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Assessment of Drought Tolerance in Mung Bean [*Vigna radiata* (L.) Wilczek] Through Phenology, Growth, Protein Yield, and Cluster Heatmap Analysis

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Mung bean [*Vigna radiata* (L.) Wilczek] cultivation is challenging under the changing climate as less precipitation during the summer season, which is becoming increasingly common, results in soil moisture deficiency and affects production especially at the early growth stages. Hence, this study was conducted to assess the phenology, growth, and protein yield traits of mung bean related to drought tolerance. Eight mung bean genotypes were grown in pots inside a rain-out shelter under well-watered (WW) and water-deficit (WD) conditions. These genotypes included four water stress-tolerant genotypes: G-1 (BARI Mung-8), G-2 (BMX-010015), G-3 (BMX-08010-2), and G-4 (BMX-08009-7), as well as four sensitive genotypes: G-5 (BARI Mung-1), G-6 (BARI Mung-3), G-7 (BU Mung-4), and G-8 (BMX-05001). Soil moisture content was maintained at 22 ± 0.5% (30% deficit of the available water) under WW condition. Water deficit was maintained at WD condition during the entire life cycle and irrigation was applied after symptoms of wilting were observed. Results showed that WD stress significantly influenced the phenology, growth, and protein yield traits in all genotypes. For days to flowering and days to maturity, G-3 took two and four days less in WD than in WW conditions, while G-5 took eight and 19 days less. Because G-3 was revealed as the most drought-tolerant genotype and G-5 as the most vulnerable, G-3 showed the smallest decrease in shoot dry weight and root dry weight under WD stress, whereas G-5 showed the biggest decrease. Likewise, G-3 experienced the smallest reductions in seed yield (35%) and protein yield (32%) under WD condition compared to WW conditions, while G-5 showed the largest reductions (80% and 76%, respectively). The hierarchical clustering analysis using two-dimension heat map also displayed the G-3 genotype as a potential and stable to water deficit stress. These findings show that the studied parameters can be useful in evaluating mung bean tolerance to drought stress and in screening for drought stress-tolerant mung bean genotypes, especially if there are no facilities to determine biophysiological and molecular traits.

Keywords: cluster heatmap, correlation, drought stress, mung bean, root-shoot ratio, root dry weight, shoot dry weight

INTRODUCTION

Mung bean [*Vigna radiata* (L.) Wilczek] is a crop with a short lifespan and can be easily adapted to major cropping systems. The acreage and production of mung bean are steadily rising due to the development of short-duration varieties that acclimatize well to existing cropping patterns (PRC-RARS 2014). It is a rich protein source, providing up to 25% of its seed's dry weight, and is a staple food for many Asian populations (Khattak et al. 2001). Various recipes also use mung beans for dal soup, bean sprouts, noodles, bean curd, and dal cake (Islam 2001). Additionally, it is rich in phosphorus, calcium, and vitamins

(Ahmed et al. 2000). It also provides a significant amount of nitrogen grown under rice-oriented cropping system (Sharma and Prasad 1999). Mung bean is mainly grown in Bangladesh from March to May (Kharif-1 season) when very scanty rainfall occurs. However, irrigation facilities are not readily available. Climate change and global warming have deleterious effects on crop production, as high temperatures and less rainfall impact mung bean growth and yield (Islam et al. 2024; Islam et al. 2021; Sarkar et al. 2017). Drought also remains to be a significant limiting factor for crop production (Bashandi and Poehlman 1974). Globally, only about 10% of arable land is free from stress (Levitt 1980), with drought stress accounting for 26% of the total stress (Mirzaei et al. 2014). In summer (Kharif-1), the crop suffers from soil moisture stress due to insufficient rainfall or drought. At the same time, increasing temperatures (> 35°C) are likely to decrease mung bean production through suppression of seedlings and vegetative and reproductive growth, along with shedding of flowers (Sinha 1997; Rainey and Griffiths 2005; Kumar, Kaur et al. 2011). Inadequate water availability is the single most crucial agroecological factor for mung bean cultivation (Kramer and Boyer 1995). Water shortage during the seedling stage hinders the development of healthy seedlings, ultimately reducing seed yield. It also hampers photosynthesis, which controls the growth and yield of crop plants (Athar and Ashraf 2005). According to previous studies, reduced leaf area and dry matter accumulation are the causes of crop photosynthetic reduction and water stress in plants (Pandey et al. 1984; Kriedemann 1986; Hamid et al. 1990). A well-developed root system is vital in addressing the effects of drought stress through water uptake from both the shallow and the deep layers of the soil (Gaur et al. 2008), which contributes positively to seed yield (Kashiwagi et al. 2006; Singh and Bell 2021).

Drought stress can be alleviated through drought management or development of drought-resistant cultivars. However, development of trait-specific variety is controlled by inherent genetic makeup and environmental factors, while the majority of crop varieties do not perform optimally across all environments. The yield ranking of genotypes often differs when they are compared with one another, primarily due to the interaction between the genotype and the environment (Al-Otayk 2010). This makes the combination of resilient cultivars with proper operational strategies a promising approach to cultivation in a stressful environment. Hence, tolerant genotypes play a significant role in the production of mung bean, especially in adverse agroclimatic conditions. Phenological changes, growth, and morphological trait mechanisms under water stress indicate drought tolerance and have been used to determine drought-tolerant genotypes. Therefore, this study was conducted to assess the phenology, growth, and protein yield traits of mung bean related to drought tolerance, which may help in screening for drought stress-tolerant mung bean genotypes.

