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THE IMPACT OF SEED SIZE ON INITIAL DROUGHT STRESS RESILIENCE AND YIELD IN WHEAT CULTIVATION

CHATTHA MB, MAQSOOD MZ*, JAVID II, ALI S, ABBAS MA, ANAS M

Department of Agronomy, Faculty of Agricultural Sciences, Quaid e Azam Campus, University of the Punjab, Lahore, Pakistan
**Correspondence Author Email Address: zubairmaqsood034@gmail.com*

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Abstract *Wheat yield is affected severely by drought in this era of changing climate patterns, including high temperatures and altered precipitation patterns. Drought is among the most challenging environmental stressors, limiting wheat cultivars' growth, productivity, and performance. The current study was conducted during the rabi season 2022 at the Research Area, Department of Agronomy, University of the Punjab, Lahore, Pakistan. Therefore, the present study evaluated the potential of diverse seed sizes to advance wheat crop growth, development, and yield when subjected to different drought levels. The study comprised two experiments. The first was a lab experiment that included different drought levels (DL), DL_0: 0.0 bar, DL_1: -2 bar, DL_2: -4 bar, and DL_3: -6 bar (drought levels were induced by solutions of PEG-6000 at different concentrations) and three wheat seed size classes, i.e., bold grain (>38 g), medium grain (<33 g), and small grain (<25 g). In the field experiment, drought stress levels were DL_0 (regular irrigation), DL_1 (first irrigation at 30 days), DL_2 (first irrigation at 45 days), and DL_3 (first irrigation at 60 days). Seed sizes included W_1 (bold >38 g), W_2 (medium <33 g), and W_3 (small <25 g). Drought severity increased with DL_1 to DL_3. The outcomes of the field experiment revealed that varying levels of drought stress and seed size classes significantly affected parameters such as emergence time, growth traits, biomass allocation, tiller count, plant height, and grain and biomass outcomes. Bold seeds contributed to higher biomass and grain yield, while severe drought decreased yields. Notably, the Harvest Index was affected, indicating bold seeds allocate more biomass to grains. In conclusion, proper seed size selection, favouring bold seeds, can enhance resilience to drought, benefiting wheat cultivation in water-scarce regions.*

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Keywords: *Drought stress; Bold seed; Growth; Yield; Wheat*

Introduction

Drought is a predominant environmental pressure that affects around 32% of the total area of 165 million hectares under wheat cultivation (Rajaram, 2000). The productivity of global crops faces a substantial threat from drought, which stands as one of the foremost abiotic stresses that severely inhibits growth and affects physiology and economic production (Ali et al., 2022; Kadam et al., 2014). Throughout the crop growth cycle, drought may emerge at any time. Nevertheless, two periods are anticipated: the early period, which may have an impact on seedling emergence, vegetative phase, and germination, and the later terminal drought, which may have an impact on pollination, seed growth, and overall formation (Watts & El Mourid, 1988). Thus, in wheat, the stages of stem elongation to booting are more affected negatively by drought than by flowering or grain-filling. Drought stress in wheat occurs during the early vegetative stages of leaf development, shoot elongation, and tillering, primarily due to reduced carbon dioxide assimilation caused by decreased transpiration, gaseous exchange, stomatal conductance, and restricted plant water supplies (Cossani & Reynolds, 2012).

Plants exhibit various drought tolerance mechanisms significantly in biomass distribution to the various organs (Asch et al., 1999). Semi-dwarf stature, early vigorous growth, tillering capacity, and early maturity serve as responsive strategies for environments characterized by terminal heat and drought stresses (Morgan, 1995; Van Ginkel et al., 1998; Kirkegaard et al., 2001; Bai et al., 2004 and Álvaro et al., 2008). Therefore, high-yielding, large-seeded, short-statured genotypes could offer substantial advantages to farmers in harsh environments like drought-pronounced areas. (Trethowan et al., 2001).

