Lowered irrigation levels decreased SA content regardless of PGPR inoculations. The greatest ABA and SA

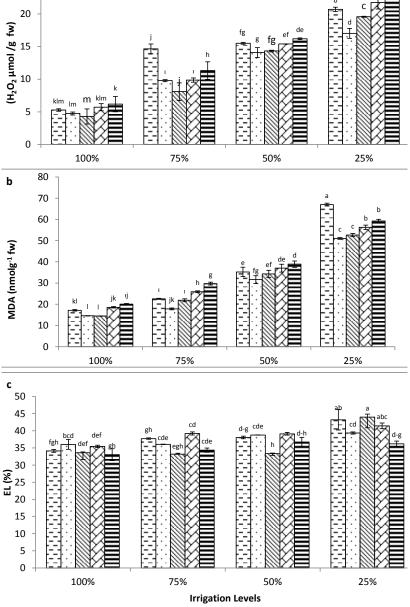
production was reached in combination of TV-6D+RK-92+TV-53D inoculated in I4 (Fig. 1c, d).



**Fig. 1:** Hormonal contents (Indole Acetic Acid-IAA (a), Gibberellic Acid-GA (b), Abscisic Acid-ABA (c), Salicylic Acid-SA (d)) of cabbage seedlings in response to PGPR treatments under different irrigation levels. Different letters on top of bars indicate differences (LSD, p < 0.05). Data are reported as means (n = 4). Vertical bars indicate the mean  $\pm$  SE

#### H<sub>2</sub>O<sub>2</sub>, MDA, EL

The MDA,  $H_2O_2$  and EL of cabbage seedlings were significantly (P < 0.05) influenced by PGPR treatments in different irrigation levels (Fig. 2a, b and c). Lower irrigation levels caused an increase in MDA,  $H_2O_2$  and EL. However, PGPR inoculations generally decreased these parameters of the cabbage seedlings grown under lower irrigation levels. TV-6D significantly (P < 0.05) reduced MDA,  $H_2O_2$  and EL compared to uninoculated control under deficient irrigated conditions inoculated (Fig. 2a, b and c). All bacterial treatments decreased  $H_2O_2$  compared to non-inoculated treatment in I2, but not in further deficient conditions. PGPR inoculated plants grown under severe drought conditions (I4) had lower MDA content than non-inoculated plants. Combination treatments of TV-6D+RK-92+TV-53D lowered EL values under water deficient conditions compared to control.

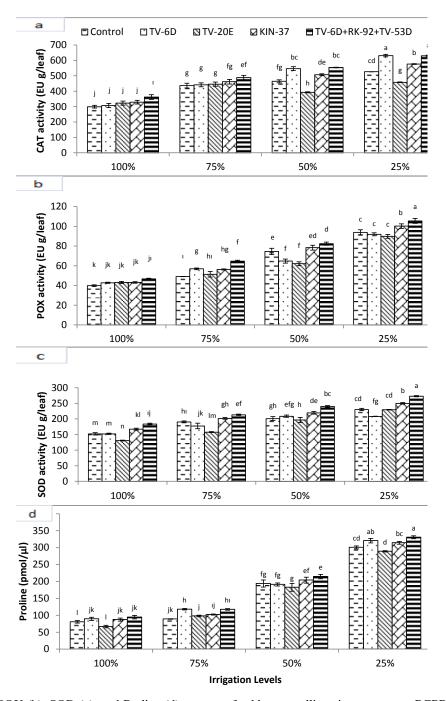


**Fig. 2:**  $H_2O_2$  (a), MDA-Malondialdehyde (b) and EC (Electrical conductivity) (c) of cabbage seedlings in response to PGPR treatments under different irrigation levels. Different letters on top of bars indicate differences (LSD, p < 0.05). Data are reported as means (n = 4). Vertical bars indicate the mean  $\pm$  SE

### **Antioxidant Enzymes and Proline**

The influence of PGPR treatments on the appropriate enzymatic system of cabbage seedlings in response to different irrigation levels was investigated by evaulating CAT, POX, and SOD activities (Fig. 3a, b and c). The CAT, POX, and SOD activities generally elevated with decreasing irrigation levels. TV-6D and combination of TV-6D+RK-92+TV-53D inoculations further increased CAT compared to non-inoculation control. Combination of TV-6D+RK-92+TV-53D treatment was more efficient than the other treatmens for POX and SOD activities under lower irrigation levels. POX content showed a similar trend with 1.13-fold increase for TV-6D+RK-92+TV-53D inoculated at I4 teratments when compared with uninoculated plants. Applications of PGPR caused to an increase in the SOD content in 25% irrigated teratments. The SOD activity peaked at TV-6D+RK-92+TV-53D inoculated plants (272.91 EU).

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**Fig. 3:** CAT (a), POX (b), SOD (c), and Proline (d) content of cabbage seedlings in response to PGPR treatments under different irrigation levels. Different letters on top of bars indicate differences (LSD, p < 0.05). Data are reported as means (n = 4). Vertical bars indicate the mean  $\pm$  SE

The proline accumulation started increasing in water deficiencies. Especially, under severe drought conditions this accumulation was drastic. When cabbage seedlings were treated with TV-6D+RK-92+TV-53D and TV-6D under lower irrigation levels, proline content significantly (p<0.05) raised compared to the control plants. The rate determination of proline content was 4.09-fold increase with TV-6D+RK-92+TV-53D inoculated at 25% irrigated

teratments when compared with uninoculated fully irrigated plants (Fig. 3d).

