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### Cover Page Footnote

The corresponding author is thankful to department of Botany, GCU Faisalabad for providing Financial support to complete this research work

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## **CAPPARIS DECIDUA EDGEW (Forssk.) STEM EXTRACT ALLEVIATES THE WATER STRESS PERTURBATIONS IN WHEAT (*TRITICUM AESTIVUM* L.) AT EARLY GROWTH STAGE**

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### **ABSTRACT**

Water shortage is one of the major limiting factors of crop productivity worldwide. This study was planned to explore the impact of *Capparis decidua* stem extract on wheat varieties (FSD-2008, 12066 and 13348) under water stress condition. Water deficit condition negatively impacted the seedling germination, growth, photosynthetic pigments and ions uptake capacity. Water limited condition significantly ( $p \leq 0.05$ ) increased time to attain 50% germination, lipid peroxidation,  $H_2O_2$  and enhanced the antioxidant mechanism to overcome the oxidative stress. Different concentrations of *C. decidua* extract i.e. Not Soaking (NS), 10%, 20%, 30% and 40% were used for seed priming. It improved the negative effects of water stress and amended plant germination and growth by enhancing photosynthesis and reactive oxygen species (ROS) scavenging mechanism. The 20% and 30% *C. decidua* stem extract remained effective in mitigating the induced water stress effects in the tested cultivars. It was found that *C. decidua* stem extract significantly ( $p \leq 0.05$ ) improved germination, growth  $Mg^{+2}$  and  $Fe^{+2}$  uptake in the wheat varieties under study. It is concluded that *C. decidua* phytoextract is an influential agent to ameliorate water stress effects in wheat at early growth stage.

**Keywords:** Oxidative stress, wheat, antioxidant mechanism, phytoextract.

### **INTRODUCTION**

Wheat (*Triticum aestivum* L.) is one of the leading food crops and it has been predicted that 30% increase in its production is indispensable to feed the ever increasing population of the world by 2050 (Daryanto et al., 2016). Water deficiency is a serious abiotic stress with adverse impacts on crop plants (Wu et al., 2014). Wheat is worldwide cultivated food crop and most sensitive to drought condition and can result up to 70% yield losses under water stress as compared to normally irrigated condition. Water limiting environment is more challenging for cultivating areas of South Asia including Pakistan where arid and semi-

arid areas are under wheat cultivation. The water deficit conditions affect wheat yield more adversely in rain-fed area primarily due to reduced germination. The rain-fed area is about 25% of the cultivated land in Pakistan and it is anticipated that yield of wheat, rice, corn, millet and mustard will be reduced in future due to limited water supply. These anticipated yield losses especially in wheat will be more dangerous for agriculture based economies like Pakistan where it is the major source of food security for people of rural areas (Mahmood et al., 2019).

Water limited condition results in retardation of photosynthesis which is directly related to reduction in chlorophyll contents, membranes stability and

impairment of many physiological processes that ultimately reduce plant productivity (Ma et al., 2017).

Various strategies are being adopted to mitigate drought stress, and seed priming is one of them. It is an important, cheap and reasonably effective measure to introduce drought tolerance in plants (Ashraf and Foolad, 2005). The priming of seeds is a pre sowing procedure which involves moderate hydration of seeds to turn on the pregerminative metabolism (Cheng et al., 2017; Savaedi et al., 2019). Seed priming enhances germination rate and seedling establishment when environmental conditions are not favorable. In agricultural practices, germination efficiency and seedling vigor are of prime importance to increase plant productivity under unfavorable conditions (Paparella et al., 2015). The metabolic and physiological improvements in plants during germination and early seedling stage are linked with seed priming which activates protective enzymes and accumulates osmoprotectants (Farhad et al., 2011). In water based seed priming seeds are hydrated but emergence does not occur. Seed priming may include treatment of simple water, solution of different chemicals and phytoextract of different origins (Chen et al., 2011).

In the current study we used *C. decidua* stem extract of varying concentrations for making seeds prime. The *C. decidua* is a medicinal plant and belongs to family *Capparaceae*. It is found in dry areas specially deserts, common in Cholistan, Pakistan (Hameed et al., 2011). It contains significant amount of active antioxidants, phenolics and is used for the treatment of various kinds of infectious diseases (Yadav et al., 1997). The current study was based upon the hypothesis that *C. decidua* stem extract may evoke the antioxidant defense system in tested wheat genotypes under water limited environment.

## MATERIALS AND METHODS

### *Experimental Design and Ecological Conditions*

Mature and healthy seeds of three wheat varieties 12066, 13348 and FSD-2008 were collected from Ayub Agricultural Research Institute (AARI) Faisalabad, Pakistan. Experiment was conducted in Plant Bioenergy and Biotechnology Lab, Department of Botany, Government College University Faisalabad, Pakistan. The collected seeds were washed with distilled water following surface sterilization with 01% sodium hypochloride solution for 05 min. Before placing in petri plates seeds were blotted dry and primed for 12 h in various concentrations (10, 20, 30 and 40%) of *C. decidua* stem extract prepared in distilled water. The *C. decidua* stems were locally collected from surroundings of Government College University Faisalabad, Pakistan. Stem extract was obtained by grinding and squeezing the stem pieces. Air dried wheat seeds were placed in petri plates containing double layered filter paper. Ten seeds were placed in petri plate (100 x 20 mm) with three replicates and experiment was laid in completely randomized design (CRD). Petri plates were kept in growth chamber (100 W, Guangdong PHILIPS Co., Guangdong, China). Primed seeds containing petri plates were divided in two groups (I) control and (II) drought which was induced using polyethylene glycol (PEG-6000). About 10 ml of PEG-6000 solution and Hoagland's solutions was given to all petri plates under same conditions of light and humidity. The water level was maintained on daily basis. After measuring germination attributes thinning was performed on 7<sup>th</sup> day of sowing and the seedlings were harvested after 20 days from the 1<sup>st</sup> day of experiment. All other morphological and physio-biochemical analysis were

completed from harvested wheat seedling later.