Drought caused a more incredibly significant amount of dry weight in stems rather than roots and leaves for high-yielding wheat genotypes (Veneklas & Peacock, 1994). According to Mason & Spaner (2006) and Andrew et al. (2015), plant height, early growth, and tillering capacity stand out as the most critical genotypic features that provide wheat and other cereals their competitive abilities. Opting for a tall, low-yielding wheat genotype over a semi-dwarf, high-yielding genotype of wheat under stress conditions raises concerns due to the negative and adverse relationships between plant height and yield components, along with potential (Austin et al., 1980; Rebetzke & Richards, 2000). Thus, Semi-dwarf height, early growth vigour, and early maturity are adaptive traits for habitats facing drought pressures (Kirkegaard et al., 2001; Bai et al., 2004; Lvaro et al., 2008). Decreased height is the primary option in hot, dry situations with no yield penalty (Ania et al., 2016).

Similarly, earlier seedling establishment is vital for the successful farming of crops in dryland farming systems. Different seed sizes have different amounts of starch and other food reserves that may be the factor to control early germination and establishment in crop plants (Wood et al., 1977). Large-seeded varieties are also associated with improved growth of early leaf area in wheat under moisture deficiency conditions (Richards & Lukacs, 2002; Rebetzke et al., 2008). Size- and weight-based seed grading is a collaborative effort to control seed germination and subsequent seedling growth. Studies regarding climatic change and global warming confirmed that Pakistan is on the list of countries more susceptible to climatic changes (Hussain et al., 2020). The rainfall distribution pattern has changed in Mediterranean environments in recent decades (Araus et al., 2002). Due to insufficient rainfall distribution variation, drought is the most common environmental challenge for achieving successful wheat production in these areas(Golla, B., 2021). Moreover, drought perceived in irrigated areas is due to insufficient rainfall and canal closure during early vegetative growth of the wheat crop. The rainfall pattern has also changed because now we record rainfall in February, while previously, those rains were in December and January (Ali et al., 2021). However, improved wheat varieties having appropriate seed size and plant stature could offer significant advantages to farmers in mitigating early drought conditions (Khadka et al., 2020). So, considering the shifts in climate trends, the present study aimed to determine the role of seed size for successful stand establishment under early vegetative drought conditions and to get the maximum return per unit of land in rain-fed areas of Pakistan.

Material and methods

Wheat germination and seedling growth parameters were examined using polyethylene glycol (PEG-6000) to examine the impact of drought stress. Three levels of polyethylene glycol (PEG-6000) stress (0.0 bar (control), -2 bar, -4 bar, and -6 bar named DL_0, DL_1, DL_2, and DL_3, respectively) were produced (Kaufmann et al., 1971). To make PEG 6000, the necessary quantity of PEG 6000 was dissolved in distilled water at 25°C. Wheat grains were subjected to a disinfection process for 3 minutes using a 5% Sodium hypochlorite (NaOCl) solution. Then, the seeds went through three rounds of washing with distilled water. In Petri plates, two filter papers were set up to initiate the germination process of 20 grains for each seed size. The appropriate PEG6000 treatment was used to wet the filter sheets and incubate Petri dishes for 20 days at 27.2 °C with parafilm covering and sealing the lids to prevent moisture evaporation. Germinated seeds (with a minimum 2 mm radical extension) were subjected to data collection and observed after every 24 hours. The following growth parameters were counted:

Agronomic Attributes

Time to 50% emergence time (days)(E_{50}), Mean Emergence Time (days) (MET), Coleoptile length (cm), Root/shoot length (cm), Root/shoot fresh weight (g), Root/shoot dry weight (g).

Yield Parameters

Number of productive tillers per Plant, plant height (cm), number of spikelets per spike, number of grains per spike, 1000-seed weight, *B*iological yield, Grain yield, and Harvest Index %.

The laboratory procedures were conducted at the Agronomy Lab, Department of Agronomy, University of the Punjab, Lahore, Pakistan.

