ASSESSMENT OF SPRING WHEAT ON PHYSIO-MORPHIC ATTRIBUTES UNDER WATER DEFICIT CONDITIONS AT SEEDLING STAGE AND MATURITY

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ABSTRACT

Wheat is one of the pivotal cereal due to its nutritious value and used all over the world as food for human. Drought is effecting grain yield up to 30-40% in wheat. The initial move towards improvement of drought tolerance in wheat is to screen existing germplasm. The experiment conducted in in wheat Research Institute Faisalabad in 2020-2021 and 2021-2022. One hundred wheat genotypes assessed at seedling stage on physio-morphic traits. Wheat line 9860 had maintained root/shoot ratio, relative water contents and excised leaf water loss under 50% and 100% field capacity. Seven best performing wheat genotypes further evaluated for drought tolerance and grain yield in field with regular irrigation and drought. The physiological traits included excised leaf water loss, cell membrane thermostability, photosynthetic rate, stomatal conductance and chlorophyll contents recorded at anthesis and yield related traits days to flowering, days to maturity, plant height, number of grains per plant, 1000-grain weight and grain yield per plant recorded at maturity indicated wheat genotype 9860 performed better in water deficit conditions and found drought tolerant. The selection criteria developed for identifying drought tolerant wheat genotypes is very efficient.

INTRODUCTION

Wheat (*Tritium aestivum* L.) is the second most important crop in the world after maize (FAO, 2023). Similarly, it grows on vast areas in Pakistan, including irrigated and non-irrigated areas (Anonymous, 2022-2023). Due to its nutritious worth, it is preferably utilized in daily food in the form of bread, Chapati, crackers, confectioneries, baked products, pasta, spaghetti etc. Wheat, based on use, has different classes for example, soft wheat is used in making cakes, pastries, and other baked goods whereas hard wheat is used for making bread. Different categories of flour are used for special types of food item, for instance all-purpose flour, whole wheat flour, semolina flour, durum flour, self-rising flour, graham flour and bread flour.

Wheat is playing a major role in the economy of Pakistan and has a strong share of GDP (Anonymous 2022-2023). The average yield of wheat in Pakistan is 2600 kg per hectare although progressive farmers are producing 8000 kg per hectare (Anonymous 2022-23). The constraints responsible for low production include poor seed quality, improper sowing method and time, climate change, poor soil and crop management. One of the major constraints of low yield is water deficit conditions or non availability of water at critical stages. Wheat yield was reduced to 50 to 90% of its irrigated potential due to water deficit (Nezhadahmadi *et al*., 2013). The situation of water is worse in Pakistan and is becoming more severe with the passage of time. The World Bank and Asian Development Bank had declared Pakistan, a water deficit country. The per capita water is below 1000 cubic meters per year in Pakistan, and the situation will be more insecure in the future. The water deficit tolerance is the most essential mechanisms of wheat yield, reliability, and its improvement is a leading concern to scientists (Nezhadahmadi *et al*., 2013).

The morphological, physiological and yield related traits have significant importance for making a selection in wheat germplasm. The genotypes were selected on the base of morphological and physiological attributes on seedling stage. The lines which performed well in water deficit condition at seedling stage were further assessed at anthesis and maturity on physiological and yield related traits.

MATERIALS AND METHODS

Seedling Experiment

The experiment conducted in in Wheat Research Institute Faisalabad in 2020-2021 and 2021-2022. One hundred genotypes planted in two-factor factorial experimental design plan with three replications and five plants for each replication kept up in L=30cm W=15 cm sand filled polythene bags on 28th of October 2020. Single plant maintained in each polythene bag. The temperature was recorded 28ºC. Pressure membrane apparatus used to appraise the field capacity of the sand. Plants under treatment 1 were under 100% field capacity watered at full limit 30 ml and plant under treatment 2 were under 50% field capacity watered at half limit 15 ml after period one day. Water withheld after 25 days and following two days data recorded for root length, shoot length, root/shoot ratio, fresh shoot weight, dry shoot weight, wilt weight, relative water contents and excised leaf water loss. Root shoot ratio was determined dividing root length by shoot length. Relative water contents were determined utilizing following formula:

