

**Evaluation of Drought tolerance in gamma
mutagenised wheat(*Triticum aestivum* L.) genotypes under
controlled condition**



THESIS SUBMITTED IN PARTIAL FULFILMENT OF THE
REQUIREMENTS FOR THE AWARD OF DEGREE OF

Master of Science (Agriculture)
in
Genetics and Plant Breeding

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Dedicated to

My Parents

Mr. Shakti Singh

Mrs. Maya Singh

&

Advisor

Prof. V.K. Mishra



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To

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Dear Sir,

We have great pleasure in forwarding the thesis entitled “**Evaluation of Drought tolerance in gamma mutagenised wheat (*Triticum aestivum* L.) genotypes under controlled condition**” submitted by **Ms. KAMINEE SINGH, I.D. No. 20412GPB012, Enrollment Number-429466**, in partial fulfilment of the requirements for the degree of **Master of Science (Agriculture) in Genetics and Plant Breeding**, Department of Genetics and Plant Breeding, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi and placing on record that she has completed the requisite residential requirements as contained in the statute of the University.

I certify that the entire scheme of investigation presented herein was planned and carried out solely by the candidate under my guidance and supervision. The data presented in the thesis, to the best of my knowledge and belief, are genuine and original.

Thanking you.

Forwarded by

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Yours faithfully,

(Prof. V.K. Mishra)
Supervisor/Advisor



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By

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Date:

Place: Varanasi

(**Kaminee Singh**)

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LIST OF ABBREVIATIONS

| | |
|---------------|------------------------|
| % | Percentage |
| Cm | Centimeter |
| i.e. | In other words |
| <i>et al.</i> | and co worker |
| R | Correlation |
| SD | Standard Deviation |
| Ph | Potential of Hydrogen |
| μ | Micronesia |
| MPa | Mega Pascal |
| °C | Degree Celsius |
| G | Gram |
| Ha | Hectare |
| SE | Standard error |
| Hrs | Hour(s) |
| ml | Mililiter |
| MT | Metric Tonne |
| STI | Stress Tolerance Index |

INTRODUCTION

Wheat (*Triticum aestivum*L.) is the dominant crop in temperate countries grown for human food and livestock feed. It is consumed mostly in the form of bread as “Chapatti”. Wheat straw is used as a feed for cattle. It is a hexaploid ($2n=42$) cereal crop belonging to the grass family ‘Poaceae’. In temperate climate it is mainly cultivated as a rainfed crop, in the subtropics with winter rainfall, in the tropics near the equator, in the highlands with altitudes of more than 1500 m and in the tropics distant from the equator where rainy season is long, it is cultivated as a winter crop. India is the 2nd largest producer of wheat in the world with annual production of 107.59 million tonnes. Major wheat growing states in India are Uttar Pradesh, Haryana, Punjab, Madhya Pradesh, Rajasthan, Bihar and Gujarat (APEDA, 2020-21). Among cereals crop wheat contains more protein and has a relatively higher content of niacin and thiamine. It contains carbohydrate 78.10%, protein 14.70%, fat 2.10%, minerals 2.10%, vitamins (thiamine and Vitamin-B) and minerals like zinc, iron and trace minerals like selenium and magnesium.

Wheat has been grown since the dawn of time. According to all available evidence, it appears to have originated in South Western Asia. Wheat grains are thought to have been brought to India by Aryans. Wheat originated in the Euphrates and Tigris, according to De Candolle, and spread from there to China, Egypt, and other regions of the world. Vavilov (1950) concluded that the durum wheat originated in Abyssinia, but the entire group of soft wheat, which includes bread wheat, originated in Pakistan, South-Western Afghanistan, and the southern regions of hilly Bokhara.

Three species of wheat are mainly grown all over the India: Bread wheat; *Triticum aestivum* L. ($2n=6x=42$) which is hexaploid, *Triticum durum* L. ($2n=28$) for macaroni wheat (tetraploid), and *Triticum dicoccum* L. ($2n=14$)(diploid) for Emmer wheat. Bread wheat contributes for 87 percent of overall wheat output, durum wheat for 12% mainly grown in central India, and dicoccum wheat for less than 1% cultivated in some parts of southern India (Ninghot, C. J. *et al.*, 2016). In Indian, wheat is a rabi season crop sown from mid October to first fortnight of December under optimum range of cardinal temperature 10-

35°Celsius when the weather is favourable for growth and yield. Temperature fluctuations and moisture stress, in addition to genotypic diversity, are important factors in wheat development and duration of stress influences drought intensity in wheat(NurunnaherAkter and M. Rafiqul Islam, 2017).

