



Original article

Influence of gibberellic acid and different salt concentrations on germination percentage and physiological parameters of oat cultivars

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ABSTRACT

Gibberellic acid (GA_3) is one of the plant growth regulators which improve salt tolerance and mitigate the salt stress impact on plants. The extant analysis was carried out to study the effect of GA_3 and different salt concentrations on seed germination and physiological parameters of oat cultivars. Oats is substantially less tolerant to salt than wheat and barley. Experimentation was conducted as factorial with Completely Randomized Block Design with three replicates. Different concentration of NaCl salt ((25, 50, 75 and 100 mM) were used in test control group and 100 and 150 ppm of GA_3 were used in two group by pre-treated (after 24 h of the seed soaking) and plants were analyzed on 15th day. Results indicate that increasing salinity would decrease the germination percentage and growth parameter in three oat cultivars. Quotes data indicating a 13%, 19.9% and 32.48% in cultivars NDO-2, UPO-212 and UPO-94 germination reduction when soil salinity reaches 50 mM. A 36.02%, 47.33% and 56.365% reduction in germination is likely when soil salinity reaches 100 mM respectively same cultivars. Seeds treated with GA_3 significantly promoted the percentage of germination, shoot and root length, total fresh and dry weight of seedling, tissue water content and seedling vigor index by NDO-2 and UPO-212 under different saline concentration. The maximum average of germination and growth parameters were observed from 150 ppm GA_3 treated seeds. But this concentration was significantly inhibited root length in sensitive cultivar UPO-94 at 75 and 100 mM salt as compared to 100 ppm. We observed that, the high concentration of GA_3 was not suitable for sensitive oat cultivars. Because the plant root are the real workforce behind any plants success. Thus, it may be concluding that, GA_3 treatment could curtail the toxic effect of salinity by increasing germination percentage and shoot and root length, total fresh and dry weight, tissue water content and seedling vigor index in tolerant cultivar.

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1. Introduction

Salinity stress is the most severe factors limiting the germination and productivity of crop plants because mostly crop plants are susceptible to salinity caused by higher concentration of salt in the soil and constitutes a issue concerned a significant segment of the plant, especially in region with hot and dry climates (Bazakos et al., 2015). It was estimated that about 20% (45 million ha) of irrigated land, producing one-third of the world's food which is salt affected (Shrivastava and Kumar, 2015). High concentrations of soil salinity adverse effects the quality and quantity of crop production (Chrysargyris et al., 2018) by inhibiting seeds germination,

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seedling growth and developmental phases owing to the collective impact of high osmotic potential and distinct ion toxicity (Gloria et al., 2016). Increased salinity caused a significant deficiency in germination percentage, length of shoot and root and fresh weight of shoot and root (El-Shaieny, 2015; Nasri et al., 2017). Plants face two basal problems in salinity environments. First of all, excess salt in soil lowers the osmotic potential of soil solution and reduced water uptake and deficit by plant. Second, increased uptake of Na⁺ and Cl⁻ ions distract the absorption of necessary minerals and impute toxicity to plants (Tester and Devenport, 2003).

GA₃ are plant growth regulators that are also known to induce different physiological responses in plants (Chauhan et al., 2016), which is unduly suited for stimulating and improving germination, plant growth and photosynthetic activity (Rout et al., 2017). Considering the tendencies of growth reduction due to salinity and progressive efficacy of exogenous GA₃ application on different morphological, physiological and biochemical activities, it can be described that application of GA₃ is useful to detract salinity stress and its effectiveness is more dynamic to salt tolerant cultivar (Misratia et al., 2015). Gibberellic acid partially mitigates the harmful effect of salinity by increasing vigor, anti oxidative enzyme activity and accumulation of osmolytes (Neelambari et al., 2018). The application of GA₃ increase the development of plant by attributing the fact that they increase the amino acid content in embryo and stimulate the syntheses of hydrolytic enzyme required for digestion of endospermic starch when seeds renew growth at seed germination (Shekafandeh et al., 2017). The objective of this study was to appraise the possible mode of interplay between different salt concentration and GA₃ application and unearth how to GA₃ can alleviate the harmful effect of salinity.

