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## Seedling invigouration by halo priming in tomato against salt stress

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### Abstract

A study on seedling invigouration in tomato against salt stress was undertaken by involving three levels of halo-priming [P (1, 2, 3% KNO<sub>3</sub>)] and two levels each of priming duration [D (24 and 48 hrs)] and salinity [S (2.5 and 5.0 EC)]. Halo-priming, priming duration and salinity individually as well as in interaction to each other influenced majority of growth, nutrient, biochemical and enzyme parameters of tomato seedlings. The interaction amongst all the factors also resulted in sufficiently good germination rate index and catalase activity (P1D2S2) under higher salinity displaying 12.09 and 0.80 µg<sup>-1</sup>, respectively. Whereas good amount of protein content (0.68 mg/g) and chlorophyll content (3.42 mg/g) were observed in P<sub>2</sub>D<sub>1</sub>S<sub>2</sub> and P<sub>1</sub>D<sub>2</sub>S<sub>2</sub>, respectively under higher salinity. It is inferred from the study that Halo-priming of tomato seeds either with 1% or 2% KNO<sub>3</sub> for 24 or 48 hrs of duration could invigourate tomato seedlings against salt stress.

**Keywords:** Halo-priming, salt stress, seedling invigouration, tomato.

### Introduction

Tomato (*Solanum lycopersicum* L.) is one of the most important vegetable belonging to family Solanaceae. Among various environmental stresses, soil salinity has become a critical problem worldwide due to its dramatic effects on plant physiology and performance. Soil salinity is a major factor limiting tomato production by affecting crop plant establishment. Germination is an important stage of seedling establishment and therefore it plays a key role in crop production. Salinity has an adverse effect on seedling growth of several crops, by creating an osmotic potential in the rhizosphere of the plant. This inhibits the absorption of water or creates toxic effect due to Na<sup>+</sup> and Cl<sup>-</sup> to the roots and the whole crop (Abraha and Yohannes, 2013) [1].

Priming is one of the most important physiological methods which improves the seed performance and provides faster and synchronized germination. Halo priming - a pre sowing seed treatment with inorganic salts like KNO<sub>3</sub>, CaCl<sub>2</sub>, KCl, NaCl, NaNO<sub>3</sub>, MnSO<sub>4</sub>, MgCl<sub>2</sub> etc. is an easy, low cost and low risk technique and an alternative approach to overcome the salinity problem in agricultural lands. It has been shown to improve germination and plant establishment under saline conditions in different plants. Priming is a valuable tool for the improvement of seedling quality in tomato seedling production (Mavi *et al.* 2006) [35]. Seed priming stimulates many of the metabolic processes involved with the early phases of germination and it has been noted that seedlings from primed seeds emerge faster, grow more vigorously and perform better in adverse conditions (Chavan *et al.* 2014 and Agawane and Parhe, 2015) [15, 5]. Duration of priming also plays an important role in strengthening the survival and vigour of seedlings under salinity conditions. Priming with potassium nitrate (KNO<sub>3</sub>), PEG or NaCl have been shown to improve the germination, seedling emergence and the initial growth of various plant species (Govinden and Levantard, 2008; Zhang *et al.* 2012) [27, 49]. Potassium nitrate has also been observed to be more effective than PEG in improving the germination speed in tomato (Frett *et al.* 1991) [25]. During sub-optimal environmental conditions like salinity and drought, the contents of compatible solutes inside the seeds such as malondialdehyde (MDA), proline and soluble sugar (SS) and the activity of protective enzymes such as superoxide dismutase (SOD), peroxidase (POD), catalase (CAT) are important indicators (Bohnert *et al.* 1999) [13], which may decide crop resistance to various stresses like salinity and drought. Therefore, the present study was planned to investigate the effect of halo priming on seedling invigouration and performance under salt stress.

### Materials and Methods

The experiment was carried out at Regional Horticultural Research Station (RHRS), ASPEE College of Horticulture and Forestry (ACHF), Navsari Agricultural University, Navsari,

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Gujarat, India, during May, 2016. The location is situated at a latitude of 20° 57' N and 72° 54' E, respectively with an altitude of about 12 m above the mean sea level. The experiment involved 12 different treatments consisting of 3 levels of halo priming (1%, 2% and 3% KNO<sub>3</sub>), 2 levels of priming duration (24 hrs and 48 hrs) and 2 of salinity (2.5 EC and 5 EC) which was laid out in a Completely Randomized Design with three replications. Seeds of tomato cv. GT-2 were surface sterilized with 5 % sodium hypochlorite solution for 5-10 minutes and rinsed three times with distilled water. Then, surface sterilized seeds were primed in 1%, 2% and 3% KNO<sub>3</sub> for different time durations *viz.*, 24 and 48 hrs. Thereafter, seeds were dried to appropriate moisture under shade and 100 seeds in each replication were sown in plug trays containing coco-peat, perlite and vermiculite (3:1:1 on volume basis). So, nutrient solution was standardized with NaCl (AR) to create salinity levels of 2.5 dS/m and 5.0 dS/m. Growth parameters like seedling survival, germination rate index, germination index, final germination percentage, leaf area index, fresh and dry weight of seedlings were recorded as per the procedure described in ISTA (1999) [30] and by Al-Mudaris (1998) [9]. Nutrient and biochemical parameters were analyzed as per the methodology described by Rnaganna (1999) [41].