### **Measurement of Germination Attributes**

Germination attributes including germination index, time to 50% germination, co-efficient of uniformity of emergence and mean germination time were measured following pre-described methods (Coolbear et al., 1984; Wiesner, 1990; Ruan et al., 2002 and Bewley and Black, 2013).

### **Plant Growth Parameters Analysis**

On 21<sup>st</sup> day of germination plants were harvested for further analysis. After harvesting root and shoots were separated. Shoot length was taken from the upper layer surface of medium in petri plate to the tip of upper most shoot. Root length was also calculated with the help of meter rod. Plant root and shoot fresh weight was calculated with digital weight balance. For dry weight shoots and roots were oven dried at 72 °C for 48 h. Root and shoot dry weight was calculated with the same digital weight balance.

### **Photosynthetic Pigments Determination**

Fresh leaf material (0.1 g) was grinded in 8 ml of 95% acetone for determination of chlorophyll and carotenoid contents. Supernatant was filtered and absorbance was recorded at 646, 663 and 450 nm using spectrophotometer (Hitachi U-2001, Tokyo, Japan). The pigment contents were calculated following the method of Arnon (1949) using following formula:

$$\text{Chl a} = [12.7 (\text{OD } 645) - 2.69(\text{OD}645)] \text{ V}/1000 \times \text{W}$$

$$\text{Chl b} = [22.9 (\text{OD } 645) - 4.68 (\text{OD } 663)] \times \frac{\text{V}}{1000} \times \text{W}$$

V = volume of extract

W = weight of fresh leaf

Total chlorophyll was calculated as;

$$\text{Carotenoids (mg ml}^{-1}\text{)} = \text{A.car/ Em } 100\% \times 100$$

$$\text{A car (carotenoid)} = (\text{OD } 480) + 0.114 (\text{OD } 663) - 0.638 (\text{OD } 645)$$

$$\text{Em } 100\% = 2500$$

For determination of anthocyanin contents, the method of Mirecki and Teramura (1984) was used. Leaf tissue (0.1 g) in 1% acidified methanol was grinded. The grinded homogenate was incubated for 1h at 4 °C, the centrifugation was done at 12,000x g at 25 °C for 5 minutes. Absorbance was taken at 530 and 657 nm using Hitachi U-2001, Tokyo, Japan, spectrophotometer.

Anthocyanin content was determined following this equation:

$$\text{Q (Anthocyanin)} = (\text{A } 530 - 0.25 \times \text{A } 657) \times \text{M}^{-1}$$

Q (Anthocyanin) = absorption value that correlates with the absorbance

M = weight of plant material (g)

### **Oxidative Stress Indicators Determination**

#### **i. Lipid Peroxidation Activity**

Malondialdehyde (MDA) contents were measured by following Heath and Packer (1968) method. For this purpose, 50 mM phosphate buffer (25 ml) having pH 7.8, containing 1% polyethene pyrrole was taken and 0.1 g frozen plant leaf was grinded in mortar at 4 °C. Centrifugation of homogenate was made at 10,000 × g for 15 min. The mixture was allowed to heat at 100 °C for 15-30 min and quickly cooled in ice bath. The absorbance of supernatant was recorded at wavelengths of 532 and 600 nm using Hitachi U-2001, Tokyo, Japan, spectrophotometer.

Using following equation MDA content was calculated.

$$\text{MDA level (nmol)} = \Delta (\text{A } 532 \text{ nm} - \text{A } 600 \text{ nm})/1.56 \times 105$$

The absorption coefficient for MDA was  $156 \text{ mmol}^{-1} \text{ cm}^{-1}$ .

### *ii. H<sub>2</sub>O<sub>2</sub> Content*

To estimate H<sub>2</sub>O<sub>2</sub> content, Velikova et al. (2000) method was employed. The plant sample was grinded in 10% TCA and centrifuged. The supernatant of sample (0.1 ml) and 0.1 ml phosphate buffer were taken in test tube, then 2 ml Potassium Iodide (KI) were added and kept for 30 min. The absorbance was taken at 390 nm.

### *iii. Flavonoids Content*

In order to determine the flavonoid content Karadeniz et al. (2005) method was followed. Fresh leaf material (0.1 g) was grinded in 80% acetone solution, then 0.3 ml of NaNO<sub>2</sub> (5%) and 3 ml of distilled water were added into pure leaf extract. After shaking the mixture, it was allowed to reset at room temperature for 5 min. Then 2 ml of 1M NaOH and 0.6 ml of 10% AlCl<sub>3</sub> were added into the mixture and volume was made up to 10 ml by adding distilled water. The absorbance was taken at 510 nm.

### *Enzymatic Antioxidants Activities*

For extraction of enzymes 0.5 g fresh leaf was grinded in 10 ml of chilled phosphate buffer having pH 7.8. The extract was centrifuged at 2000x g for 20 min, the obtained supernatant was stored at -80 °C for the determination of enzymes activity.

For determination of superoxide dismutase (SOD), the protocol devised by Giannopolitis and Ries (1977) was used. The absorbance was recorded at 560 nm by using Hitachi U-2001, Tokyo, Japan, spectrophotometer. One unit of SOD was quantified by the amount of enzyme required to cause 50% inhibition of the rate of nitroblue tetrazolium (NBT) reduction.

Peroxidase (POD) and catalase (CAT) activities were estimated using the method described by Chance and Maehly (1955) by recoding the absorbance at 240 and 70 nm, respectively. The CAT activity was determined as units ( $\mu\text{mol}$  of H<sub>2</sub>O<sub>2</sub> decomposed per min) per mg of protein while POD 01 unit activity was defined as the change of 0.01 absorbance unit per min per mg of protein.

