

## Effect of Foliar Application of Zinc and Manganese on Growth and Some Biochemical Constituents of *Brassica Juncea* Grown under Water Stress

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**Abstract:** This study was carried out to determine the response of *Brassica juncea* which was grown under different levels of drought to foliarly applied zinc and manganese at Botanical Garden of Abdul Wali Khan University Mardan, Khyber Pukhtunkhuwa, Pakistan. The experimental design was randomized complete block design with four replications. Plants were grown under four levels of drought (Control, 7 days drought stress, 14 days drought stress and 21 days drought stress). Plant were applied with zinc sulfate (3000 & 4000 mg/L) and manganese sulfate (3000 & 4000 mg/L) through leaves. Plant height, root length, number of leaves per plant, number of fruits, fresh and dry biomass of plant, some biochemical aspects (total chlorophyll and carotenoids), relative water content, leaf water loss, pollen viability and electrolyte leakage were measured. Results of statistical analysis showed that drought stress at vegetative stage led to significant reduction in above-mentioned parameters, except electrolyte leakage which showed an increase compared to control. Plants treated with different doses of  $ZnSO_4$ ,  $MnSO_4$  and  $ZnSO_4+MnSO_4$  showed an increase in plant height, biomass, root length, number of leaves and fruits per plant, chlorophyll and carotenoids of the plant. Micronutrients showed reduction in electrolyte leakage, leaf water loss and pollen viability while showed improvement in relative water content of plants. Finally, we recommended that foliar application and different concentration of micronutrients such as zinc sulfate and manganese sulfate exhibited promotory effect on plant growth and biochemical parameter of *Brassica juncea* plant grown under normal as well as drought condition.

**Key words:** Drought • Micronutrient • Pollen viability • Electrolyte leakage • Relative water content • Leaf water loss

### INTRODUCTION

Abiotic stress is the harmful effects of natural but non-living factors on the living organisms in a specific environment. Drought is one of the most common natural events that have a great negative impact on agriculture and water resources [1]. Drought increased climate change and water deficiency [2]. The presence of drought stress factors during production of fruits and vegetables crop is becoming more frequent with climate change patterns [3]. Human activity such as over farming and excessive irrigation is one of the causes of drought stress [4]. Plants stress cause disturbance of the association between membrane lipids and proteins, as well as enzymes activity and transport capacity of membranes [5]. In many plant species drought stress causes shrinkage in size of leaves, decrease in the number of stomata; thickening of

cell walls, cutinization of leaf surface and under development of conductive system, increase in the number of large vessels, submersion of stomata in succulent plants and in xerophytes, formation of tube leaves in cereals and induction of early senescence are caused by water stress [6].

Micronutrients are vital components for balanced growth and development in plants [7]. Small amount of Cu, Zn, B, Fe, Mo and Mn are essential for growth and quality of the crop as they control most of the physiological activities of the crop by interrupting the level of chlorophyll content in leaves, which ultimately influence the photosynthetic activity of the plant [8]. Zinc (Zn) is one of the most important micronutrients, which is mainly supplied as Zinc sulphate ( $ZnSO_4$ ). Zn plays an important role in cell division, cell expansion, protein synthesis and in carbohydrate, nucleic acid and

lipid metabolism [9]. It also plays an important role in auxin metabolism [10, 11]. Zinc deficiency cause biochemical changes in membranes and also cause reduction in both growth and yield [12, 13]. Zinc deficiency affects the metabolism of carbohydrates, synthesis of proteins, attributed to a sharp reduction in transcription, deformation and reduction of ribosomes [14].

Manganese (Mn) is an important micronutrient in most organisms. In plants, it plays a vital role in the structure of photosynthetic, proteins and enzymes. Its deficit is dangerous for chloroplasts as it affects the water splitting system of photosystem II (PSII), which provides the necessary electrons for photosynthesis [15]. However, its excess seems also to be damaging to the photosynthetic apparatus [16]. Thus, Mn has two roles in the plant metabolic processes: as an essential micronutrient and as a toxic element when it is in excess [17, 18]. Mn deficiency symptoms are interveinal chlorosis which appears in mid to late summer. Mn sulfate is the form most widely used. The recommended rates are 1-2.5 g/l. Because its mobility is higher, Mn corrections last longer than, say, boron or Zn [19].

*Brassica juncea* (L.) Czern, belongs to the Cruciferae (Brassicaceae) plant family, commonly known as the mustard family. [20] have shown by molecular analysis that *B. juncea* contains conserved genomes of the progenitor species. Mustard is available in the form of seeds, powders and oil. Recently, *B. juncea* has been explored for its biodiesel potential [21]. Indian mustard is reported to be an odyne, apertif, emetic, diuretic, rubefacient and stimulant and is used for arthritis, footache and rheumatism [22]. Believed to be aperient and tonic, the oil is used as a counter-irritant and stimulant. Mustard oil is used for skin eruptions and ulcers [23]. The oil is also antibacterial. In skin disease mustered oil is used as antiseptic and anti-inflammatory. The main component of mustard oil is allylisothiocyanate [24]. Although, oilseed Brassica are grown over 15% of arable land in India but their productivity is considerably hindered by various biotic and abiotic stresses [25, 26], like drought, chilling, pesticides and heavy metals. Aim of this study will be the investigation of the effect of different treatments of drought stress on growth and some biochemical aspects of *Brassica juncea* plant. Investigation will also be performed to study the effects of foliar application and different concentration of micronutrients ( $ZnSO_4$  and  $MnSO_4$ ) on growth and biochemical aspects of *Brassica juncea* plant grown under drought stress.

## MATERIALS AND METHODS

The present study was conducted to investigate the effect of zinc sulfate and manganese sulfate on growth, chlorophyll, carotenoids, pollen viability, electrolyte leakage, relative water content and leaf water loss of *Brassica juncea* grown under drought stress.

**Source of Seeds:** Seeds of *Brassica juncea* were obtained from the local market of Mardan, K.P.K.

**Growth Condition and Treatments:** This experiment was comprised of 84 pots which were divided into seven sets. Details of 7 sets were as follows:

1<sup>st</sup> Set: Without zinc and manganese comprising of control and three drought treatments (7, 14 and 21 days).