The enzymatic of catalase was measured according to method described by Aebi (1984) [2] and peroxidase activity was determined by standard procedure (Allam and Hollis, 1972) [8]. Superoxide dismutase activity was determined by measuring its ability to inhibit the photochemical reduction of nitroblue tetrazolium (NBT) using the method of Beauchamp and Fridovich (1971) [11]. Ascorbate peroxidase enzyme activity was measured from fresh extracts and was assayed as described by Costa *et al.* (2005) [18]. The observations recorded were subjected to statistical analysis through WASP-Web Agri Stat. Package 2.0 (Anon., 2016) [10].

## Results

### Main effect of halo priming

**Table 1:** Main effect of halo priming, priming duration and salinity on growth and nutrient parameters of tomato seedlings.

| Treatments          | Growth parameters |      |        |       |                 |                           |                         | Nutrient parameters |               |             |
|---------------------|-------------------|------|--------|-------|-----------------|---------------------------|-------------------------|---------------------|---------------|-------------|
|                     | Seedling survival | GRI  | GI     | FGP   | Leaf area index | Fresh weight of seedlings | Dry weight of seedlings | Nitrogen (N)        | Potassium (K) | Sodium (Na) |
| P <sub>1</sub>      | 65.25             | 8.26 | 335.92 | 69.08 | 0.61            | 1.69                      | 1.10                    | 6.97                | 26.76         | 16.43       |
| P <sub>2</sub>      | 65.75             | 9.72 | 461.67 | 89.33 | 0.61            | 1.76                      | 1.15                    | 7.59                | 29.49         | 13.17       |
| P <sub>3</sub>      | 52.75             | 5.36 | 237.42 | 48.25 | 0.59            | 1.56                      | 0.98                    | 7.58                | 27.27         | 15.55       |
| LSD <sub>0.05</sub> | 3.10              | 0.46 | 23.63  | 4.43  | NS              | 0.08                      | 0.05                    | 0.33                | 1.41          | 0.88        |
| D <sub>1</sub>      | 57.56             | 7.14 | 343.56 | 69.06 | 0.62            | 1.65                      | 1.05                    | 7.36                | 28.52         | 15.41       |
| D <sub>2</sub>      | 64.94             | 8.41 | 346.44 | 68.72 | 0.59            | 1.70                      | 1.10                    | 4.39                | 27.16         | 14.70       |
| LSD <sub>0.05</sub> | 2.53              | 0.37 | NS     | NS    | 0.03            | NS                        | 0.04                    | NS                  | 1.15          | NS          |
| S <sub>1</sub>      | 75.89             | 8.72 | 377.89 | 74.67 | 0.63            | 1.72                      | 1.12                    | 7.99                | 32.40         | 15.20       |
| S <sub>2</sub>      | 46.61             | 6.84 | 312.11 | 63.11 | 0.58            | 1.63                      | 1.03                    | 6.76                | 23.28         | 14.90       |
| LSD <sub>0.05</sub> | 2.53              | 0.37 | 19.29  | 3.62  | 0.03            | 0.07                      | NS                      | 0.27                | 1.15          | NS          |

**Table 2:** Main effect of halo priming, priming duration and salinity on biochemical and enzymatic activity of tomato seedlings.

| Treatments          | Biochemical parameters |                 |                     | Enzymatic activity |                  |                            |                            |
|---------------------|------------------------|-----------------|---------------------|--------------------|------------------|----------------------------|----------------------------|
|                     | Total chlorophyll      | Protein content | Total soluble sugar | Catalase (CAT)     | Peroxidase (POD) | Superoxide dismutase (SOD) | Ascorbate peroxidase (APX) |
| P <sub>1</sub>      | 2.41                   | 2.41            | 2.41                | 0.56               | 0.38             | 41.17                      | 4.04                       |
| P <sub>2</sub>      | 2.53                   | 2.53            | 2.53                | 0.68               | 0.50             | 51.83                      | 4.37                       |
| P <sub>3</sub>      | 1.72                   | 1.72            | 1.72                | 0.32               | 0.27             | 35.75                      | 3.28                       |
| LSD <sub>0.05</sub> | 0.15                   | 0.15            | 0.15                | 0.02               | 0.02             | 2.74                       | 0.16                       |
| D <sub>1</sub>      | 2.21                   | 2.21            | 2.21                | 0.47               | 0.38             | 41.95                      | 3.68                       |
| D <sub>2</sub>      | 2.23                   | 2.23            | 2.23                | 0.56               | 0.39             | 43.89                      | 4.12                       |
| LSD <sub>0.05</sub> | NS                     | NS              | NS                  | 0.02               | 0.02             | NS                         | 0.13                       |

