

EFFECT OF DROUGHT STRESS ON CHLOROPHYLL FLUORESCENCE, AND BIOMASS PORTIONING OF *AEGILOPS TAUSCHII* L.

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Abstract. *Aegilops tauschii* Coss. is one of the problematic weeds, which competes for resources in wheat crop. Drought stress affects productivity, growth, and development of the crops. A greenhouse experiment was conducted to evaluate the effect of different field capacity levels on biomass partitioning and chlorophyll fluorescence on *Aegilops tauschii* population of China. Dry biomass, plant height, leaf area (LA), leaf area ratio (LAR), specific leaf area (SLA), and the root-shoot ratio (RSR) fluctuated under different field capacity levels and decreased with increasing drought stress level. Maximum dry biomasses, plant height (1.71 g and 15.6 cm) were observed in 70-80% field capacity while the minimum (0.70 g and 12.7 cm) were observed under 10-20% field capacity (FC) level. In root-shoot ratio maximum (0.72 g) was observed in 50-60% field capacity level while minimum (0.60 g) was observed under 10-20% field capacity level. Similarly, in LAR and SLA the maximum values (111.67 cm, 175.45 cm and 433.59 g) were observed in 70-80% field capacity level while the minimum values (68.78 g, 42.02 g and 86.32 g) were recorded under 10-20% field capacity level. Similarly, drought stress also influences the maximum fluorescence (F_m) in the light-adapted state and has an effect on the maximum quantum yield of PSII. Similarly, the trend was observed in qP (photochemical quenching) and qN (non-photochemical quenching) like minimal and maximal fluorescence.

Keywords: *growth biomass, abiotic stress, Aegilops tauschii and field capacity, photosynthetic activity, water stress and mechanism of Aegilops tauschii*

Abbreviations: LA: Leaf area; LAR: Leaf area ratio; SLA: Specific leaf area; Fo: Minimal fluorescence; F_m: Maximum fluorescence; qP: Photochemical quenching; qN: non-photochemical quenching; PSII: Photosystem II

Introduction

Goat grass (*Aegilops tauschii* Coss.) is among one of the noxious exotic weeds (Dudnikov, 2014), with troublesome effect particularly in wheat-growing areas (Zhang et al., 2007). It is native to temperate Asia, tropical Asia and Europe (USDA, 2010). *Aegilops tauschii* was thought to be an aggressive plant that scattered across more than ten provinces (Fig. 1) of China (Wei et al., 2008, 2007; Su et al., 2013). It competes with the main wheat for capturing resources such as nutrients, light, and water. It reduced the crop's yield because it performs better under environmental stress conditions (Curtis and Halford, 2014). Under natural environmental conditions plant faces different types of environmental constraints such as drought stress, salinity stress and temperature stress (Tatrai et al., 2016; Mittler, 2006; Ibanez et al., 2010). Under different abiotic stresses, plants change their physiological, molecular and cellular conditions (Massa et al., 2013). Water is the main component and has an important role in plant growth and development. Roots have an important role among plant parts to uptake the water under drought condition and distribute in whole plant body (Wang et al., 2013).