Nakano and Asada (1981) protocol was followed for ascorbate peroxidase (APX) activity and was expressed with pattern of wavelength variation at 290 nm.

### *Non-enzymatic Antioxidants Determination*

#### *i. Ascorbic Acid (AsA)*

Leaf AsA content was determined following method described by Mukherjee and Choudhuri (1983). Leaf sample (0.25 g) was grinded in 10 ml of 6% trichloroacetic acid (TCA). After centrifugation, 2.5 ml of supernatant were mixed in 2 ml of 2% acidic dinitrophenyl hydrazine solution. Two drops of 10% solution of alcoholic thiourea were added. The whole mixture was boiled for 20 min in water bath. After cooling, 5 ml of 80% H<sub>2</sub>SO<sub>4</sub> were added and absorbance was recorded at 530 nm. The AsA content in the samples was worked out from a standard curve, prepared using a range of AsA standards.

#### *ii. Total Phenolics*

The total phenolic contents were estimated according to the method described by Julkunen-Tiitto (1985) using 0.5 g fresh leaf sample.

#### *iii. Proline*

For the estimation of proline from leaf sample, the method described by Bates et al. (1973) was employed using 0.5 g

sample. The sample was grinded in 10 ml of 3% sulfo-salicylic acid. Then, 2 ml of homogenate were mixed with 2 ml of acid ninhydrin. The mixture was incubated for 01 h at 100 °C and immediately cooled on ice. The mixture was mixed with 4.0 ml of toluene using a vortex. The toluene layer containing chromophore was separated and kept at room temperature for a few minutes and the absorbance was read at 520 nm. The proline concentration was determined using standard curve on fresh weight basis as under.

$\mu\text{mole proline g}^{-1} \text{ fresh weight} = (\mu\text{g proline ml}^{-1} \times \text{ml of toluene}/115.5) / (\text{g of sample})$

#### iv. Reducing Sugars

Reducing sugars were determined colorimetrically by using 3,5-dinitrosalicylic acid solution according to Miller (1959) using glucose as a standard.

#### Determination of Shoot and Root Nutrient Content.

The seedling roots and shoots were separated, and thoroughly washed with distilled water. They were dipped in 20 mM EDTA solution for 30 seconds. After rewashing the samples with deionized water, they were oven dried at 105 °C for 24 h. Acid digestion of material was made in HNO<sub>3</sub>: HClO<sub>4</sub> (7:3 v/v). Sample was diluted with distilled water and final volume was made 50 ml following filtration. Using Atomic absorption spectrophotometer nutrient content (Fe<sup>2+</sup> and Mg<sup>2+</sup>) were determined.

#### Statistical Analysis

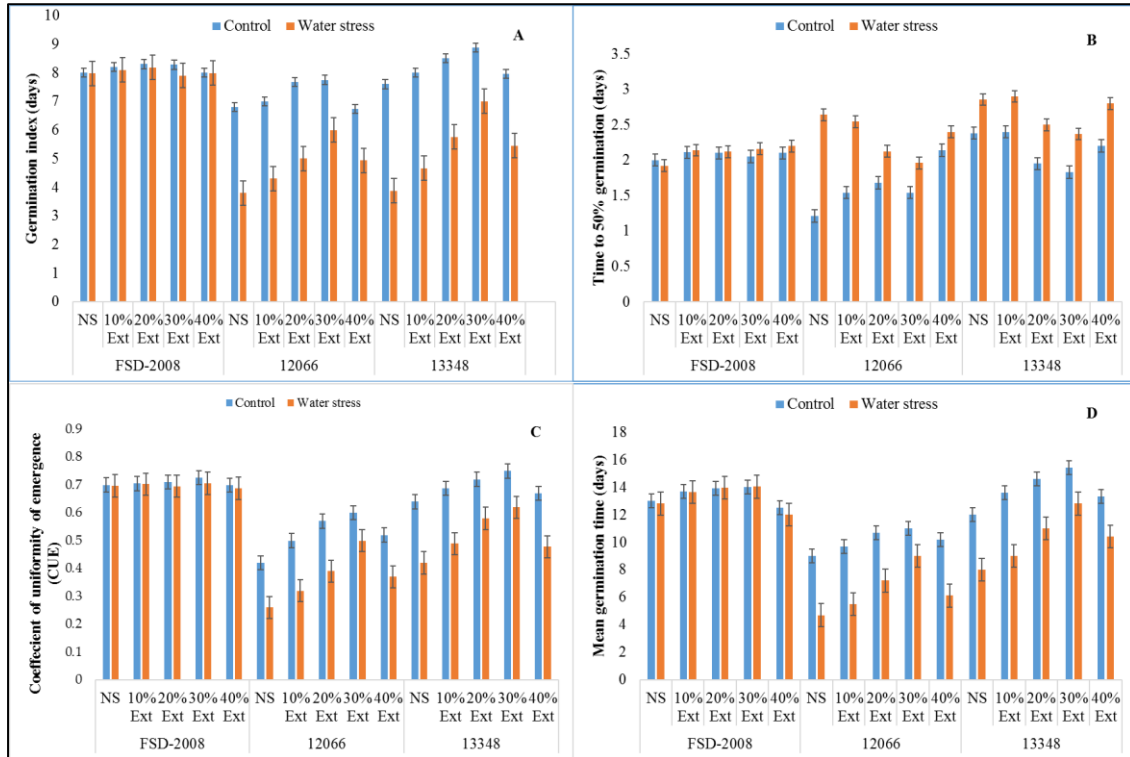
Collected data were subjected to an analysis of variance (ANOVA) using the statistical software Co-Stat version 6.2. The treatment means were equated by the

least significant difference method (Fisher's LSD) at p value of  $\leq 0.05$  level.

