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Physiological parameters for evaluating drought tolerance in durum wheat varieties grown in the fields in Syria

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ABSTRACT

Wheat is necessary to ensure a stable food style everywhere in the world. During the past few years wheat production in Syria fluctuated enormously, one important reason was climate change and more precisely drought. A physiological approach can complement empirical breeding and can enhance the rate of yield improvement under drought condition by identification important physiological traits associated with drought tolerance. This investigation was carried out under field conditions in the 1st and 2nd settlement zone in Syria (under water limited condition) with some drought tolerant and susceptible durum wheat varieties. Membrane stability index, relative water content, chlorophyll content and chlorophyll fluorescence were measured at vegetative, anthesis and post anthesis stage. Our findings showed that all varieties were superior in all physiological parameters under study as grown in the 1st settlement zone compared with the 2nd zone at all growth stages. More reduction in all physiological characters was recorded in drought susceptible variety in bohouth, which showed also more reduction in yield and yield components. Drought tolerant variety in douma were more stable and showed better adaptation and comparatively better physiological performance and higher yield under drought stress condition. This study lays emphasis on the importance of these traits particularly for breeder for improving drought stress tolerance.

Key words: Drought, wheat, chlorophyll, membrane stability, relative water content, Fv/Fm

1. INTRODUCTION

Wheat (*Triticum aestivum L.)* is a staple food for more than 35% of the worlds population and it is also the first grain crop in most of the developing countries (1). In arid/semi-arid farming regions the major constraint limiting wheat production is inadequate rainfall reducing average yield up to 50% and over (2). Drought is the most common environmental stress affecting about 32% of the 99 million hectares which is under wheat cultivation in developing countries and at least 60 million hectares under wheat cultivation in developed countries (3). Water stress reduces plant growth and manifests several morphological, physiological and biochemical alterations leading to massive loss in yield (4) Water stress tolerance is seen in almost all plant species but its extent varies from species to species and even within species due to differences in phonological, morphological, biochemical, physiological and molecular adaptive mechanisms (5). Drought tolerance does not exist as a unique and easily quantifiable plant attribute, it is a complex physiological, morphological and molecular character connected with relative water content (RWC), relative water loss (RWL), chlorophyll fluorescence, cell membrane stability (CMS) (6). According to the previous studies, there is a link between various physiological responses of crop plants to drought and their tolerance mechanisms such as high relative water content and water potential (7) membrane stability (8) and pigment content stability under stress (9). The high RWC and low excised leaf water loss (RWL) have been suggested as important indicators of water status (9, 10). Al Meselmani et al., (11) reported that membrane stability index is the best indicator for screening wheat varieties for drought stress particularly at advance stages of plant growth. Total chlorophyll content and the Chl a/b ratio were found to reduce under water stress conditions. A decrease in this index was faster in drought sensitive than in drought tolerant genotypes (12). According to (13) chlorophyll, fluorescence measurement appears very promising for screening of genotypes for improved tolerance to drought and high temperature. The improvement of tolerance to drought has been a principal goal of the majority of breeding programmes for a long time, as a water deficit in certain stages of wheat growth is common for many wheat growing regions of the world (14). To overcome the low response to direct selection for yield under drought conditions, substantial efforts have been targeted on manipulation of physiological traits influencing drought resistance through an escape, avoidance and tolerance mechanism (15). The aim of this study was to evaluate the physiological behavior of wheat varieties differ in their drought tolerance at varies stages of plant growth under drought stress condition.

2. MATERIALS AND METHODS

2.1. Plant materials and growth conditions

Three drought tolerant, moderately tolerant and susceptible durum wheat varieties viz., douma3, sham9 and bohouth9 were used in this study. Seeds were obtained from Crop Research Directorate, General Commission for Scientific Agricultural Research, Syria, and sown under rain fed conditions in the field on 20th Nov. 2011 in the first settlement zone (Jellen Research Station, annual rainfall 400mm) and second settlement zone ((Izra Research Station, annual rainfall 291mm)). Crops were sown at an adjusted rate of 300 viable seeds/m^2 in three replications. Normal agronomic practices were performed and relevant metrological parameters were obtained from the observatory at each research station and daily minimum and maximum temperature and rainfall were recorded. Chlorophyll content (chl), membrane stability index (MSI), relative water content (RWC), chlorophyll fluorescence Fv/Fm were estimated on the first fully expanded leaf (third from top) at vegetative stage and flag leaf at anthesis and post anthesis stage.

2.2. Chlorophyll Content Estimation

The chlorophyll meter (SPAD meter) was used for chlorophyll estimation. The meter makes instantaneous and non-destructive readings on a plant based on the quantification of light intensity (peak wavelength: approximately 650 nm: red LED) absorbed by the tissue sample. A second peak (peak wavelength: approximately 940 nm: infrared LED) is emitted simultaneously with red LED to compensate the leaf thickness.

2.3. Membrane stability index

Membrane stability index was determined by recording the electrical conductivity of leaf leachates in double distilled water at 40 and 100 ^{o}C (16). Leaf samples (0.1 g) were cut into discs of uniform size and taken in test tubes containing 10 ml of double distilled water in two sets. One set was kept at 40 ^{o} C for 30 min. and another set at 100 ^{o} C in boiling water bath for 15 minutes and their respective electric conductivities were measured by Conductivity meter.

2.4. Relative Water Content

Relative water content was determined by the method described by Barr and Weatherly. 100 mg leaf material was taken and kept in double distilled water in a petri dish for two hours to make the leaf tissue turgid. The turgid weights of the leaf materials were taken after carefully soaking the tissues between the two filter papers. Subsequently this leaf material was kept in a butter paper bag and dried in oven at 65 ^{o} C for 24 hours and their dry weights were recorded and RWC was calculated.