RWC = [(Fresh shoot weight–Dry shoot weight) / (Turgid weight–Dry shoot weight)] × 100

Excised leaf water loss calculated using the following formula

ELWL = (Fresh weight – wilted weight) / Dry weight

Field Experiment

In the subsequent year, 2021-2022 selected lines at seedling stage additionally assessed in field on base of physiological and yield related parameters. Ten wheat genotypes selected. Trial conducted in tunnel with two variable water conditions. Split plot design utilized with three replications in RCBD. The distance between the plants was and rows was 22 cm. Main plot was for irrigation factor while genotypes were in subplot. The plot size was 1.8 m^2 for each genotype. Water withheld in drought plot by controlling regular irrigation and rain water by covering tunnel with polythene during precipitation. In normal irrigation treatment, plants consistently irrigated and tunnel kept without cover of polythene. The physiological parameters assessed at anthesis stage. In the field conditions test taken from five plants for each replication and methodology received to record ELWL, which had utilized at seedling stage. Cell membrane thermostability estimated following the technique proposed by

Saadalla et al., [9] and altered by Petcu and Ciuca, [10]. The completely extended leaf tests taken from the chose plants during the anthesis stage as leaf discs of 0.5mm measurement in falcon tubes with six discs in each tube. Tubes were loaded up with 20 ml deionized water and kept at room temperature for two hours and subsequent to shaking an initial conductance (C1) reading was noted. At that point, tests were autoclaved at 121°C for 15 minutes and kept at room temperature for overnight to take second conductance (C2) perusing.

Membrane injury index calculated by formula:

$$\% \text{ injury} = (C1/C2) \times 100$$

Cell membrane thermo-stability calculated using the formula:

$$CMT = 1 - \% \text{ injury} = 1 - (C1/C2) \times 100$$

Where C1 and C2 are the first and the second reading of conductance respectively. Photosynthetic rate (μmol m⁻2 s⁻1), stomatal conductance (μmol m⁻2 s⁻1) and CO_2 (μmol s⁻1) assimilation recorded at anthesis, during a sunny day data recorded at 11:00 am using CIRAS-3 Portable Photosystem II (USA). Chlorophyll contents recorded from guarded plants for all treatments in three replications using a SPAD-502 plus chlorophyll meter (nmol/cm^2) (USA) during sunny day at 11:00 am. Yield related traits including days to flowering, days to maturity, plant height (cm), number of grains per plant, 1000- grain weight (g) and grain yield per plant (g) recorded at maturity.

RESULTS AND DISCUSSION

The analysis of variance confirmed significant variation among 100 wheat genotypes for all the considered seedling traits. The performance of genotypes in two different treatments i.e. normal and water deficit conditions had significant variation. The genotype × treatment interaction also had significant variability. Root shoot ratio is very sensitive to drought stress and it increases during water deficit conditions. The genotypes which maintained root/shoot ratio under at 50% FC declared as water deficit tolerant. Results indicated that lines 9860, Manthar 2003, 9787, Aas 2011, 9864, Chakwal 86, 9860 and 9846 showed highest resistant to change in their root/shoot ratio in both at 50% FC and 100% FC. The genotypes 9526, 9610, 9505, 9521 and 9495 showed highest change in root/shoot ratio. Comparison of mean performance of genotypes for root/shoot ratio under normal and water deficit conditions presented in Fig 1. Conclusions were in agreement with (Tavakol and Pakniyat, 2007 and Siddig *et al.*, 2013). It was documenter that root shoot ratio increased in water shortage and genotypes which had high root shoot ratio performed better in water deficit conditions (Naeem *et al.*, 2016; Khan *et al.*, 2010a and Baloch *et al.*, 2012). During water deficit conditions plant uses all its reserves to increase in length of root in search of water. The shoot does not grow and plant indicates small shoot length. Consequently, root/shoot ratio increased. The shoot length is equally important for plant as much as root length. The shoot provides sources for the plant (leaves) which made food. The food sinks into shoot. When plant get mature and starts grain formation the store food moves to the grain. The plant use shoot as source for food and store in the grain. If shoot length remains stunted, the source of the plant will be smaller at maturity which ultimately effect the grain size and weight. The grain yield per plant will be reduced. Therefore, along with long root length, shoot length is also important. The lines which maintained root/shoot ratio under normal and water deficit conditions have the capacity to tolerate water shortage. The root/shoot ratio of the selected plant for water deficit tolerant should be close to 1 at seedling stage. Which will indicate the equal growth of root length and shoot length.