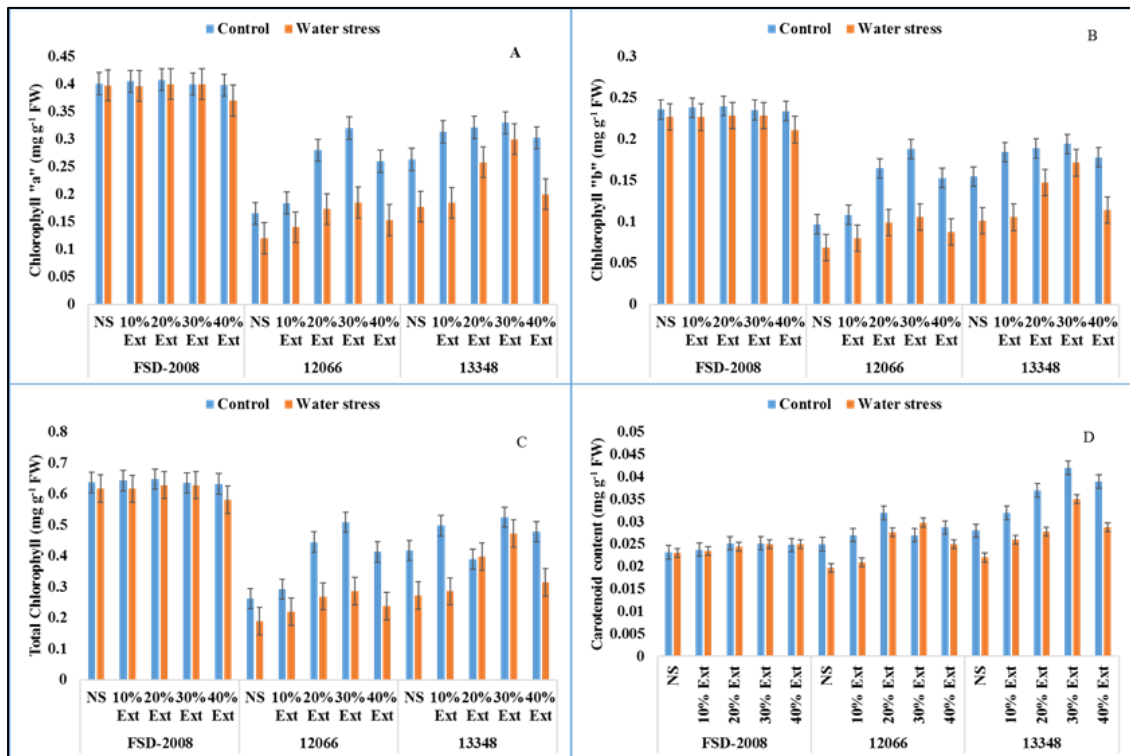
## RESULTS

### *Variations in Germination Attributes*

Drought is one of the significant abiotic stresses that drastically reduce the germination, growth and yield of crop plants. Arid and Semi-arid areas are facing water scarcity and considered poor for agricultural practices. Among the various practices used for crop improvement seed priming is considered as one of the best approaches. Priming with various chemicals has been used from a long time. Phyto-extracts are also used for priming purpose. In the current study we applied *C. decidua* stem extract due to its key chemical composition and environment friendly nature. Among the germination attributes germination index (A), coefficient of uniformity of emergence (c), mean germination time (d), reduced under water stress condition. FSD-2008 a drought tolerant variety experimentally proved from Ayub Agricultural Institute of Research, Faisalabad, Pakistan was not affected, and quite similar results were observed in seeds treated with *C. decidua* extracted solutions. The PEG-induced water stress significantly ( $p \leq 0.05$ ) reduced the described germination parameters in variety 12066 when compared with v variety 13348. The primed seeds also exhibited slight increase in the germination attributes. An increasing and decreasing trend observed in both wheat cultivars (12066 and 13348), germination attributes accelerated with increasing concentrations of extract, but increase was not continuous while 20% and 30% solutions significantly reduced the water stress effects. Time increased for reaching up to 50% germination under induced water stress condition except in FSD-2008 (Fig.1).



**Figure 1: Effect of *C. decidua* stem extract solutions on germination attributes (A=Germination index, B=Time to 50% germination, C=Coefficient of uniformity of emergence, D=Mean germination time, in wheat varieties (FSD-2008, 12066 and 13348) under PEG induced water stress.**



**Figure 2: Effect of *C. decidua* stem extract solutions on photosynthetic pigments (A=Chlorophyll a, B=Chlorophyll b, C= Total Chlorophyll, D=Carotenoids, in wheat varieties (FSD-2008, 12066 and 13348) under PEG induced water stress**



**Table 1. Effect of *C. deciddua* extract on growth parameters of wheat (*Triticum aestivum* L.) varieties (FSD-2008, 12066 and 133489) under PEG induced water stress.**