2.5. Chlorophyll fluorescence

For the estimation, the polyphasic rise of fluorescence transients of intact leaves of non-stressed and water stressed plants were measured by a Plant Efficiency Analyzer (PEA, Handsatech Instruments Ltd., King's Lynn, UK) (17). On mid Jun plants harvested from m^2 and used for recording biological and grain yield, number of tillers/m^2, seed number per ear, 1000 grain weight. Data analyzed statistically and analysis of variance (ANOVA) for factorial design at each stage was work out using CoStat6.311 Cohort software and LSD values was measured.

3. RESULTS AND DISCUSSION

In this study some physiological parameters were examined in the field under rain fed condition of the most important tolerant and susceptible durum wheat varieties grown in Syria. Total rainfall was well distributed up to anthesis stage (March/2012) indicated that enough water was available for fast and rapid emergence of seeds. The total amount of the rainfall in the 2nd zone was 19% less than that in the 1st zone i.e., 292.9mm and 358.8mm respectively (Figure 1). Only 42.8mm were received at the most sensitive stage (anthesis and grain filling stage) in the 2nd zone compared with 89.5mm in the 1st zone, which may have adverse effect on growth and productivity particularly in the susceptible varieties. In general terminal drought stress experienced by the varieties in both zones, however the drought was more sever in case of 2nd settlement zone and enough water was available in the soil for the varieties in the 1st zone for good tilling and spike emergence.

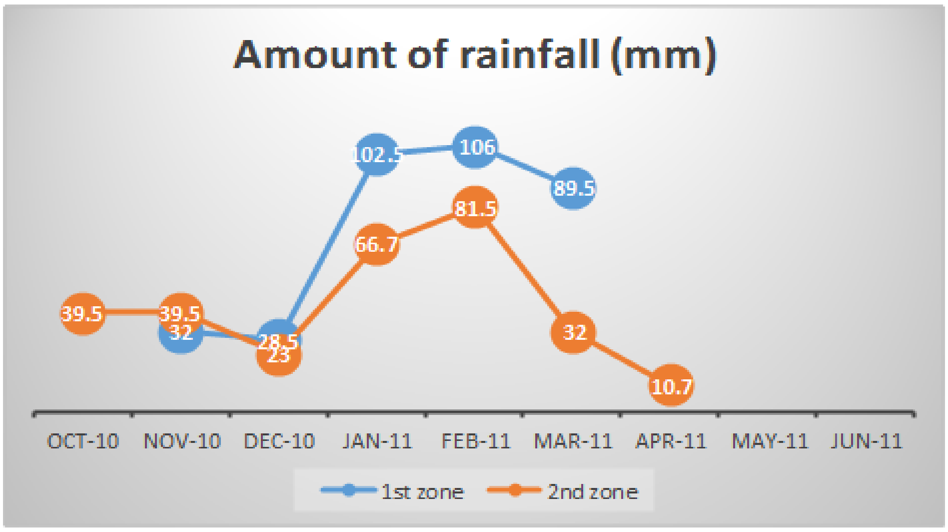


Figure 1. Total amount of rainfall (mm) in the 1st and 2nd settlement zone during the growing season

It is well documented that maintenance of cell membrane integrity and stability under water deficit conditions is major component of drought tolerance in plants (18) Membrane stability is a widely used criterion to assess crop drought tolerance, since water stress caused water loss from plant tissues which seriously impairs both membrane structure and function (19). Our results indicated significant differences in MSI between all wheat varieties at all growth stages in both zones; however, MSI values were higher in the 1st zone compared to the 2nd zone. Drought susceptible wheat variety bohouth9 showed highest MSI values at vegetative, anthesis and post anthesis stage in the 1st zone i.e., 85.4, 78.1 and 70.3 respectively, while in the 2nd settlement zone drought tolerant variety doum3 were more superior and showed highest MSI values at all growth stages i.e., 78.3, 75.9 and 65.6 respectively (Figure 2 a, b and c). Drought tolerance variety showed 2, 3 and 5% reduction in MSI at vegetative, anthesis and post anthesis stage respectively in the 2nd zone compared with the 1st zone, while drought susceptible variety bohouth9 showed 14, 10 and 12 % reduction respectively at various growth stages. The results from electrolyte leakage measurements in our experiment showed that membrane integrity was conserved for tolerant compared to susceptible varieties, this is in agreement with the conclusion of Al meselmani et al., (11) that electrolyte leakage was correlated with drought tolerance.