Results indicated that, 9860, 9846, Chakwal 86, Manthar 2003, 9864, Aas 2011 and 9787 had maintained highest, and 9610 had minimum relative water contents at 50%FC and 100% FC. Means values of genotypes for RWC presented in Fig 2. Similar results were also observed in previous studies (Lugojan and Ciulca, 2011; Soleimani *et al.*, 2014 and Khakwani *et al.*, 2011). Wheat cultivars retained highest relative water contents are most tolerant against water deficit conditions (Arjenaki *et al.*, 2012). Water deficit cause water losses within plants and resulted in reduction of relative water contents. In this sense RWC are the reliable and widely used source to check the sensitivity and tolerance of plant against drought (Liu *et al.*, 2013).

Water shortage is decline of available water, in response plant concentrated available solutes i.e. carbohydrates and proline to take water and maintain water potential through osmotic regulation (Martin *et al.*, 1993). Osmotic regulation helps plant in growth and development in water deficit conditions (Pessarkli, 1999). Decrease of RWC resulted in closing of stomata which ultimately decrease in rate of photosynthesis (Cornic, 2000). High percentage of relative water contents increase chances of survival for plant in water deficit environment (Schonfeld, *et al.*, 1988). Moderate to rigorous water deficit conditions effect plant's morphological and physiological traits. Bilal *et al.* (2015) confirmed relative water contents as excellent criteria for selection against drought stress. The ability of plant to survive in water deficit conditions depends on its relative water contents (Larabi and Meliche, 2004). Relative water contents proposed as most important indicator of plant water status as compared to any other (Almeselmani *et al.*, 2011).

The genotypes manifested lowest value of ELWL selected as water deficit tolerance. Investigation revealed that genotypes 9860 and 9864 indicated, lowest value of excised leaf water loss and 9495 and 9521 manifested highest excise leaf water loss at 50% FC and 100% FC. Mean values of genotypes for ELWL presented in Fig 4.14. The results were also very relevant with (Babgohari *et al.*, 2017, Naeem *et al.*, 2016 and Saleem *et al.*, 2016). Mossa *et al*. (2016) and Kaur *et al*. (2016) found that genotypes which indicated very low value of ELWL performed better at 50% FC. The selected lines also performed better in the field under water deficit environment. The characterization of wheat genotypes for water deficit tolerance on the base of ELWL is very effective. The stomata of excised leaf semi closed after some time and moisture contents retained in excised leaf for hours. The wilt weight recorded after 6hrs. The water loss from incomplete open stomata. This parameter can help the plant to recover from water deficit stress condition. The value of ELWL is directly proportional to water retention capacity of plant. The lower value of ELWL ensures about the high water retention capacity of plant under water deficit conditions.