2. Materials and methods

The certified seeds of oat (NDO-2, UPO-212 and UPO-94) were obtained from different sources. The seeds of UPO-94, NDO-2 and UPO-212 were procured from Pant University of Agriculture and Technology, Pantnagar (Uttarakhand). Oat seeds choosed for germination behavior under growth regulator (100 and 150 mg/L GA₃) at different salt levels to confirm the seedling growth execution for salinity tolerance among oat cultivars.

2.1. Treatment and experimental design

To study the response of oats cultivars to GA₃ during salt stress a pot experiment was carried out. The experiment was set up with Randomly Complete Block Design (RCBD) with three replicates, the first factor include under 4 NaCl levels (25, 50, 75 and 100 mM) were prepared by U.S. Salinity Laboratory Staff Handbook (1954). The second and third factor included two concentration of gibberellic acid (GA₃) i.e. 100 and 150 ppm under four NaCl concentrations. The second factor with 100 ppm and third factor with 150 ppm GA₃ application were treated after 24 h of the soaking

of seed. Seeds of test cultivars were surface sterilized with 0.01% HgCl₂ (Mercuric chloride) for one minutes to avoid microbial and fungal infection and taken out immediately, then washed repeatedly with distilled water (DW). Twenty-five seeds of uniform size for each cultivar were germinated in small plastic pots (diameter of 20.22 cm and 22.10 cm in depth). After this, 40 ml aqueous solution of different salt concentrations was adding in the salt-treated pot on the first day for moister.

*Germination percentage was conducted as described in previous works (Chauhan et al., 2016). Physiological studies (shoot and root length and fresh and dry weight) was measured after 15 days.

***Germination Percentatge (%)**: It was calculated based the International Seed Testing Association (ISTA) method.

$$\text{Germination \%} = \left(\frac{\text{Number of Germinated Seeds}}{\text{Number of Total Seeds}} \right) \times 100$$

***Tissue Water Content (TWC)**: TWC was calculated by the following formula (Black and Pritchard, 2002).

$$\text{TWC} = \frac{\text{Fresh Weight} - \text{Dry weight}}{\text{Fresh Weight}} \times 100$$

***Seedling Vigor Index (I)** = SVI (I) was calculated based on the formula used by Abdalbaki and Anderson (1970).

$$\text{SVI(I)} = \text{Germination Percentage} \times [\text{Root Length(cm)} + \text{Shoot Length(cm)}]$$

***Seedling Vigor Index (II)** = SVI (II) was calculated by using this formula (Hossein and Kasra, 2011).

$$\text{SVI(II)} = \text{Dry Weight of Seedling(mg)} \times \text{Germination Percentage}$$

2.2. Statistical analysis

The data were statistically analyzed following RCBD design by SPSS Statistical Computer Package (SPSS for WINDOWS, Standard Version 20.0). Basic statistical parameters such as mean, standard deviation (SD) were computed along with one way analysis of variance (ANOVA).

3. Results and discussion

All tested observations (ANOVA) shows that the salinity has significant effect on sensitive cultivar (UPO-94). The cultivar NDO-2 shows a high interaction between salinity and GA₃ treatment at all parameters except germination (Table 1).

3.1. Seed germination

In Present study, we observed that seed germination were decrease by increase salinity concentrations, and salt-stress-induced suppression of seed germination was alleviated by GA₃ (Fig. 1a and Table 2). Salinity, in usual case has the inhibitory impact on germination of seeds in wheat, (Zhang et al., 2013) in

Table 1

ANOVA results for the effect of salt stress and application of GA₃ on germination and seedling growth parameters of three cultivars.