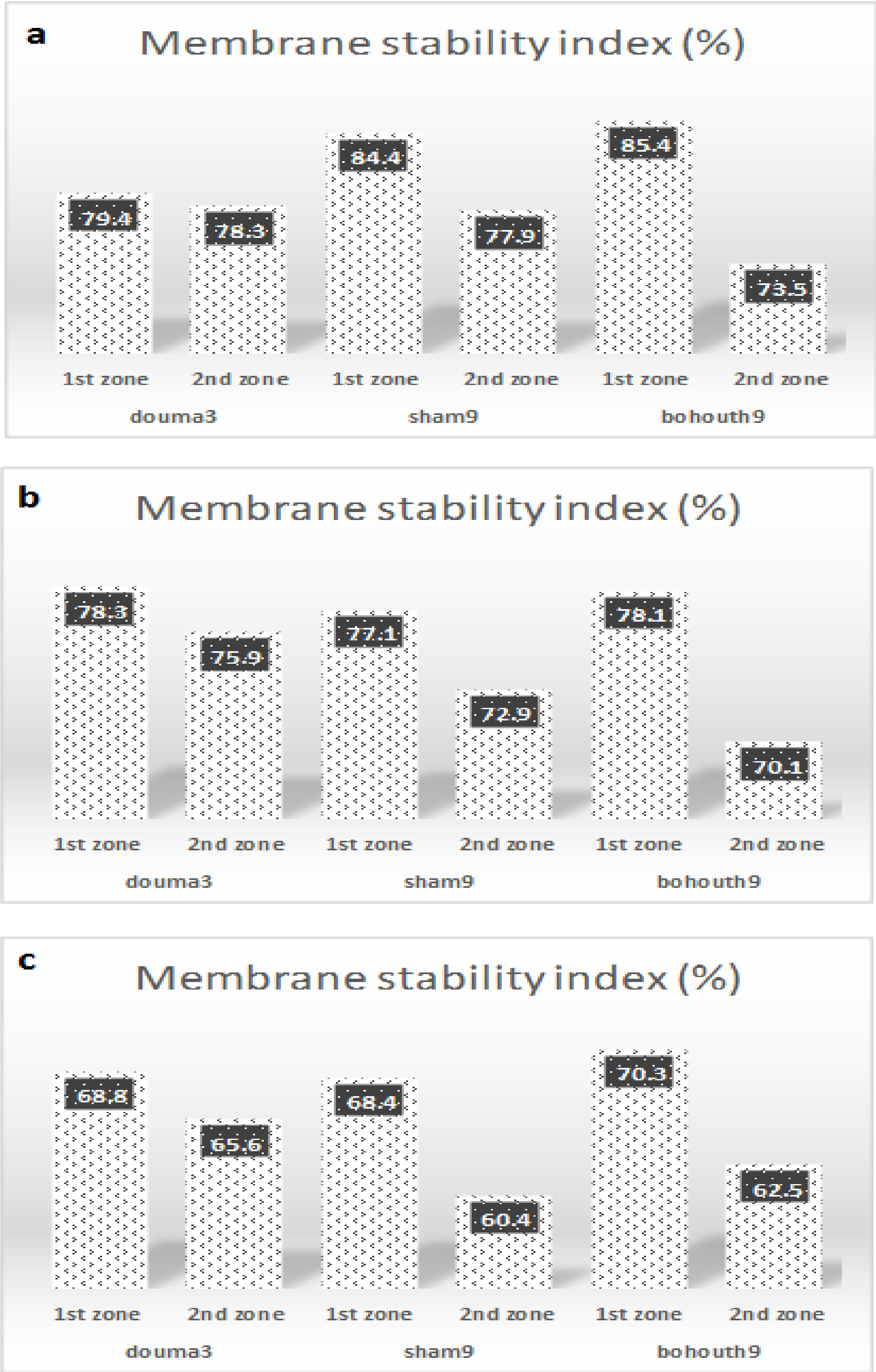
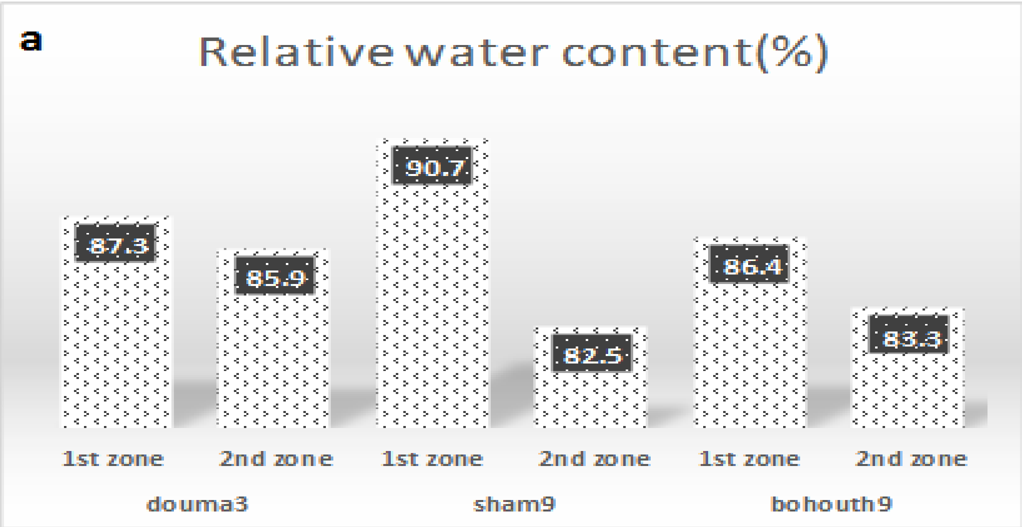
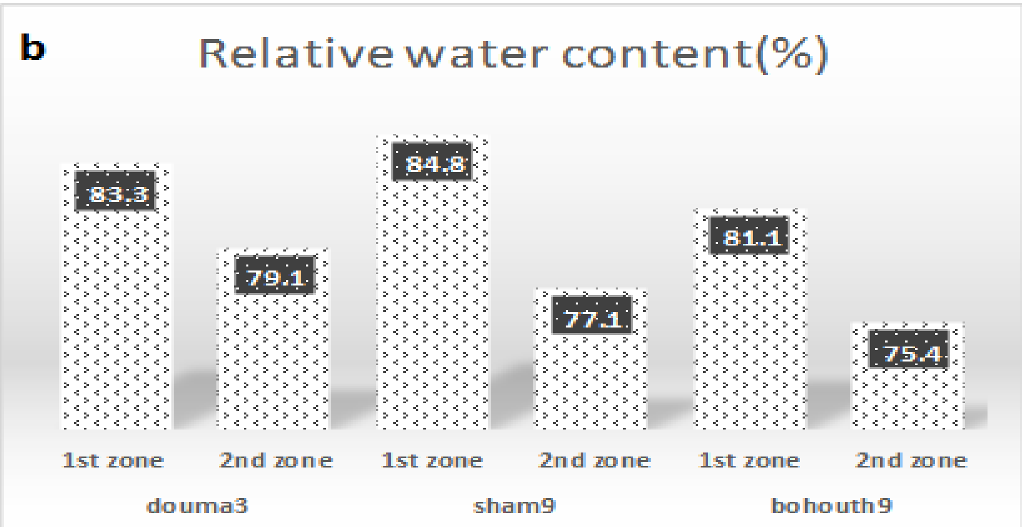