2<sup>nd</sup> Set: Zinc provided as zinc sulfate @ 3000 mg/L, the set comprises of control and three drought treatments (7, 14 and 21 days).

3<sup>rd</sup> Set: Zinc provided a zinc sulfate @ 4000 mg/L, the set comprises of control and three drought treatments (7, 14 and 21 days).

4<sup>th</sup> Set: Manganese provided as manganese sulfate 3000 mg/L, the set comprises of control and three drought treatments (7, 14 and 21 days).

5<sup>th</sup> Set: Manganese provided as manganese sulfate @ 4000 mg/L, the set comprises of control and three drought treatments (7, 14 and 21 days).

6<sup>th</sup> Set: Zinc and manganese provided as zinc sulfate and manganese sulfate in combination @ 3000 mg/L, the set comprises of control and three drought treatments (7, 14 and 21 days).

7<sup>th</sup> Set: Zinc and manganese provided as zinc sulfate and manganese sulfate in combination @ 4000 mg/L, the set comprises of control and three drought treatments (7, 14 and 21 days).

These plastic pots of 17.5 cm in diameter and 6.5 cm deep having basal outlet for drainage. Out of 84 pots 12 pots present in each set and 3 replicates were maintained for each treatment i) control (non-saline), ii) 7 days drought, iii) 14 days drought and iv) 21 days drought. Every pot was filled with 1 Kg of thoroughly washed sandy loam soil. Soil in each pot was saturated with full strength Hoagland's solution. Approximately uniform size and equal number of seeds were surface sterilized with 0.1% mercuric chloride for one minute and then washed with distilled water. Five (5) seeds were sown in each pot. They were then daily irrigated with an equal amount i.e.,

50 ml of tap water. When seedlings were reached at 3 leaves stage they were thinned out as one seedling per pot. All these 84 pots were then arranged in a completely randomized design (CRD) in the Botanical Garden of Department of Botany, Abdul Wali Khan University, Mardan. Drought treatment was give and then each pot was irrigated with 1.5L of tap water twice a week. When drought treatment was completed in pots different concentrations of zinc sulfate and manganese sulfate were applied foliarly different sets.

**Growth Analysis:** Plant height, number of leaves and branches, root length, fresh and dry biomass, number of pods per plant were recorded in harvested plants at termination of experiment.

#### Biochemical Analysis

**Chlorophyll Content:** Chlorophyll concentration (Chl) was determined in fresh leaves following the protocol of Maclachlan and Zalik, [27].

**Tetrazolium Chloride Test for Pollen Viability:** Pollen grains were collected from plants raised under different intervals of drought in irrigation water and their viability was observed as outlined by Dafni, [28].

**Electrolyte Leakage (EL):** EL was measured in fresh leaves as described by Lutts *et al.* [29] with a few modifications.

**Relative Water Content (RWC):** RWC was determined using the method described by Mata and Lamattina [30].

**Leaf Water Loss (LWL):** LWL was measured as described by Clark and Caige, [31].

**Experimental Design and Statistical Analysis:** The experimental design was completely randomized Design (CRD) with three salt levels and three replicates. Collected data was analyzed statistically by using SPSS to analysis of variance (ANOVA) and the means compared by Duncan's multiple range test ( $P < 0.05$ ).

## RESULTS AND DISCUSSION

**Plant Height:** In our experiment plants treated with different levels of drought showed significant ( $P < 0.001$ ) decrease in plant height Fig. 1. Our observations are in correspondence with the findings of [32]. Nour, [33] and Parvez *et al.* [34] also reported that drought stress

showed significant reduction in plant height. Similar reduction of plant height in corn seedlings due to drought was reported by Del Rosario *et al.* [35]. Our results also showed that  $ZnSO_4$  treated plants showed significant ( $P < 0.001$ ) increase in plant height compared to control plants. Our findings are in correspondence with observation of Zohreh *et al.* [36] on sunflowers. Plants treated with different doses of  $ZnSO_4$  and  $MnSO_4$  showed significant ( $P < 0.001$ ) increase in plant height in our observation that show resemblance with the result of Majid *et al.* [37] on *Cuminum cyminum* plant. Mostafavi *et al.* [38] also reported that simultaneous consumption of Zn and Mn led to an increase of wheat plant height.

**Root Length:** In our observation drought stress showed significant ( $P < 0.001$ ) decrease in root length as compared control plants (Fig. 2). Similar results were reported by Pace *et al.* [32], who pointed that drought stress inhibited root growth. Our results also showed that plants treated with different doses of  $ZnSO_4$  (3000mg/l, 4000mg/l) showed a significant ( $P < 0.05$ ) increase in this trait. Elif and Nuray, [39] reported that the root growth of seedlings exposed to only Zn and Zn with Cd is significantly promoted. Mark and Zdenko, [40] also stated that root length in all diameter classes increased significantly ( $P = 0.05$ ) with increasing Mn rate from 0 to 500 Mn in the culture solution.

**Number of Leaves:** In our experiments drought stress showed significant ( $P < 0.001$ ) decrease in number of leaves per plants compared to control plants (Fig. 3). Our results are in correspondence with the results of Parvez *et al.* [34]. Akkas *et al.* [41] also reported less number of leaves in drought treated plants compared to control plants on *Zea mays* (L) cultivars. Similar decrease in number of leaves in corn seedlings due to drought was reported by Eck, [42]. Plants treated with different doses of  $ZnSO_4$  and  $MnSO_4$  (3000mg/l, 4000mg/l) showed a significant ( $P < 0.001$ ) increase in this parameter in control, as well as in treated plants. Our observations are in the correspondence with the findings of Hasani *et al.* [43], who reported in his experiments that plants treated with micronutrients showed an increase in number of shoots and leaves.