| Varieties | Sources of variation | df | F-ratio | | | | | | | |
|-----------|---------------------------|----|--------------------|--------------------|--------------------|----------------------|----------------------|--------------------|---------------------------|----------------------------|
| | | | Germ-nation | Shoot length (cm) | Root length (cm) | TDW of seedling (mg) | TFW of seedling (mg) | TWC of seedling | Seedling vigour index (I) | Seedling vigour index (II) |
| UPO-94 | (S) | 4 | 38.2** | 3.23** | 6.23** | 4.58* | 31.80** | 41.97** | 10.63** | 11.18** |
| | (GA ₃ T) × (S) | 2 | 0.38 ^{ns} | 4.50* | 2.17 ^{ns} | 3.23 ^{ns} | 0.44* | 0.33 ^{ns} | 1.26 ^{ns} | 1.24 ^{ns} |
| UPO-212 | (S) | 4 | 33.29* | 8.00** | 2.71* | 2.00* | 10.51** | 13.57** | 7.13* | 6.76** |
| | (GA ₃ T) × (S) | 2 | 5.83* | 1.77* | 5.45 ^{ns} | 7.48** | 1.50* | 1.00 ^{ns} | 2.00 ^{ns} | 2.16* |
| NDO-2 | (S) | 4 | 2.28 ^{ns} | 0.57 ^{ns} | 1.00 ^{ns} | 0.77 ^{ns} | 3.88* | 13.58** | 1.14 ^{ns} | 1.08 ^{ns} |
| | (GA ₃ T) × (S) | 2 | 6.43** | 9.46** | 14.80** | 19.19** | 3.48** | 2.75* | 12.01** | 13.01** |

(ns) Not-significant, (*) significant at 5%, (**) significant at 1%, (df) degree of freedom, (S) salinity and (T) treatment.

oat species and (Dheebea et al., 2015) in black gram. The cultivar UPO-212 exhibited highest germination percentage at all levels of salinity inclusive control, but the lowest reduction was achieved by cultivar NDO-2 in all treatment. At high salinity levels (75 and 100 mM), higher germination percentage (62.10–52.67%) was achieved in cv. UPO-212 followed NDO-2 (50.22–43.80%) and least percentage (33.00–27.10%) in UPO-94. Present results indicate that the percentage germination reduced significantly with an increase in saline concentration in oat cultivars. Similar results were reported earlier by Benliglu and Ozkan (2016) in oats.

Under 150 ppm GA₃ concentrations, tolerant cultivar NDO-2 revealed higher germination percentage whereas sensitive cultivar UPO-94 showed low germination as compared to 100 ppm GA₃ concentration under different salinity levels. GA₃ has reported to increase germination percentage and pull off the preventive effects of the salinity stress on germination (Patel and Mankad, 2014)). The growth in germination percentage with GA₃ might be due to involvement of GA₃ in the activation of cytological enzymes along with increase in cell wall plasticity and improve water absorption (Padma et al., 2013).