The major wheat-producing nations have diverse and vulnerable agroclimatic conditions, but wheat yield and quality losses are commonly restricting under heat and drought-prone agricultural region exclusively when both stresses occur together, threatening world food security.

Drought is a complicated and recurring climate phenomena that is the least understood of all the climatic risks. It is commonly described as a "serious water deficit." By generating a hydrological imbalance and persistent dry weather conditions resulting in water shortages can leads to natural disasters, such as agricultural drought (Du *et al.*, 2013). Drought may be classified into four categories: meteorological, hydrological, agricultural, and socioeconomic. Agricultural drought is considered to be the most important of them because of its influence on food security and socioeconomic stability (Mishra and Singh, 2010; Berhanet *al.*, 2011; Rasul *et al.*, 2012). Globally impact of heat and drought stress on wheat production is estimated to be 5.5% and 12% respectively. The weather forecast predicts rise in global temperature by 0.6-2.5°C by 2050 and 1.4–5.8°C by 2100, accompanied by increased drought severity, the frequency and extent of these losses may grow (Intergovernmental Panel on Climate Change (IPCC). Fourth Assessment Report: Climate Change. Geneva; 2007).

Wheat responds differently to growth stages and hence this could be used for effective screening of water-stress resistant genotypes.Stresses can be applied throughout the tillering, booting, and grain development stages(Farooq *et al.*, 2009; Ahmad and Prasad, 2011).Water loss can reduce leaf water potentials, resulting in decreased turgor pressure, stomatal conductance, and photosynthesis, as well as reduced growth and yields. There are a number of physiological characteristics that may be used to have been found that contribute to sustained development under water stress (Garcia del Moral *et al.*, 2003).

Through the generation of beneficial and heritable variation, mutation induction has the potential to widen the genetic basis of qualitatively and quantitatively inherited characteristics, such as drought tolerance. It has previously been used to create better and

well-adapted variations from elite germplasm of diverse crops (Shu *et al.* 2012). Wheat variety HD2967 is well known for its good yield and adaptability all over the country. By using gamma rays HD2967 is mutagenized and its progenies were screened for drought tolerance performance and best performing genotype is further evaluated through various parameters for water stress.

Since 18th century, hydroponics, or the 'water culture' of plants, has been employed in both scientific and commercial cultivation system. Hydroponics was established as part of early plant nutrition research and is currently utilized effectively on a wide scale by commercial farmers of fast-growing horticultural crops such as lettuce, strawberries, tomatoes, and carnations. Hydroponics is an excellent research tool for studying plant responses to abiotic conditions such as salt, boron and aluminum toxicity, nutrient shortages, and drought. Hydroponics has the benefit of being able to carefully regulate treatments, consistently assess plant responses. Since maintaining controlled condition could be accessible to grow the crops irrespective of season, hydroponics was found as excellent tool for research studies in seasonal field crops. The roots of hydroponically grown plants are freely accessible, enabling for the examination of morphological features, short-term uptake studies, and the collection of root exudates for investigation. However, it's important to understand that hydroponics is a distinct, man-made technique for growing plants, not a replacement for soil. (Anshoriet *al.*, 2018).

Root traits are critical in the production of novel wheat genotypes with increased drought tolerance, nutrient and water absorption efficiency, lodging resistance, and mineral toxicity tolerance. With rising demand for wheat production at a time when water and soil nutrients are becoming limited resources worldwide, these features are especially important for wheat adaptation to marginal habitats (Manske, G. G., and Vlek, P. L., 2002).

Depending on the environmental conditions, the traits and QTLs related with root-system architecture gives a perspective on root system design in response to water scarcity (Varshney *et al.*, 2014). It combines genetic and genomic methodologies to examine quantitative trait loci (QTLs) related with root system architecture (RSA) and beneficial root characteristics can be exploited in crop improvement program for maximal yield under water stress condition. Crop productivity has been enhanced through various research programmes in which root quality traits are introgressed into best performing genotype. (Varshney *et al.*, 2016).