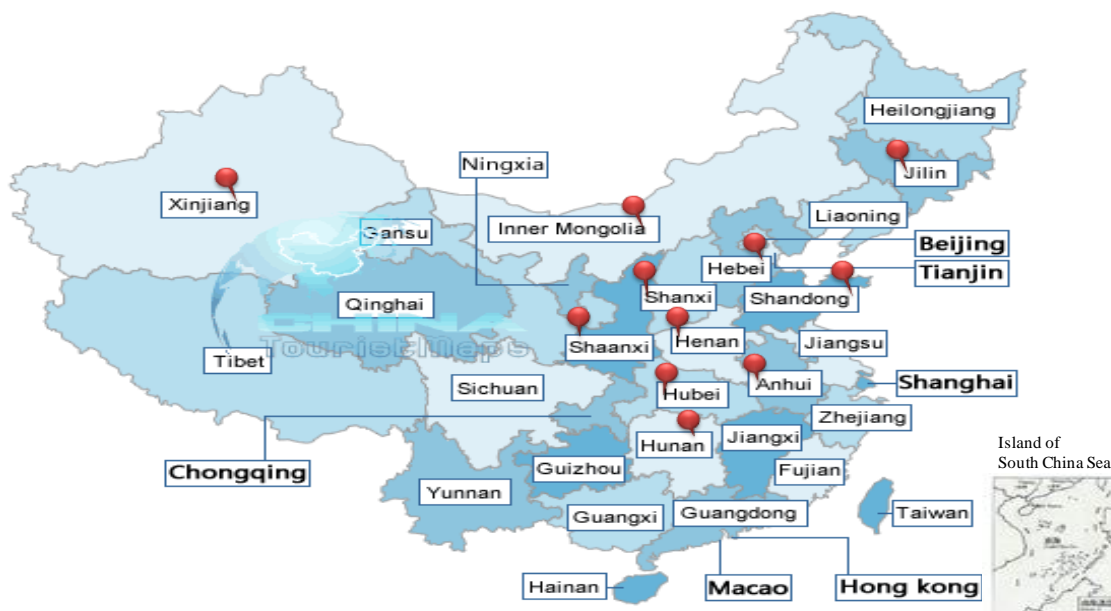


Figure 1. Geographic zones of *Aegilops tauschii* and red dot indicating the affected provinces

Plant response to limited water is fundamental for implementing crop management strategies (Chaves et al., 2003). Under drought stress levels plants stimulate phenological and biochemical changes which decrease plant growth (Benjamin et al., 2014). Plant growth parameters (plant height, leaf area, dry weight biomass, and chlorophyll fluorescence) are affected under drought condition (Guo et al. (2016). In addition the photosynthetic activity is directly affected under water stress condition (Wu et al., 2010). The crop vulnerability under drought stress depends upon the duration of growth stages, plant species, and stress intensity (Singh et al., 2013). One of the prominent techniques for attaining comprehensive estimates of photosynthetic activities is chlorophyll fluorescence. Chlorophyll fluorescence is an important and fast technique for the examination of photosynthetic activities (Sajbidorova et al., 2015). Plant biochemistry and function of PSII are affected by different abiotic stresses (Yang, 2013). Drought stress significantly effects maximal fluorescence (F_m), maximum fluorescence in the light-adapted state ($F'm$), the maximum quantum yield of PSII, photochemical quenching qP and non-photochemical quenching qN (Dias and Bruggemann, 2010). Similarly, Cendrero-Mateo et al., 2015 along with Van der Tol et al., 2009 reported that accessibility of light, nutrition, and drought stress manipulate the relationship between photosynthesis, chlorophyll fluorescence, and non-photochemical quenching.

In general, under different abiotic stress conditions chlorophyll fluorescence is a dominant technique for photosynthetic activity (Barbagallo et al., 2003; Proctor et al., 2003; Gottardini et al., 2014). This technique has been used in the study of wheat stress response under different abiotic stress conditions, such as water stress (Eduardo et al., 2002), heat stress (Lu et al., 1999, 2001; Chaerle et al., 2007), cold stress (Rizza et al., 2001; Ying et al., 2002), salt stress (Lu et al., 2003), and nitrogen deficiency (Lu et al., 2001; Shangguan et al., 2000) (Sampol et al., 2003). As we know *Aegilops tauschii* is a rich source of genetic material and perform better under biotic and abiotic stress condition (Assefa and Fehrmann, 2000; Hsam et al., 2001; Colmer et al., 2006).

Some studies investigated the biological characteristic in different crop plant species under different abiotic stresses (Ma et al., 2010). In order to ascertain control strategies for resistant weeds, problem-solving approaches of the species characters should be constructed (Beffa, 2012). Chlorophyll fluorescence was observed on barley leaves when the carbon assimilation was affected (Quick and Horton, 1984). It started an era for stress detection on photosynthesis with chlorophyll fluorescence measurement. In weed science, herbicide stress on Photosystem II (PSII) was used for the dose optimization by measuring photosynthetic efficiency (Kempenaar et al., 2010). Chlorophyll fluorescence was used to quantify the agar-based Syngenta ‘RISQ’ test and detected the herbicide resistance in *Alopecurus myosuroides* depending on the dose response analysis (Kaundun et al., 2011; Wang et al., 2015). Due to the fast-growing habit under the stressful environmental condition, we design an experiment to evaluate the effect of different field capacity levels on biomass partitioning and chlorophyll fluorescence of *Aegilops tauschii*. The objective of this study is to develop a method capable to be used in the field for measuring chlorophyll fluorescence quantum yield of PSII (Fv/Fm) under different water capacity level. The results would contribute to the strategic decision to understand the behavior of *Aegilops tauschii* under different field capacity level.