MATERIALS AND METHODS

Plant Resources and Study Site

Four water stress-tolerant genotypes [G-1 (BARI Mung-8), G-2 (BMX-010015), G-3 (BMX-08010-2), and G-4 (BMX-08009-7)] as well as four sensitive genotypes [G-5 (BARI Mung-1), G-6 (BARI Mung-3), G-7 (BU Mung-4), and G-8 (BMX-05001] were produced at well-watered (WW) and water-deficit (WD) conditions within pots. These genotypes were selected from a laboratory screening experiment with 33 mung bean genotypes based on their germination indices, seedling development performance, and their comparative performance under WD stress induced by PEG-6000 (Islam 2020). During the Kharif-1 season in 2016, the experiment was conducted in a rain-out shelter in the research field of the Department of Agronomy at Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur District, Bangladesh. The experimental site was situated under the Old Himalayan Piedmont Plain (Agro-Ecological Zone-1) in Bangladesh. The area was situated between 25°38´ N latitude and 88°41´ E longitude, and 38.20 m asl. Agroclimatic information, namely temperature, relative humidity, and sunshine hour during the experimental period is shown in Fig. 1.

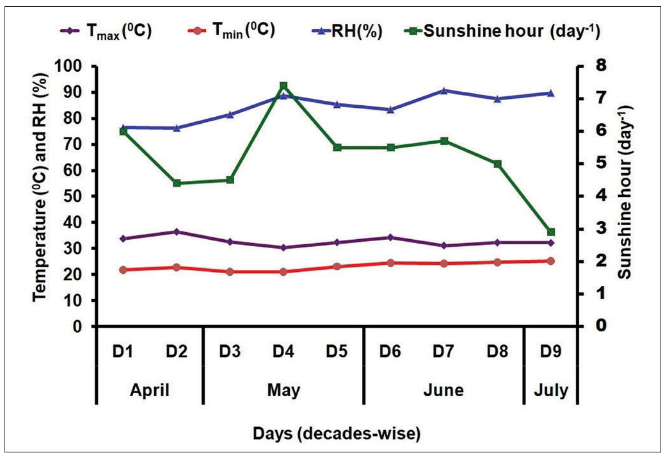


Fig. 1. Decades-wise mean temperature, relative humidity (RH), and sunshine hour (day^{-1}) during the experiment (T_{max} = maximum temperature, T_{min} = minimum temperature).

Soil Characteristics

The soil used in the experiment had a sandy loam texture with a field capacity of 25.8%, a bulk density of 1.49 g/cc, and a permanent wilting point of 11.6%. Its initial composition included 5.01 pH, 0.68% organic matter, 0.03% total nitrogen, 11.53 µg/mL available phosphorus, 0.26 meq/100 g available potassium, 17.53 µg/mL sulfur, 0.15 µg/mL boron, and 0.88 µg/mL zinc.

Experimental Design and Treatments

The research was carried out using a two-factor completely randomized design replicated four times. The treatments were done on four susceptible and four tolerant mung bean genotypes evaluated under WW and WD conditions.

Preparation of Pots for Seed Sowing

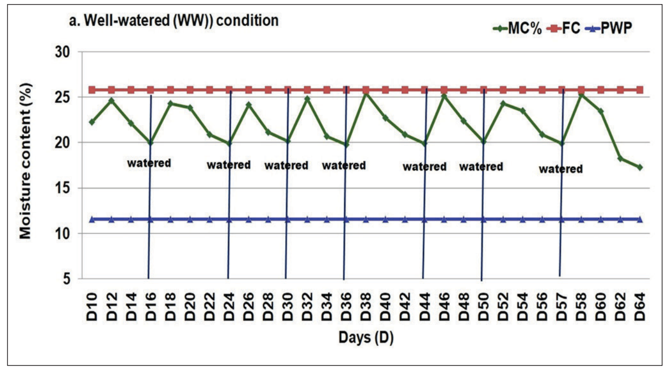
The experiment was conducted using plastic pots that had an interior diameter of 23 cm, a height of 23 cm, and a base volume of 17 cm. Ten kilograms of well-pulverized, air-dried soil combined with compost in a 4:1 ratio was placed into each pot. Urea, triple superphosphate, muriate of potash, and boric acid were evenly applied at rates of 0.103, 0.088, 0.093, 0.046, and 0.007 g per pot, or 20-17-18-10-2 kg of N, P, K, S, and B per hectare, respectively (Azad et al. 2017). The seeds were rinsed well under running water after being treated for 2 min with a 0.1% mercuric chloride solution (Dutta and Bera 2008; Saminathan 2013; Swathi et al. 2017). Each pot was then seeded with 20 seeds, maintaining a seeding depth of 2–3 cm. Five robust and steady seedlings were kept in each pot to reach maturity following emergence and establishment.