The main effect of halo-priming was observed to be significant for majority of parameters and P<sub>2</sub> level of halo-priming (2% KNO<sub>3</sub>) recorded maximum seedling survival (65.75%), growth rate index (9.72), germination index (461.67), final germination percentage (89.33%), fresh weight of seedlings (1.76 g), dry weight of seedlings (1.15 g), nitrogen (7.59 mg/g), potassium (29.49 mg/g), total chlorophyll (2.53 mg/g), protein (0.63 mg/g), soluble sugar content (19.71 mg/g), catalase (0.68 µg<sup>-1</sup>), peroxidase (0.50 µg<sup>-1</sup>), superoxide dismutase (51.83 µg<sup>-1</sup>) and ascorbate peroxidase activity (4.37 µg<sup>-1</sup>). Whereas the maximum sodium content of 16.43 mg/g was observed in P<sub>1</sub> level of halo-priming (Table 1 and 2).

### Main effect of priming duration

The main effect of priming duration recorded maximum seedling survival (64.94%), growth rate index (8.41), dry weight of seedlings (1.10 g), protein content (0.42 mg/g), catalase (0.56 µg<sup>-1</sup>), peroxidase (0.39 µg<sup>-1</sup>) and ascorbate peroxidase (4.12 µg<sup>-1</sup>) activity in D<sub>2</sub> level (48hrs) of priming duration. Whereas the maximum leaf area index (0.62 cm<sup>2</sup>) and potassium content (28.52 mg/g) were observed in D<sub>1</sub> level of priming duration (24hrs) (Table 1 and 2).

### Main effect of salinity

The main effect of salinity recoded maximum seedling survival (75.89%), growth rate index (8.72), germination index (377.89), final germination percentage (74.67%), leaf area index (0.63 cm<sup>2</sup>), fresh weight of seedlings (1.72 g), nitrogen (7.99 mg/g), potassium (32.40 mg/g), total chlorophyll content (2.65 mg/g), protein (0.44 mg/g), soluble sugar content (19.32 mg/g), catalase activity (0.56 µg<sup>-1</sup>), superoxide dismutase activity (46.22 µg<sup>-1</sup>) and ascorbate peroxidase (4.06 µg<sup>-1</sup>) were observed under S<sub>1</sub> level (2.5 EC) of salinity, whereas the maximum root-shoot ratio (0.75) and peroxidase activity (0.41 µg<sup>-1</sup>) were observed under S<sub>2</sub> level (5 EC) of salinity (Table 1 and 2).

|                     |      |      |      |      |      |       |      |
|---------------------|------|------|------|------|------|-------|------|
| S <sub>1</sub>      | 2.65 | 2.65 | 2.65 | 0.56 | 0.36 | 46.22 | 4.06 |
| S <sub>2</sub>      | 1.79 | 1.79 | 1.79 | 0.48 | 0.41 | 39.61 | 3.74 |
| LSD <sub>0.05</sub> | 0.10 | 0.10 | 0.10 | 0.02 | 0.02 | 2.24  | 0.13 |

### Interaction effect

Halo-priming and priming duration interacted positively and P<sub>1</sub>D<sub>2</sub> recorded maximum seedling survival (74.33%). Whereas, the higher germination rate index (10.88), dry weight of seedlings (1.18 g), total chlorophyll content (2.87 mg/g), soluble sugar content (20.98 mg/g), catalase activity (0.78 µg<sup>-1</sup>), peroxidase activity (0.53 µg<sup>-1</sup>), superoxide dismutase activity (55.33 µg<sup>-1</sup>) and ascorbate peroxidase activity (4.69 µg<sup>-1</sup>) were observed in P<sub>2</sub>D<sub>2</sub> and the maximum leaf area index (0.64 cm<sup>2</sup>) in P<sub>1</sub>D<sub>1</sub>. However, the maximum potassium (31.69 mg/g) and protein content (0.63 mg/g) was observed in P<sub>2</sub>D<sub>1</sub> (Table 3 and 4).

Under higher salinity, interaction between halo-priming and salinity (P<sub>1</sub>S<sub>2</sub>) observed maximum sodium content (17.60 mg/g) and performed well for seedling survival (54.17%) and dry weight of seedlings (1.08 g), whereas P<sub>2</sub>S<sub>2</sub> expressed sufficiently good germination rate index (7.87), germination index (431.67), final germination percentage (86.67%), potassium (26.68 mg/g), protein content (0.59 mg/g) and catalase activity (0.65 µg<sup>-1</sup>), which had at par performance with P<sub>2</sub>S<sub>1</sub> for final germination percentage (Table 3 and 4).