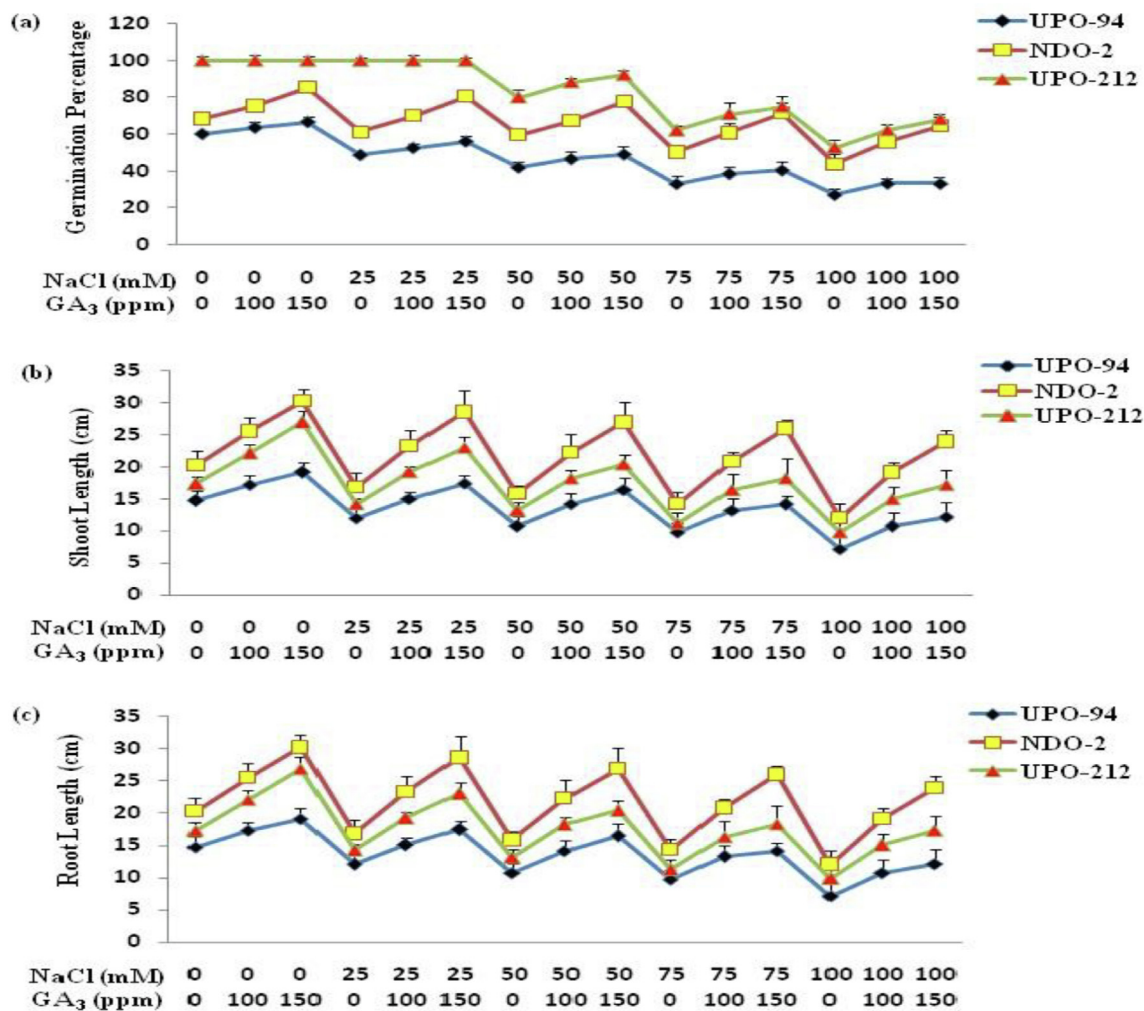


Fig. 1. Effects of NaCl and GA₃ concentrations on (a) seed germination and (b) length of shoot and (c) length of root of three oat cultivars.

Table 2
Average comparison of salt stress and applications of GA₃ on germination and growth parameters.

| Varieties | Treatments | Germination (%) | Shoot length (cm) | Root length (cm) | TDW of seedling (mg) |
|-----------|-------------------------------|-----------------|-------------------|------------------|----------------------|
| UPO-94 | (S) | 42.18 ± 13.00 | 10.84 ± 2.80 | 6.52 ± 2.55 | 12.45 ± 4.31 |
| | (S) × 100 ppm GA ₃ | 46.84 ± 11.92 | 14.05 ± 2.41 | 8.34 ± 1.95 | 15.33 ± 3.84 |
| | (S) × 150 ppm GA ₃ | 49.16 ± 13.44 | 15.81 ± 2.75 | 9.21 ± 3.12 | 18.05 ± 4.26 |
| UPO-212 | (S) | 78.97 ± 21.58 | 13.11 ± 2.87 | 8.37 ± 2.24 | 16.86 ± 5.65 |
| | (S) × 100 ppm GA ₃ | 84.26 ± 17.16 | 18.16 ± 2.74 | 10.75 ± 2.26 | 20.00 ± 4.97 |
| | (S) × 150 ppm GA ₃ | 87.06 ± 14.71 | 21.13 ± 3.89 | 12.56 ± 2.07 | 27.81 ± 4.99 |
| NDO-2 | (S) | 56.64 ± 9.68 | 15.59 ± 2.71 | 10.76 ± 2.29 | 19.25 ± 4.58 |
| | (S) × 100 ppm GA ₃ | 65.87 ± 7.68 | 22.15 ± 2.43 | 14.41 ± 2.31 | 26.38 ± 4.20 |
| | (S) × 150 ppm GA ₃ | 76.03 ± 8.16 | 27.07 ± 2.40 | 17.20 ± 2.61 | 37.29 ± 5.09 |

Results were representing the average of five replicates ± SD, (S) salinity, (TDW) total dry weight.