Water Regimes

To ensure consistent germination and seedling establishment, irrigation was provided to both the WW and WD stress treatments after seeding, and then again five days after sowing (DAS). From 10 DAS onward, the WW treatment was maintained by regularly providing adequate water to each pot to support optimal plant growth and development throughout the growing period, with soil moisture content maintained at approximately 22 ± 0.5%, equivalent to 20% moisture content (MC) and a 30% deficit of available water (Fig. 2a). In the WD treatment, water stress was maintained for the entire growth period, and irrigation was only performed when wilting symptoms became apparent (Fig. 2b), with soil moisture at that stage around 50% of field capacity. A digital soil moisture meter (Model PMS-714) was used to continuously measure the soil moisture levels for both treatments, and water was administered to bring the soil moisture levels back to field capacity. The net water requirement per irrigation was determined using Michael's (1978) formula:

$$d=\frac{FC-MC}{100}\times p\times D$$

Where, FC = soil's field capacity used in the pot (%)
MC = soil's moisture content at the time of watering (%)
p = soil's bulk density (g cm-3)
D = depth of the root zone (cm)



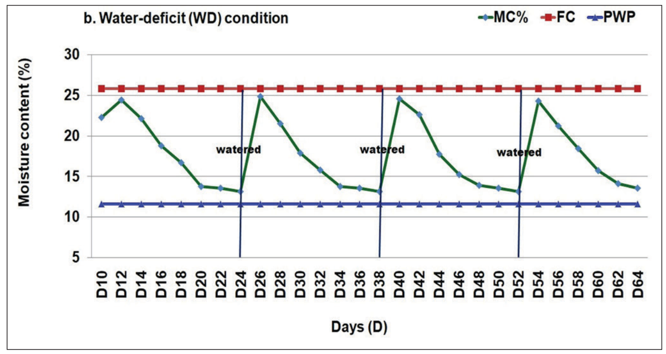


Fig. 2 (a, b). Differences in soil moisture content between WW and WW conditions. MC = moisture content; FC = field capacity; PWP = permanent wilting point.*
**This content is adapted from 'Drought Tolerance in Mung Bean is Associated with the Genotypic Divergence, Regulation of Proline, Photosynthetic Pigment and Water Relation' by Islam et al. (2023), published on TSP Website, and licensed under CC-BY 4.0.*

Cultural Management and Crop Outcome

Weeding was done as needed during the entire the growth span. Imidacloprid (Imitaf 20 SL) insecticide was applied at a concentration of 0.5 mL per liter of water during the peak flowering and full podding stages to safeguard the crop from flower thrips. Spraying with lambda-cyhalothrin (Karate 2.5 EC) was also done twice at a rate of 1 mL per liter of water, first at 100% podding and again after a seven-day interval to control pod borer, which affects the crop from the flowering stage onward.

Data Collection

Phenology, growth, and seed yield of the mung bean genotypes were measured for drought tolerance from five plant samples of each pot in the growing period. For seed yield (SY), the matured pod was collected thrice by handpicking. After the final pod harvest from each pot per treatment, the stems of all plants were cut at the soil surface using secateurs and collected to measure the aboveground shoot dry weight, including the leaves. To determine the root dry weight (g plant^{-1}), the roots from each pot were carefully separated from the potted plants by turning the pots over. Soil contaminants were then removed by gently washing the roots with running water. The samples were dried in an electric oven at 80°C for 72 h, after which they were weighed to determine the dry weight. The weights of the shoot and root of the individual plant were recorded. To compare the performance of the samples' plant traits and yield, the relative performances (RP) of the traits were calculated using the following formula as described by Asana and Williams (1965):

$$RP = \frac{\text{Data obtained of a trait in water-deficit condition}}{\text{Data obtained of a trait in well-watered condition}}$$

Assessment of the Protein and Nitrogen Content of Mung Bean Seeds

The Micro-Kjeldahl method (Bremner and Mulvaney 1982) was used to assess the N content of the corresponding mung bean genotypes under WW and WD conditions. Each sample's N content was determined using a standard curve and reported as a percentage. The protein yield (PY) and seed protein content were then computed using the formulae described by Habibzadeh and Moosavi (2014) and Thalooth et al. (2006):

Protein content of seed (%) = 6.25 × total N (%)

Protein yield (g plant-1) = protein (%) × seed yield (g plant-1).

Data Analysis

Using computer-based R software (R Core Team 2016), the data were assembled and statistically analyzed in accordance with the fundamental process described by Gomez and Gomez (1984). Correlation analysis was done using the 'Agricolae' package (Mendiburu 2023). The least significant difference (LSD) test was done at a 5% probability level to separate the means. Functional associations within the various traits as influenced by drought stress were formed through regression analysis using Microsoft Excel.

RESULTS AND DISCUSSION

Distribution Pattern of Agroclimatic Elements

Agroclimatic elements such as temperature, relative humidity, and sunshine hours are significant factors that contribute to crop growth and development. The ideal temperature range for the growth and maximum yield of mung bean is 28°C–30°C (Kaur et al. 2015). However, increasing temperatures above 35°C negatively affect summer-sown mung bean genotypes, hindering both vegetative and reproductive growth (Kumar, Karajol et al. 2011). In this study, a fluctuating pattern was observed in these weather parameters throughout the growing period of mung bean (Fig. 1). During the crop season (April to July), the maximum temperature (T_{max}) was 30.4°C–36.4°C, while the minimum temperature (T_{min}) was 21°C–25.2°C. In the growing season, the 2nd decade showed the highest T_{max} and the 4th decade exhibited the lowest T_{min}. Moreover, the relative humidity varied from 76.3% to 90.7%. However, the maximum sunshine hours were also found at the 4th decade of the crop lifespan. Data for T_{max} was above-range and T_{min} was below-range throughout the crop's growing period (Fig. 1).