Treatments	FSD-2008 (control)	FSD-2008 (water stressed)	12066 (control)	12066 (water stressed)	13348 (control)	13348 (water stressed)
<b>Shoot fresh weight (g)</b>						
Ns	0.140	0.141	0.110	0.080	0.160	0.097
10% Ext	0.143	0.142	0.120	0.081	0.189	0.125
20% Ext	0.144	0.140	0.130	0.090	0.196	0.144
30% Ext	0.148	0.139	0.148	0.091	0.210	0.164
40% Ext	0.143	0.140	0.113	0.045	0.156	0.102
<b>Root fresh weight (g)</b>						
Ns	0.150	0.147	0.120	0.080	0.160	0.097
10% Ext	0.152	0.148	0.130	0.081	0.179	0.125
20% Ext	0.154	0.148	0.140	0.090	0.180	0.144
30% Ext	0.158	0.150	0.145	0.091	0.190	0.164
40% Ext	0.155	0.150	0.134	0.070	0.1560	0.120
<b>Shoot dry weight (g)</b>						
Ns	0.112	0.113	0.088	0.064	0.128	0.078
10% Ext	0.114	0.114	0.096	0.065	0.144	0.100
20% Ext	0.115	0.112	0.104	0.072	0.157	0.115
30% Ext	0.118	0.111	0.118	0.0728	0.168	0.131
40% Ext	0.114	0.112	0.090	0.036	0.125	0.082
<b>Root dry weight (g)</b>						
Ns	0.120	0.118	0.096	0.064	0.128	0.078
10% Ext	0.122	0.118	0.104	0.065	0.144	0.100
20% Ext	0.123	0.118	0.112	0.072	0.144	0.115
30% Ext	0.126	0.120	0.116	0.073	0.152	0.131
40% Ext	0.124	0.120	0.107	0.056	0.125	0.096
<b>Shoot length (cm)</b>						
Ns	11.430	11.360	9.000	6.400	12.000	8.000
10% Ext	11.400	11.380	9.500	6.780	13.500	9.800
20% Ext	11.443	11.400	10.700	7.400	13.945	11.000
30% Ext	11.500	11.500	9.600	7.890	14.700	12.300
40% Ext	11.460	11.450	10.000	6.340	12.568	10.000
<b>Root length (cm)</b>						
Ns	8.000	7.870	6.000	4.300	7.800	5.800
10% Ext	8.050	7.590	6.400	4.700	8.400	6.340
20% Ext	8.094	7.790	7.000	5.000	8.945	7.020
30% Ext	8.100	7.930	7.200	5.800	9.400	7.800
40% Ext	8.000	7.875	6.870	5.030	8.000	7.000

### ***Growth Variations***

Plant growth attributes, including root and shoot length along with the plant fresh and dry biomass significantly reduced in seedlings grown under water stress as compared to control condition. Table 1 shows data regarding shoot and root fresh weight, shoot and root dry weight, shoot length and root length. Among wheat varieties, FSD-2008 exhibited excellent growth because this was the experimentally proved drought resistant wheat variety, reduction in the growth parameter of variety 12066 and variety 13348 was observed under stress. Seed priming treatments positively increased the plant growth and the peak growth measurement was noticed at 20% and 30% treatment levels.

### ***Photosynthetic Pigment Contents***

The PEG-induced water stress decreased the plant photosynthetic pigment contents. However, the cultivar FSD-2008 seedlings were not much affected with induced stress. Application of *C. decidua* extract solution increased the listed parameters, but the increase was not in continuous manner, a decreasing trend was noted in seedlings under treatment of 40% extract (Fig.2). The wheat cultivar 13348 showed better improvement and resistance as compared to cultivar 12066 under water stress conditions. The 20% and 30% treatment levels proved more efficient in reducing drought effects in both the cultivars.

### ***Lipid Peroxidation Activity, H<sub>2</sub>O<sub>2</sub>, Anthocyanin and Flavonoids Contents***

Lipid peroxidation activity was estimated by measuring the MDA content under water stress and *C. decidua* extract applications. PEG-induced water stress significantly ( $p \leq 0.05$ ) increased the lipid peroxidation activity and caused breakage

of lipid contents of cellular membranes (Fig.3). However, *C. decidua* extract solutions of varying concentrations reduced the lipid peroxidation level along with concentration of H<sub>2</sub>O<sub>2</sub>.

The cultivar FSD-2008 showed resistant to the induced drought stress, while 12066 and 13348 showed relative effects against drought and given treatments. Anthocyanin and flavonoids contents were significantly affected by *C. decidua* extract applications under water stress as compared to control. The 20% and 30% extract solutions minimized the stress effects significantly ( $p \leq 0.05$ ).

### ***Enzymatic Antioxidants***

Plant possess antioxidant mechanism to deal with the various abiotic stresses. Stress levels may be estimated by calculating the activity of enzymatic and non-enzymatic antioxidants. Figure 4 represents the activity of various enzymatic antioxidants and the effects of drought and seed priming effects with various concentrations of *C. decidua* stem extract solutions. PEG-induced water stress condition activated enzymatic antioxidants activity in wheat seedlings. Wheat seedlings primed with different regimes of *C. decidua* stem extract showed a clear difference in comparison to seedlings without priming treatments even in drought. Cultivar difference was also found as represented in Fig.4. In case of treatment levels 20% and 30% extract solutions performed well under water stress conditions.

### ***Non-enzymatic antioxidants, sugars and proline content***

The data presented in figure 5 elaborates the effects of drought and applied treatments on phenolic, reducing sugars, proline and ascorbic acid in wheat seedlings grown under PEG