Identification of wheat genotypes that can withstand low water conditions is critical for increasing wheat output, which can only be accomplished by maximising genetic potential from existing wheat germplasm. Understanding character associations for seedling qualities under water deficiency situations is also crucial for determining yield limiting variables(Ahmad M.*et al.*,2013). The goal of this study was to find wheat genotypes that could withstand high levels of water stress.

Screening genotypes based on seedling attributes is simple, inexpensive, and less time-consuming. Seedling attributes, on the other hand displays moderate to high variability with additive gene action across environments (Rauf *et al.*, 2008), allowing for efficient selection at an early stage. In deciding the production of a crop in a short period of time, a vigorous seedling will give significant results. (Noorka and Khaliq, 2007).

Understanding the importance of early crop establishment and seedling development conferring resistance to water stress "Screening of gamma mutagenised wheat germplasm for drought tolerance" was conducted under controlled conditions with the following goals in mind:

- a) To assess the performance of several wheat genotypes when exposed to control and water stress condition.
- b) To identify drought-tolerant mutant wheat genotypes.



REVIEW OF LITERATURE

Wheat is one of the most cultivated crops all over the world and its production is affected with several biotic and abiotic factors. Maximum yield loss in wheat is due to drought as a result of prolonged moisture stress.

Study was conducted to assess climate change and productivity of wheat productivity in India's Punjab province from 1970 to 1999 (Vyankatrao, 2017; Guntukula, 2019; Kumar and Sidana, 2019; Praveen and Sharma, 2019a; Praveen and Sharma, 2019b; Singh, 2020; Kumar *et al.* 2021b). Minimum and maximum temperatures showed detrimental impact on wheat yields. The findings suggested that highest temperature has a detrimental impact, whereas the lowest temperature and rainfall had a favorable impact on wheat production (Bhardwaj *et al.*, 2022).

Hexaploid nature of wheat and redundancy in gene functioning make genetic advancement of desired phenotype time-consuming and in some situations impossible due to gene linkage or gene drag (Li *et al.*, 2020). Enhancing the resilience of wheat production and minimizing environmental fluctuations is critical to ensuring global food and ecosystem security.

The most common methodology for screening of seedlings at early stage for drought tolerance is by inducing moisture stress through poly ethylene glycol (PEG 6000) in the solution media (Rauf *et al.*, 2006). PEG 6000 molecules are inert, non-ionic, and almost impermeable chains that have been employed to cause water stress in agricultural plants without inflicting major physiological harm (Carpita *et al.*, 1979). Polyethylene glycol (PEG) alter the osmotic potential of solution media culture and can be used to cause plant water deficit in a regulated way, which is ideal for experimental methods (Lagerwerff *et al.*, 1961). Earlier research aimed at identifying drought-tolerant wheat genotypes using varying PEG 6000 concentrations

revealed considerable changes in seedling characteristics (**Rauf *et al.*, 2006; Singh *et al.*, 2008**).

2.1 Water stress in wheat and its performance

Karim M. *et al.* (2000) have found that water stress treatment did not influence the dynamics of grain moisture content. The smaller grain in water stressed plants was presumably due to shortened linear grain growth phase as well as slower grain growth. Water stress had almost no effect on grain setting, but it did have a significant impact on grain weight. When compared to irrigated plants, the grain production of stressed plants was reduced by 65 percent.

Shams K. *et al.* (2018) has revealed that drought has a significant impact on plant growth, grain yield and quality causing molecular, biochemical, physiological, and morphological changes (**Zarafshar *et al.* 2014**). Wheat production suffers from a severe moisture deficit from seedling to full maturity when subjected to drought stress (**Bilal *et al.* 2015**).

Del Pozo *et al.* (2016) investigated various physiological traits as essential factors linked to yield potential and water stress performance. In wheat, increased yield potential and stress adaptability have mostly been achieved by empirical grain yield selection. However, there is evidence that phenotyping with physiological characteristics, in addition to agronomic variables, can aid in the identification of selected features that speeds up breeding for yield potential and drought performance (**Araus *et al.*, 2002, 2008; Fischer, 2007; Foulkes *et al.*, 2007; Cattivelliet *et al.*, 2008; Fleury *et al.*, 2010**). Wheat stress accelerates chlorophyll breakdown, resulting in a reduction in leaf area and canopy photosynthesis. Evergreen genotypes with delayed leaf senescence have been shown to perform better under drought condition (**Rivero *et al.*, 2007; Lopes and Reynolds, 2012**).