Figure 2. Membrane stability index (%) of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones at vegetative (a) and anthesis (b) and post anthesis (c) stages, LSD values at vegetative: 0.947, anthesis: 0.757 and post anthesis stage 0.863 respectively

In general highest RWC were recorded at vegetative stage in all variety in both zones, and all variety grown in the 1st zone were more superior compared with that grown in the 2nd zone. Moderately drought tolerant variety sham9 were superior at vegetative and anthesi stage and drought susceptible variety bohouth9 at post anthesis stage i.e., 90.7, 84.8 and 67.8 in the 1st zone, while in the 2nd zone douma3 were more superior at all growth stages i.e., 85.9, 79.1 and 64.2 respectively (Figure 3 a, b and c). Lowest values for RWC were recorded in the 2nd zone in drought susceptible variety bohouth9 at all growth stages i.e., 83.3, 75.4 and 58.1 respectively. Drought susceptible variety bohouth9 showed highest percent of reduction in RWC at all growth stages in the 2nd compared to 1st zone i.e., 4, 7 and 14% respectively. A decrease in the RWC in response to drought stress has been noted in wide variety of plants (20). It is reported that high relative water content is a resistant mechanism to drought, and that high RWC is the result of more osmotic regulation or less elasticity of tissue cell wall (21). RWC of the leaves is very responsive to drought stress and has been shown to correlate with drought tolerance (22). Al meselmani et al., (23) reported that RWC indicates the water status of the cells and has significant association with yield and stress tolerance.





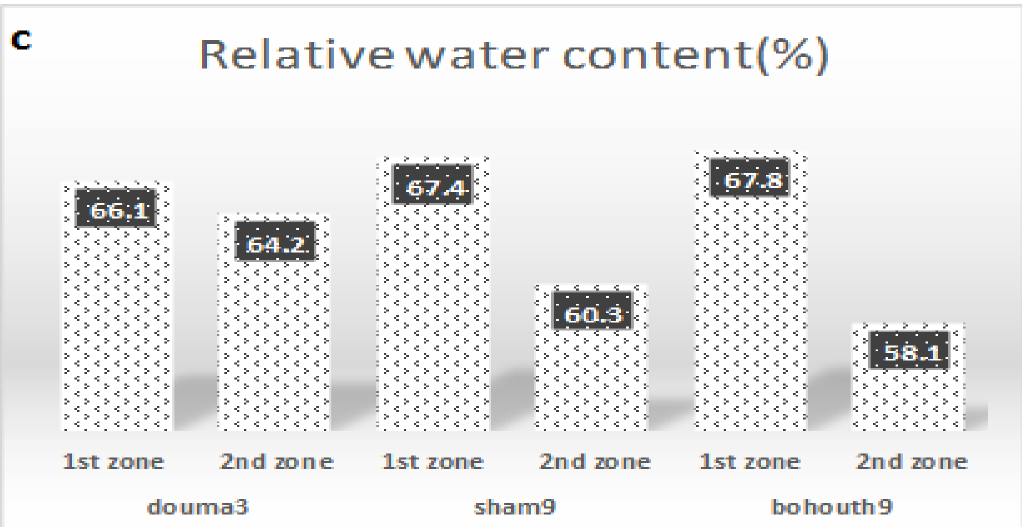


Figure 3. Relative water content (%) of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones at vegetative (a) and anthesis (b) and post anthesis (c) stages, LSD values at vegetative:1.2, anthesis:0.9 and post anthesis stage: 0.9 respectively

With regard to chlorophyll content highest values were recorded at vegetative stage in both zones in all varieties. In the 1st zone highest values were recorded in sham9 at all growth stages i.e., 65.5, 58.1 and 56.4 respectively. While in the 2nd zone highest values were recorded in douma3 i.e., 50.2, 45.8 and 43.2 respectively (Figure 4 a, b and c). However, drought susceptible variety bohouth9 showed highest percent of reduction in chlorophyll content in the 2nd zone compared with the 1st zone at all growth stages i.e., 19, 23 and 26% respectively. Chlorophyll concentration has been known as an index for evaluation of source (24), therefore decrease of chlorophyll can be considered as a non-stomata limiting factor under drought stress conditions. Chlorophyll content is one of the major factors affecting photosynthetic capacity. Reduction or no-change in chlorophyll content of plant under drought stress has been observed in different plant species and its intensity depends on stress rate and duration (25). This is supported by the findings of (26) noting the inhibition of chlorophyll synthesis and inability of sensitive wheat to withstand water deficit. Chlorophyll maintenance is essential for photosynthesis under drought stress and higher chlorophyll content and lower percent of reduction under stress in tolerant genotype of wheat has also been reported (16, 27, 28).

