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Morphological and physiological variation in drought tolerance of wheat landraces originated from southeast Türkiye

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ABSTRACT

Drought stress, which is the most important abiotic stress factor affecting arable land in the world, causes serious crop losses. These crop losses reach up to 70% in some agricultural plants. Understanding the complex drought stress response is very important to develop a strategy against this form of stress. Although some progress has been achieved with the previous studies, the desired targets have not been reached up to now. Therefore, using resistant varieties in environmental conditions has become a widely used strategy in combating drought stress today. In this study, a total of 23 cultivars (16-landraces and 7 modern wheat cultivars) were used. The aim of this study was to reveal the drought tolerance degrees of 16 landraces by comparing them to 7 modern wheat cultivars. For this purpose, 23 cultivars were exposed to drought stress for seven days by withholding watering. After that, stem length, MDA and proline content of cultivars were determined and compared. According to our results, MDA and proline contents of sensitive modern cultivars were found to be high, while tolerant cultivars were found to be low. It has also been determined that some of the landraces exhibit a similar profile to the cultivars known to be tolerant. Among these cultivars, especially 88, 90 and 108 cultivars have low MDA and proline content under stress, which may indicate that these cultivars are potentially drought tolerant.

1. Introduction

The world's population has been dramatically increasing and is expected to hit 9 billion by 2050, food demand is also increasing accordingly. Wheat is one of the most cultivated agricultural products in the world and the demand for wheat is increasing day by day (Dixon et al. 2009). Drought has become one of the major challenges for crop productivity, as a result of altered precipitation patterns and insufficient rainfall (Toker et al. 2007; Mir et al. 2012). In agricultural areas, drought stress is often accompanied by high temperatures, which directly affects the kinetics of photosynthesis. The fact that photosynthesis kinetics are affected by environmental stresses causes significant decrease in crop yield (Tuberosa and Salvi 2006).

Plants have developed different mechanisms to cope with drought stress. Insufficient understanding of the physiological basis of drought stress has made it difficult to improve drought-tolerant crop varieties (Sinclair 2011). The seedling stage is critical to coping with drought stress. Physiological and morphological characteristics such as MDA content, proline, the water content in leaf, root and stem length provide important data to understand the drought tolerance state of wheat during the seedling stage (Polania et al. 2017; Mwenye et al. 2018; Chun et al. 2018; Dien et al. 2019).

Food security will rely on improved resistance cultivars to drought (Borlaug 2007; Tester and Langridge 2010; Chapman et al. 2012). Breeders need large variations in wheat that are responsible for drought resistance traits. Southeast Anatolia, part of the Fertile Crescent is the origin and site of genetic diversity for wheat. Durum and bread wheat landraces have been cultivated since ancient times in southeast Anatolia (Gökgöl 1939; Özkan et al. 2011). Wheat landraces have a trait that is resistant to biotic and abiotic stress (Nevo et al. 2002). Responses of wheat landraces under drought stress provide an opportunity to understand the mechanism of drought tolerance and gene discovery related to drought tolerance. Researchers depicted that landraces cultivated under biotic and abiotic stress conditions have better performance compared to modern wheat varieties in terms of vigor in the seedling stage, for some morphological parameters and grain yield (Aktaş et al. 2017; Aktaş et al. 2018).

Wheat landraces have the ability to adapt to arid and semiarid areas due to their genetic variation. Domestication of wheat took place in southeast Anatolia and that part of the Fertile Crescent has rich genetic diversity in terms of wheat landraces. Plant responses to environmental stresses can be studied by evaluation of traits at morphological, physiological and molecular levels (Praba et al. 2009). It is important to carry out such studies, as the characterisation of landraces that have a wide variation in terms of phenotypic and physiological defense mechanisms will contribute to the development of model plant varieties for plant breeders. The response of wheat landraces to drought stress provides unique information because of their huge genetic diversity. We aimed to determine the drought tolerance level of wheat landraces from the Fertile Crescent by comparing them to modern wheat cultivars under drought stress. For this purpose, malondialdehyde (MDA) and proline content, and shoot length were evaluated under drought stress in this study. 16 major landraces which originated from the Fertile Crescent plus 2 sensitive (Atik, Güney Yildizi) and 5 drought tolerant modern wheat cultivars were used. Genotypes used in the study may be useful material candidates for cultivation and for a better understanding of the mechanisms of drought tolerance for the seedling stage in wheat.