Table 3
Average comparison of salinity stress and applications of GA₃ on seedling growth and vigour index.

| Varieties | Treatments | TFW of seedling (mg) | TWC of seedling | SVI (l) (mg) | SVI (l) (mg) |
|-----------|-------------------------------|----------------------|-----------------|------------------|------------------|
| UPO-94 | (S) | 116.14 ± 41.22 | 105.36 ± 41.45 | 796.73 ± 454.40 | 567.93 ± 358.54 |
| | (S) × 100 ppm GA ₃ | 130.09 ± 36.57 | 118.24 ± 36.89 | 1089.05 ± 479.44 | 754.37 ± 378.12 |
| | (S) × 150 ppm GA ₃ | 139.10 ± 36.40 | 126.05 ± 36.73 | 1286.43 ± 611.46 | 928.60 ± 459.87 |
| UPO-212 | (S) | 131.88 ± 36.79 | 119.08 ± 36.85 | 1777.05 ± 848.26 | 1404.86 ± 713.84 |
| | (S) × 100 ppm GA ₃ | 150.54 ± 29.75 | 135.85 ± 29.85 | 2499.24 ± 878.64 | 1903.62 ± 733.42 |
| | (S) × 150 ppm GA ₃ | 164.37 ± 28.35 | 146.16 ± 28.28 | 2997.06 ± 979.73 | 2398.88 ± 733.42 |
| NDO-2 | (S) | 155.14 ± 37.53 | 142.71 ± 37.59 | 1530.39 ± 535.35 | 1125.52 ± 444.13 |
| | (S) × 100 ppm GA ₃ | 178.87 ± 28.50 | 164.13 ± 28.49 | 2437.17 ± 593.44 | 1763.43 ± 479.69 |
| | (S) × 150 ppm GA ₃ | 201.19 ± 23.06 | 183.95 ± 22.77 | 3398.30 ± 734.46 | 2669.43 ± 624.40 |

Results were representing the average of five replicates ± SD, (S) salinity, (TFW) total fresh weight, (TWC) tissue water content and (SVI) seedling vigor index.