Field Experimental Site, Climatic Conditions, and Soil Descriptions

The experiment was conducted during the rabi season 2022 at the Research Area, Department of Agronomy, University of the Punjab, Lahore. A randomized complete block design (RCBD) with a factorial arrangement conducted the trial with three replications. The climate at the research site is subtropical, and (Fig. 1) shows the weather patterns throughout the growing season. Soil samples collected from various areas of the experimental field with the help of an auger underwent testing to identify various physio-chemical parameters by Homer and Pratt's (1961) recommended practices. The soil was a clay loam with a pH of 8.1, an electrical conductivity of 0.79 dS/m, 0.49% organic matter, a bulk density of 1.19 g/cm³, 58% saturation, and contains 5.9 mg/kg of available P and 130 mg/kg of available K.

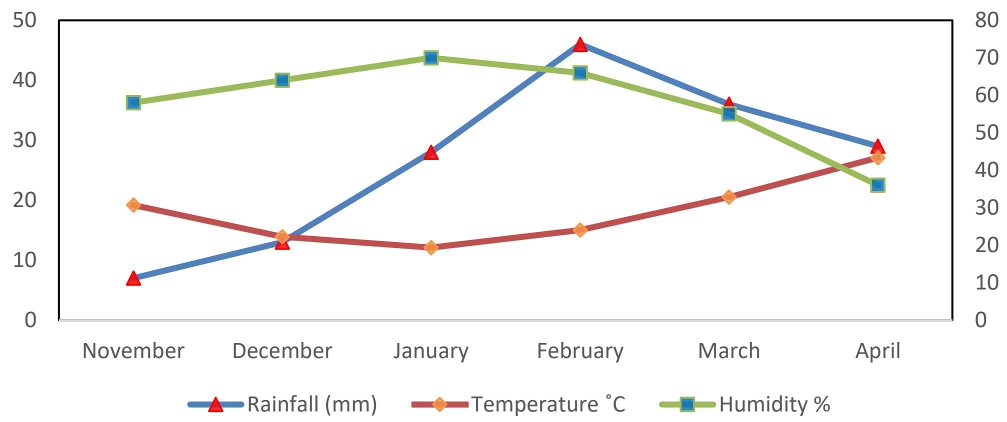

Figure 1: Weather conditions during the crop growth period at the experimental site during the study

Treatments and Agronomic management

In this study, the performance of three wheat seed size classes—bold grain (>38 g), medium grain (<33 g), and small grain (<25 g)—was examined in terms of their establishment, subsequent growth, and yield under normal field conditions. Early drought stress was induced without rain shelters, and any occurring rainfall during the stress period was recorded. The experiment employed a main plot factor involving varying levels of early drought stress: DL_0 (regular irrigation throughout the crop period), DL_1 (first irrigation at 30 days after sowing followed by subsequent irrigation as per crop need), DL_2 (first irrigation at 45 days after sowing followed by subsequent irrigation as per crop need), and DL_3 (first irrigation at 60 days after sowing followed by subsequent irrigation as per crop need). Additionally, a sub-plot factor consisted of seed size classes based on 1000-seed weight after grading: W1 (bold grain >38 g), W2 (medium grain <33 g), and W3 (small grain <25 g).

Statistical Analysis

The recorded findings from the lab and field experiments were analyzed using Statistics 8.1 software. The analysis involved applying an F-test, and significant differences among treatment means were subsequently determined through the LSD test at a 5% probability level.

Crop husbandry

The field was twice cultivated, then planked to establish the final seedbed. Using a manual drill and rows spaced 22.5 cm apart, the appropriate seeding density of 125 kg/ha^{-1} was applied. Following a soil analysis, fertilizer was administered at the following rates: 100, 80, and 60 kg NPK ha^{-1}. For fertilization, urea (46% N), single superphosphate (SSP) (18% P), and sulfate of potash (50% K) were used. The P, K, and approximately one-third of the nitrogen fertilizers were applied as the base dosage; the remaining N was applied in two applications (one during the tillering and the other at the booting stage). For improved growth, the crop received a total of four irrigations.