2. Materials and Methods

2.1. Plant material and stress treatment

In the present study, 23 different wheat genotypes (Table 1) were used. After the seeds were sterilized, they were planted in pots containing a 5:3:2 soil:peat:sand mixture. The field capacity of the prepared soil mixture was determined before sowing and the seeds were sown in this soil mixture which was irrigated up to the field capacity. Seeds irrigated once a week were grown for three weeks after germination, and half of the plants were exposed to drought stress for 7 days by withholding watering. At the end of this period, all the plants were harvested and placed in liquid nitrogen and stored in a deep freezer until the analysis.

2.2. Measurement of stem length

Stem length was measured from 5 plants of each variety. The main stem length was measured with a ruler. Average values were calculated for each variety.

Table 1. Wheat cultivars used in the study

2.3. Determination of MDA content

Lipid peroxidation was determined by measuring the malondialdehyde (MDA) level according to Ohkawa et al. (1979). Leaf tissue (0.25 g) was homogenised 2 mL (5%) trichloroacetic acid (TCA) solution. The homogenate was centrifuged for 10 min at 8000 rpm. After that, supernatant, thiobarbituric acid and TCA solutions were mixed in equal volumes in tubes and tubes were incubated at 96°C for 25 minutes. The tubes were placed in an ice bath to terminate the reaction and centrifuged at 6000 rpm for 5 min. The mixture was measured at 532 and 600 nm. The MDA content was calculated by using the extinction coefficient.

2.4. Determination of free proline content

Free proline content was determined according to Bates et al. (1973). Leaf tissue (0.5 g) was homogenised in 3% sulfosalicylic acid. The homogenate was centrifuged for 3 min at 3000 rpm and then the supernatant mixed well with acid ninhydrin and glacial acetic acid in equal volumes and incubated at 100°C for 1 hour. The reaction was terminated by adding cold toluene (4 mL) to the tubes. The toluene phase was evaporated and analysed by spectrophotometry at 520 nm. The proline level was determined from a standard curve.

2.5. Statistical analysis

Variance analyse was performed by using GENSTAT 12th (GENSTAT 2009) statistical program and the difference between the mean of the data.

Number/Cultivar	Origine	Traits	Drought Tolerance
8	Diyarbakır	High Plant Height	Tolerant
16	Diyarbakır	High Plant Height	Tolerant
25	Diyarbakır	High Plant Height	Tolerant
29	Diyarbakır	High Plant Height	Tolerant
Sorgül	Diyarbakır	High Plant Height	Tolerant
30	Adıyaman	High Plant Height	Tolerant
46	Adıyaman	High Plant Height	Tolerant
58	Adıyaman	High Plant Height	Tolerant
70	Adıyaman	High Plant Height	Tolerant
73	Mardin	High Plant Height	Tolerant
85	Mardin	High Plant Height	Tolerant
87	Mardin	High Plant Height	Tolerant
88	Mardin	High Plant Height	Tolerant
90	Şırnak	High Plant Height	Tolerant
108	Şırnak	High Plant Height	Tolerant
109	Şırnak	High Plant Height	Tolerant
Atik	Private Company	Medium Plant Height	Sensitive
Güney Yıldızı	Research Inst.	Medium Plant Height	Sensitive
Firat-93	Research Inst.	Short Plant Height	Tolerant
Aydın-93	Research Inst.	High Plant Height	Tolerant
Sümerli	Research Inst.	Medium Plant Height	Medium Sensitive
Sarıçanak	Research Inst.	Medium Plant Height	Medium Sensitive
Svevo	Italy	High Plant Height	Tolerant

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3. Results and Discussion

Drought stress increased the Malondialdehyde (MDA) content in all wheat cultivars used in this study. The five cultivars whose MDA content increased the most under drought stress were 73, Atik, 8, 46 and 30, respectively, while those with the least increase were Firat, Svevo, Sariçanak, 88 and Sümerli (Figure 1). MDA, the end product of lipid peroxidation, is one of the important indicators of oxidative stress. The MDA content reflect the degree of damage under adverse conditions (Yang and Deng 2015). High MDA content is known to be an indicator of membrane damage caused by oxidative stress (Gawel et al. 2004; Morales and Munné-Bosch, 2019).