Phenological Behavior of Mung Bean Genotypes

The tested mung bean genotypes showed a semi-determinate habit regarding flowering and pod maturity, and notable differences were found in WW and WD conditions (Table 1). Under WD stress, the duration of flowering and maturity was reduced. The range of days to flowering was 38–49 d under WW condition, whereas the range was 34–44 d under WD condition. Similarly, the range of days to maturity was 65–80 d under WW condition and 57–70 d under WD condition (Table 1). Due to WD stress, G-5 flowered and matured earlier by 8 and 19 d, respectively. The genotype G-3 attained flowering and maturity stages 2 and 4 d earlier, while G-4 attained these stages 3 and 5 d earlier. The longer time to maturity (70 d) observed in the G-3 genotype during WD stress might be due to the delayed initiation of first flowering (44 d). These results also conform with the study of Nunez Barrios (1991), who found that water stress delayed the onset of flowering in common bean (*Phaseolus vulgaris*), which enhanced to reduce the number of flowers resulted low pod setting. Levitt (1980) also reported that environmental stresses reduce the crop maturity period due to premature leaf senescence, limiting available resources that support the sink. Under WD stress, plants may be able to avoid unfavorable stress circumstances by blooming a few days sooner, which could lead to early flowering and pod maturity. The results also agree with those published by Ahmed et al. (2008) for mung bean and De Costa and Shanmugathasan (1999) for faba bean. In addition, the days to flowering and maturity in WD stress are likely primarily influenced by the genetic characteristics of the mung bean genotypes. On the contrary, grain growth duration (days to reproduction) also varied considerably from 26–31 d and 17–26 d under WW and WD conditions, respectively. The longest duration for grain growth (26 d) under WD condition was recorded in G-3 which was 2 d less than the other genotypes, while the shortest period (17 d) was observed in G-5 which was 11 d less than other genotypes.

Growth Parameters of Mung Bean in Response to Water-deficit (WD) and Well-watered (WW) Conditions

Shoot Dry Weight

Shoot dry weight (SDW) was measured at harvest and was significantly reduced under WD stress. The extent of the decline in SDW varied among the genotypes (Fig. 3). At WW condition, the G-2 genotype produced the highest SDW (1.95 g), followed by G-5 (1.73 g). The lowest SDW (1.30 g) was in G-6, whereas other genotypes had SDW values at 1.57–1.62 g. Under WD stress condition, the G-2 genotype had the highest SDW (1.16 g), followed by G-3 (1.13 g). The lowest SDW was observed in G-6 (0.64 g), while the SDW for the other genotypes were at 0.65–0.88 g. The G-3 genotype exhibited the best performance, with the highest relative value of 0.70, followed by G-4 at 0.67. In contrast, G-5 showed the lowest relative value of 0.38, indicating the poorest performance under drought stress. These results indicate that the highest relative reduction of SDW (62.43%) was found in G-5 due to WD stress, while the lowest was from G-3 (30.25%). Similar reductions in SDW due to WD stress have been reported in common bean (Ramos et al. 1999) and soybean (Gadallah 2000). This may be because plants under WD stress have lower cell turgor which causes prolonged cell elongation, resulting in reduced plant growth and development and lower shoot dry weight (Amira and Qados 2014; Suresh et al. 2015). The translocation of assimilates towards the roots significantly contributes to SDW, but this process is impaired by water stress (Ghebremariam et al. 2013).

Root Dry Weight

Root physiology and architecture are essential for a plant's ability to cope with soil moisture and nutrient deficiencies. Although roots do not directly participate in the process of reproduction or carbon accumulation, they are crucial to the plant's sustainability and play a key role in stress tolerance (Jenks and Hasegawa 2005). The WD stress significantly reduced root dry weight (RDW) in all the mung bean genotypes (Fig. 4). Under WD stress, G-3 had the highest RDW (0.32 g plant^{-1}) followed by G-2 (0.28 g plant^{-1}), while the lowest RDW (0.15 g plant^{-1}) was recorded in G-5. However, G-3 produced the highest relative value (0.81) while G-5 had the lowest relative value (0.46), indicating drought tolerance and susceptibility. Wasaya et al. (2018) reported that root characteristics such as thin root thickness, root length efficiency, root surface area, root orientation, and root length per unit volume influence a plant's productivity under drought conditions. Especially during the initial growth stage, these are important in

Table 1. Phenological changes of selected mung bean genotypes under WW and WD conditions.

Genotypes	Days to Flowering			Days to Maturity (60%–70%)			Days of Reproduction		
	Well-watered Condition	Water-deficit Condition	Difference Over Stress	Well-watered Condition	Water-deficit Condition	Difference Over Ctress	Well-watered Condition	Water-deficit Condition	Difference Over Stres
G-1	38	34	4	65	57	8	27	23	4
G-2	49	43	6	75	64	11	26	21	5
G-3	46	44	2	74	70	4	28	26	2
G-4	44	41	3	70	65	5	26	24	2
G-5	46	38	8	74	55	19	28	17	11
G-6	39	35	4	70	57	13	31	22	9
G-7	39	35	4	67	56	11	28	21	7
G-8	49	42	7	80	67	13	31	25	6
LSD (5%)	4.63			5.82			7.05		
CV (%)	6.48			5.04			16.08		
LS	ns			*			ns		

Where: LS = level of significance; ns = non-significant at $P = 0.05$; *significant at $P = 0.05$.