**Number of Fruits:** Drought stress showed significant ( $P < 0.001$ ) decrease in number of fruits compared to control (Fig. 4). Our observations are in correspondence with the findings of Parvez *et al.* [34], who treated tomato plants

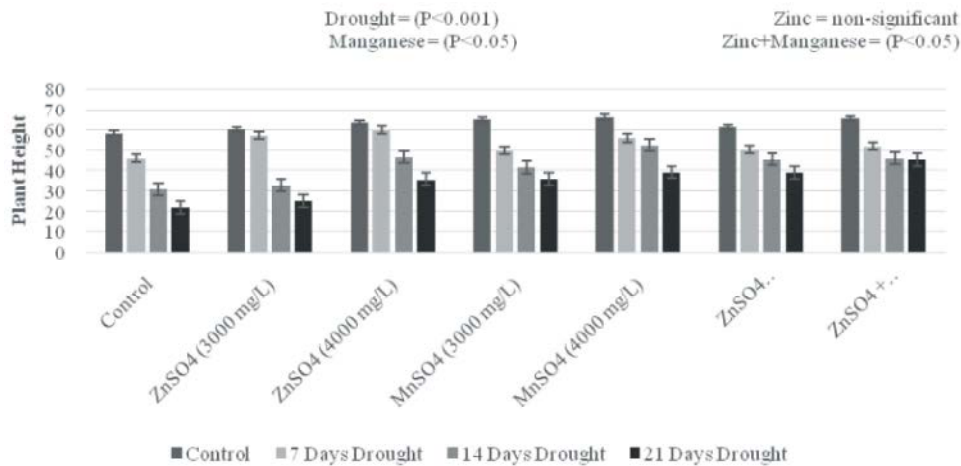


Fig. 1: Influence of micronutrient (Zinc and Manganese) on plant height (cm) of *Brassica juncea* grown under drought stress

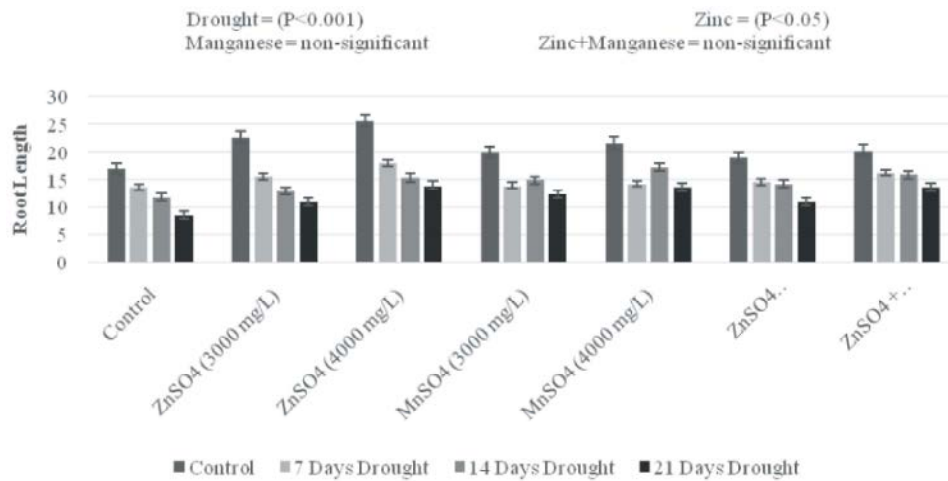


Fig. 2: Influence of micronutrient (Zinc and Manganese) on root length (cm) of *Brassica juncea* grown under drought stress

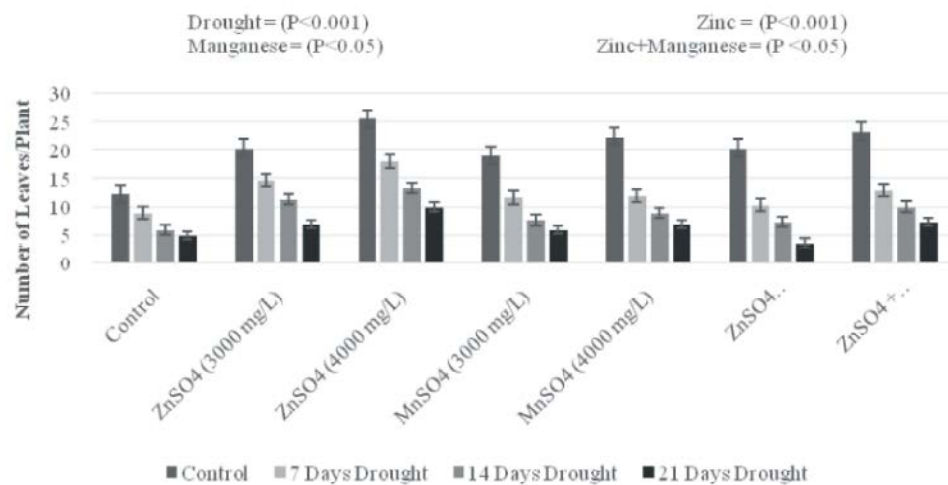


Fig. 3: Influence of micronutrient (Zinc and Manganese) on number of leaves/plant of *Brassica juncea* grown under drought stress

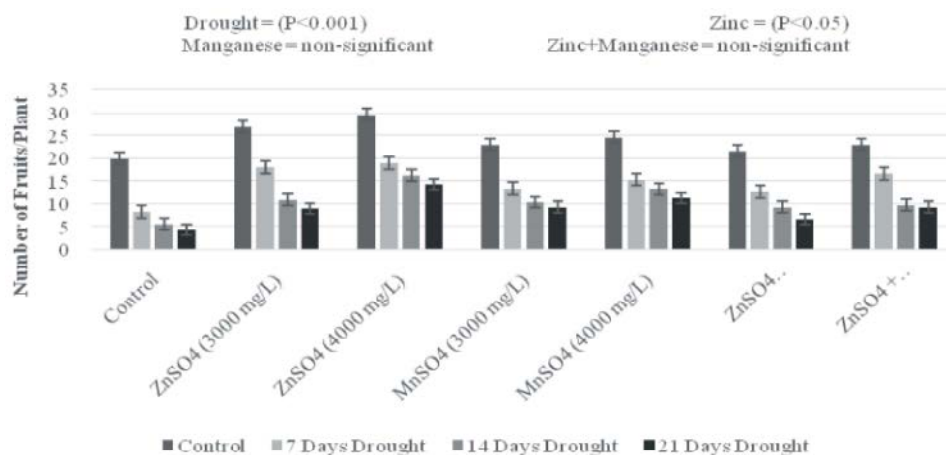


Fig. 4: Influence of micronutrient (Zinc and Manganese) on number of fruits/plant of *Brassica juncea* grown under drought stress