Meena *et al.* (2015) shown that moisture stress is a key restriction in wheat production across India. Climate change and erratic rains aggravate the issue in moisture-stressed areas. Wheat genotypes that can provide higher yields across a wide range of optimal and sub-optimal water availability are preferred. The purpose of this

study was to evaluate various moisture stress selection indices and their applicability in identifying drought tolerant wheat genotypes that can adapt to various moisture stressed environments in India's 11 wheat growing zones, which include the North Western Plain Zone, North Eastern Plain Zone, Central Zone, and Peninsular Zone. Different irrigation regimes were used to assess a range of wheat genotypes under moisture stress conditions.

2.2 Drought tolerance at early stages of seedling establishment

Blum A. *et al.* (1980) reported significant variation among wheat cultivars under water stress at seedling stage. Seedling growth tolerance was not connected to germination responses under stress across cultivars. The use of PEG-containing nutrient solutions to screen for water stress tolerance in developing seedlings was shown to be effective.

Dhandaset *al.* (2012) indicated that seed vigour index and shoot length among the most susceptible traits to drought stress, followed by root length and coleoptiles length. With an increase in osmotic stress, the rate of seed germination and ultimate germination percentage, as well as the amount of water absorbed by the seeds, were all significantly reduced.

Rauf M. *et al.* (2007) studied wheat genotypes response to water stress in terms of germination, seedling elongation, seedling fresh weight and dry weight, and emergence over the entire establishment period. Further investigated the relationship between water stress level and drought tolerance mechanisms. Under control condition, seedling development has been regarded as an appropriate growth stage for testing drought tolerance in wheat. It might be postulated that the presence of elevated concentrations of PEG during seedling growth impairs the developmental properties and survival of wheat seedlings.

Sapra, V. T. *et al.* (1991) observed that during the seedling test, a significant association was with plant height, fresh weight, and dry weight stress indicators. Under stress, cultivars that grew taller accumulated more dry matter and had higher

germination and water absorption stress indices, demonstrating that height is a reliable predictor of cultivar performance under such conditions.

Bibi, A. et al. (2012) showed that the roots detect a water deficiency and begin to synthesize ABA within an hour of the water stress induction. Within minutes to hours, ABA is transferred from roots to leaves via xylem. In plant, variation in root length is an essential characteristic for drought resistance. In general, cultivars with longer root and lateral growth are drought resistant.

2.3 Root system architecture for drought tolerance in wheat

Root system architecture (RSA) has been the target of wheat research and breeding to develop drought tolerant cultivars.

Kulkarni, M. et al. (2017) investigated drought tolerance mechanisms in wheat cultivars which include deeper root system, higher biomass, improved stomatal conductance over transpiration (Chipilskiet al., 2012), improved osmo-protective and antioxidant response and most importantly, a better coordination of positive and negative gene expression regulation. Plant developmental reaction to drought stress expresses itself in increased root growth and reduced shoot growth, resulting in a higher root: shoot ratio (Sharp et al., 2004; Yamaguchi and Sharp, 2010; Xu et al., 2013). Under drought stress, zinc finger proteins (ZFPs) with a QALGGH conserved domain have been linked to gene expression regulation (Cheuk and Houde, 2016). The role of ZFPs in drought resistance has been extensively established in wheat (Cheuk and Houde, 2016), rice (Zhang et al., 2014), are known to help plants cope with drought stress (Huang et al., 2012; Luo et al., 2012).

Pask, A., and Reynolds, M. (2013) reported transpiration efficiency is connected to root system depth and both of these attributes must be improved at the same time simultaneously. Crop cultivars that can derive moisture from deeper root zones (60–120 cm) have higher stomatal conductance, allowing the canopy temperature to be kept at moderate range. Wheat cultivars with a mix of deep root biomass and Trans-
evapotranspiration traits will be more drought resistant.