Mehmood et al. (2020) determined a good correlation between MDA and H_2O_2 level. This finding may indicate that the increase in ROS that occurs under stress conditions causes membrane damage and that the MDA content increases as an indicator of this situation. In many studies, it has been shown by various researchers that the MDA content increases with drought stress in different plants (Pandey et al. 2010; Yildizli et al. 2018; Khaleghi et al. 2019). However, it has been reported that cultivars with lower MDA content under drought (Ma et al. 2015; Mihaljević et al. 2021) and salt (Kiran et al. 2019) conditions have higher tolerance to these stresses. Based on this information in the literature, it can be said that the Fırat, Svevo, Sarıçanak, 88 and Sümerli varieties used in our study are more tolerant in terms of MDA content under drought conditions. In many studies comparing cultivars with known tolerance under environmental stress conditions, it has been shown that tolerant cultivars have a much stronger antioxidant system and, accordingly, suppress oxidative damage caused by stress more rapidly and more strongly than sensitive cultivars (Sultan et al. 2012; Amoah et al. 2019). This information provides clues that the antioxidant systems of these five cultivars may be stronger than other cultivars. The data obtained may shed light on the molecular studies planned to be conducted on this subject.

Proline content of all cultivars also increased under drought stress. The five cultivars with the highest increase in Proline content were 87, 46, Atik, 25 and 73, while those with the least increase were 70, Sarıçanak, 90, Fırat and 109 under drought stress (Figure 2). When the proline results are examined, it can be seen that the proline contents of Fırat and Sarıçanak varieties, which have low MDA content under drought conditions, are also lower than other varieties. The increase in Proline content under



Figure 1. MDA content in leaves of wheat plants under drought and control conditions. Data are showed as mean \pm SE of three independent biological replicates. Different letters indicate significant differences between groups P < 0.05 (Drought LSD 0.175, Control LSD 0.110).



Figure 2. Proline content in leaves of wheat plants under drought and control conditions. Data are showed as mean \pm SE of three independent biological replicates. Different letters indicate significant differences between groups *P* < 0.05 (Drought LSD 0.173, Control LSD 0.011).

various environmental stresses has been shown in many studies (Johari-Pireivatlou 2010; Ahmed et al. 2017; Chun et al. 2018). However, when studies using cultivars with different tolerances are examined, it can be seen that there are different results in the literature in terms of proline content under stress. In some of these studies, proline content was found to be higher in tolerant cultivars (Solanki and Sarangi 2014; Mwadzingeni et al. 2016; Dien et al. 2019), while in some others it was found to be higher in sensitive cultivars (Duangpan et al. 2007; Ergen et al. 2009; Marček et al. 2019). This situation may occur depending on the species or varieties. Marček et al. (2019) emphasized that proline should not be shown as a specific drought tolerance indicator for wheat varieties. The best-known feature of proline is that it is a good osmoprotectant. It is thought that the amount of waterrelated stress increases and contributes to the protection of water under stress. It is also reported to have antioxidant properties (Arteaga et al. 2020). In our study, the high proline content of cultivars with high MDA content (Atik, 46 and 73) may be an indication of high oxidative stress in these cultivars. The low proline content of cultivars with low MDA content (Firat, Sarıçanak, Svevo and Sümerli) may be an indication that these cultivars are exposed to a lower oxidative stress compared to cultivars with high proline content. However, it is thought that this stress response can be better understood by investigating the changes in the activities and amounts of antioxidant system elements under drought conditions. Isoenzyme analysis may be the best indicator to show antioxidant responses of these cultivars under drought stress.

Drought stress decreased stem lengths (Figure 3) in all cultivars. However, these reductions were not statistically significant between cultivars in this study. When the literature is examined, it is seen that there are many studies showing that drought stress reduces stem lengths (Rauf et al. 2007; Polania et al. 2017; Mwenye et al. 2018). Studies show that different results can be obtained depending on the onset time of stress and the application period.

4. Conclusion

When all the results were evaluated together, it was seen that both MDA and proline content of the Atik variety, which is known to be sensitive, were high, and stem length was also suppressed by drought stress. On the contrary, other cultivars known to be tolerant to drought were found to have particularly low MDA and proline content. However, it has been determined that there are local cultivars that exhibit a profile similar to the cultivars known to be tolerant. Among these cultivars, especially 88, 90 and 108 cultivars have low MDA and proline content under stress, which may indicate that these cultivars are potentially drought tolerant.



Figure 3. Shoot length of wheat plants under drought stress.

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