Fig. 1: Mean comparison of selected wheat genotypes for root/shoot ratio at 50% FC and 100%FC

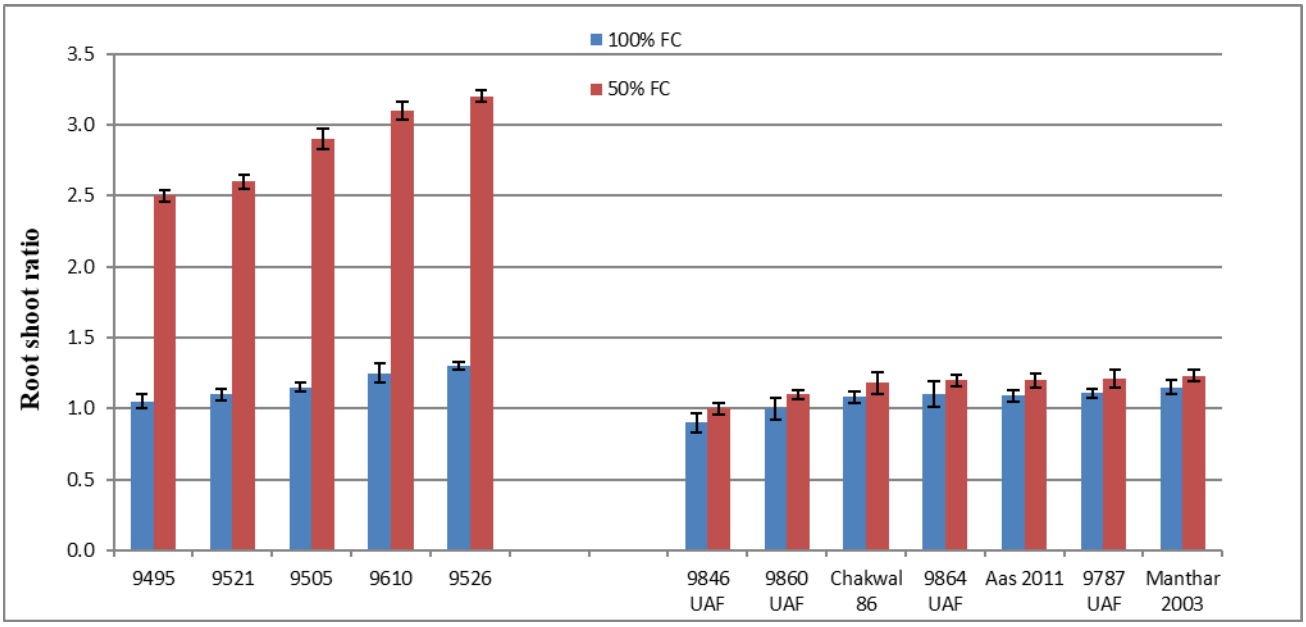


Fig. 2: Mean Comparison of selected wheat genotypes for RWC at 50% FC and 100% FC.