with drought stress. The effect of drought stress on fruit number in *Brassica* plant is in agreement with the findings of Goksoy *et al.* [44]. Increasing drought stress and decreasing in Zn sulfate level resulted in a decrease in fruit number per plants. Hatwar *et al.* [45] and Dongre *et al.* [46] also reported that ZnSO<sub>4</sub> (3000mg/l, 4000mg/l) has significantly ( $P<0.05$ ) positive influence on number of fruits compared to control plants. Zohreh *et al.* [36] on sunflower also reported that Zn sulfate treated plants give more fruits compared to control and drought treated plants. Results of Hasani, [43] showed resemblance with our findings that Mn sprays at both levels significantly increased fruit yield. Similar results had been reported that foliar spray of Mn increased fruit yield in ‘Ganesh’ pomegranate by Bambal *et al.* [47] and ‘Valencia’ orange by Labanauskas *et al.* [48]. Mn sprays increased number of fruit/tree and fruit average weight, although the increase was not statistically significant, therefore the increase in fruit yield caused by Mn could be due to the increase in number of fruit/tree as well as fruit average weight.

**Biomass:** In our experiment different levels of drought stress showed significant ( $P<0.001$ ) decrease in dry weight of plants (Fig. 5 and 6). Control plants had comparatively higher weight of the plants and drought treated plants has less weight. Our observations are in correspondence with the findings of Pace *et al.* [32]. Similar results were reported by Majid *et al.* [37], on *Cuminum cyminum*, Parvez *et al.* [34] on tomato and Ibrahim, [49] on chick pea. Our results also showed that plants treated with ZnSO<sub>4</sub> and MnSO<sub>4</sub> (3000mg/l, 4000mg/l) showed non-significant increase in plant biomass. Our findings are in agreement with those

obtained by Kenneth, [50], who reported that Zn and Mn treatments showed an increase in biomass or productivity of plants.

**Chlorophyll:** In our observations plants treated with drought stress showed significant increase in chlorophyll content as compare to control plants (Fig. 7, 8 and 9). Similar results were reported by Mehdi *et al.* [51] that the most amount of chlorophyll fluorescence was recorded in drought treated plants. Ebrahim, [52] also reported that total chlorophyll content remained increase in drought treated plants as compare to untreated plants. There was significant positive correlation between DSI and total chlorophyll, chlorophyll *a* and chlorophyll *b*. Drought resistant genotypes had high value for these parameters in both conditions or at least had a least reduction in stress condition as reported by Reza *et al.* [53] on chickpea (*Cicer arietinum*) plant. Chlorophyll content and the chlorophyll stability index decreased up to day 40 of drought and increased later in *Ctenanthe setosa* plant [54]. Higher chlorophyll content has also been associated with the stress tolerance of plants [55]; Sairam, [56]; Kraus *et al.* [57].

It was also reported in our experiment that ZnSO<sub>4</sub> and MnSO<sub>4</sub> (3000mg/l, 4000mg/l) treated plants showed non-significant increase on total chlorophyll level. Results of Mehdi *et al.* [51] also showed resemblance to our observation that the maximum amount of leaf chlorophyll content was obtained in foliar application Zn and Mn as sole treatment or foliar application of Zn +Mn, respectively compared to control. These results showed that manganese and zinc have a main effect on chlorophyll biosynthesis in plant. Marschner, [12] stated that manganese is important in chlorophyll biosynthesis.

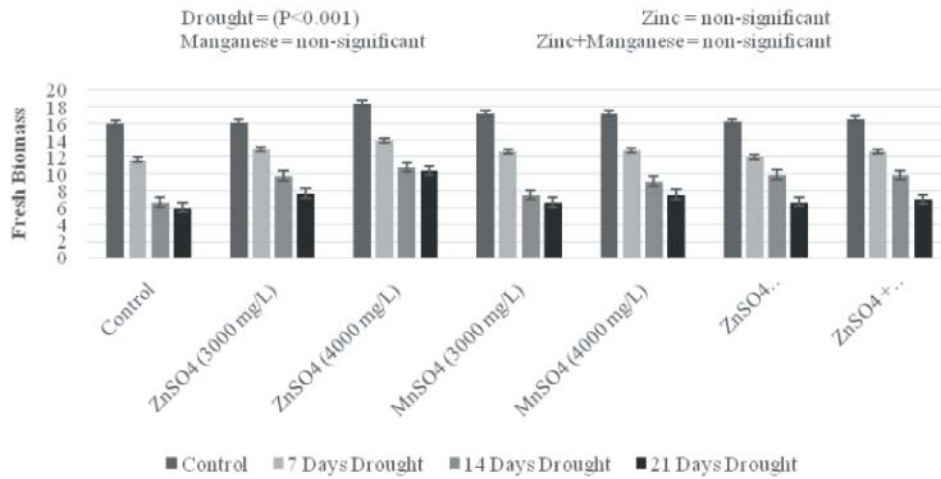


Fig. 5: Influence of micronutrient (Zinc and Manganese) on fresh biomass (g) of *Brassica juncea* grown under drought stress

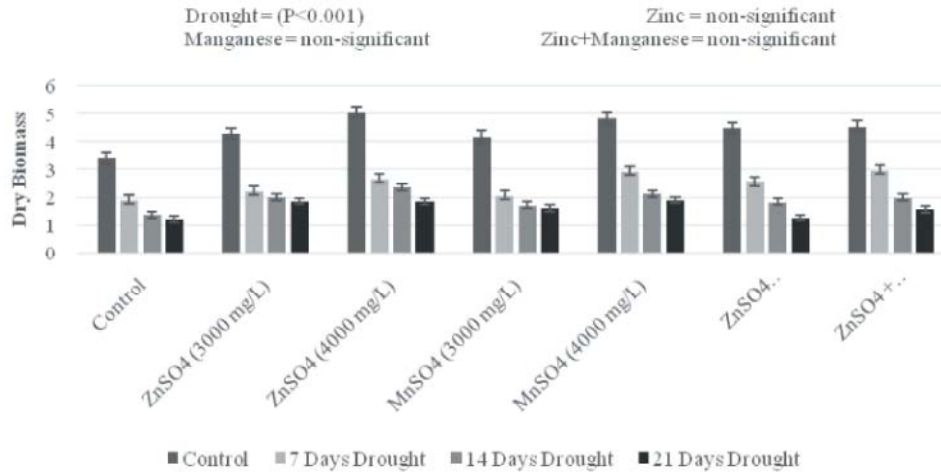


Fig. 6: Influence of micronutrient (Zinc and Manganese) on dry biomass (g) of *Brassica juncea* grown under drought stress