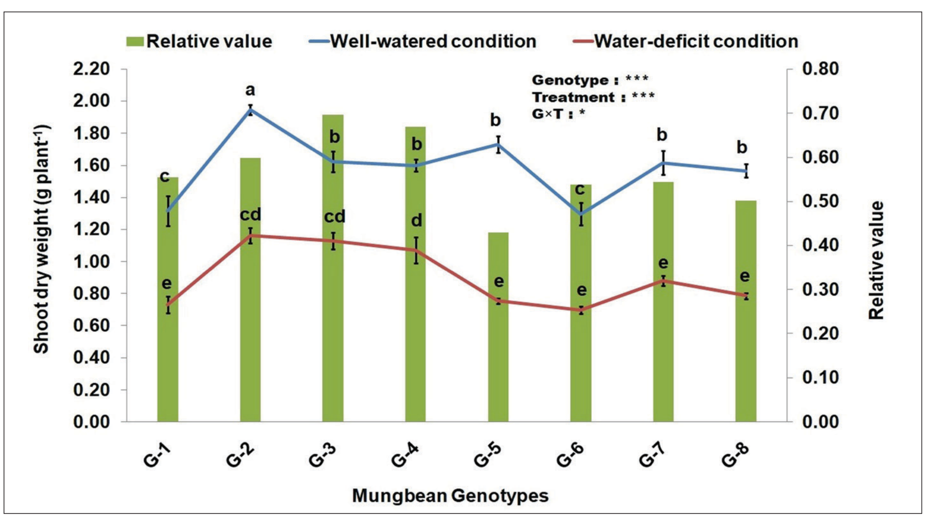


Fig. 3. Shoot dry weight of selected mung bean genotypes in response to WW and WD conditions.

avoiding soil moisture stress and play a key role in drought tolerance for lentils and other food legumes (Kashiwagi et al. 2005; Sarker et al. 2005; Gaur et al. 2008; Vadez et al. 2008; Asfaw and Blair 2012). The development of vigorous shoot and root contributes to drought avoidance and tolerance, which has been reported in mung bean (Kashiwagi et al. 2005; Asfaw and Blair 2012), chickpea (Gaur et al. 2008), and lentil (Sarker et al. 2005). Well-developed roots also support the growth of seedlings in soils where the surface dries rapidly, ensuring that adequate moisture remains in deeper layers. Thus, better knowledge about plant reactions against abiotic stresses can aid in selecting more tolerant genotypes (Den Herder et al. 2010).

Root-Shoot Ratio

Studying root traits is essential for determining water stress-tolerant genotypes (Suresh et al. 2015). Considerable variation was observed in the root-shoot ratios (RSR) among the different mung bean genotypes under WW and WD conditions (Fig. 5). Nonetheless, in all genotypes, RSR values were higher under WD condition than under WW condition (Fig. 3). Less available water may have caused the roots to expand in search of moisture, which led to more assimilates to be directed towards the roots. Moreover, plants under WD stress condition frequently reduced dry matter assimilation in the shoot and produced higher dry weight in the roots, resulting in a higher RSR than under WW condition. The highest RSR values (0.243 and 0.283) were observed in G-3 both in WW and WD conditions, respectively, while the lowest ratios (0.187 and 0.200) were observed in G-5. It has been reported that high values of root-shoot ratio represent a species' drought tolerance (Lopes et al. 2011; Greco et al. 2012), which is also observable in the results of this study. Water stress also induced an increase in root-shoot ratio in mung bean (Uddin and Parvin 2013), maize (Sharp et al. 2004), cowpea (Itani et al. 1992), and chickpea (Ali et al. 2005).

Functional Relationship Between Shoot Dry Weight and Root Dry Weight

A regression analysis was done to evaluate and quantify the relationships among the selected traits. In both WW and WD conditions, a positive linear relationship between SDW and RDW ($Y = 0.216X + 0.004$, $R^2 = 0.69$; $Y = 321X - 0.075$, $R^2 = 0.92$) was observed. The co-efficient of determination (R^2) was much higher at WD than WW conditions, showing a comparatively weak relationship among traits in WW condition (Fig. 6). Shoot dry weight increased at the rate of 0.216 g and 0.321 g plant-1 along with an increase of 1 g plant-1 for RDW in WW and WD conditions. The R^2 values of 0.69 and 0.92 indicate that SDW for WW and WD conditions were 69% and 92%, respectively. Previous studies also showed a positive linear regression among plant height, number of pods plant^{-1}, and shoot and root biomass with seed yield of mung bean (Islam 2008) and French bean (Choudhury 2009) under well-watered and water-stress conditions.

Seed Yield

There was a significant reduction in seed yield in response to water stress in all the studied mung bean genotypes (Table 2). Under WW condition, G-3 had the highest SY (2.23 g plant^{-1}), followed by G-5 (2.15 g plant^{-1}) and G-1 (2.12 g plant^{-1}); under WD condition, G-3 also had the highest SY (1.46 g plant^{-1}). The relative decrease in SY in WD compared with WW conditions ranged from 0.20 to 0.65, depending on the genotype. The highest relative value (0.65) was observed in G-3 and the lowest was observed in G-5 (0.20). The increased relative seed yield of G-3 compared with the other genotypes was likely due to its high relative values of 100-seed weight and pod number. These findings further confirm the superior water stress tolerance of G-3. Previous studies have also shown that