Materials and method

Plant materials, growth conditions, and stress treatments

A greenhouse experiment was conducted at The Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China. *Aegilops tauschii* seeds were collected from five different parts of China. After drying for one week in open sunlight, deformed and damaged seeds were discarded, and apparently healthy seeds were air-dried and then stored at 4 °C. Before sowing seeds were soaked in water for 24 h. A complete randomized design was applied under four replication with four treatments of different field capacity levels (10-20% (D1), 30-40% (D2), 50-60% (D3) and 70-80% (D4). Seeds of similar size were sown in plastic pots (15 cm in diameter and 13 cm in height) with seven seeds per pot in June 2017. Each pot contained similar volumes of sandy loam soil, and hummus in a ratio of 3:1. Initially, pots were watered thoroughly, with soil moisture maintained at about 80–100% field capacity, to ensure the seed germination. Fifteen days after sowing different field capacity levels were maintained in the pots according to the experimental layout and continued until 60 DAS treatment instigation. Soil moisture was measured by using a TDR soil moisture meter (TRIMEPICO, Germany).

Growth parameters

Plant height was measured with a meter rod from the base of the plant to the leaf tip. The height of five chosen plants was measured in each pot and was averaged. Leaf area was measured by a portable leaf meter, and five plants from each pot were selected for further analysis. For recording dry biomass allocation data five plants were removed carefully and washed for the further procedure; the plants were separated into root and shoot put into an oven and dried for 48 h, dry weight was measured with a digital weight balance. Leaf area ratio was determined as follows: total leaf area divided by dry weight biomass. Specific leaf area (SLA) is measured as the ratio of leaf area to dry mass.

Chlorophyll fluorescence

Chlorophyll fluorescence was measured with an imaging-PAM Mini-Series (IMAG-K5, Walz Germany) (Fig. 2) following the procedure described by van Kooten and Snel (1990). Chlorophyll fluorescence imaging-PAMs are highly sensitive research instruments which give quantitative information on the quantum yield of photosynthetic energy conversion. Selected plants of *Aegilops tauschii* were placed in the darkroom for 30 min before measuring the Chlorophyll fluorescence under the different field capacity levels. This imaging-PAM mini-series was used to measure minimum fluorescence (F_o'), maximum fluorescence ($F'm$), the quantum yield of PSII (Φ PSII), photochemical quenching (qP) and non-photochemical quenching (qN). The photochemical quenching (qP) (Eq. 1) and non-photochemical quenching (qN) (Eq. 2) was determined by the following formula shown in Equations 1 and 2.