Djanaguiraman, Met al. (2019) reported that drought-stressed plants with large root systems can increase water intake and reduce the effects of drought stress. Because roots are a primary sink for assimilates, reduced root biomass may increase the availability of assimilates for aboveground parts, particularly grains. Seed set percentage, individual seed weights, and seed yields per plant all were higher in drought-tolerant lines with longer lateral roots. Although total root length was linked to all of the above factors, the link between total root length, seed yield per plant was higher than the others, indicating that these features may be physiologically related. Deep, wide-spreading, much-branched root system is one of the important properties of drought tolerance. Plants adapt to increased rooting depth and root biomass in order to gain access to more accessible soil moisture.

Nielsen et al. (1997) concluded that drought stress causes root system flexibility by increasing the amount of fibrous roots, lowering lateral root diameter, and causing changes in root biomass, in addition to deep and proliferative rooting. Root system architecture (RSA) is critical for crop performance when water and nutrients are scarce (Paez-Garcia and colleagues, 2015). Wheat roots are divided into two types: seminal (embryonal) roots and nodal (crown or adventitious) roots (Chochoiset al., 2015). In cultivated wheat, seminal roots contain one primary root, two pairs of symmetric roots and in rare cases, a sixth central root. At the tillering stage, nodal roots are usually visible (Esau, 1965). Seminal roots go deeper into the soil than nodal roots and are active throughout the plant's life cycle, aiding in the absorption of moisture from deeper soil layers (Manschadi et al., 2013).

2.4 Physiological and molecular mechanism imparting drought

Lal, M. K., et al. (2021) concluded that germination, seedling establishment, source-sink activity, leaf area, shoot and root development are morphological factors that are influenced heat stress. Heat has an impact on physiological factors such as photosynthesis, respiration, leaf senescence, water and nutrient relations. Heat stress causes the production of reactive oxygen species, which disrupt the thylakoid, chloroplast, and plasma membrane systems. The formation of photo assimilates and their distribution are affected by photosystem deactivation, reduced photosynthesis

and inactivation of rubisco. Further anthesis, grain filling, grain size and maturity of wheat were adversely affected reducing crop output. The interaction of many systems, including antioxidants and hormones is critical in conferring heat stress resistance in wheat. To boost the efficiency of water metabolism, wheat flag leaves undergo morphological and physiological modifications such as leaf rolling (Hasanuzzaman *et al.* 2013). The denaturation of vital enzymes involved in photosynthesis and carbon assimilation such as Ribulose-1,5-bisphosphate (RuBP) carboxylase/oxygenase (Rubisco), sucrose phosphate synthase, PEP carboxylase and invertase, occurs when cells are exposed to extreme high temperatures for seconds or minutes. Prolonged heat exposure causes organ damage and death, as well as leaf loss and flower abortion (Kumar *et al.* 2019a).

Fleitaset *al.* (2020) studied seed germination, root and leaf emergence, stem growth, floral initiation, pollination, fertilization, seed production, and seed quality are all affected by moisture stress during wheat growth and development.

Bresticet *al.* (2012) investigated PSII thermostability and acclimation effects in 30 winter wheat genotypes utilising rapid chlorophyll fluorescence kinetics before and after dark heat treatment. Based on this, all genotypes were categorised into low, middle, and high thermostability groups, and it was established that the lipid content of thylakoid membranes is critical for heat thermostability.

Shah and Paulsen (2005) observed under stress reduction in vegetative shoot plant development and accelerated leaf senescence in wheat was due to reduced photosynthetic ability. There was structural alterations in the chloroplast under heat stress in wheat plants, which were followed by vacuolar collapse and eventually, loss of plasma membrane integrity, causing cellular homeostasis to be disrupted.

Bowne *et al.* (2012) investigated that drought has a significant impact on the physiology of grain crops. In many cases, both at the cellular and metabolic levels. The concentration of chemicals is raised for osmo-protective actions to avoid desiccation. By enhancing the enzymatic activity, resulted in lowering reactive oxygen molecules in the cell. They used a targeted GC-MS methodology to find chemicals that differ in

three ways. The bread wheat cultivars have variable levels of tolerance to a variety of stresses.