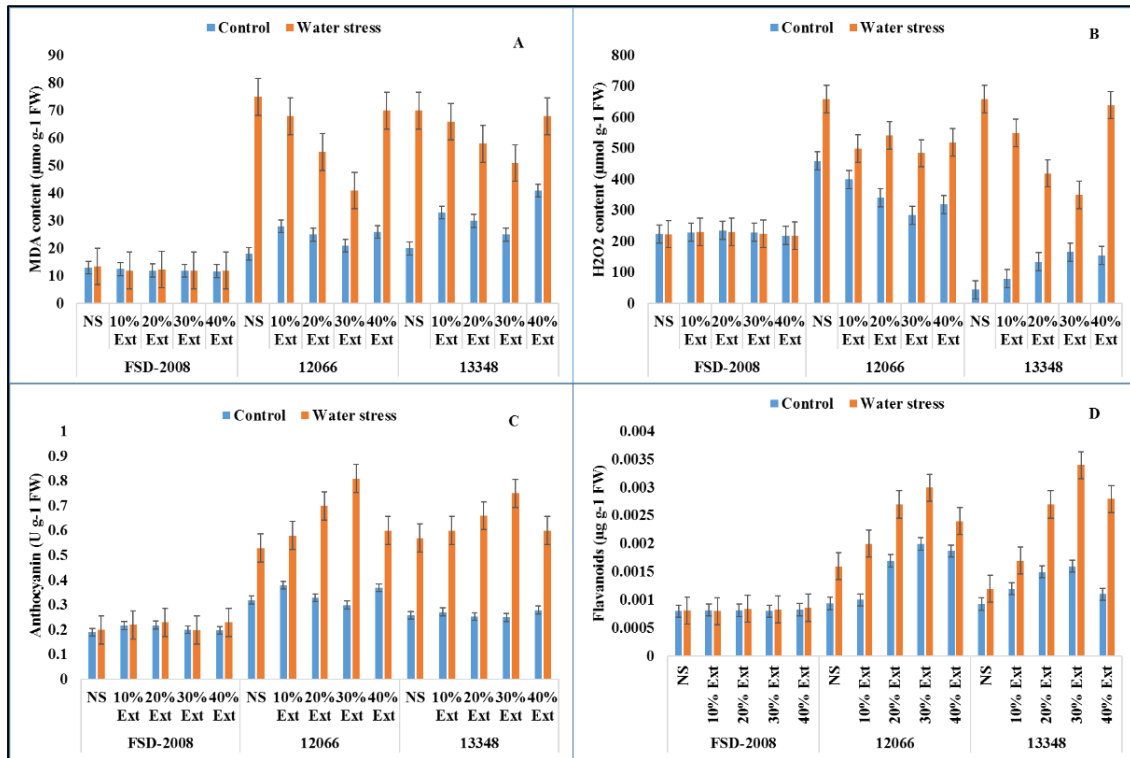


Figure 3: Effect of *C. decidua* stem extract on A=MDA, B= H<sub>2</sub>O<sub>2</sub>, C= Anthocyanin, D=Flavonoids, in wheat varieties (FSD-2008, 12066 and 13348) under PEG induced water stress.

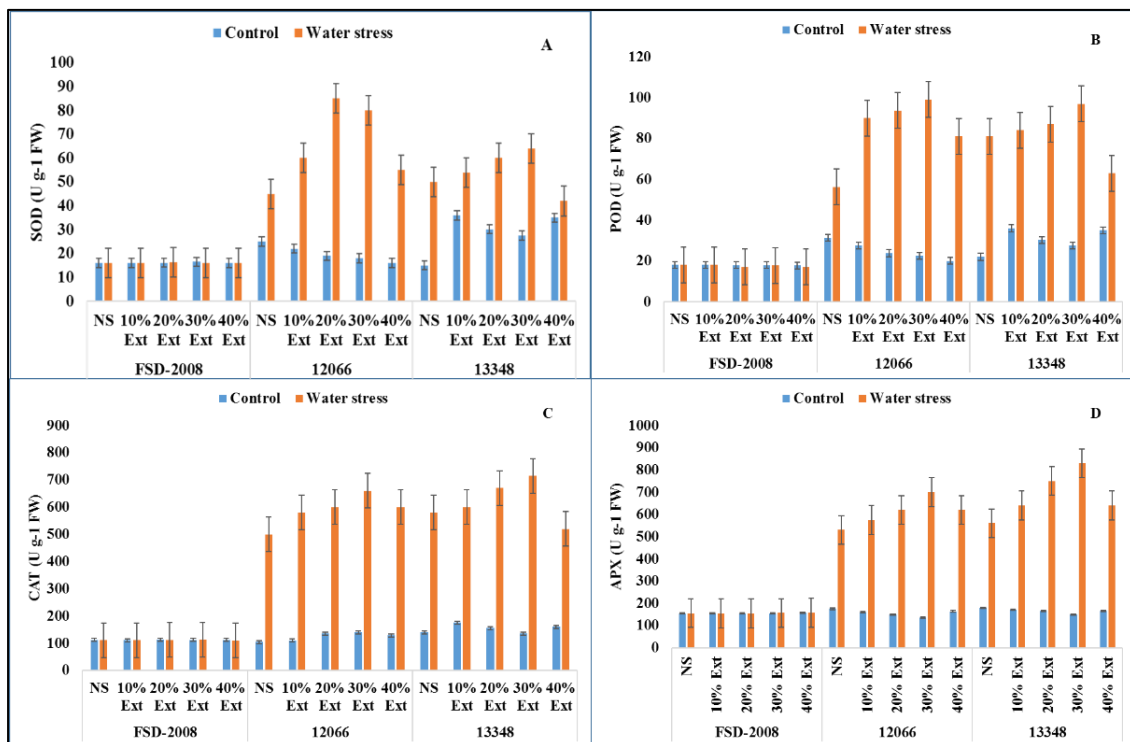


Figure 4: Effect of *C. decidua* stem extract on enzymatic antioxidants (A=SOD, B= POD, C= CAT, D=APX) in wheat varieties (FSD-2008, 12066 and 13348) under PEG induced water stress.