Observations and Data Collection

Once the seeds had been sown, the plots were inspected daily to determine the emergence time. Until the final harvest, tillers were tallied, and each plot had a unit area (m^2) marked on it. Ten plants from each plot were also picked, and their heights were averaged after being measured. The length and number of grains of ten spikes were measured and tallied. After harvest, the crop was weighed to determine the biological and grain yield, expressed in t ha^{-1}.

Results and discussion

Agronomic attributes

Results indicated that different seed sizes and drought levels remarkably affected mean emergence time (MET). Maximum MET (18.9 days) was taken by Drought level 3, and lowest MET (9.9 days) was taken by Control. Similarly, the maximum (15 days) MET was observed in small seeds, and bold seeds showed a minimum (11.5 days) MET. Regarding Time to 50% emergence (E50), the highest mean (12.6 days) was observed by Drought level 3, and Drought level 0 showed a minimum E_{50} (6.656); seed sizes also showed a significant difference in E50 small seeds had a maximum E50 (10.042) however bold seeds had minimum E50 (7.667) (Table 1). The above outcomes of emergence percentage correlate with the findings of Hussain et al. (2018). Among seed sizes, Bold seeds exhibited a higher coleoptile length (8.541 cm), while the minimum coleoptile length was 5.208 cm, observed in tiny seeds. Similarly, Coleoptile length (Table 1) varied significantly among various drought levels. DL_0 had a maximum Coleoptile length (7.8333 cm), while DL_3 showed a shorter coleoptile (5.9444 cm). Results also indicated that the maximum root length (9.1562 cm) and shoot length (9.1562 cm) observed in Bold seeds (Table 1) and small seeds showed minimum root length (6.9375 cm) and shoot length (6.9375 cm); similar results are found in different drought levels DL_0 had maximum root (9.45 cm) and shoot length (2.1605 cm) while DL_3 had minimum root (5.8333 cm) and shoot lengths (1.4568 cm).

Table 1: Effect of different seed sizes and drought levels on Agronomic attributes; Time at 50% emergence(E_{50}), mean emergence team(MET), coleoptile length(CL), root length(RL), shoot length(SL), shoot fresh weight(SFW), root fresh weight(RFW), shoot dry weight(SDW), and root dry weight(RDW)

Seed Size (SS)	E_{50}(Days)	MET(Days)	CL (cm)	RL (cm)	SL (cm)	SFW	RFW	SDW	RDW
(W1) Bold	7.67b	11.5b	8.60a	9.15a	12.16a	4.01a	2.96a	2.22a	1.56a
(W2) Medium	9.41a	14.15a	7.12b	7.93b	10.84b	3.31b	2.45b	1.84b	1.29b
(W3) Small	10.04a	15.06a	5.20c	6.93c	9.83c	2.96c	2.19c	1.65c	1.17c
LSD ≤0.05 P	**1.056**	**1.585**	**0.255**	**0.291**	**0.447**	**0.166**	**0.123**	**0.092**	**0.065**
Drought Level (DL)									
DL_0 (Control)	6.65c	9.98c	7.83a	9.45a	12.61a	3.89a	2.88a	2.16a	1.51a
DL_1 (-4 bar)	8.4b	12.67b	7.33a	9.12a	12.16a	3.66b	2.71b	2.04b	1.42b
DL_2 (-6 bar)	8.46b	12.68b	6.72b	7.62b	10.16b	3.54b	2.62b	1.97b	1.37b
DL_3 (-8 bar)	12.61a	18.91a	5.94c	5.83c	7.78c	2.62c	1.94c	1.45c	1.02c
LSD ≤0.05 P	**1.220**	**1.830**	**0.506**	**0.613**	**0.817**	**0.216**	**0.160**	**0.120**	**0.084**
SS × DL	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**