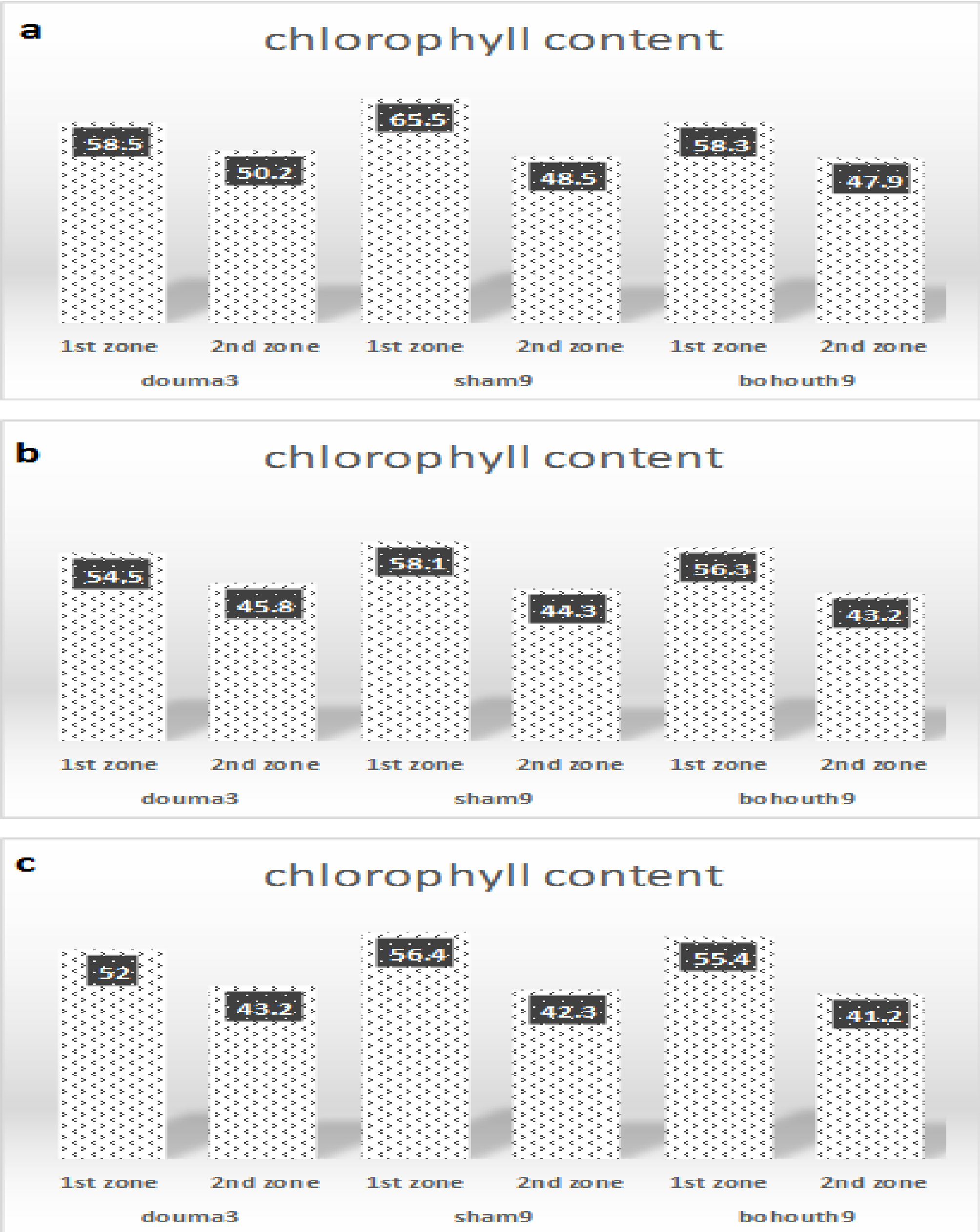


Figure 4. Chlorophyll content (SPAD reading) of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones at vegetative (a) and anthesis (b) and post anthesis (c) stages, LSD values at vegetative:1.1, anthesis:2.3 and post anthesis stage: 1.2 respectively

Use of a chlorophyll fluorescence technique as a tool to investigate drought tolerance of different wheat genotypes has been reported by (23). According to (29) in the assessment of effects caused by high temperature or water deficit on the photosynthetic activity, chlorophyll fluorescence may be a safer indicator than net photosynthesis rate, because it is a practical and precise method. Our results showed that chlorophyll fluorescence readings decreases as plant advances in age and highest values were recorded in the 1st zone in all varieties compared to the 2nd zone. Highest values were recorded in douma3 at vegetative and anthesis stage and in sham9 at post anthesis stage i.e., 0.83, 0.79 and 0.703 respectively. While in the 2nd zone highest values were recorded in douma3 at all growth stages i.e., 0.82, 0.73 and 0.663 respectively (Figure 5 a, b and c). Highest reduction in chlorophyll fluorescence readings were recorded in bohouth9 in the 2nd zone compared to 1st zone at all growth stages i.e., 5, 6 and 10%. Dark-adapted values of *F*v/*F*m reflect the potential quantum efficiency of PSII and are used as a sensitive indicator of photosynthetic performance, with optimal values of around 0.832 measured from most plant species (30) Flagella et al., (31) and Al meselmani et al.,(23) also reported that drought tolerant cultivars showed a smaller decrease in photosynthetic efficiency (*F*v/*F*m ratios).