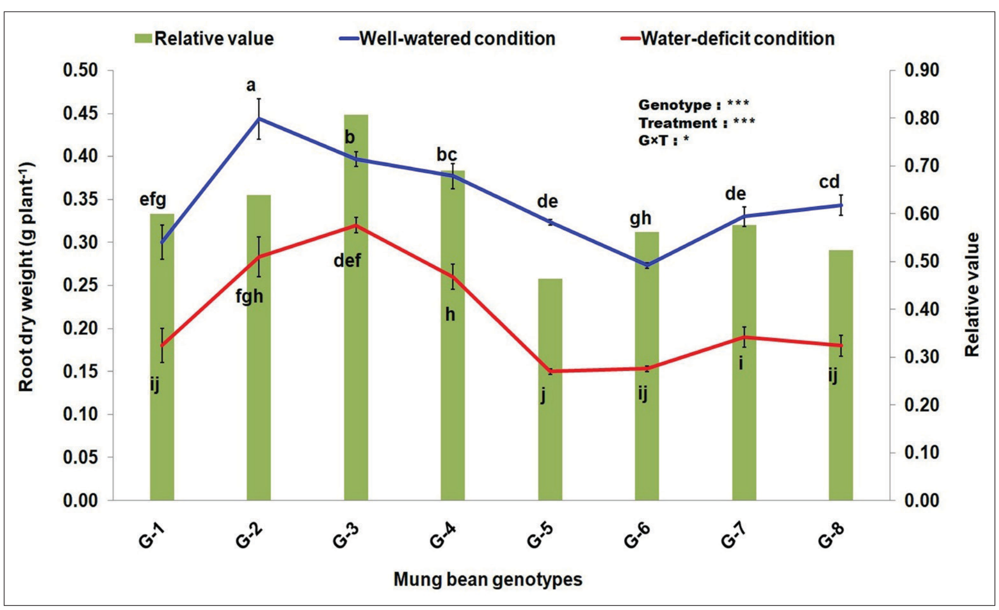


Fig. 4. Root dry weight of selected mung bean genotypes in response to WW and WD conditions.

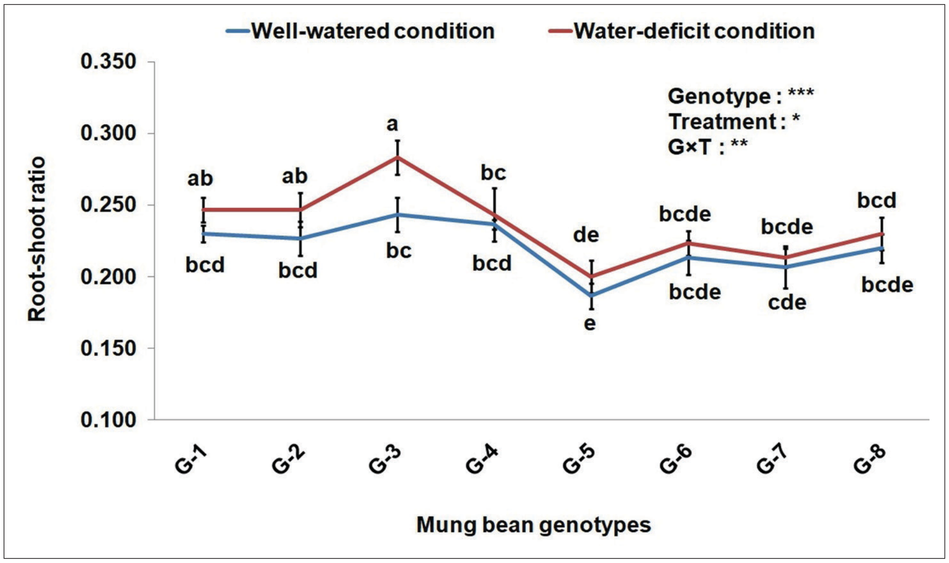


Fig. 5. Root-shoot ratio of selected mung bean genotypes in response to WW and WD conditions.

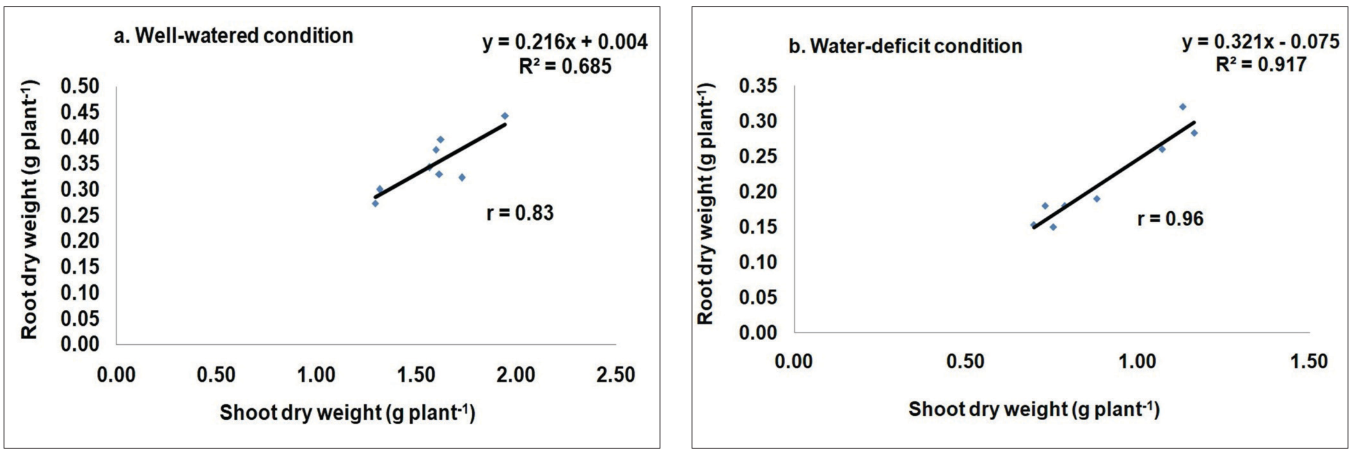


Fig. 6. (a, b). Association of shoot dry weight with root dry weight in response to WW and WD conditions (significant at *P* = 0.01; ***significant at *P* = 0.001).**

Table 2. Seed yield of selected mung bean genotypes in response to WW and WD conditions.