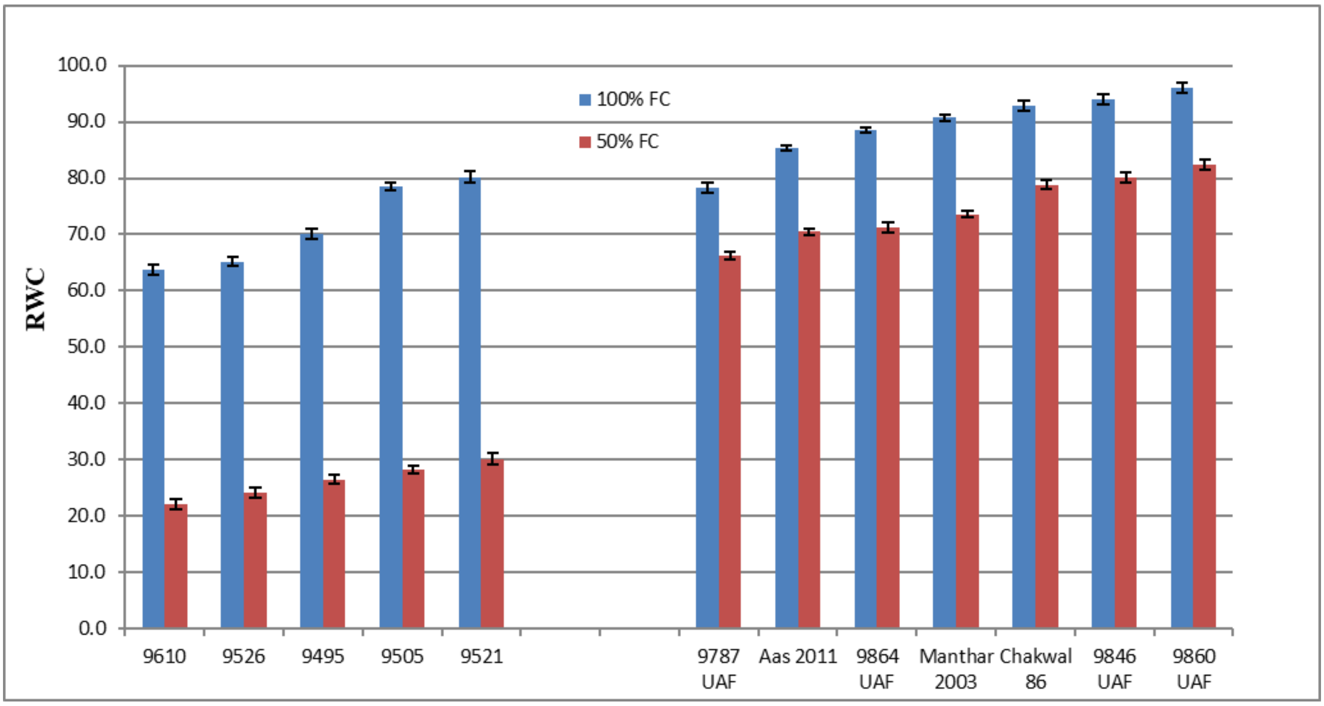
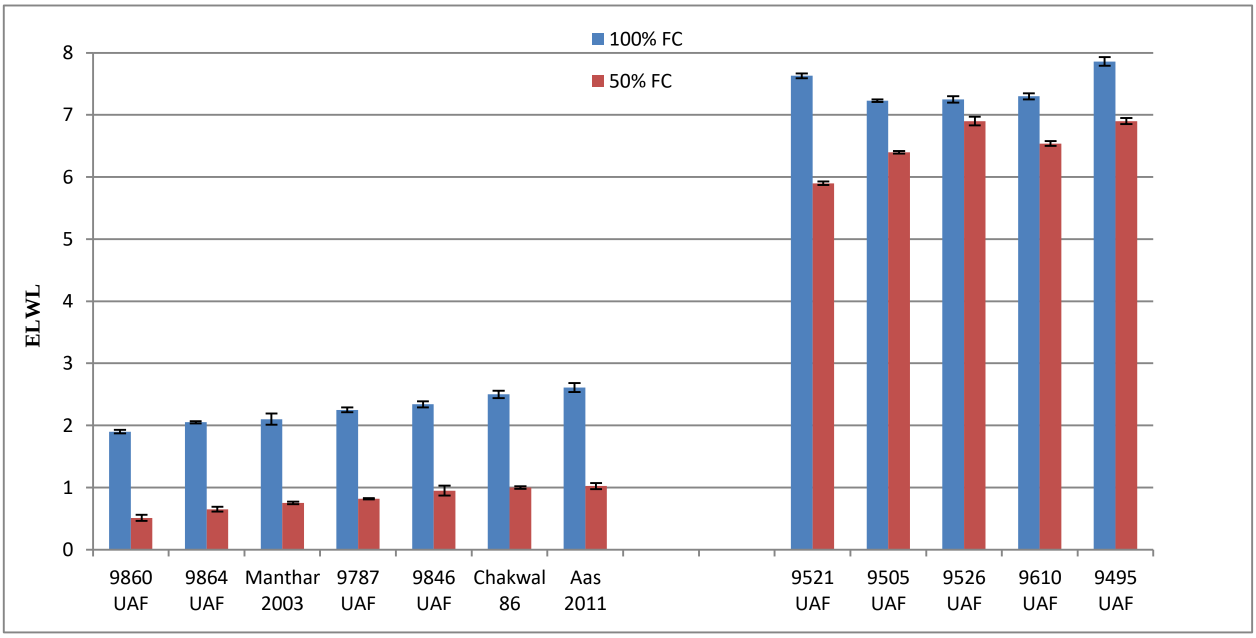


Fig.3: Mean comparison of selected wheat genotypes for ELWL at 50% FC and 100% FC.