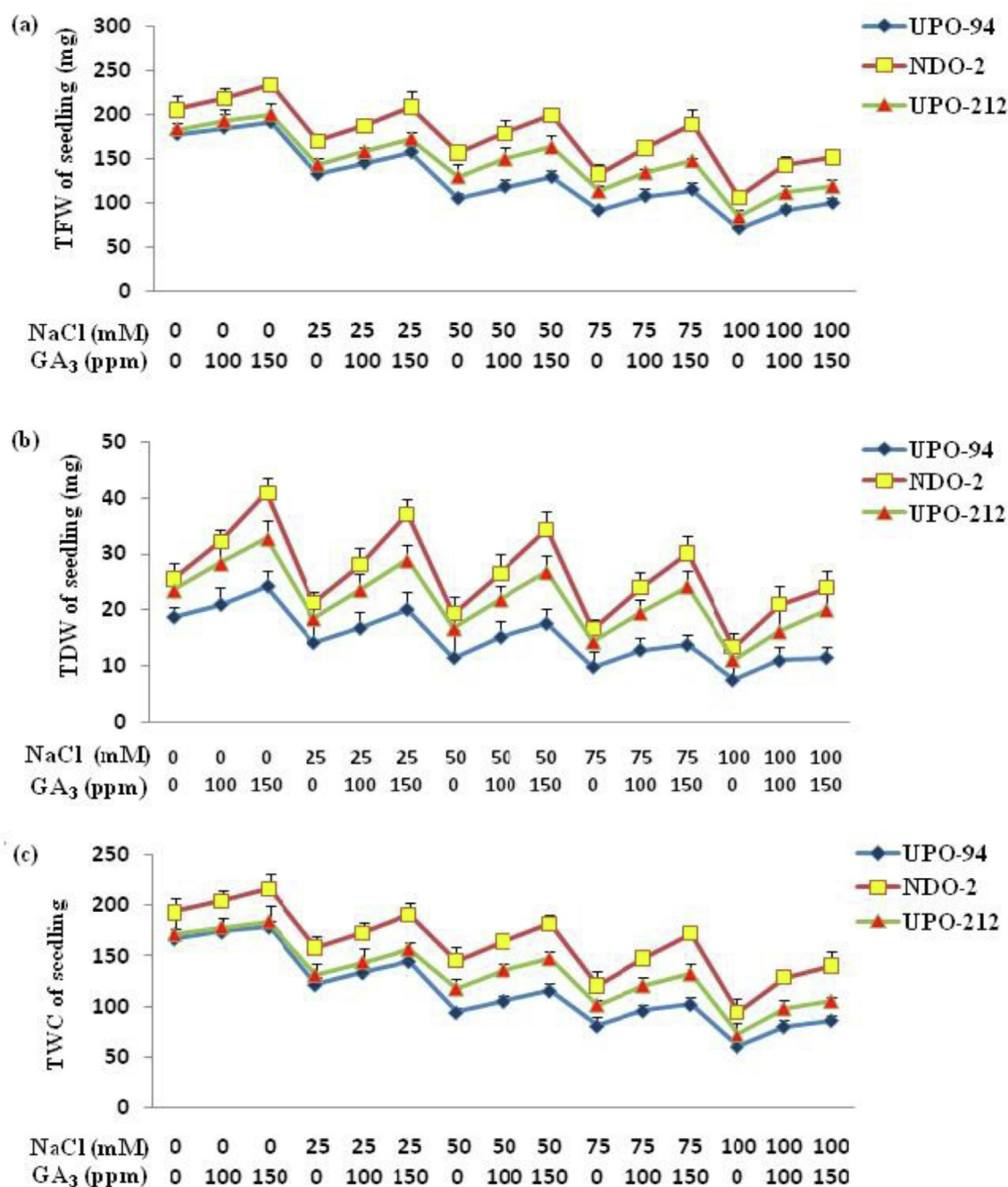


Fig. 2. Combined effects of NaCl and GA₃ concentrations on (a) TFW of seedling, (b) TDW of seedling and (c) TWC of seedling of three oat cultivars. Application of GA₃ (150 ppm) significantly promoted TFW, TDW and TWC of seedling as compared to 100 ppm GA₃ under different salt concentrations.

3.2. Length of shoot and root

Both concentrations of Gibberellic acid strongly promotes shoot and root growth in the cultivar NDO-2 and UPO-212 but 150 ppm concentration of GA₃ reduced root growth in sensitive cultivar UPO-94 at 75 and 100 mM (Fig. 1b and c). Chen et al. (2014) reported that GA₃ (100 μM) significantly diminished root length (and root dry mass) through it promoted above-ground growth in the dwarf lines, suggesting that GA₃ had negatory effects on seedling root length in the Rht12dwarf in bread wheat plants. Salt concentration significantly reduced the length of shoot and root in three oat cultivars. The higher reduction (51.01%) and (57.76%) was observed in cultivar UPO-94 and lower reduction (37.60%) and (42.82%) in cultivar NDO-2 was recorded respectively in the shoot and root length at 100 mM NaCl.

The negatory effect was more pronounced in root than shoot. Salt stress reduced the capability of plants to absorb water which leads to the reduction in growth (Rajput et al., 2015)). On the basis of Table 2, application of 150 ppm GA₃ improved the growth of three oat cultivars as compared to 100 ppm GA₃ and alleviated the negative effects of salinity since the above mentioned characters were significantly increased. 150 ppm GA₃ application significantly increased the length of shoot and root in tolerant cultivar NDO-2 at high salinity concentration (100 mM). Shaddad et al. (2013) reported a alike increase in the development of shoot and root of soybean and plants in respond to GA₃ treatment. Bejaoui (1985) has suggested that the effects of exogenously applied GA₃ in the abolition of salinity stress may be caused by activation of special enzymes which participate in RNA and protein synthesis. The higher GA₃ concentration reduced root length in sensitive cultivar UPO-94 (Fig. 1C).