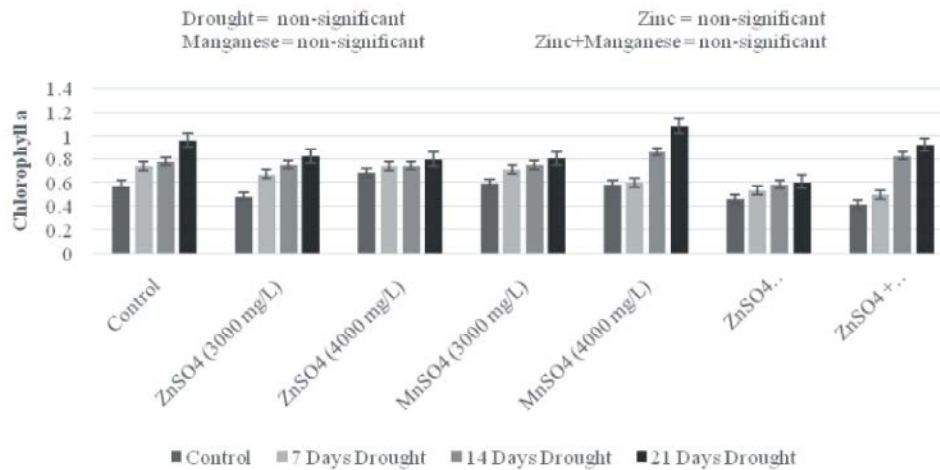


Fig. 7: Influence of micronutrient (Zinc and Manganese) on chlorophyll a (mg/g FW) of *Brassica juncea* grown under drought stress

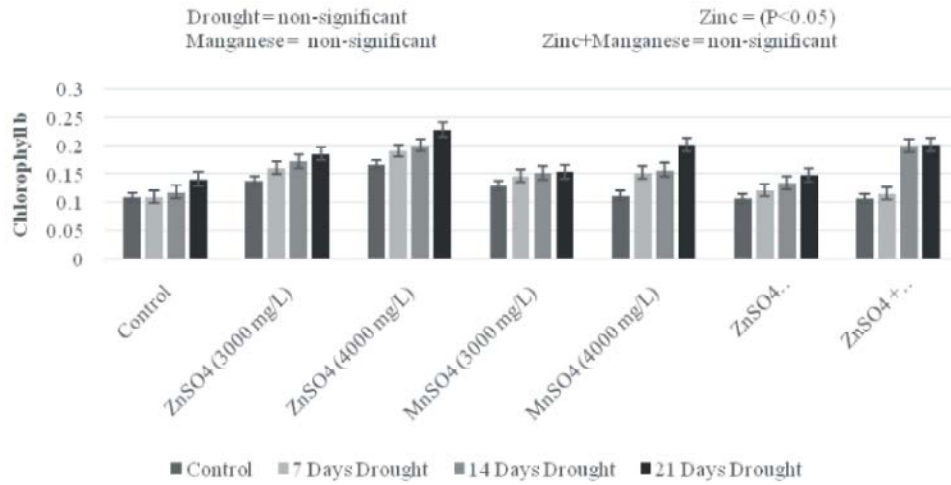


Fig. 8: Influence of micronutrient (Zinc and Manganese) on chlorophyll b (mg/g FW) of *Brassica juncea* grown under drought stress

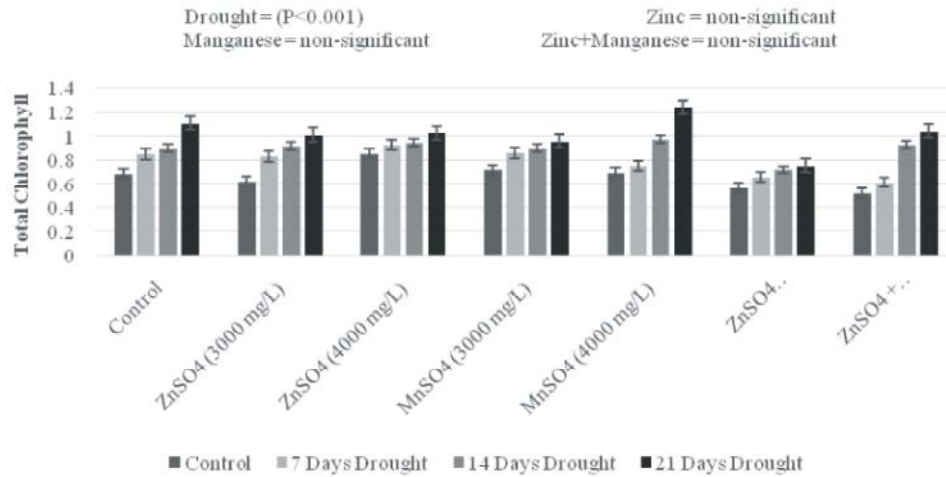


Fig. 9: Influence of micronutrient (Zinc and Manganese) on total chlorophyll (mg/g FW) of *Brassica juncea* grown under drought stress