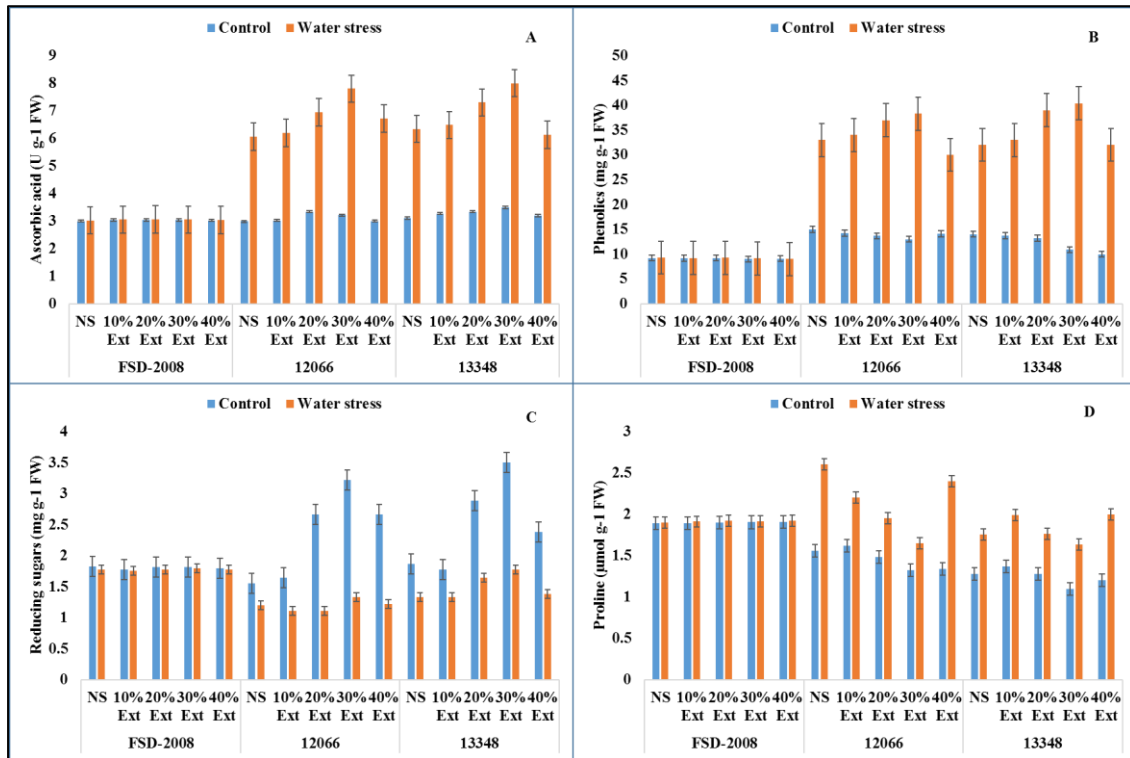


Figure 5: Effect of *C. deciddua* stem extract on A=ascorbic acid, B= phenolics, C= reducing sugars, D=proline contents in wheat varieties (FSD-2008, 12066 and 13348) under PEG induced water stress.

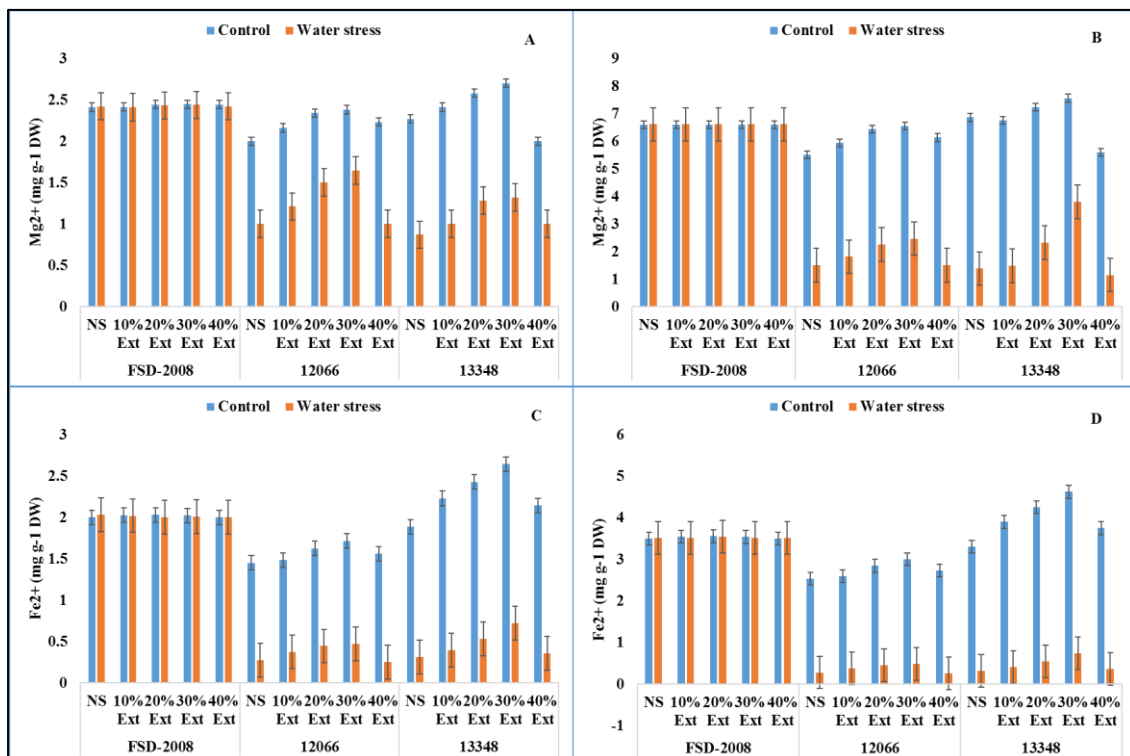


Figure 6: Effect of *C. deciddua* stem extract on Mg<sup>+2</sup> (A=root, B= shoot) and Fe<sup>+2</sup> uptake (C= root, D=Shoot) in wheat varieties (FSD-2008, 12066 and 13348) under PEG induced water stress.

induced water stress. It exhibited reduction in the contents of listed components as compared to non-stress condition. All the tested wheat cultivars showed significant effects of drought and priming treatments except FSD-2008. With increasing concentration of *C. decidua* extract the contents of listed components also increased but further increase in concentration after 30% caused reduction again. In case of proline, stress increased the proline contents. Priming effectively mitigated the drought effects and reduction level was significant in wheat seedlings primed with 20% and 30% extract (Fig.5).