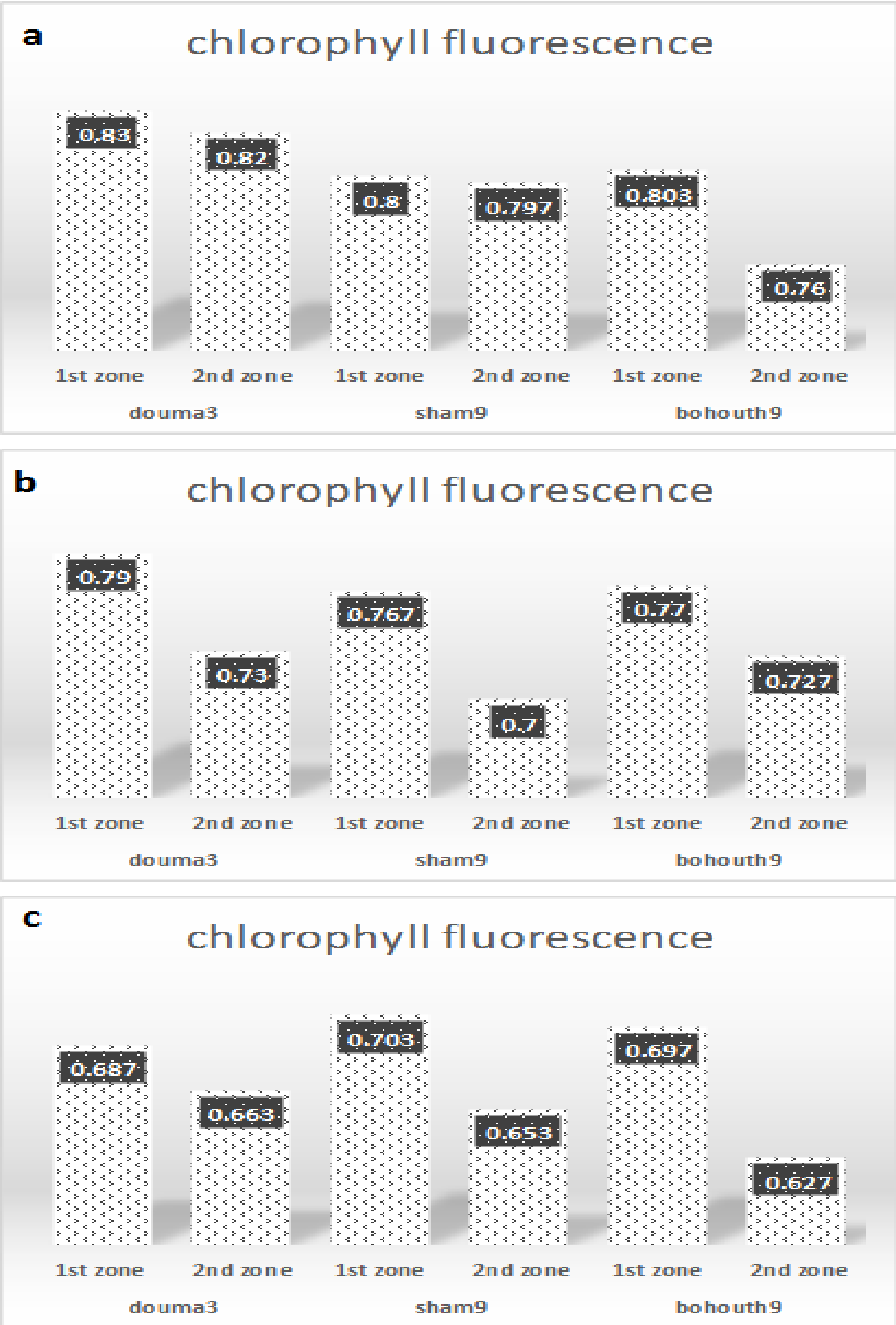


Figure 5. Chlorophyll fluorescence (*Fv/F*m ratios) of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones at vegetative (a) and anthesis (b) and post anthesis (c) stages, LSD values at vegetative: 0.011, anthesis: 0.013 and post anthesis stage: 0.0103 respectively

The negative effect of drought stress on yield and yield performance has been well documented as a major problem in many developing countries of the world (32). Under 1st zone, yield and yield components in all varieties were much better and showed higher values compared with that in the 2nd zone. Significant differences in biological yield were recorded between varieties in both zones and biological yield decreased significantly in the 2nd compared with the 1st zone in all varieties under study. Drought susceptible variety bohouth9 showed highest value in the 1st zone i.e., 1528g, while in the 2nd zone highest value were recorded in douma3 i.e., 844g (Figure 6). The percentage of reduction in biological yield in the 2nd compared to 1st zone was 39, 50 and 52% in douma3, sham9 and bohouth9 respectively. Ashraf, (33) indicated that plant produces their maximum biomass under adequate water supply, whereas moisture stress causes a marker decrease in plant biomass production.

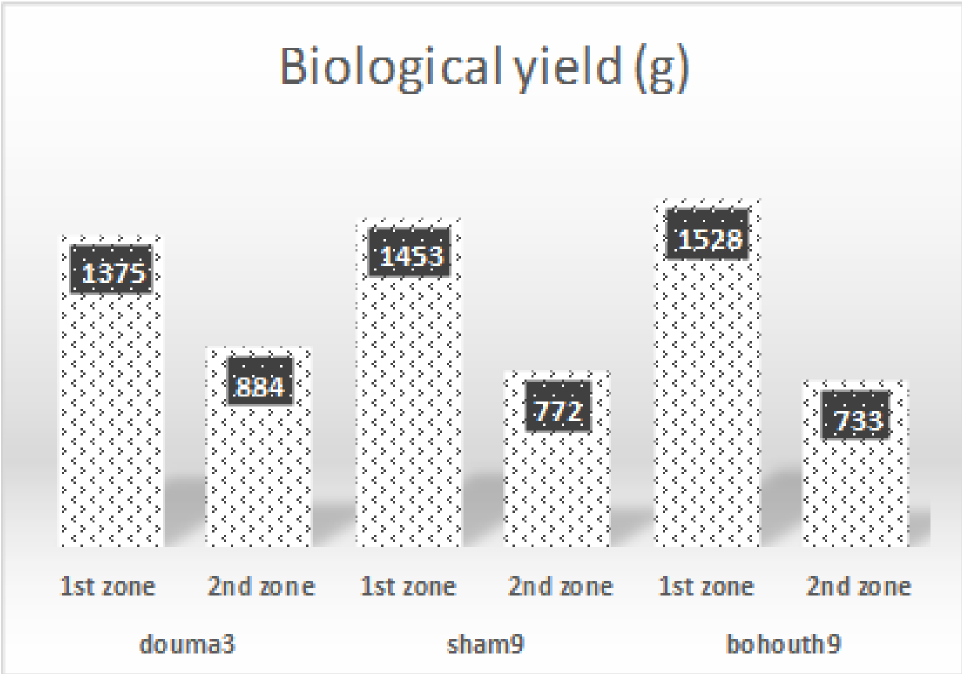


Figure 6. Biological yield (g) of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones, LSD value: 20.7

In case of grain yield highest value were recorded in bohouth9 in the 1st zone i.e., 701g, while douma3 showed highest value in the 2nd zone i.e., 437g and highest reduction in grain yield in the 2nd compared with the 1st zone were recorded in bohouth 9 i.e., 50% (Figure 7).