Fahad *et al.* (2019) observed water intake and transpiration regulate the temperature of plant tissue, resulting in a stable water content in the tissue. However, when there is a scarcity of water, a rise in temperature might be fatal. Leaf relative water content (LRWC), leaf water potential (LWP), rate of transpiration and stomatal conductance (SC) are all regulated by leaf and canopy temperature under heat stress (Farooq *et al.* 2011)

Sairam *et al.* (2000) studied the impact of plant antioxidant systems for drought tolerance in three wheat genotypes. After anthesis, water stress increased lipid peroxidation and reduced membrane stability, as well as chlorophyll content and carotenoid levels. Antioxidant enzymes such as ascorbate peroxidase, glutathione reductase, and non-specific peroxidase all increased dramatically in response to water stress. The susceptible genotype WH 542 had the lowest antioxidant enzyme activity, membrane stability, and chlorophyll and carotenoids content under water stress, whereas the genotype PBW175, which had the highest ascorbate peroxidase, glutathione reductase, and peroxidase activity, had the lowest lipid peroxidation (malondialdehyde content), the highest membrane stability, and the highest chlorophyll.

2.5 Osmotic adjustment

Drought resistance is thought to be mostly dependent on osmotic adjustment (OA) (e.g. Ludlow and Muchow, 1990). Morgan and Condon (1986) established that wheat cultivars have persistent genetic variances in OA and that high OA cultivars perform better under drought stress than low OA cultivars. Despite this evidence, verbal arguments were occasionally made that the results were confined to the genetic materials and test circumstances utilised in the research.

Blum, A. *et al.* (1999) demonstrated that wheat cultivars varied regularly in terms of OA and that these variations might be linked to variances in plant output during pre-

flowering drought stress. As a result, it is argued that OA can be a major component of drought tolerance in wheat when grown in the right climate.

Gupta, N. et al. (2014) reported that to develop drought-tolerant crops, osmolyte accumulation (OA), also known as osmotic adjustment, has long been advocated as a selection criteria in conventional breeding or through marker assisted selection and genetic engineering programmes (Ludlow and Muchow 1990; Zhang *et al.* 1999). Despite the fact that the role of osmolytes in the physiological process is unknown, Osmolyte accumulation (OA) is frequently associated with osmotic adjustment. Furthermore, OA has been linked to preservation of turgor as well as dehydration tolerance. All of these tactics were supposed to improve plant resilience to abiotic conditions such as water deficiency.

2.6 Mutagenesis approach for drought tolerance in wheat

Sen A. et al. (2017) found that for the development of agricultural crops, mutation breeding is an alternate strategy. To develop drought-tolerant mutant lines, a large number of Sagittario bread wheat seeds (*Triticum aestivum* L.) were treated to gamma ray irradiation (200 Gy). Polyethylene glycol (PEG) 6000 was administered to the M2 and M3 individuals in vitro to assess drought tolerance. Drought stress was generated by a 50% reduction in irrigation water supplied to M3 and M4 plants in a greenhouse, except in well-watered (100%) control plants.

Rai, M. et al. (2011) investigated that the physiological and biochemical aspects of mutant plants under stress are just as significant as their molecular characterization. Studying the constituent of stress-related metabolites and antioxidant enzymes has shown to be a beneficial approach for tracking stress-related changes. These adaptation processes can take time to get activated after drought stress, resulting in ideal cellular conditions for normal plant development (Rai and colleagues 2011).

Kontz, B. et al. (2009) concluded that drought resistance in mutated winter wheat lines was tested by withholding water for 3 weeks after 9 weeks of normal development and observing their reactions after re-watering. 73 percent of the 146 mutants

produced died or recovered poorly owing to water stress, whereas 27 percent recovered better than the wildtype line. This experiment allowed candidates to be chosen for future examination in order to learn more about the genetic basis of drought resistance in winter wheat.

Hallajian, M. T. (2016) reviewed that breeding for drought resilience is mainly done by recombination, mutation and molecular breeding methods. Out of which, mutation breeding is an alternative method for developing agriculturally important crops. A large set of Sagittario bread wheat seeds (*Triticumaestivum* L. cv.) were exposed to gamma ray irradiation (200 Gy) to obtain drought-tolerant mutant lines. To study drought tolerance, polyethylene glycol (PEG) 6000 was applied to the M₂ and M₃ individuals under in vitro conditions. Except in well-watered (100%) control plants, drought stress was triggered by a 50% decrease in the irrigation water applied to M₃ and M₄ plants in a greenhouse.