#### Discussion

The lower irrigation levels drastically affected the growth of cabbage seedlings as indicated by stunted sedling growth, reduced stem diameter and leaf area and low chlorophyll reading value (Table 1 and 2). Plant growth, hormone content, antioxidant enzymes and proline accumulation increased in seedlings inoculated with PGPRs under drought stress as compared without PGPR teratment. In fact, inoculation with PGPR has been found effective under drought stress environment to increase plant growth and development, hormone content, nutrient content (Sahin *et al.*, 2015). No previous information reports the effects of TV-6D and combination of TV-6D+RK-92+TV-53D inoculations on cabbage seedlings under drought stress.

To explaine the positive effect of PGPR inoculations on plant growth under drought stress, differential mechanisms have been documented by previous studies. The positive role of PGPR plant growth under drought stress can be caused by nutritional, physiological, and cellular effects (Saravanakumar et al., 2011). In the present research the improved seedling growth parameters in response to inoculanted TV-6D compared with the control indicates the beneficial role of these rhizobacteria. Shoot and root fresh weight, stem dimension and leaf area of cabbage seedlings increased especially in inoculated seedlings with TV-6D under deficient irrigations. PGPR inoculations have been reported to enhance plant tolerance to drought by increasing their water content, which can be attributed to enhancement of root growth because of IAA produced by bacteria (Marulanda et al., 2009). Furtheremore, studies suggested that PGPR could ameliorate the deleterious effect of stress conditions on plant growth by producing ACC deaminase, IAA, GA and cytokinins (Turan et al., 2014). In fact, PGPR strains used in this study have been reported to produce ACC deaminase, IAA, SA and GA (Kotan et al., 2014; Turan et al., 2014).

Drought stress has been reported to stimulate production of reactive oxygen species (ROS), causing membrane injuries, protein degradation, enzyme inactivation and thus induce oxidative stress (Zlatev and Lidon, 2012). The present research revealed that seedling inoculation with TV-6D resulted in decreased EL, MDA and  $H_2O_2$  content in deficient irrigations. The electrolyte leakage EL lower in plants exposed to drought has been considered indicative of a relative tolerance to water stress. Naveed et al. (2014) suggested that bacterial inoculation did help plants to decrease their EL% as compared with uninoculated plants in drought stress. Drought stress is accompaniment of the foundation of reactive oxygen species such as O2, H2O2 and OH, which damage membranes (Mittler, 2002). The result of this study showed that lower irrigation levels caused an increase in H<sub>2</sub>O<sub>2</sub>, MDA and EL. However, PGPR treatments decreased these parameters. Abiotic stress conditions result in an increase accumulation of oxygen free radicals in plants. PGPR can protect the host plants by scavenging ROS and increasing the antioxidant enzyme activity. PGPR treated against membrane damages demonstrate the tolerance capability of the plants to combat or abide under water limited conditions where inoculation can be subsidiary. In our study, it was

observed that PGPR inoculations elevated the activities of SOD, POX, and CATcompared to the control. CAT, SOD and POX are essential enzymes to scavenge  $H_2O_2$  and in coping with oxidative stress caused by drought stress conditions. PGPR used in the study significantly stimulated the CAT activity in cabbage plants under drought stress. The PGPR treatments increased SOD content in bean under water deficit conditions (Sarma and Saikia, 2014). Kohler *et al.* (2008) pointed out that PGPR inoculation improved CAT accumulation and resulted in drought stress amelioration in lettuce.

Drought-stressed plants accumulate various molecules such as proline thereby protecting enzyme activity (Saravanakumar *et al.*, 2011). Proline as stress responsive molecule is often synthesized by plants in response to various abiotic stress conditions (Naveed *et al.* 2014). Kohler *et al.* (2008) declared that PGPR elevated proline content of lettuce under water deficit conditions, which was an important indicator for drought stress alleviation. In the present study, except for I2 irrigation levels, the proline content was higher in the uninoculated plants than in the TV-6D and TV-6D+RK-92+TV-53D-inoculated plants under stress exposure. Proline, an amino acid, is a compatible solute involved in cell osmotic adjustment (OA) and protection of cell components during dehydration (Zhang *et al.*, 2009).

Lower irrigation levels caused a reduction in IAA, GA and SA content but increased ABA content. Mia *et al.* (2012) suggested that growth promoting effects of PGPR on plants could be attributed to production of hormone strategies. The present research shows that TV-6D+RK-92+TV-53D inoculation elevated the IAA, GA and SA content under lower irrigation levels, which showes its particular ability to stimulate plant growth under abiotic stress conditions. PGPR strains used in the study might afect root hormone concentrations by producing plant hormones in the rhizosphere, which were then absorbed by the root (Turan *et al.*, 2014).

Researchers have recently identified cytokinin, gibberellin, auxin and ACC deaminase accumulation by PGPR (Timmusk and Wagner, 1999). PGPRs have been reported to stimulate plant growth regulator accumulation, having a positive effect in plant growth (Bent *et al.*, 2001). GA, SA, IAA production of PGPR can alleviate negative influence of drought stress on cabbage seedlings (Yuwono *et al.*, 2005). Positive effects of PGPR on plant growth can be attributed to their manipulating plant hormone pathways and chnaging plant stress response pathways (Atkinson and Urwin, 2012). The investigation on Bacillus pumilus TV-67C proved its IAA, GA and SA production capacity under drought stress.

#### Conclusion

The PGPR inoculations could modulate biochemical and physiological characteristics of cabbage seedlings under

water deficit conditions. Based on our results we concluded that application of TV-6D+RK-92+TV-53D and TV-6D was effective to improve physiology, membrane integrity and biomass of cabbage seedlings under reduced irrigation levels. The improved osmolyte accumulation and production of ROS scavenging enzymes ultimately leads to reduce negative influences of water deficit on growth of cabbage. It should be conducted experiments to understand performance in the field trails of the interaction of PGPRs and cabbage plants under drought stress.

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