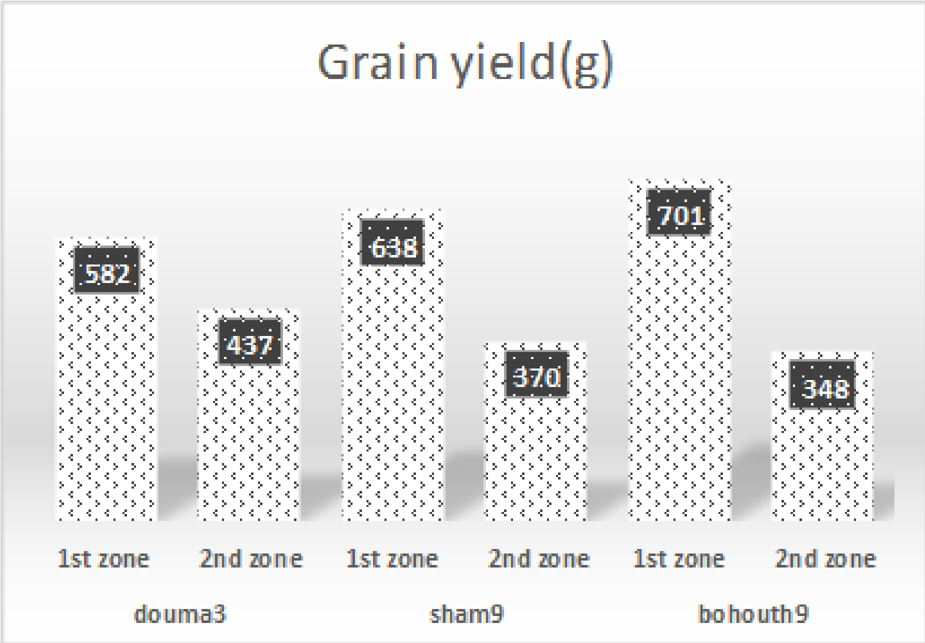


Figure 7. Grain yield (g) of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones, LSD value: 21.8

Bohouth9 showed highest number of tiller/m^2 in the 1st zone i.e., 333 and douma3 were superior in the 2nd zone i.e., 217; however, bohouth9 showed highest reduction in tiller number/m^2 in the 2nd compared to the 1st zone i.e., 30% (Figure 8).

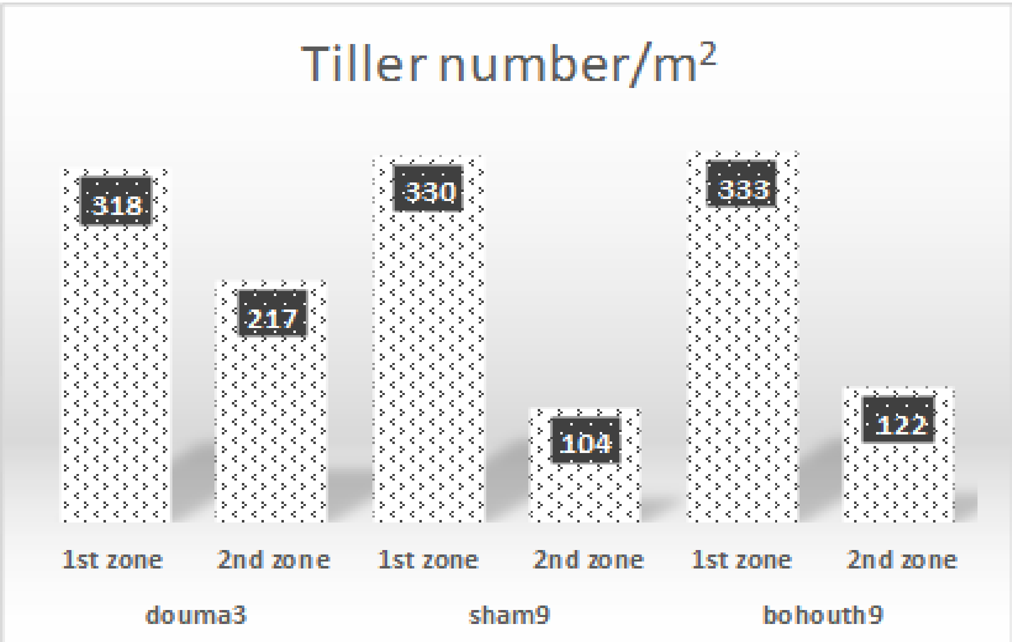


Figure 8. Tiller number/m^2 of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones, LSD value: 6.6

With regard to grain number per ear highest value were recorded in douma3 in both zones i.e., 51.4 and 38.7 respectively, however grain number per ear decreased in the 2nd zone compared to the 1`st zone in all variety and bohouth9 detained lowest number of grain per year in both zones i.e., 47 and 27.6 respectively (Figure 9).