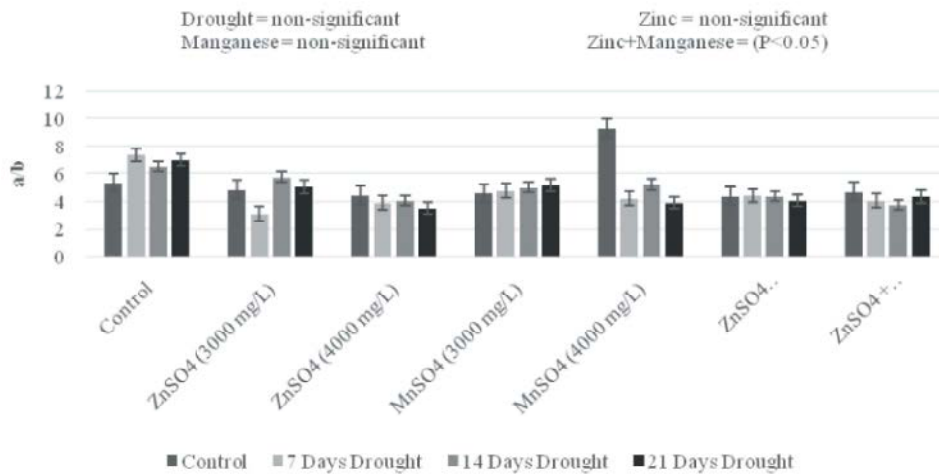


Fig. 10: Influence of micronutrient (Zinc and Manganese) on chlorophyll a/b ratio of *Brassica juncea* grown under drought stress



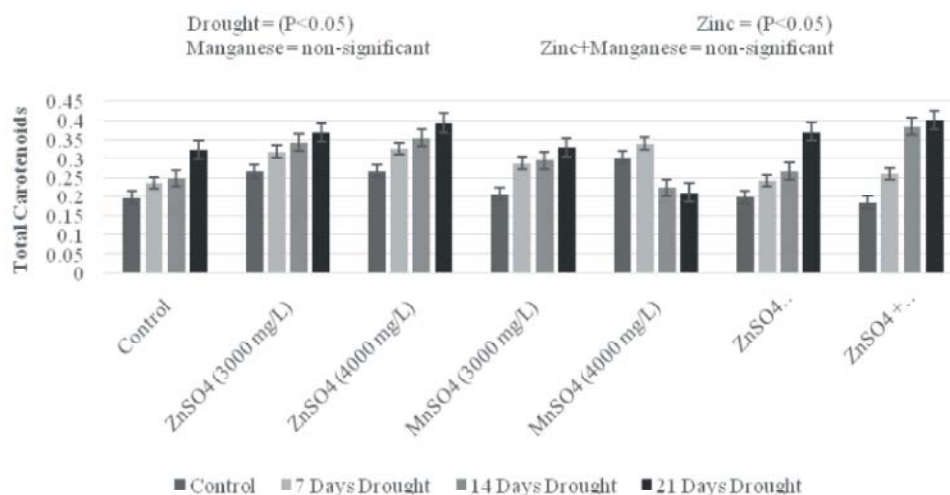


Fig. 11: Influence of micronutrient (Zinc and Manganese) on total carotenoids (mg/g FW) of *Brassica juncea* grown under drought stress.

In our observation drought stress showed non-significant increase and sometime decrease in a/b ratio of plants (Fig. 10). Plants treated with different doses of  $\text{MnSO}_4 + \text{ZnSO}_4$  showed significant ( $P < 0.05$ ) decrease and  $\text{ZnSO}_4$  showed non-significant decrease in a/b ratio of the plants. This result is in agreement with those obtained by Shakya *et al.* [58], who reported that the effects of the heavy metals copper (Cu), zinc (Zn) and lead (Pb) on the chlorophyll content of two mosses *Thuidium delicatulum* (L.) Mitt. and *T. sparsifolium* (Mitt.) Jaeg., as well as leafy liverwort *Ptychanthus striatus* (Lehm. and Linderb). These plants were treated with different concentrations of Cu, Zn and Pb, in isolation and in combination under experimental conditions. The ratio of chlorophyll a/b was decreased more rapidly in both *Thuidium* species, with higher concentrations occurring when Cu+ Zn+ Pb ions were together than when Cu, Zn, or Pb ions were alone.

**Carotenoids:** In our experiment drought treated plants showed significant ( $P < 0.001$ ) increase in carotenoids (Fig. 11). Higher level of carotenoids concentration in drought-tolerant genotypes has also been reported by Deng *et al.* [59]; Kalefetoglu and Ekmekci, [60]. Drought-tolerant genotypes accumulated more carotenoids than susceptible genotypes. Accumulation of carotenoids for osmotic regulation in drought-stressed leaves in many crops has been reported by Khan *et al.* [61]; Gunes *et al.* [62]. Carotenoid content decreased up to day 40 of drought and increased later, [54]. Higher carotenoids content have also been associated with the stress tolerance of plants [55, 56, 57]. In our observation foliar spray of  $\text{ZnSO}_4$  and  $\text{MnSO}_4$  plants showed

non-significant increase in carotenoids, while plants treated with different doses of  $\text{ZnSO}_4$  (4000mg/l, 3000mg/l) showed significant ( $P < 0.05$ ) increase in this parameter as compared to control set. Our observations are in correspondence with the findings of Moustafa *et al.* [63]. Who reported that foliar application of Zn and Mn individually or in mixture form showed significant increase in carotenoids level.

**Pollen Viability:** Pollen viability exhibited significant ( $P < 0.05$ ) reduction in different levels of drought compared to control plants. A reduction in pollen viability is a common symptom in angiosperms under the stress environments [64] who also reported that drought stress induced a decrease in pollen viability and germination *in vitro*. The decrease in germination in both media suggests that water deficits imposed during floral development had a detrimental effect on the subsequent capacity of pollen to germinate and, as the pollen tube data shows, a detrimental effect on pollen tube growth *in vivo*. Flower abortion in chickpea induced by a water deficit may be attributed not only to an impairment of pollen viability, but also to an impairment of stigma/style function. Davies *et al.* [65]; Siddique *et al.* [66]; Leport *et al.* [67, 68], who reported that Chickpea cultivars that water deficits impaired both pollen and stigma/style function and the impairment of pistil function was an important factor relating to flower abortion, while, thirdly, it showed that initiation date significantly affected flower and pod development with early-initiated flowers and pods less likely to abort, while late-initiated flowers and pods largely aborted. Water stress during reproductive development