2.7 Screening of wheat lines under controlled condition

Ayalew, Het al. (2015) developed a plastic box(3000 ml) with holes of around 8 mm diameter drilled in the lids were used to create a hydroponic system that allowed plant growth on the surface of a solution. The boxes were filled with water/solutions, and the lids were perforated and lined with filter paper to keep the plants in place and the surface moist. A total of 838 wheat lines were tested, including control types. The Australian winter cereals collection provided bread and durum wheat genotypes, as well as wild cousins from six. A seven-block (planting time) augmented full block design with six stress and control types was adopted. Seeds were germinated for 48 hours in Petri plates coated with filter paper saturated in distilled water before being transferred to the hydroponic system. PEG 6000 was used to induce osmotic stress of 0.82 MPa. The pH of the solution was adjusted to 5.5–5.7 and the relative humidity was 65–70 % while the temperature was 25/22 °C day/night. Light intensity of 300 micro mol m⁻²s⁻¹ was supplied using cool florescent lamps in 10/14 dark and light timing using an automatic timer. The solution was being constantly aerated using an electric air bubbler. Data were recorded on root length 14 days after planting (seven days after stress treatment) using a graduated ruler.

Michael and Kaufman (1973) reported that seeds were first treated for 15 minutes with a 1.5 percent sodium hypochlorite solution. Then thoroughly washing the seeds with distilled water, any residual chlorine was removed. Fifty seeds were deposited on two layers of Whatman no. 2 filter paper with a 25 mm diameter and reproduced three times under an osmotic potential of 0 and – 10 bars established using a polyethylene glycol solution (PEG-6000). He found that under osmotic stress seed vigour index and shoot length is the most sensitive traits followed by root length and coleoptiles length whereas germination % and root to shoot ratio remain quite stable under stress.

Tavakkoli, E. et al. (2012) developed an experimental system supported by hydroponic system. Plants were cultivated in a series of tubs containing 42 PVC tubes filled with cylindrical black polycarbonate pellets (about 2–4 mm length and 1–2 mm in diameter) in cylindrical PVC tubes (4 cm diameter 32.8 cm depth). A single tank containing 80 litres of nutritional solution with two tubs were used to load tubs with 25 litres of nutritional solution every 30 minutes. The following ingredients were employed in a modified Hoagland's solution (Tavakkoliet al., 2010a): NH_4NO_3 (0.2); KNO_3 (5); $\text{Ca NO}_3.2$ (2); MgSO_4 (2); KH_2PO_4 (0.1); Na_2SiO_3 (0.5); NaFe (III) –hydroxyethylethylenediaminetriacetic acid (HEDTA) (0.05); H_3BO_3 (0.01); MnC (0.0005), and Na_2MoO_3 (0.0001). Solutions were changed in every 7th day and the pH was adjusted to 6.0. The experiment was conducted in a temperature-controlled growth chamber with day/night temperatures of approximately 23/19°C.

Dadshani, Set al. (2019) established the system for studying the reactions to salinity (Na^+ and Cl) in a hydroponics experiment. A totally randomised design with four replication was used to set up a factorial experiment with a control (0 mM NaCl) and two amounts of NaCl (75 and 150 mM). A supported hydroponic system was employed in the experiment. Plants were cultivated in a succession of 15 L tubs filled with cylindrical black polycarbonate pellets (about 2–4 mm long and 1–2 mm in diameter) in cylindrical polyvinyl chloride (PVC) tubes (4 cm diameter 28 cm depth) with a mesh bottom. These tubs each included 24 PVC tubes and were put on top of and linked to another tub with 24 L of nutritional solution. Nutrient solution was pumped from the bottom tub into the top tub every 30 minutes, where it stayed for another 30 minutes until the pump was turned off and the nutrient solution in the top

tub was emptied back into the bottom tub. Between the 'on' and 'off' cycles, the roots stayed wet, and there was no sign of water stress on the developing seedlings.

2.8 Evaluation of mutant line parameters for selecting drought tolerant lines wheat

Noorka, I. R. *et al.* (2013) investigated that when water supply was withheld, resulting in the death of the majority of the seedlings. Following that, the plants were re-watered to recover, and each replication which survived was carried on for re-growth. Different researchers measured the number of living seedlings and the number of dead seedlings on a daily basis.

Desiccation tolerance index: The seedlings were adequately hydrated until they reached the 2-3 leaf stage, which ISTA recommends for seedling assessment (Anon., 1997).

Emergence Index (EI): This is a measure of seedling emergence rate determined using the method defined by the AOSA Association of Official Seed Analysis (Anon., 1983).