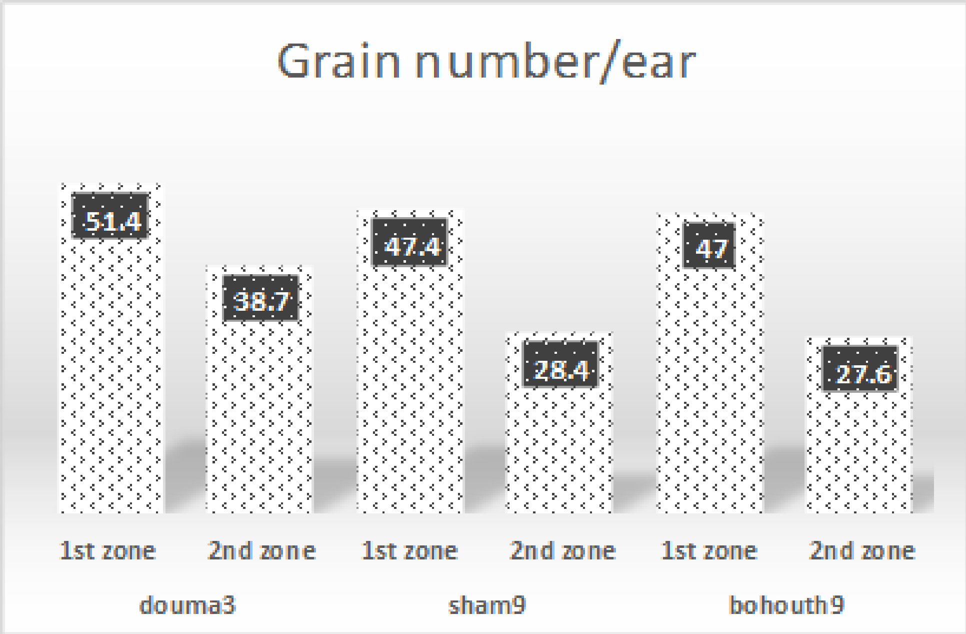


Figure 9. Grain number/ear of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones, LSD value: 0.68

Highest 1000 grain weight were recorded in bohouth9 i.e., 55.4 in the 1st zone, while in the 2nd zone douma3 showed highest value i.e., 41.1 (Figure 10).

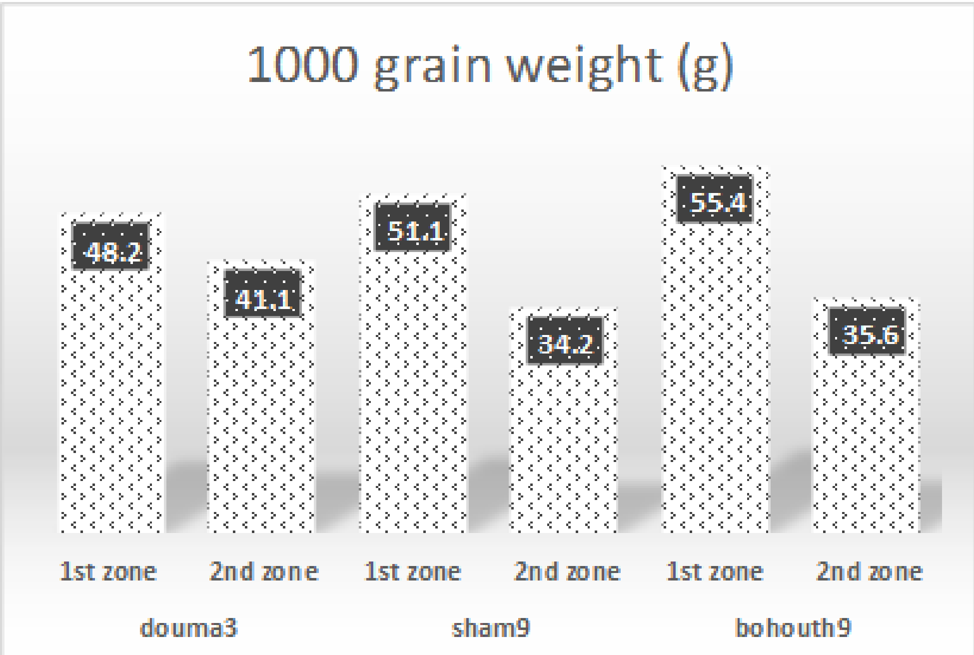


Figure 10. 1000 grain weight (g) of drought tolerant, moderately tolerant and susceptible wheat varieties in 1st and 2nd settlement zones, LSD value: 0.487

Drought stress reduced the number of gain/spike and grain yield (34), while kernel weight is negatively influenced by high temperatures and drought during ripening (35, 36) and Qadir et al., (37) who observed that 1000 grain weight of wheat was reduced mainly due to increasing water stress. The decrease in 1000 grains weight may be due to disturbed nutrient uptake efficiency and photosynthetic translocation within the plant (38) that produced shriveled grains due to hastened maturity. This is likely due to the shortage of moistures which forces plant to complete its grain formation in relatively lesser time (39). Under water stress, the decrease in seed set and grain growth in wheat has been reported by several workers (40-43).

4. CONCLUSION

Among all the factors limiting wheat productivity, drought remains the single most important factor affecting the world security and sustainability in agricultural production. For improving yield under dry land conditions, the development of new wheat cultivars with high grain yield potential through identifying drought tolerance mechanism is of great significance. Measurements of different physiological processes of plant response to drought is an important information on the reactions of the plant intended to remove or to reduce the harmful effects of water deficit and targeting of specific physiological characters that is associated with maintained better yield may be more effective than direct selection for yield. Our findings indicated that all studied parameters have important role in drought tolerance and could be used effectively for selection drought tolerant varieties and lines particularly at reproductive stage.

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AUTHORS CONTRIBUTION

This work was carried out in collaboration among all authors.

CONFLICT OF INTEREST

The authors declared no potential conflicts of interests with respect to the authorship and/or publication of this article.

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