The subsequent year 2021-2022 selected drought tolerant wheat genotypes further evaluated on the base of physiological and yield related parameters. Experiment conducted in an open tunnel. The treatment under normal irrigation kept open for rain and regular irrigation while other treatment under drought had not irrigated and saved from rainwater and covered tunnel during rain. Ultimately, plant in drought treatment kept completely without moisture until maturity. Physiological parameters recorded at anthesis stage and a comparison made between plants under normal regular moisture condition and plant under drought conditions. The mean performance of physiological parameters observed during studies in presented in Table 1. Mean values of physiological parameters reveled that genotype 9860 indicated lowest value of excised leaf water loss, high cell membrane thermostability, high photosynthetic rate, low value of stomatal conductance, high rate of CO2 assimilation and high chlorophyll contents (Salma *et al*., 2016). The decrease in excised leaf water loss enhanced ability of plant to survive under drought conditions. Cell membrane thermostability measures to estimate the percentage of injury during drought conditions (Chaudhry *et al*., 2017). The low injury index indicated stability and resilience ability of plant under water shortage. Photosynthetic rate decreases during drought conditions due to non-availability of water (Rashid *et al*., 2019). Maintenance of photosynthetic rate during normal and drought conditions indicating drought tolerance (Devasirvatham *et al*., 2018). One of the plant's defense system against water shortage is closing of stomata (Pouresmael *et al*., 2012). The increased in water shortage triggered the release of abscisic acid which get deposited in guard cells of stomata and force the stomata to close. The low stomatal conductance during drought conditions enabled plant to maintain its turgor pressure and moisture level can be maintained. The process of fixing inorganic carbon into organic carbon builds the capacity of plant for high photosynthetic rate (Rezai *et al*., 2015). High value of CO_2 assimilation under drought conditions is directly proportional to photosynthetic and an indicator of drought tolerance. Chlorophyll contents are indicator of plant health and presence of nitrogen in plants. The stability of chlorophyll contents under normal and drought conditions is the replica of drought tolerance (Maqbool *et al*., 2017). Yield and yield related traits recorded at anthesis and maturity Table 2. Results indicated that due to drought conditions plant flowers and can mature early (Sabaghpour *et al*., 2006). This phenomenon is termed as drought escape. In this drought tolerance, mechanism plant try to avoid drought and complete it maturity before it is too harsh for the plant to deal with it (Ashraf *et al*., 2005). According to observation wheat genotypes under drought conditions started flowering and get matured 10 to 12 days before as compared with normal irrigation conditions. The plant height, number of grains per plant, 1000-grain weight and grain yield per plant also effected due to water shortage (Ismail *et al*., 2017). Plants under drought conditions had 15 cm less height as compared with plant under normal irrigations. The number of grains per plant 25% reduced under drought and 1000 grain weight reduced to 15% and grain yield per plant 10-15% decreased under drought conditions. Wheat line 9860 indicated satisfactory levels of physiological traits and had maintained yield and yield traits. The lines found drought tolerant and can be further investigated in yield trials. These lines can also used in breeding programs for development of drought tolerant high yielding lines.