### ***Variation in Essential Ions uptake Capacity***

Ion uptake in root and shoot reduced under given PEG-induced water stress. The figure 6 is representing data related to essential ions ( $Mg^{2+}$  and  $Fe^{2+}$ ) uptake ability of wheat varieties under stress and control condition with seed priming of *C. decidua* extract of varying concentrations. FSD-2008 respond toward ion up take was not much disturbed due to its drought resistant mechanism. However, 12066 and 13348 cultivars could not overcome the stress and ion uptake reduced significantly under water stress condition. Essential ion uptake ability enhanced with increasing the treatment levels but cultivar difference in ion uptake ability was also observed.

### **DISCUSSION**

Drought, high temperature and salinity are the significant inevitable abiotic stresses that significantly ( $p \leq 0.05$ ) reduce plant growth, germination and yield (Jaleel et al., 2009) and all of these are interrelated with each other (Brenchley et al., 2012). Various abiotic stresses cause 50% crop loss, among them drought and heat contribute 10% and 20% respectively (Kajla et al., 2015). In this study negative drought effects and seed priming efficiency was explored in commercial

wheat varieties (FSD-2008, 12066 and 13348). Water stress condition reduced germination properties (GI and CUE). Our study clearly showed that water stress significantly reduced germination index, coefficient of uniformity of emergence. Seed priming with *C. decidua* stem extract significantly improved the germination (Figure.1). This study is in strong relation with previous reports (Yasmeen et al., 2013). Seed priming with *Moringa* leaf extract increased germination attributes in wheat in a study conducted by Afzal et al. (2008). The same results were reported in maize (Basra et al., 2011). Our study was in agreement with all these previous studies.

Drought caused decline in growth attributes as indicated in table.1. However, seed priming with *C. decidua* increased the plant growth attributes. Our study was making a direct connection with (Sundaria et al., 2019), where seeds primed with iron oxide nanoparticles triggered the plant growth in wheat under drought. Seed priming with silicon nanoparticles enhanced plant biomass, growth, chlorophyll and carotenoid contents of wheat when exposed to Cd stress (Hussain et al., 2019). Current study revealed when seeds of wheat cultivars soaked in different concentrations of *C. decidua* extract quite similar increasing and decreasing trend was observed in both wheat cultivars (12066 and 13348). Seeds primed with SA significantly enhanced the plant biomass, chlorophyll content, total soluble sugars, total soluble protein and reduced free amino acid content by reducing the adverse effects of drought in wheat under salinity (Azeem et al., 2018), the findings were in strong relation with this study. Phenolics content of leaf also increased in primed seeds like currently conducted study. The Si NPs priming reduced the oxidative damage by reducing MDA content and enhanced the enzymatic antioxidant (SOD, POD, and CAT) activity up to 38%, 56% and 63% respectively (Hussain et al., 2019). The

findings of current study were in line with previous report (Shah et al., 2019), where seed priming improved the activity of enzymatic anti-oxidants (POD, SOD and CAT), and 20% and 30% treatment levels exhibited significant positive impacts in alleviating the drought effects.

Nucleic acids, proteins, membrane lipids and photosynthetic pigments are oxidized due to the drought-induced high reactive oxygen species (ROS) production. It leads towards modification of cellular redox status ultimately retarding physiological and biochemical activities in plant cells (Saleem et al., 2020). Low water levels in the soil cause ultra-structural alterations, oxidative stress, increased electrolyte leakage and MDA concentrations. As a result, the antioxidant enzyme activities such as superoxide dismutase (SOD), peroxidase (POD) and catalase (CAT) and ascorbate peroxidase (APX) are altered in cells (Kamran et al., 2019). As reported by Taha et al. (2020) antioxidants, such as ascorbic acids and glutathione contents, additional ROS may be scavenged and therefore increased plant tolerance against harsh conditions may be achieved (Kamran et al., 2019). Furthermore, osmo-protectants also constitute redox active molecules which hamper the production of ROS as well as participate in the ascorbate-glutathione cycle (Rana et al., 2020). In the present study, the contents of MDA and H<sub>2</sub>O<sub>2</sub> was increased in the plants which were grown in the water deficient environment compared to those plants which were grown in the control treatment. The increase in the concentration of reducing sugars in drought-stressed plants showed that improved cell osmotic adjustment would help to maintain higher water content in plant cells and lower electrolyte leakage (Anjum et al., 2017).

The uptake of common minerals is inhibited under drought conditions reducing the plant productivity (Nawaz et al., 2020). Nutrient absorption and water up take are different phenomenon but

correlation of water with plant growth and nutrient transportation make these process closely related (Waraich et al., 2011). Water acts as a medium for nutrient ions up take by root cells and ultimate transportation. The water limited environment reduced the nutrient mobility in soil thus inhibiting absorption and transportation of these nutrient ions by root. Thus, plants under drought stress have low absorption of nutrients and ion homeostasis mechanism is destroyed (Waraich et al., 2011). Suitable mineral absorption is vital for the conservation of plant structural and functional integrity and any deviations in mineral commitment may negatively affect plant metabolism. Compared to the plants grown in the control treatment, the contents of various minerals (Mg<sup>2+</sup> and Fe<sup>2+</sup>) in the roots and shoots of *T. aestivum* seedlings were decreased significantly ( $p \leq 0.05$ ) in the plants grown in the water deficient condition. Although the plant would take up sufficient quantities of essential nutrients to control plant structure and composition and many other biological processes of a plant's life cycle and any decreased in nutrient uptake, not only impaired plant metabolism but also decreases plant growth and yield related attributes (Liang et al., 2018). It has been concluded by this study the stem extract of *C. decidua* may be applied as seed priming of wheat cultivars specially developed for arid and semi-arid regions to increase growth and yield under water limited conditions.

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