$$qP = (F'm - F_s) / (F'm - F_0) \quad (\text{Eq.1})$$

$$qN = (F_m - F'm) / F'm \quad (\text{Eq.2})$$

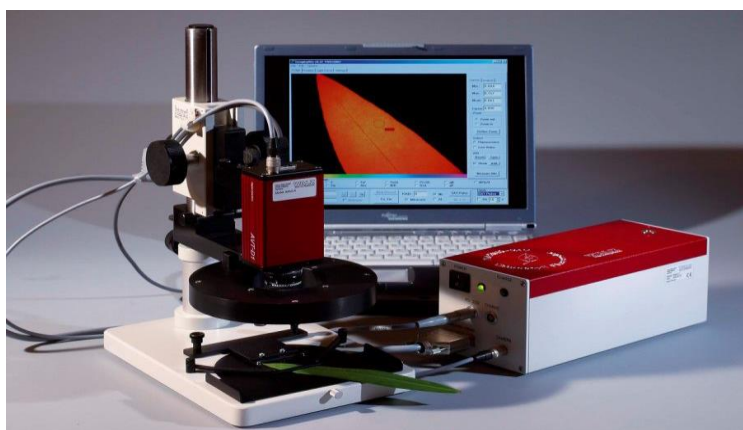


Figure 2. PAM M-series used for chlorophyll fluorescence parameters

Statistical analysis

Statistix 8.1 software (Analytical software, Statistix; Tallahassee, FL, USA, 1985-2003) was used for statistical data extrapolation. Analysis of variance (ANOVA) for plant dry weight biomass, root biomass, and leaf weight biomass was used to find out the effect of treatments and Tukey's honest significance difference (HSD) test was used at 0.05 probabilities to compare the differences among the treatment means. All the results are expressed in standard error (SE) and shown in the figures.

Results

Effect of different field capacity level on growth parameters

Drought stress significantly affects all the parameter under study about biomass partitioning and chlorophyll fluorescence. Chlorophyll fluorescence and growth parameter significantly decreased with increasing drought stress level. Data regarding dry weight biomass of *Aegilops tauschii* under different field capacity level (Table 1)

indicated that dry weight biomass was significantly ($p < 0.05$) influenced by different field capacity level, maximum dry weight biomass (1.71 g) was recorded under 80% field capacity level, and the minimum (0.70 g) was recorded in lower field capacity level (10-20%). The maximum reduction (59%) in dry weight biomass was recorded in (10-20%) field capacity level respectively. In this study, maximum plant height (15.6 cm) was recorded under 70-80% field capacity level while minimum (12.7 cm) was recorded in lower field capacity level. The maximum reduction in plant height (18.5%) was under 10-20% field capacity level. Regarding root-shoot data, the maximum value (0.72 g) was recorded in 70-80% field capacity level while the minimum (0.60) was recorded under 10-20% field capacity level. The reduction with decreasing water field capacity was (16.6%) under 10-20% field capacity level. Regarding leaf area data the maximum (Fig. 3) (111.67 cm) was recorded under 80% field capacity level while the minimum (68.78 cm) was recorded under lower field capacity (10-20%) level. The maximum reduction in leaf area (38%) was recorded through lower field capacity level. Leaf area ratio (LAR) and specific leaf area (SLA) were significantly affected under field capacity levels, the maximum LAR and SLA (175.45 and 433.59) was recorded under 80% field capacity level while the minimum (42.02 and 86.32) was recorded under lower field capacity level (10-20%). The maximum reduction (76% and 80%) in LAR and SLA was recorded through a decreasing field capacity level.

Table 1. Dry weight biomass, plant height, leaf area (cm), leaf area ratio, specific leaf area and root shoot ratio of *Aegilops tauschii* under different field capacity levels

Field capacity	Dry weight	Plant height (cm)	Leaf area	LAR	SLA	R/S ratio
D1	0.70 C	12.7 C	68.78 B	42.02 B	86.32 B	0.60 A
D2	0.98 C	13.5 BC	92.22 AB	77.76 B	147.63 B	0.71 A
D3	1.19 B	15 AB	99.95 A	102.10 B	239.5AB	0.72 A
D4	1.71 A	15.6 A	111.67 A	175.45 A	433.59A	0.68 A

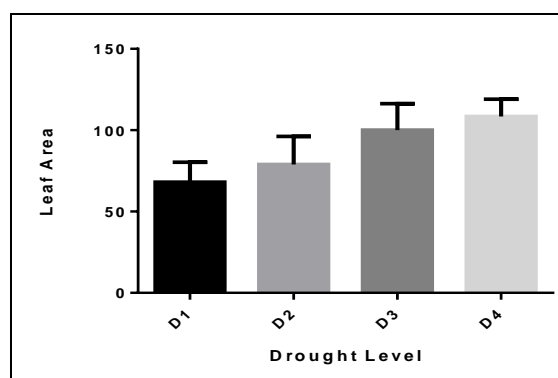


Figure 3. Effect of different capacity levels on leaf area (m^2) of *Aegilops tauschii* L.