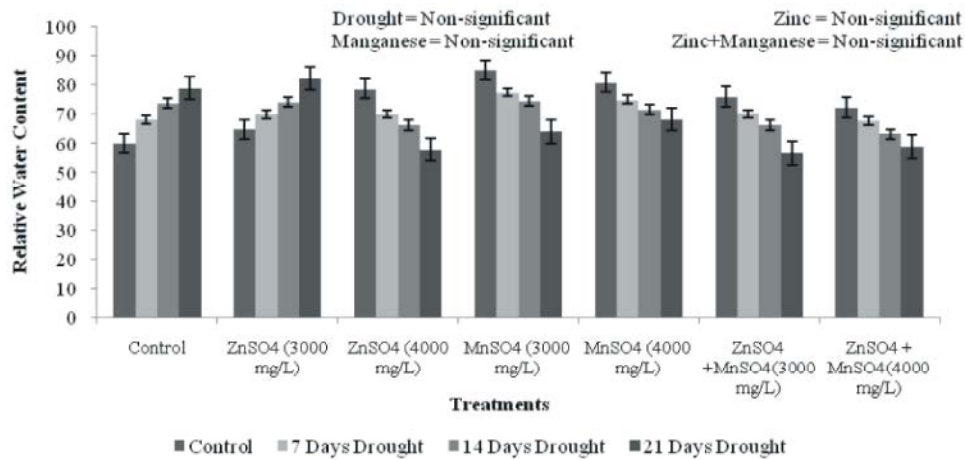


Fig. 1: Interactive effects of drought and micronutrients (Zinc and Manganese) on relative water content (%) of *Brassica juncea*.

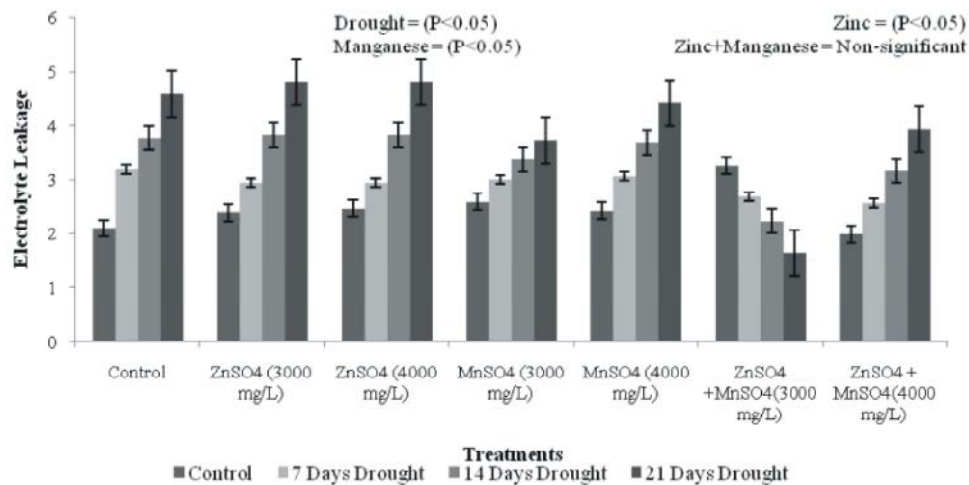


Fig. 2: Interactive effects of drought and micronutrients (Zinc and Manganese) on electrolyte leakage (%) of *Brassica juncea*.

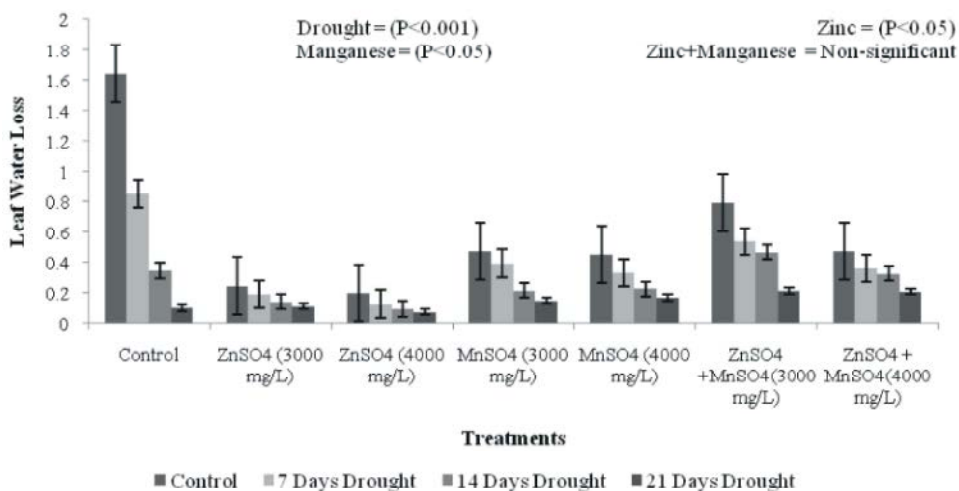


Fig. 3: Interactive effects of drought and micronutrients (Zinc and Manganese) on Leaf water loss (%) of *Brassica juncea*.

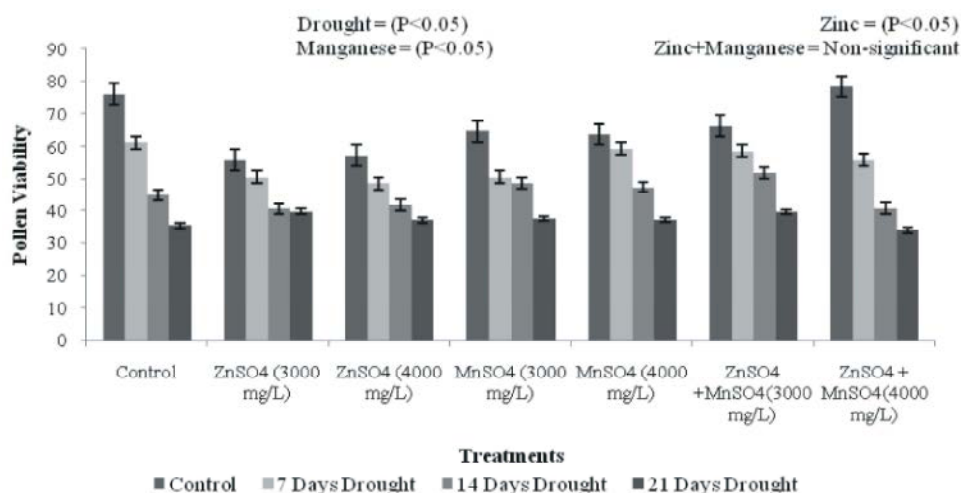


Fig. 7: Interactive effects of drought and micronutrients (Zinc and Manganese) on pollen viability (%) of *Brassica juncea*.