Genotypes	Seed yield (g plant^{-1})		
	Well-watered condition	Water-deficit condition	Relative value
G-1	2.12	1.08	0.51
G-2	1.43	0.66	0.46
G-3	2.23	1.46	0.65
G-4	1.17	0.63	0.54
G-5	2.15	0.44	0.20
G-6	1.74	0.57	0.33
G-7	1.89	0.62	0.33
G-8	1.03	0.46	0.45
LSD (5%)	0.21		
CV (%)	9.97		
LS	***		

Where: LS = level of significance; *significant at *P* = 0.05; ***significant at *P* = 0.001.

mung bean yield decreased by 64% and 34% under WD stress of 75% and 50% of field capacity, respectively (Ambachew et al. 2014). The results are consistent with previous research on mung bean (De Costa and Shanmugathasan 1999), faba bean (Ricciardi et al. 2001), and common bean (Simsek et al. 2011), all of which reported that water stress during the reproductive stages, especially at flowering and pod formation, greatly diminished grain yield. Previous studies have also shown that the reduced number of pods per plant was due to a higher rate of reproductive organ abortion, which led to lower seed yield under drought stress (Graham and Ranalli 1997; Kokubun et al. 2001; Terán and Singh 2002; Liu et al. 2003; Liu et al. 2004). Based on the results of this study, G-3 has shown itself to be a relatively drought-tolerant mung bean genotype due to its superior performance in the studied traits, while G-5 can be considered the most drought-susceptible genotype.

Seed Protein Content and Protein Yield

The seeds' protein content and protein yield were measured at harvest. Both traits showed significant differences among the studied genotypes and stress levels (Table 3). Protein content was higher under WD condition, with the highest values observed in G-4 (30.50%) and G-8 (30.53%). The highest differences were recorded in G-5 (3.35) and G-6 (2.09), while the lowest differences were recorded in G-3 (1.02) and G-4 (1.27). This means that G-5 and G-6 were more affected by water stress than G-3 and G-4. Nonetheless, G-3 was the most tolerant and G-5 was the most susceptible genotype due to the respective values of seed protein content. These are in line with the results of previous studies on spotted bean (Bayat et al. 2010), red bean (Mohammadzadeh et al. 2011), mung bean (Habibzadeh and Moosavi 2014), and in black gram (Baroowa and Gogoi 2015), where it was observed that the amount of seed protein increased significantly under water deficit stress. In addition, protein contributes to a plant's resistance under water deficit stress (Mathur and Vyas 1995).

Protein yield significantly decreased under WD condition (Table 3). Among the genotypes, G-5 showed the highest relative reduction in PY (75.90%), while G-3 had the least relative reduction in PY (31.95%). Therefore, G-3, which had a higher PY under WD condition, can be modified to withstand drought and be used as a drought-tolerant material for future crop enhancement initiatives. The results are consistent with earlier studies on mung bean (Allahmoradi et al. 2011), black bean (Nielsen and Nelson 1998), and common bean (Rosales-Serna et al. 2004), where it was found that protein yield decreased as water deficit stress increased.

Table 3. Protein content and protein yield of seeds of selected mung bean genotypes under WW and WD conditions.

Genotypes	Protein content (%)			Protein yield (g plant^{-1})		
	Well-watered condition	Water-deficit condition	Difference over stress	Well-watered condition	Water-deficit condition	% reduction
G-1	26.26	27.56	1.30	55.62	29.68	46.64
G-2	25.73	27.13	1.40	36.81	17.91	51.34
G-3	26.33	27.35	1.02	58.75	39.98	31.95
G-4	29.23	30.50	1.27	34.35	19.09	44.43
G-5	26.60	29.95	3.35	57.19	13.78	75.90
G-6	28.36	30.45	2.09	49.35	17.31	64.92
G-7	25.03	26.54	1.51	47.17	18.95	59.83
G-8	28.95	30.53	1.58	29.76	12.83	56.89
LSD (0.05)	1.58			4.90		
CV (%)	3.26			8.40		
LS	ns			***		

Where: LS = level of significance; ns = non-significant at P = 0.05; ***significant at P = 0.001.

Correlation of Seed Yield with Phenology, Growth, and Protein Yield Traits

Correlation was computed for seven phenology, growth, seed yield, and protein yield traits to assess the possible relationship between any two traits (Fig. 7). The correlation analysis showed positive and negative correlations among the traits. Days to maturity (DM), SDW, and RDW all exhibited a strong and positive relationship with days to flowering (DF), while DF was found to have a negative insignificant association with PY and SY and a less positive insignificant relationship with the RSR. Positive non-significant correlations were found between the DM and the SDW and RSR, negative non-significant correlations with the PY and SY, and positive and significant correlations with the RDW. There was a positive significant association between the SDW and the DF and RDW, a negative non-significant correlation with the PY and SY, and a positive non-significant correlation with the RSR. The trait RDW exhibited significant positive correlations with DF, DM, and RSR, while showing non-significant positive correlations with PY and SY. The RSR and other remaining qualities were found to have positive non-significant correlations, while PY exhibited positive correlations with RDW and RSR. Understanding the relationships between various parameters is essential for the simultaneous improvement and selection of these traits; for example, if two traits are positively associated, selection for one trait will consequently benefit the other (Varma 2016). A significant correlation among number of pods per plant (NPPP), number of seeds per pod (NSPP), hundred seed weight (HSW), and SY of mung bean was also reported in treatments under control and water stress conditions (Islam 2008). Hence, correlation analysis provides valuable insights into the nature and extent of the relationships among various traits. It is also helpful in selecting suitable genotypes for any crop improvement program.