Effect of different field capacity levels on chlorophyll fluorescence

Photosynthetic activity (Chlorophyll Fluorescence) was significantly affected under different capacity levels. In different field capacity levels the maximum minimal fluorescence (F_o) was recorded under 20% field capacity and gradually decreased with

increasing field water capacity, and reached a 0.12 value at maximum field capacity (70-80%). Maximal fluorescence (F_m) (Fig. 4b) was recorded (0.32) in 70-80% field capacity and minimum fluorescence (Fig. 4a) (0.20) was recorded under 10-20% field capacity. Under different field capacity levels values of the quantum yield of PSII (F_v/F_m) reaction centre were recorded. Maximum PSII (Fig. 5a) (0.44) was recorded under 70-80% field capacity level while minimum (0.29) was recorded under 10-20% field capacity level. Similarly, trend was observed in effective quantum yield on Photosystem II (Fig. 5b). The photochemical quenching ingenuousness of the center of PSII determines the proportion of energy engrossed by photosystem pigments, which is used for photosynthesis (Van Kooten and Snel, 1990). In addition, the results showed that photochemical quenching qP (Fig. 6a) was increased under 70-80% field capacity and reached a maximum (0.52) and decreased gradually with decreasing field water capacity and reached a minimum (0.39) level. The non-photochemical quenching (qN) (Fig. 6b) is an important mechanism to protect the plant. In non-photochemical quenching $qN = (F_m - F'_m)/F'_m$ directly correlate with field capacity level. An increase in non-photochemical quenching was observed with an increase of field capacity level. Maximum non-photochemical quenching (0.79) was recorded under 70-80% field capacity level and reached a minimum (0.61) at 20% capacity level.

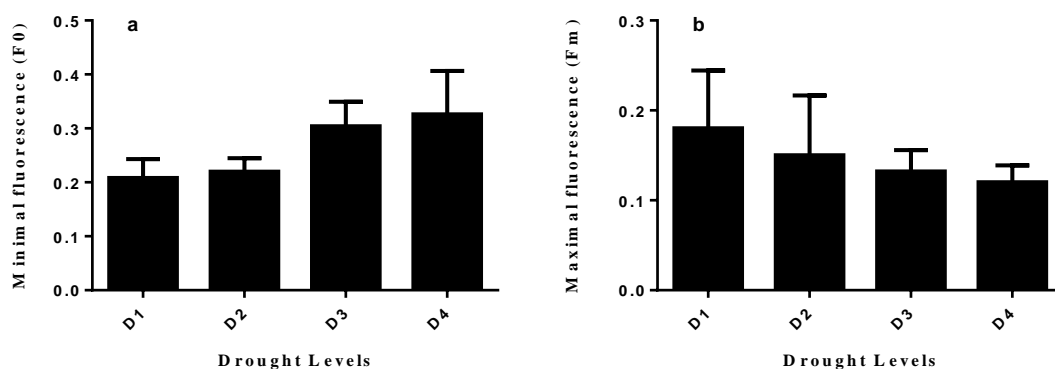


Figure 4. Effect of different capacity levels on minimal fluorescence and maximal fluorescence of *Aegilops tauschii* L. Bars are expressed as \pm SE

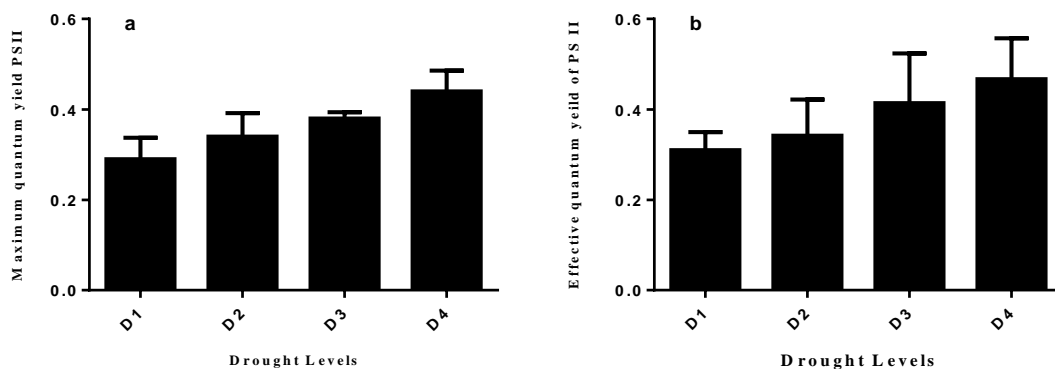


Figure 5. Effect of different capacity levels on the maximum quantum yield on Photosystem II and effective quantum yield on Photosystem II under different field capacity levels