in common bean (*Phaseolus vulgaris*) resulted in reduced pollen viability, reduced pollen germination and an abnormal exine with deeply pitted and smooth regions [69]. A considerable reduction in pollen viability might affect seed set if fewer viable pollen grains are deposited on the receptive stigma and if infertile pollen grains interfere with pollen tube growth. It is also possible that pollen from stressed plants might have a shorter lifespan and have reduced vigour; pollen tube growth may begin, but fail to reach the ovule so that no fertilization takes place [70]. However, this study has also shown that the water deficit had a greater effect on pollen growth in the pistil than on pollen viability *in vitro* and that the effect of a water deficit is greater on the stigma/style function than on pollen viability and germination [71, 72]. ZnSO<sub>4</sub> and MnSO<sub>4</sub> exhibited significant ( $P<0.05$ ) increase compared to control.

**Relative Water Content:** Relative water content exhibited reduction in different levels of drought compared to control. Takkar *et al.* [73] studied that decreasing of relative water content is an indication of decrease of swelling pressure in plant cells and causes to decrease the growth. Depletion of soil water caused lower water potential in the root region, so the plant should be decreased transpiration rate for remaining consistent water potential of inside. ZnSO<sub>4</sub> and MnSO<sub>4</sub> showed non-significant decrease and usually but not always in drought condition as compared to control. The results showed that leaf relative water content (RWC) strongly was affected by water deficit stress and with intensifying stress decreased significantly. Our results were

concordant with those of obtained by Jiang and Huang [74]. Difference among leaf relative water contents was significant at Zn levels so that the plants which received a higher amount of Zn, had more leaf relative moisture. Maintenance of high RWC in drought resistant cultivars has also been reported to be an adaptation to water stress in several crop species [75], who also reported the recovery of water stress was markedly and consistently improved by micronutrient application, particularly Zn and Mn.

**Electrolyte Leakage:** Electrolyte leakage exhibited significant ( $P<0.05$ ) promotion in different levels of drought compared to control. According to Prasil and Zamecnik [76], electrolyte leakage showed significant ( $P<0.05$ ) increase. Who analyze that electrolyte leakage from plant tissue in deionized water is a function of time. A rapid leakage would occur from the intercellular free space regions followed by slower releases across the plasma membrane and then tonoplast. Such a pattern would be expected to follow a triple exponential function [77]. This implies that electrolyte leakage subsequently quantified in deionized water should be considered as a consequence of hyperosmotic to hypo-osmotic transition rather than a consequence of hyperosmotic stress. Plants membranes are subject to changes often associated with the increases in permeability and loss of integrity under environmental stresses [78]. Therefore, the ability of cell membranes to control the rate of ion movement in and out of cells is used as a test of damage to a great range of tissues. Valentovic *et al.* [79] reported that the electrolyte leakage of the sensitive maize cultivar

increased but the increase in ion leakage of tolerant cultivar was not so high. Quan *et al.* [80] also found higher electrolyte leakage in drought stressed maize (*Zea mays* L.) plants than in plants grown under well watered conditions. ZnSO<sub>4</sub> and MnSO<sub>4</sub> showed significant (P<0.05) increase in drought condition as compared to control. The main site of attack by drought in a plant cell is usually the cell membrane [81]. Zn treatment reduced ion leakage, Fridovich [82] reported that elevated levels of H<sub>2</sub>O<sub>2</sub> and O<sub>2</sub><sup>-</sup> caused by stress facilitate the formation of highly active hydroxyl radicals (OH<sup>•</sup>).

**Leaf Water Loss:** Leaf water loss parameter exhibited significant (P<0.001) reduction in different levels of drought compared to control. The lowest leaf water loss values associated with a high drought tolerance was recorded in cultivars: Riniker, Compact, Salemer, Plaisant, Dana. In case of cultivars Andrei, Orizont, Ogra, Gerbel, Precoce, high values of this parameter attests a low drought tolerance [83]. The rate of excised-leaf water of chickpea cultivars grown under optimal, pre- and post-anthesis drought stress conditions. The cultivars in this treatment were grown in optimal conditions, so the leaf water loss either were unaffected, decreased, or increased depending on cultivars. The data showed significant (P<0.05) decrease in different growth stages, it can be concluded that as the amount of irrigation water decreases and the plant adjust themselves gradually to the stressful condition, they would suffer less damage. In the blistering stage, plants completely try to overcome the environmental stresses in order to fill their seeds [85]. Generally, compared to severe stress, moderate stress has a more intensified effect on maize. The difference between normal and moderate stressful condition is much more than that of moderate and severe stressful conditions that maize is vulnerable to drought stress [85] LWL is a useful index to screen the varieties capable of growing under drought stressful conditions [86]. Drought vulnerable varieties have a higher LWL and compared to these conditions, their LWL is higher under normal conditions [31]. Drought stress decreased the LWL. The level of this decrease was not significant, but under moderate and severe drought stressful conditions, vulnerable varieties showed a higher decrease than tolerant ones [87]. ZnSO<sub>4</sub> and MnSO<sub>4</sub> exhibited significant (P<0.05) decrease as compared to control.

Different levels of drought stress showed inhibitory effect on different growth parameter i.e. (plant height, root length, number of leaves, number of fruits per plant, fresh

and dry biomass) and some biochemical parameters i.e. (chlorophyll a, b and total chlorophyll, a/b ratio, carotenoids, reducing and non-reducing sugar, total carbohydrates, total proteins). Foliar application and different concentration of micronutrients such as zinc sulfate and manganese sulfate exhibited promotory effect on plant growth and biochemical parameter of *Brassica juncea* plant grown under normal as well as drought condition.

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