Under higher salinity, the interaction between priming duration and salinity (D<sub>2</sub>S<sub>2</sub>) performed well for seedling survival (52.33%), germination rate index (8.03), sodium (14.92 mg/g), total chlorophyll content (2.12 mg/g), soluble sugar content (18.54 mg/g) and catalase activity (0.53 µg<sup>-1</sup>), which had at par performance with D<sub>1</sub>S<sub>1</sub> for sodium. While, D<sub>1</sub>S<sub>2</sub> expressed sufficiently good leaf area index (0.62 cm<sup>2</sup>) and potassium content (24.84 mg/g), which had at performance with D<sub>2</sub>S<sub>1</sub> for leaf area index (Table 3 and 4).

The treatment combination P<sub>2</sub>D<sub>2</sub>S<sub>1</sub> showed maximum germination rate index (12.09), chlorophyll content (3.42 mg/g), protein content (0.68 mg/g) and catalase activity (0.80 µg<sup>-1</sup>) and the maximum potassium content (33.24 mg/g) was observed in P<sub>3</sub>D<sub>2</sub>S<sub>1</sub>, which was statistically at par with P<sub>2</sub>D<sub>1</sub>S<sub>2</sub>. The treatment combination P<sub>2</sub>D<sub>2</sub>S<sub>2</sub> performed well under higher salinity and showed sufficiently good germination rate index (9.67) and catalase activity (0.75 µg<sup>-1</sup>). However, P<sub>2</sub>D<sub>1</sub>S<sub>2</sub> showed good amount of protein content (0.61 mg/g) and P<sub>1</sub>D<sub>2</sub>S<sub>2</sub> showed good chlorophyll content (2.75 mg/g) under higher salinity (Table 5 and 6).

**Table 3:** Interaction effect between halo priming, priming duration and salinity on growth and nutrient parameters of tomato seedlings.

| Treatments                    | Growth parameters |       |        |       |                 |                           |                         | Nutrient parameters |               |             |
|-------------------------------|-------------------|-------|--------|-------|-----------------|---------------------------|-------------------------|---------------------|---------------|-------------|
|                               | Seedling survival | GRI   | GI     | FGP   | Leaf area index | Fresh weight of seedlings | Dry weight of seedlings | Nitrogen (N)        | Potassium (K) | Sodium (Na) |
| P <sub>1</sub> D <sub>1</sub> | 56.17             | 7.24  | 338.67 | 69.50 | 0.64            | 1.64                      | 1.05                    | 6.97                | 26.79         | 17.33       |
| P <sub>1</sub> D <sub>2</sub> | 74.33             | 9.28  | 333.17 | 68.67 | 0.57            | 1.75                      | 1.16                    | 6.96                | 26.73         | 15.53       |
| P <sub>2</sub> D <sub>1</sub> | 63.17             | 8.56  | 453.67 | 88.83 | 0.61            | 1.71                      | 1.11                    | 7.58                | 31.69         | 13.35       |
| P <sub>2</sub> D <sub>2</sub> | 68.33             | 10.88 | 469.67 | 89.83 | 0.62            | 1.81                      | 1.18                    | 7.60                | 27.28         | 12.99       |
| P <sub>3</sub> D <sub>1</sub> | 53.33             | 5.63  | 238.33 | 48.83 | 0.62            | 1.59                      | 1.00                    | 7.54                | 27.08         | 15.54       |
| P <sub>3</sub> D <sub>2</sub> | 52.17             | 5.09  | 236.50 | 47.67 | 0.57            | 1.54                      | 0.97                    | 7.62                | 27.46         | 15.57       |
| LSD <sub>0.05</sub>           | 4.38              | 0.65  | NS     | NS    | 0.04            | NS                        | 0.07                    | NS                  | 2.00          | NS          |
| P <sub>1</sub> S <sub>1</sub> | 76.33             | 9.05  | 397.50 | 82.67 | 0.63            | 1.72                      | 1.13                    | 7.48                | 32.16         | 15.26       |
| P <sub>1</sub> S <sub>2</sub> | 54.17             | 7.46  | 274.33 | 55.50 | 0.58            | 1.67                      | 1.08                    | 6.46                | 21.36         | 17.60       |
| P <sub>2</sub> S <sub>1</sub> | 84.00             | 11.57 | 491.67 | 92.00 | 0.63            | 1.85                      | 1.22                    | 8.16                | 32.30         | 13.21       |
| P <sub>2</sub> S <sub>2</sub> | 47.50             | 7.87  | 431.67 | 86.67 | 0.60            | 1.67                      | 1.07                    | 7.01                | 26.68         | 13.14       |
| P <sub>3</sub> S <sub>1</sub> | 67.33             | 5.54  | 244.50 | 49.33 | 0.63            | 1.59                      | 1.01                    | 8.33                | 32.74         | 17.14       |
| P <sub>3</sub> S <sub>2</sub> | 38.17             | 5.18  | 230.33 | 47.17 | 0.56            | 1.54                      | 0.95                    | 6.82                | 21.80         | 13.97       |
| LSD <sub>0.05</sub>           | 4.38              | 0.37  | 33.41  | 6.27  | NS              | NS                        | 0.07                    | NS                  | 1.15          | 1.25        |
| D <sub>1</sub> S <sub>1</sub> | 74.22             | 8.65  | 377.67 | 75.33 | 0.63            | 1.70                      | 1.11                    | 7.90                | 32.20         | 15.93       |
| D <sub>1</sub> S <sub>2</sub> | 40.89             | 5.64  | 309.44 | 62.78 | 0.62            | 1.59                      | 1.00                    | 6.82                | 24.84         | 14.89       |
| D <sub>2</sub> S <sub>1</sub> | 77.56             | 8.79  | 378.11 | 74.00 | 0.63            | 1.73                      | 1.13                    | 8.08                | 32.60         | 14.47       |
| D <sub>2</sub> S <sub>2</sub> | 52.33             | 8.03  | 314.78 | 63.44 | 0.54            | 1.66                      | 1.07                    | 6.70                | 21.72         | 14.92       |
| LSD <sub>0.05</sub>           | 3.58              | 0.53  | NS     | NS    | 0.04            | NS                        | NS                      | NS                  | 1.63          | 1.02        |