Table 1: Mean values of wheat genotypes of physiological traits under normal and drought conditions

Genotypes	Trt	ELWL m±S.E	CMT m±S.E	PR m±S.E	SC m±S.E	CA m±S.E	CC m±S.E
9860	N	3.1±0.08	81.1±0.01	27.3±0.61	0.7±0.06	1.3±0.07	26.8±0.45
	D	1.0±0.06	72.1±0.04	25.2±0.57	0.6±0.04	1.2±0.05	26.5±0.58
9846	N	3.2±0.06	72.8±0.05	27.2±0.41	0.7±0.05	1.3±0.04	23.5±0.62
	D	2.2±0.06	71.1±0.07	22.2±0.24	0.6±0.07	1.2±0.06	22.1±0.48
Chakwal 86	N	4.0±0.06	70.3±0.06	25.3±0.59	0.7±0.07	1.3±0.08	24.5±0.42
	D	1.0±0.04	70.4±0.09	21.1±0.61	0.5±0.05	1.2±0.04	20.3±0.42
Manthar 2003	N	5.2±0.07	66.4±0.05	22.2±0.64	0.7±0.06	1.3±0.06	22.3±0.84
	D	1.4±0.05	57.3±0.04	20.2±0.52	0.5±0.04	1.2±0.04	20.3±0.87
9864	N	5.4±0.04	56.0±0.04	24.1±0.47	0.7±0.06	1.3±0.04	21.4±0.89
	D	1.5±0.04	56.3±0.06	20.2±0.53	0.6±0.04	1.2±0.06	19.3±0.78
Aas 2011	N	3.1±0.06	74.1±0.04	25.3±0.64	0.7±0.03	1.3±0.05	35.8±0.80
	D	1.5±0.04	74.1±0.08	24.1±0.47	0.5±0.07	1.2±0.08	33.8±0.74
9787	N	3.6±0.08	73.1±0.07	25.2±0.54	0.7±0.07	1.3±0.07	35.8±0.81
	D	2.0±0.09	69.1±0.04	23.3±0.58	0.6±0.04	1.2±0.05	32.2±0.86

Table 2: Mean values of wheat genotypes of yield and yield related traits under normal and drought conditions

Genotypes	Trt	DOF m±S.E	DOM m±S.E	PH (cm) m±S.E	NOG P^{-1}	100 GW (g) m±S.E	GYP^{-1} (g) m±S.E
9860	N	99.6±0.98	152.1±0.91	109.3±0.59	79.6±0.74	26.8±0.45	7.5±0.81
	D	90±0.89	146.6±0.85	91.4±0.88	47.3±0.54	18.5±0.58	5.7±0.89
9846	N	98±0.88	153±0.87	109.2±0.84	78.6±0.54	23.5±0.62	7.4±0.54
	D	92.3±0.7	145.3±0.82	90.3±0.59	46.3±0.87	16.1±0.48	5.5±0.52
Chakwal 86	N	91.3±0.85	154.3±0.75	108.6±0.51	78.3±0.52	24.5±0.42	6.9±0.56
	D	87.6±0.87	143.6±0.74	89.76±0.62	45.3±0.87	15.3±0.42	4.9±0.58
Manthar 2003	N	84.3±0.75	156.6±0.75	107.5±0.67	77.6±0.53	22.3±0.84	6.8±0.56
	D	83.6±0.68	145.3±0.76	88.7±0.62	44.3±0.87	15.3±0.87	4.8±0.58
9864	N	91.3±0.74	157±0.78	107.6±0.63	65.3±0.52	21.4±0.89	6.7±0.56
	D	86.3±0.68	144.6±0.79	88.1±0.57	43.3±0.87	15.3±0.78	5.4±0.35
Aas 2011	N	98.3±0.47	151.3±0.71	111.7±0.91	81.6±0.57	35.8±0.80	7.6±0.64
	D	95.6±0.52	145.3±0.73	95.4±0.54	54.3±0.53	26.8±0.74	6.8±0.56
9787	N	99.6±0.57	153±0.72	110.6±0.52	77.6±0.53	35.8±0.81	7.5±0.69
	D	94.6±0.53	149.6±0.71	95.2±0.54	53.3±0.87	25.5±0.86	6.7±0.56

Conclusion

Wheat line 9860 indicated satisfactory levels of physiological traits and had maintained yield and yield traits. The lines found drought tolerant and can be further investigated in yield trials. These lines can also use in breeding programs for development of drought tolerant high yielding lines. The selection criteria developed for identifying drought tolerant wheat genotypes is very efficient.

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