3.3. Total fresh and dry weight of seedling

The effect of GA₃ treatments on seedling fresh and dry weight of oat under different salt concentrations in oat cultivars have been

shown in Fig. 2(a) and (b). The highest seedling fresh and dry weight was obtained in tolerant cultivar NDO-2 and lowest in sensitive cultivar UPO-94 at all salinity levels. At 100 mM salt concentrations, the fresh and dry weight was highly reduced in cultivar UPO-94. It ranged (48.03%) and (59.64%) in fresh weight and (46.455) and (56.47%) in dry weight respectively. The similar results reported by Dheeba et al. (2015) who showed that salinity reduced the fresh and dry weight of plants. Deficiency in dry and fresh biomass at higher concentration might be due to poor absorption of water from the growth medium due to physiological drought (Ramezani et al., 2011).

The maximum average of total fresh and dry weights was observed in 150 ppm GA₃ followed by 100 ppm in NDO-2 (Tables 3 and 2). According to Fig. 2(e), less difference was recorded for the total dry weight of seedling between both concentration 100 and 150 ppm GA₃ at higher salinity level 75 and 100 mM. Under GA₃ treatment (150 ppm), the cultivar UPO-94 show greater reduction (47.80% and 42.78%) while lesser (25.99% and 29.90%) was achieved in cultivar NDO-2 on seedling fresh and dry weight respectively, at 75 and 100 mM NaCl. This may be responsible to GA₃ enhanced the fresh and dry weights of seedlings, which also improved the rate of photosynthesis. Exogenous application of GA₃ was promoted, generally growth parameters of oat plants, thus alleviating to some extent the oppressive effect of salt stress (Abd El-Samad and Shaddad, 2013).

3.4. Tissue water content (TWC)

TWC in leaves was higher in cultivar NDO-2 than in UPO-212 and salinity reduced TWC in three oat cultivars (Fig. 2c). Tissue water content in cv. NDO-2 show lower reduction followed by UPO-212 and higher in UPO-94, which was reduced by 51.18%, 57.95% and 63.81% for 100 mM salt concentrations, respectively. Similarly, results by Khosravinejad et al. (2008) were also reported to the TWC decreased significantly with salt stress. Furthermore, salt stress reduced the relative water content in seedling of