**Table 4:** Interaction effect of halo priming x priming duration, halo priming x salinity and priming duration x salinity on biochemical and enzymatic activity of tomato seedlings.

| Treatments                    | Biochemical parameters |                 |                     | Enzymatic activity |                  |                            |                            |
|-------------------------------|------------------------|-----------------|---------------------|--------------------|------------------|----------------------------|----------------------------|
|                               | Total chlorophyll      | Protein content | Total soluble sugar | Catalase (CAT)     | Peroxidase (POD) | Superoxide dismutase (SOD) | Ascorbate peroxidase (APX) |
| P <sub>1</sub> D <sub>1</sub> | 2.16                   | 0.29            | 18.19               | 0.43               | 0.33             | 38.17                      | 3.52                       |
| P <sub>1</sub> D <sub>2</sub> | 2.67                   | 0.48            | 18.87               | 0.69               | 0.44             | 44.17                      | 4.56                       |
| P <sub>2</sub> D <sub>1</sub> | 2.19                   | 0.63            | 18.43               | 0.58               | 0.47             | 48.33                      | 4.06                       |
| P <sub>2</sub> D <sub>2</sub> | 2.87                   | 0.62            | 20.98               | 0.78               | 0.53             | 55.33                      | 4.69                       |
| P <sub>3</sub> D <sub>1</sub> | 2.29                   | 0.26            | 18.50               | 0.41               | 0.32             | 39.33                      | 3.46                       |
| P <sub>3</sub> D <sub>2</sub> | 1.15                   | 0.16            | 16.89               | 0.23               | 0.22             | 32.17                      | 3.10                       |
| LSD <sub>0.05</sub>           | 0.17                   | 0.02            | 0.96                | 0.03               | 0.03             | 3.88                       | 0.23                       |
| P <sub>1</sub> S <sub>1</sub> | 2.79                   | 0.40            | 19.19               | 0.59               | 0.36             | 43.17                      | 4.14                       |
| P <sub>1</sub> S <sub>2</sub> | 2.04                   | 0.37            | 17.87               | 0.53               | 0.41             | 39.17                      | 3.95                       |
| P <sub>2</sub> S <sub>1</sub> | 3.04                   | 0.67            | 20.15               | 0.70               | 0.48             | 56.83                      | 4.52                       |
| P <sub>2</sub> S <sub>2</sub> | 2.02                   | 0.59            | 19.27               | 0.65               | 0.52             | 46.83                      | 4.22                       |

|                               |      |      |       |      |      |       |      |
|-------------------------------|------|------|-------|------|------|-------|------|
| P <sub>3</sub> S <sub>1</sub> | 2.12 | 0.24 | 18.62 | 0.38 | 0.24 | 38.67 | 3.52 |
| P <sub>3</sub> S <sub>2</sub> | 1.31 | 0.18 | 16.77 | 0.25 | 0.31 | 32.83 | 3.04 |
| LSD <sub>0.05</sub>           | NS   | 0.02 | NS    | 0.03 | NS   | NS    | NS   |
| D <sub>1</sub> S <sub>1</sub> | 2.97 | 0.42 | 19.35 | 0.52 | 0.34 | 45.56 | 3.84 |
| D <sub>1</sub> S <sub>2</sub> | 1.45 | 0.36 | 17.40 | 0.42 | 0.41 | 38.33 | 3.52 |
| D <sub>2</sub> S <sub>1</sub> | 2.33 | 0.45 | 19.29 | 0.62 | 0.37 | 46.89 | 4.27 |
| D <sub>2</sub> S <sub>2</sub> | 2.12 | 0.39 | 18.54 | 0.53 | 0.42 | 40.89 | 3.96 |
| LSD <sub>0.05</sub>           | 0.14 | NS   | NS    | 0.05 | NS   | NS    | NS   |