Cluster Heatmap Analysis

A heatmap is a two-dimensional data visualization technique presented in the form of a diagram where values are displayed as color intensities. This is useful in determining genotypes that are tolerant to water deficit stress, since the cluster heatmap analysis also groups various traits between genotypes (Nazari and Pakniyat 2010; Dehbalaei et al. 2013; Liu et al. 2015; Sakinah et al. 2021). In this study, the analysis was performed with the relative values of the studied traits using the kmeans clustering method. Then, two types of dendrograms, namely a horizontally directed genotype dendrogram and a vertically directed trait dendrogram, were partitioned (Fig. 8). The subsequent clustering of the selected genotypes clearly demonstrates the trait variability for each genotype according to relative trait values of the WW and WD treatments. A light-to-dark-red color intensity with positive values in all studied

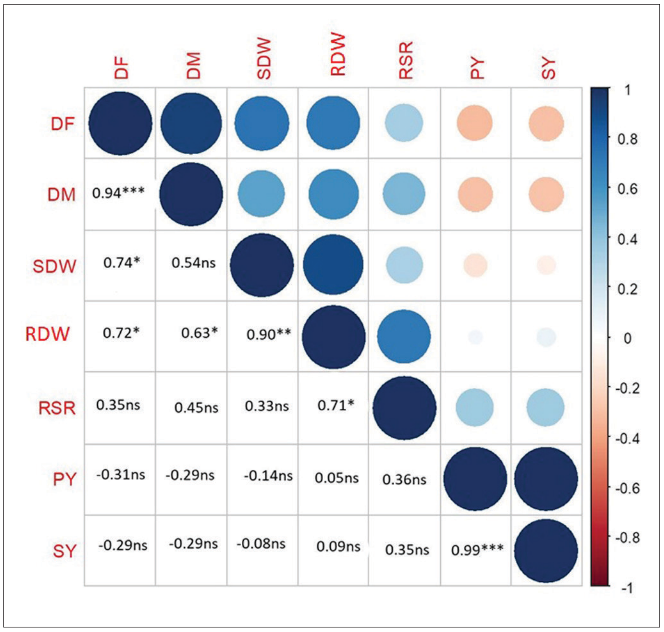


Fig. 7. Correlation for yield and yield-related traits of selected mung bean genotypes. DF = days to flowering; DM = days to maturity; SDW =shoot dry weight; RDW = root dry weight; RSR = root-shoot ratio; PY = protein yield; and SY = seed yield.

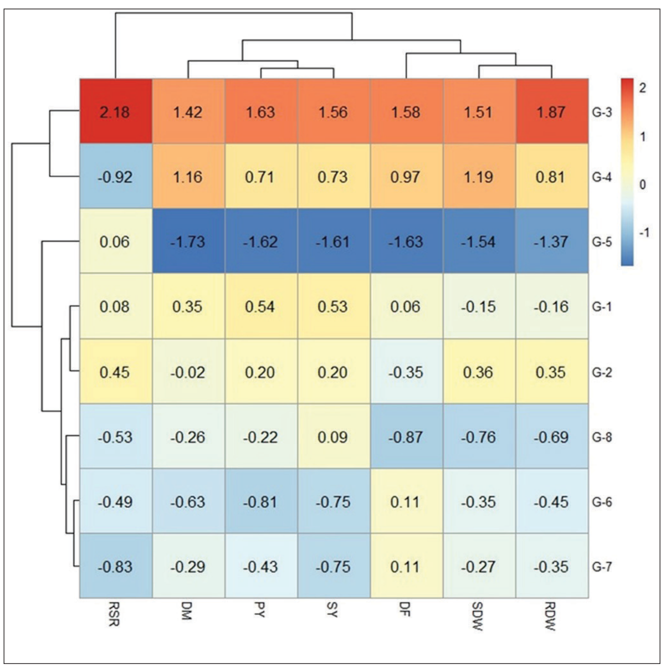


Fig. 8. Correlations among the selected mung bean genotypes, classified as either sensitive or drought-tolerant, and measured variables. Relative values obtained from the studied traits were normalized and clustered. The heatmap's color intensity, which represents a normalized relative scale from -1 to +2, illustrates the link between genotypes and traits. Darker red denotes resilience to water deficit stress, while darker blue denotes sensitivity.

traits is displayed for G-3, indicating WD stress tolerance. This is followed by G-4 which obtained positive values in most of the traits except for RSR. A darker blue color intensity with higher negative values for most of the studied traits was found in G-5, representing a higher susceptibility to WD stress than other genotypes. These results are similar to earlier findings where G-3 was found to be water stress-tolerant as indicated by the heatmap generated from the hierarchical cluster analysis, which was performed on morphological, physio-biochemical, and plant water status traits. Conversely, G-5 was found to be susceptible (Islam et al. 2023).

CONCLUSION

This study assessed the phenology, growth, and protein yield traits of mung bean related to drought tolerance by observing eight mung bean genotypes grown under well-watered (WW) and water-deficit (WD) conditions. Results showed that G-3 (BMX-08010-2) had the highest tolerance to drought due to its delayed flowering and maturation stages. Under WD condition, it also had the smallest decrease in shoot dry weight and root dry weight and had the highest seed yield and protein yield. Conversely, G-5 (BARI Mung-1) was identified as the most drought-susceptible genotype, exhibiting opposite trait outcomes. The hierarchical clustering analysis using a two-dimension heatmap further confirmed the drought tolerance and susceptibility of these genotypes. Hence, G-3 is recommended for inclusion in varietal improvement programs targeting water-deficient farming areas.

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