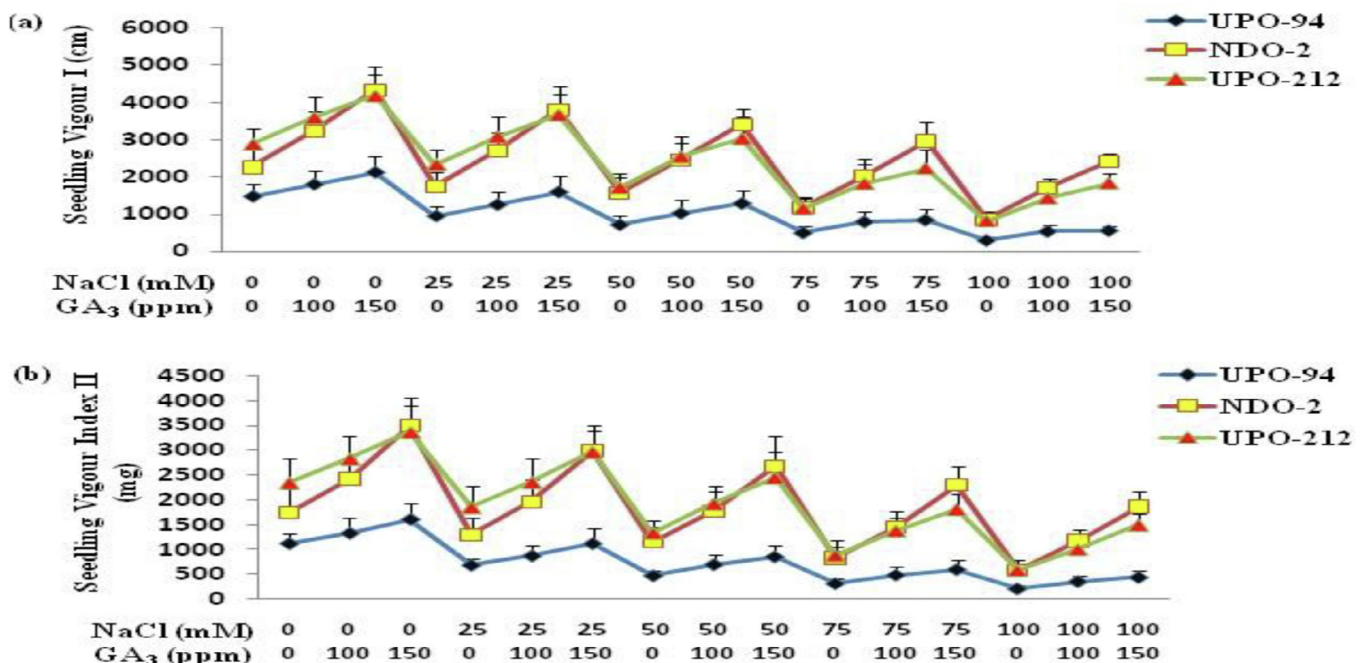


Fig. 3. Combined effects of NaCl and GA₃ concentrations on (a) seedling vigour index (I) and (b) seedling vigour index (II) of three oat cultivars. Application of GA₃ (150 ppm) significantly promoted seedling vigour index (I & II) as compared to 100 ppm GA₃ under different salt concentrations.

salt-sensitive cultivars. Application of GA₃ (150 ppm) highest increased TWC in NDO-2 and lowest in UPO-94 at high level of salt 100 mM.

The cultivar NDO-2 showed a minimum reduction (27.72%) and maximum (51.92%) was obtained by UPO-94 at 100 mM. Present results showed that priming with 100 ppm concentration of GA₃ had low effect on tissue water content; however, in 150 ppm concentration of GA₃ was more increase fresh and dry weight and TWC (Table 3). Ghodrat and Rosta (2012) also reported a similar increase in the TWC of corn plants in response to GA₃ treatment. Priming with suitable concentration of GA₃ plays an important role in the induction of tolerance to salinity stress and overcome limitations created by the environmental stress such as nutritional imbalance, osmotic effects and ion toxicity (Jamil and Rha, 2007).

3.5. Seedling vigor index (SVI)

Increasing salinity caused the significant reduction in the seedling vigor index have been shown in Fig. 3(a) and (b). The highest vigor index was recorded (2274.24 cm) and (1745.73 mg) for SVI (I & II) in NDO-2 and lowest vigor index recorded (1483 cm) for SVI (I) and (1125 mg) for SVI (II) in UPO-94 at control. Present results are in agreement with (Rajput et al., 2015) who showed that SVI is related to the special effect of ions and reduction of environmental water potentiality in presence of salinity. Vigor index-I & II were found to be highest in seeds receiving GA₃ 150 ppm in NDO-2 followed by UPO-212 and lowest was observed in UPO-94 under different saline conditions. Mosavian and Eshraghi-Nejad (2013) expressed that probably reduced percentage of germination and vigor index under low osmotic potential due to endosperm material decompose or slower transfer of this material to the seedling. Present results indicated, application of GA₃ enhanced seedling vigor index under different saline condition. Vigor index in the GA₃ treatment might have resulted in copious production of photosynthates resulting in high vigor index (Dhinesh babu et al., 2010).

4. Conclusion

In the present investigation, application of GA₃ mitigates the effects of salinity stress, and improved the seed germination and growth parameters of salt-stressed plants. The cultivar NDO-2 proved to have better seedling growth, vigor index and high tissue water content when seedling was treated with 150 ppm GA₃ under different salinity conditions. But this concentration of GA was not suitable for sensitive oat cultivars because 150 ppm GA₃ concentration reduced root growth at high salt levels. Because root growth is most important parameter for all plants in any surviving condition. This study helps to find out the concentrations of GA₃ which have the higher influence on the growth components which also economically helpful for farmers.

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