**Table 5:** Interaction effect of halo priming, priming duration and salinity on growth and nutrient parameters of tomato seedlings.

| Treatments                                   | Growth parameters |       |        |       |                 |                           |                         | Nutrient parameters |               |             |
|--|-------------------|-------|--------|-------|-----------------|---------------------------|-------------------------|---------------------|---------------|-------------|
|  | Seedling survival | GRI   | GI     | FGP   | Leaf area index | Fresh weight of seedlings | Dry weight of seedlings | Nitrogen (N)        | Potassium (K) | Sodium (Na) |
| P <sub>1</sub> D <sub>1</sub> S <sub>1</sub> | 69.00             | 9.00  | 411.67 | 84.33 | 0.67            | 1.68                      | 1.09                    | 7.47                | 32.16         | 16.87       |
| P <sub>1</sub> D <sub>1</sub> S <sub>2</sub> | 43.33             | 5.48  | 265.67 | 54.67 | 0.61            | 1.60                      | 1.01                    | 6.47                | 21.41         | 17.79       |
| P <sub>1</sub> D <sub>2</sub> S <sub>1</sub> | 83.67             | 9.11  | 383.33 | 81.00 | 0.60            | 1.75                      | 1.16                    | 7.48                | 32.16         | 13.64       |
| P <sub>1</sub> D <sub>2</sub> S <sub>2</sub> | 65.00             | 9.45  | 283.00 | 56.33 | 0.55            | 1.74                      | 1.15                    | 6.44                | 21.30         | 17.42       |
| P <sub>2</sub> D <sub>1</sub> S <sub>1</sub> | 83.33             | 11.05 | 476.67 | 91.33 | 0.59            | 1.81                      | 1.20                    | 8.00                | 32.20         | 13.58       |
| P <sub>2</sub> D <sub>1</sub> S <sub>2</sub> | 43.00             | 6.08  | 430.67 | 86.33 | 0.62            | 1.61                      | 1.02                    | 7.16                | 31.18         | 13.12       |
| P <sub>2</sub> D <sub>2</sub> S <sub>1</sub> | 84.67             | 12.09 | 506.67 | 92.67 | 0.66            | 1.89                      | 1.24                    | 8.33                | 32.39         | 12.83       |
| P <sub>2</sub> D <sub>2</sub> S <sub>2</sub> | 52.00             | 9.67  | 432.67 | 87.00 | 0.58            | 1.73                      | 1.11                    | 6.87                | 22.18         | 13.16       |
| P <sub>3</sub> D <sub>1</sub> S <sub>1</sub> | 70.33             | 5.89  | 244.67 | 50.33 | 0.62            | 1.62                      | 1.03                    | 8.23                | 32.24         | 17.33       |
| P <sub>3</sub> D <sub>1</sub> S <sub>2</sub> | 36.33             | 5.36  | 232.00 | 47.33 | 0.62            | 1.55                      | 0.96                    | 6.84                | 21.92         | 13.75       |
| P <sub>3</sub> D <sub>2</sub> S <sub>1</sub> | 64.33             | 5.18  | 244.33 | 48.33 | 0.63            | 1.56                      | 1.00                    | 8.43                | 33.24         | 16.95       |
| P <sub>3</sub> D <sub>2</sub> S <sub>2</sub> | 40.00             | 4.99  | 228.67 | 47.00 | 0.50            | 1.52                      | 0.94                    | 6.80                | 21.69         | 14.19       |
| LSD <sub>0.05</sub>                          | NS                | 0.92  | NS     | NS    | NS              | NS                        | NS                      | NS                  | 2.82          | NS          |

**Table 6:** Interaction effect of halo priming, priming duration and salinity on biochemical and enzymatic activity of tomato seedlings.