Means with different letters differed by 0.05 P level. NS: non-significant

When exposed to elevated PEG concentrations, a marked decrease in the coleoptile, shoot, and root lengths was observed compared to the control group. This reduction in root lengths and shoots could be attributed to the advancement in osmotic potential resulting from intensified drought conditions. This heightened osmotic potential contributes to dehydration and creates an ionic imbalance within transpiring leaves, reducing meristem activity and cellular elongation (Table 1). (Zhu et al., 2001); (Munns, R. 2005); (Huang et al., 2006). DL_0 demonstrated maximum values for shoot fresh weight (3.88 g) and shoot dry weight (2.16 g) among the groups, whereas DL_3 exhibited minimum values for both parameters. Furthermore, Bold seeds displayed the most excellent shoot fresh weight (4.0 g) and shoot dry weight (2.22 g), whereas small seeds showed the lowest values for both shoot fresh weight (2.966 g) and shoot dry weight. DL_0 displayed the highest shoot fresh weight, shoot dry weight, root fresh weight, and root dry weight among the groups, whereas DL_3 exhibited the lowest values for all four parameters (Table 1).

Similarly, Bold seeds showed the most excellent shoot fresh weight, shoot dry weight, root fresh weight, and root dry weight, whereas tiny seeds consistently demonstrated the lowest values for these measurements (Table 1). These findings align with R.M. Trethowan et al. (2001), who state that large-seeded varieties could benefit farmers in drought-stressed environments.

Yield parameters

Results also indicated that diverse SS and DL significantly impacted both parameters, i.e., growth and yield. The highest number of productive tillers/plant (5.3a) was found in DL_0, and the lowest number (4.14c) was observed in DL_3 (Table 2). Likewise, bold seeds had the highest number of productive tillers/plant (5.6a), while small seeds had minimum productive tillers/plant (4.2b) (Table 2). Bold seeds also had a more significant number of tillers compared to other seed types. While tiny seeds exhibited lower vigour and have weak germination, which results in inferior tillering, Gan and Stobbe (1995) found that bold seeds maintain enough plant populations throughout a wide variety of field conditions and, as a result, significantly boost the output of tillers. Bold seeds also exhibited the tallest plants (78.62 cm) and a maximum number of grains per spike (51.317), while the shortest plants (60.57 cm) were observed in small seeds and the lowest number of grains per spike (33.183) (Table 2). Drought stress at the anthesis stage led to a decline in the number of grains per spike in wheat genotypes, resulting in a decreased yield potential (Shokat et al., 2020). Similarly, DL_0 had the tallest plants (79.889 cm) and a maximum number of grains per spike (46.933), while the shortest plants (60.968 cm) were observed in DL_3 and the lowest number of grains per spike (36.433) (Table 2). Bold seeds emerged faster in comparison to all other seed types. According to Mustafa et al. (2018), bold seeds are believed to exhibit high vigour and possess an extensive reservoir of stored reserves, enhancing the metabolic process and contributing to improved and earlier emergence. The results highlighted that various seed sizes and drought levels had significantly impacted the plant height and number of productive tillers.

These findings are aligned with Delong et al. (2012), who found that drought stress significantly impacted plant height in wheat, with a higher drought stress coefficient at the jointing stage. The maximum 1000-grain weight (49.128 g) was noted in bold seeds, while the lowest (40.718 g) was observed in small seeds (Table 2). Treatment DL_0 demonstrated the maximum 1000-grain weight (54.233 g), whereas treatment DL_3 showcased a minimum weight (38.073 g) (Table 2). Bold seeds also contributed to a higher 1000 seed weight due to healthier seedlings from bold seeds, which helped the better production of assimilates and ultimately resulted in more weighted seeds (Shahwani et al., 2014). Bold seeds resulted in the maximum biological yield (7.8375 t ha^{-1}), surpassing the yield obtained from tiny seeds (5.6427 t ha^{-1}) (Table 2). Similarly, DL_0 displayed the highest biological yield (8.216 ha^{-1}), whereas DL_3 showed the lowest biological yield (5.1977 t ha^{-1}) (Table 2). Drought stress significantly impacts the biomass yield of wheat. The physio-chemical processes of the plant are modified as a reaction to moisture deficiency, resulting in a decrease in yield attributing traits such as biomass yield, harvest index, and grain yield (Ankita et al., 2020). Plants originating from bold seeds displayed the highest grain yield (4.005 t ha^{-1}), outperforming those grown from small seeds, which had the lowest yield (2.1932 t ha^{-1}) (Table 2).