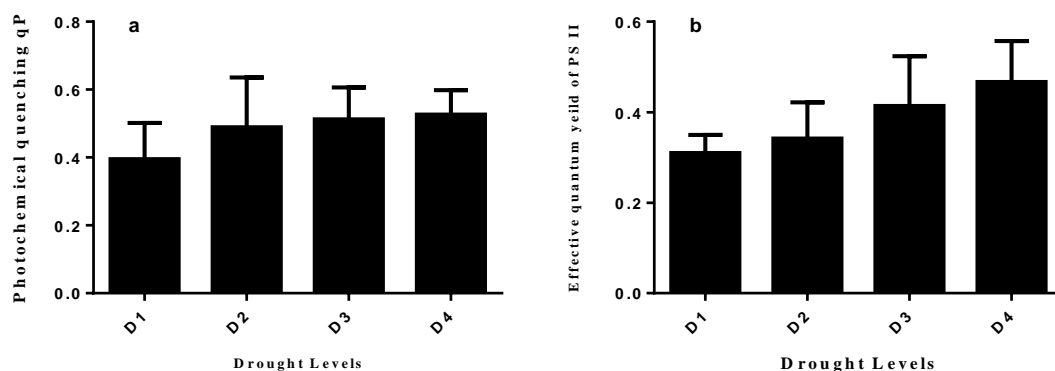


Figure 6. Effect of different capacity levels on photochemical quenching (qP) and non-photochemical quenching (qN) of *Aegilops tauschii* L. Bar are expressed as \pm SE

Under drought stress duration non-photochemical quenching (qN) showed a decline, at 70-80% field capacity a qN value of 0.79 was recorded, which was higher than at 10-20% field capacity (0.61). This showed that the degree of ingenuosness of PS II and reaction center under drought stress condition were very low as compared to high field capacity (70-80%), which affect the photosynthetic activity in plants. In our study plant leaves and photosynthetic activity severely damaged under lower field capacity which led to decrease in Fm' , Fv/Fm , $\Phi PSII$, QP , and qN . While *Aegilops tauschii* performed better under drought stress condition.

Discussion

In the present study, we analyzed the effect of different field capacity levels on the biomass partitioning and chlorophyll fluorescence on *Aegilops tauschii*. The plants were irrigated after three days interval. The *Aegilops tauschii* was surviving better under high water stress at 20% field capacity. Under lower field capacity *Aegilops tauschii* shows aggressiveness and perform better. According to the above results, under higher field capacity *Aegilops tauschii* growth is higher than in lower field capacity. This competitive utilization of water in *Aegilops tauschii* in an early stage help to utilize more water content of the soil and initiates more rapid growth as compared to the wheat crop. *Aegilops tauschii* can uptake more water as compared to another crop.

However, lower field capacity level (10-20%) negatively affected the leaves, root, and plant dry biomass of *Aegilops tauschii*. Water is an important component for growth, development, and production (Kabbas et al., 2018). Likewise, Bortolheiro and Silva (2017) and Yamaguchi-Shinozaki and Shinozaki (2006) reported that accessibility and unavailability of proper water seriously affect the biochemical processes, physiology, and morphology of crop plants. Plant growth and biomass allocation are affected under drought stress (Li et al., 2008) and with the variation in availability of water vary the growth and biomass of the plants (Padilla et al., 2009). Chuhan (2013) reported a decrease in biomass production of switch grass under lower field capacity level.