| Treatments                                   | Biochemical parameters |                 |                     | Enzymatic activity |                  |                            |                            |
|--|------------------------|-----------------|---------------------|--------------------|------------------|----------------------------|----------------------------|
|  | Total chlorophyll      | Protein content | Total soluble sugar | Catalase (CAT)     | Peroxidase (POD) | Superoxide dismutase (SOD) | Ascorbate peroxidase (APX) |
| P <sub>1</sub> D <sub>1</sub> S <sub>1</sub> | 2.99                   | 0.30            | 18.83               | 0.45               | 0.31             | 39.00                      | 3.57                       |
| P <sub>1</sub> D <sub>1</sub> S <sub>2</sub> | 1.32                   | 0.27            | 17.55               | 0.40               | 0.35             | 37.33                      | 3.47                       |
| P <sub>1</sub> D <sub>2</sub> S <sub>1</sub> | 2.59                   | 0.49            | 19.55               | 0.73               | 0.40             | 47.33                      | 4.70                       |
| P <sub>1</sub> D <sub>2</sub> S <sub>2</sub> | 2.75                   | 0.47            | 18.19               | 0.65               | 0.47             | 41.00                      | 4.43                       |
| P <sub>2</sub> D <sub>1</sub> S <sub>1</sub> | 2.66                   | 0.65            | 19.10               | 0.60               | 0.45             | 53.67                      | 4.16                       |
| P <sub>2</sub> D <sub>1</sub> S <sub>2</sub> | 1.72                   | 0.61            | 17.76               | 0.55               | 0.50             | 43.00                      | 3.96                       |
| P <sub>2</sub> D <sub>2</sub> S <sub>1</sub> | 3.42                   | 0.68            | 21.19               | 0.80               | 0.52             | 60.00                      | 4.89                       |
| P <sub>2</sub> D <sub>2</sub> S <sub>2</sub> | 2.32                   | 0.56            | 20.78               | 0.75               | 0.54             | 50.67                      | 4.49                       |
| P <sub>3</sub> D <sub>1</sub> S <sub>1</sub> | 3.26                   | 0.31            | 20.12               | 0.52               | 0.27             | 44.00                      | 3.80                       |
| P <sub>3</sub> D <sub>1</sub> S <sub>2</sub> | 1.31                   | 0.21            | 16.88               | 0.30               | 0.37             | 34.67                      | 3.12                       |
| P <sub>3</sub> D <sub>2</sub> S <sub>1</sub> | 0.98                   | 0.17            | 17.12               | 0.25               | 0.20             | 33.33                      | 3.24                       |
| P <sub>3</sub> D <sub>2</sub> S <sub>2</sub> | 1.31                   | 0.15            | 16.66               | 0.20               | 0.24             | 31.00                      | 2.95                       |
| LSD <sub>0.05</sub>                          | 0.24                   | 0.03            | NS                  | 0.05               | NS               | NS                         | NS                         |

## Discussion

### Growth parameters

The decrease in survival percentage in present investigation is probably either due to the increase in osmotic pressure or due to the toxicity of Na<sup>+</sup> ions. The germination was directly related to the amount of water absorbed and the delay in germination due to the salt concentration in the root zone, which ultimately responded to provide survival of seedlings (Demir and Oztokat, 2003) [19]. Similar kind of observations was also observed by Mahdi and Idris (2013) [34] who reported to have good survival of seedlings under higher level of salinity. The accelerated germination rate due to priming under salt stress may be due to an increase in water uptake rate to achieve the important moisture content required for germination or could be due to the acceleration of the rate of the cell division as calcium plays an important role in cell wall structure and cell division (Singh, 1984 and Bittencourt *et al.* 2005) [45, 12]. These findings are in accordance with the finding of earlier worker namely Nagarajan and Pandita (2001) [38], Zhang *et al.* (2012) [49], Ebrahimi *et al.* (2014) [22] and Ahmed *et al.* (2017) [7] who also observed higher

germination rate index as a result of halo priming of tomato seeds.

The germination was directly related to the amount of water absorbed and the delay in germination due to the salt concentration in the root zone (Farooq *et al.* 2005) [23]. The results of the current study are parallel to the findings of Haigh (1988) [28], Pill *et al.* (1991) [40], Nawaz *et al.* (2011) [39] and Ebrahimi *et al.* (2014) [22] in tomato and Khan *et al.* (2009) [32], Dutta and Singh (2015) [21] in hot pepper.

The decrease in the rate of photosynthesis is the result of the toxic effect of salt (NaCl) at higher salinity levels, which damage the roots and decrease their ability to absorb the water and nutrients which cause marked effects in leaf area index (Demir and Oztokat, 2003 and Farooq *et al.* 2005) [19, 23]. These findings are in agreement with Chen *et al.* (2009) [16]. Improved seedling fresh weights might be due to increased cell division within the apical meristem of seedling roots, which causes an increase in plant growth (Agrawal, 1993; Sivritepe *et al.* 2003 and Ghassemi *et al.* 2010) [6, 47, 26]. These findings are in agreement with Nawaz *et al.* (2011) [39] and Zhang *et al.* (2012) [49]. Improvement in seedling dry weight

has affected by increased the activities of dehydrogenases (an indicator of seed viability) and peroxidase (free radical scavenging enzyme). (Coolbear and Grierson, 1979; Agrawal, 1993 and Sivritepe *et al.* 2003) [17, 6, 47]. The results were also supported by the earlier findings of Afzal *et al.* (2008) [4], Nawaz *et al.* (2011) [39] and Zhang *et al.* (2012) [49].