Table 2: Effect of different seed sizes and drought levels on the Number of productive tillers per Plant (NPT), plant height (PH), number of spikelets per spike (NSPS), number of grains per spike (NGS), 1000-seed weight (TSW), biological yield (BY), grain yield (GY), harvest index %(HI)

Seed Size (SS)	NPT	PH	NSPS	NGS	TSW	BY	GY	HI %
Bold	5.61a	78.60a	25.71a	51.32a	49.12a	7.83a	4.05a	50.4a
Medium	4.60b	70.52b	21.92b	40.41b	44.14b	7.07b	2.99b	41.6b
Small	4.21b	60.51c	19.61c	33.24c	40.71c	5.64c	2.19c	36.7b
LSD ≤0.05 P	**0.495**	**3.534**	**0.11**	**2.193**	**0.618**	**0.347**	**0.257**	**5.718**
Drought Level (DL)								
DL_1 (Control)	5.3a	54.2a	25.2a	46.9a	54.2a	8.26a	4.60a	56.16a
DL_2	5.12ab	44.6b	23.32b	43.47b	44.6b	7.59b	3.36b	43.88b
DL_3	4.7b	41.6c	21.34c	39.7c	41.6c	6.39c	2.36c	35.34c
DL_4	4.14c	38.07d	19.9d	36.4d	38.07d	5.19d	1.97d	36.26c
LSD ≤0.05 P	**0.572**	**0.714**	**0.127**	**2.532**	**0.714**	**0.401**	**0.297**	**6.603**
SS × DL	**NS**	**NS**		**NS**	**NS**	**NS**	**NS**	**NS**

Means with different letters differed by 0.05 P level. NS: non-significant

Concerning drought levels, the maximum grain yield (4.606 t ha^{-1}) was attained under the DL_0 (no drought) condition, whereas the minimum yield (1.9711 t ha^{-1}) was observed under the DL_3 (severe drought level) **(**Table 2). The study also highlighted a decrease in grain yield under drought stress than normal field conditions because the water-deficit condition may reduce the number of grains per spike (Ankita et al., 2020) and also produced light-weighted grain, which ultimately contributed to lower yield during drought stress (Yordanovet al., 2003). Grain yield serves as a direct selection criterion for identifying the performance of drought-resistant varieties. The efficacy of using Grain Yield as a direct selection criterion for drought tolerance has been successfully demonstrated by the experimental work and the results of the findings by Liu et al. (2017). Maximum Harvest Index (HI) was noted in DL_0 (no drought) condition (56.167%), whereas the lowest Harvest Index (36.226%) was observed in DL_3 (severe drought level) (Table 2). Regarding Seed size, Bold seeds showcased the highest harvest index (50.412%), signifying a more significant proportion of the total biomass allocated to grains. These findings are consistent with the conclusions drawn by Shoaib et al. (2022), who found that the highest harvest index is related to bold seeds. Conversely, tiny seeds exhibited the lowest harvest index (36.706%) (Table 2**).** The subsequent reduction in HI may be due to lower grain yield during drought stress (Mukti et al., 2020). Additionally, the bold seeds had the highest HI, which may be due to their increased grain and biological yield (Marisa et al., 1993).

Conclusion and recommendations

Bold seeds displayed the highest grain yield, potentially due to increased tiller production, improved grains/spike, longer spikes, and greater grain weight than other SS classes. Based on our current research, we recommend using larger seed sizes (bold seeds) to enhance wheat performance in drought conditions, highlighting the importance of seed size selection for improved yields in water-deficit environments.

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Declarations

Declaration of Interest Statement

We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work.

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Author's contributions

MBC and SA conducted the field trials and planned the experiment. MZM analyzed the data. IIJ and MA assisted with data collection. MAA proofread the manuscript. All authors have read and approved the final manuscript.

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Not applicable

Consent for Publication

Not applicable