Plants under different drought stress conditions decreased the dry biomass and distorted the biomass allocation from root to leaves. We also conclude that under lower

field capacity dry biomass significantly increases and *Aegilops tauschii* survive better. Similarly, results were found in *Prunus mongolica* in root to shoot ratio under different field capacity levels (Guo et al., 2015; Liu et al., 2017). Similarly, in our study different field capacity levels affected *Aegilops tauschii* growth. Rehimy et al. (2010) said that *Plantago ovata* and *P. psyllium* are significantly affected under drought stress condition. Azevedo (2011) reported under lower field capacity *Carapa guianensis* has maximum growth and biomass. In our study, *Aegilops tauschii* was suppressed under lower field capacity (20 to 40%) level. This may be due to a survival mechanism and growth under different conditions. Under lower water field capacity plant height decreased due to a decline in cell enlargement (Li et al., 2008; Manivannan et al., 2007; Wang et al., 2013). Likewise, Moran and Shower (2005) said that lower field capacity affects or decrease the fresh weight and shoot weight (35% and 35%) of *A. palmeri* S. Continuous water stress affect the smaller leaf of dwarf plants (Haffani et al., 2014).

Under different abiotic stress conditions, chlorophyll fluorescence is an important and common method to determine the leaves photosynthetic activity (Resco et al., 2008). Moreover, Lichteuthaler and Miede (1997) said that plant leaves are sensitive in nature under drought stress condition, decreased the photosynthetic rate at Photosystem II and increased the non-photochemical quenching. In our experiment, lower field capacity greatly affected the chlorophyll fluorescence of *Aegilops tauschii* and considerably decline with a decreasing of field capacity level. The date related to chlorophyll fluorescence of *Aegilops tauschii*. Minimal fluorescence (F_0) increased, while maximal fluorescence (F_m) decreased with increasing field capacity level. This showed that the transportation energy absorbed by Photosystem II partially suppressed the photochemical reaction.

The increase in energy by fluorescence and thermal deactivation loss evidently reduced the photosynthesis energy. Under lower field capacity, photosynthetic activity of *Aegilops tauschii* was damaged that gradually decreased the F_v/F_m value. This showed that the initial reaction of photosynthesis in *Aegilops tauschii* is concealed and the exchange competence of light energy was decreased by inhibiting the potential active center. Chlorophyll fluorescence (F_0) is a powerful component under drought stress condition (Percival and Sheriffs, 2002). The photochemical quenching imitates to an increase in minimal fluorescence and a decrease in maximum fluorescence (F_m) or a ratio of F_v/F_m (Thomas and Turner, 2001). In *Aegilops tauschii*, photochemical quenching and non-photochemical quenching was decreased under lower field capacity level. An important component of Photosystem II is photochemical quenching (q_P) (Li et al., 2013), and protection system under stress condition is non-photochemical quenching (q_N). It showed the proportion of energy absorbed by the PSII pigment but debauched as a heat (Maxwell and Johnson, 2000). *Aegilops tauschii* photosystem II was significantly damaged due to decreased non-photochemical quenching (q_N) and trouncing of heat indulgence potential. This showed that long-term lower water availability affects the photosynthetic process in *Aegilops tauschii* which partially suppressed the photochemical reactions. Similarly, this kind of results have been reported in different crops, such as sugar beet (Sikuku et al., 2010) *Coronilla varia* (Yang et al., 2013) In *P. mongolica* and *L. ruthenicum* (Guo et al., 2015) it was showed that chlorophyll fluorescence parameters are extensively affected under different field capacity levels.

Conclusion

This study revealed the survival mechanism of *Aegilops tauschii* under different field capacity levels. In conclusion, our results showed that lower field capacity affects the biomass partitioning and chlorophyll fluorescence of *Aegilops tauschii*. Furthermore, all growth-related parameters start declining under 20 to 40% field capacity, the growth and survival of *Aegilops tauschii* were adversely affected under higher field capacity of 60 to 80%. Likewise, lower field capacity level damages *Aegilops tauschii* leaves, photosynthetic activity that leads to a decrease in minimal fluorescence (F₀), maximal fluorescence (F_m), and maximum quantum yield of PSII (F_v/F_m), photochemical quenching q_P and non-photochemical quenching q_N, therefore, lower field capacity level severely affects *Aegilops tauschii*.

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Conflict of interests. The authors declare that they have no conflict of interests.

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