#### Nutrient parameters

The water-soluble inhibitors in the seeds are nitrogen components, therefore halo priming increase leaching inhibitors during priming period (Smith, 1976) [48]. The results of current study were also in agreement with the findings of Chen *et al.* (2009) [16], Singh *et al.* (2012) [46] and Lara *et al.* (2014) [33] in tomato. Increased NaCl concentration amplifies Na<sup>+</sup> and Cl<sup>-</sup> contents in shoot and roots and K<sup>+</sup> contents get decreased at the same time. The high saline concentration increases Na<sup>+</sup> content and decreases K<sup>+</sup> content in affected plants. This is due to an antagonistic effect between sodium and potassium (Kaya *et al.* 2002 and Bybordi *et al.* 2010) [31, 14]. The results were also supported by the findings of earlier researchers Singh *et al.* (2012) [46].

#### Biochemical parameters

Salinity increase in photosynthetic tissues causes accumulation in adjacent grana membranes, shrinking of the thylakoid membranes and break-off of chlorophyll. Poor germination and less chlorophyll contents in tomato seedlings raised from seeds primed with higher concentration of salts (CaCl<sub>2</sub> or KNO<sub>3</sub>) might be due to increased uptake of toxic mineral elements such as Cl<sup>-</sup> or NO<sub>3</sub><sup>+</sup> (Franco *et al.*, 1993; Afzal *et al.* 2008 and Khan *et al.* 2009) [24, 4, 32]. The results of present study were also supported by Afzal *et al.* (2011) [3] and Saleethong *et al.* (2011) [42]. The positive effects of priming on seed germination of many species are attributed to the induction of biochemical mechanisms of cell repair. The resumption of the metabolic activity can restore cellular integrity through the synthesis of nucleic acids (DNA and RNA), proteins and the improvement of the antioxidant defense system (Di Girolamo and Barbanti, 2012) [20]. It appears that not only hydration but protein, sugar and RNA content got increased in halo primed seeds. Similar kind of observations was also observed by Lara *et al.* (2014) [33]; Mahdi and Idris (2013) [34] who reported to have good protein content under higher level of salinity. A strong association of earlier germination with increased hydrolytic enzyme activity and total sugars was reported in tomato (Handa *et al.* 1983) [29]. The present findings of maximum soluble sugar content in primed seeds over unprimed seeds are supported by the findings of Nawaz *et al.* (2011) [39].

#### Enzymatic activity

Catalase and superoxide dismutase are the main enzymes involved in the detoxification of the deleterious oxygen species. Alleviation of oxidative damages and maintaining integrity of the cellular membranes is a key mechanism in salinity tolerance. It is evident that priming can increase the activities of free radical scavenging enzymes, *i.e.*, catalase (CAT), ascorbate (APX) and peroxidase (POD) in seeds (Saxena, 1976; Saxena, 1980; Singh, 1984 and Mittova *et al.* 2003) [43, 44, 45, 37]. Whereas, Saxena (1976) [43]; Saxena (1980) [44] and Singh (1984) [45] were of the opinion that improvement in germination is not only due to hydration but enzymatic activation of catalase, peroxidase, amylase and invertase. The results of the current study are parallel to the findings of Lara *et al.* (2014) [33]; Mahdi and Idris (2013) [34]. Superoxide

dismutase (SOD) is the main enzyme involved in the oxidative stress that leads to the formation of reactive oxygen species as a result of anaerobic metabolism. The results of present study were supported by Lara *et al.* (2014) [33]; Mahdi and Idris (2013) [34]. Although priming with KNO<sub>3</sub> increased the activity of the SOD and CAT in tomato seeds, the reduction in the activity of APX was observed. The reduction of APX activity could be related to small concentration of H<sub>2</sub>O<sub>2</sub> required for its operationality, which is owned to its high affinity for substrate (Mittler and Zilinskas, 1991) [36]. The results of current study are parallel to the findings of Lara *et al.* (2014) [33] who observed the similar kind of activity of APX under salinity.

On the basis of present finding it can be concluded that different combinations of halo-priming, priming duration and salinity has significantly increased growth, nutrient, biochemical as well as enzymatic activities. Halo-priming of tomato seeds either with 1% or 2% KNO<sub>3</sub> for 24 or 48 hrs of duration could invigourate tomato seedlings against salt stress and may be served as a better alternative for boosting up survival and performance of tomato seedlings under higher salinity conditions.

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