Mutava (2009) suggested (i) leaf rolling and wax content, which reduce leaf temperature, (ii) yield traits like seed filling length and rate, which increase seed size, and (iii) root traits like improved root development and water absorption, which increase water intake.

Ameleworket *al.* (2015) examined drought resistance mechanisms such as drought avoidance, recovery, survival, and tolerance. Plant survival and productivity are linked to these drought tolerance systems. Drought avoidance is described as a plant's capacity to preserve water at the total plant level by reducing water loss from the shoots or absorbing water more effectively from the soil. Due to significant relationship between leaf rolling and leaf water potential, breeders can use leaf rolling as a visual scoring baseline when selecting plants for drought tolerance. The rolling of leaves is generally triggered by a decrease in leaf water potential. The degree of leaf rolling, on the other hand, is determined by the plant's capacity to adapt.

Miller et al. (1958) gave the percentage damage of cell membrane permeability in leaf tissues using the following equation:

$$\text{Per cent injury} = \frac{1 - \left(1 - \frac{T_1}{T_2}\right)}{\left(1 - \frac{C_1}{C_2}\right)} \times 100$$

T1 and T2 represent the first and second conductivity measurements for the desiccation treatment, respectively, while C1 and C2 represent the first and second conductivity measurements for the control. The coefficients of variation and heritability (Burton and Devane 1953), genetic progress (Johnson et al. 1955), and phenotypic and genotypic correlations were all calculated using the mean values for distinct features

Mohi-Ud-Dinet et al. (2021) gave the formula for tissue water content (TWC) was estimated using the method in terms of the quantity of water per unit shoot or root fresh weight.

$$\text{TWC} = (\text{Fresh wt.} - \text{Dry wt.}) / (\text{Fresh wt.} - \text{Dry wt.}) / (\text{Fresh wt.} / \text{Fresh wt.})$$

The seedling vigour index (SVI) was determined using **Hellalet et al.** method's, which was

$$\text{SVI} = ((\text{Shoot length} + \text{Root length}) \times (\text{Germination percent})) / 10.$$

Leaf relative water content (LRWC) was estimated according to the procedure of Meheret et al. The dry weights of the samples were taken till a constant weight was achieved.

$$\text{LRWC (\%)} = [(\text{Fresh wt.} - \text{Dry wt.}) / (\text{Turgid wt.} - \text{Dry wt.})] \times 100$$

Shams K. et al. (2018) established that relative water content (RWC) reported highly significant results based on the analysis of variance representing changes in leaf RWC under the impacts of water deficit stress and cultivar from the statistical viewpoint (for observation fresh weight from three youngest fully expanded leaves (flag leaves) were determined within 2 hrs after excision. Turgid weight was obtained after soaking the leaves for 16 to 18 hrs in distilled water. After soaking, leaves were quickly and carefully blotted dry with tissue paper prior to determine of turgid weight. Dry weight

was obtained after drying the leaves sample for 72 hrs at 70°C. Relative water content was calculated from the following equation:

$$\text{RWC} = [(\text{fresh weight} - \text{dry weight}) / (\text{turgid weight} - \text{dry weight})] \times 100$$

Bogaleet al. (2011) studied the differential morpho-physiological response to triggered water scarcity and the association between some of these morphological and physiological features and yield components of eighteen durum wheat genotypes grown in pots under lath house conditions. Gas exchange and chlorophyll fluorescence characteristics were considerably impacted by a lack of water. At both the anthesis and grain-filling stages, it lowered the net photosynthetic rate (Pn), transpiration rate (E), and stomatal conductance (gs). Similarly, when there was a water shortage, the value of beginning fluorescence (Fo) increased while variable fluorescence (Fv), maximum fluorescence (Fm), and optimal quantum yield fluorescence (Fv/Fm) declined. No significant correlations were found between chlorophyll fluorescence parameters and grain yield under water deficit condition.

Rampinoet al. (2006) reported that water scarcity is a severe environmental stress. The most significant limitation to plant output, having a clear impact on plant growth. The goal of this study was to compare the responses of Triticum and Aegilops seedlings. Drought stress affects the physiology and chemistry. First, the relative water content was used to determine sensitive genotypes (RWC) measurement. Additional characterisation of genotypes with differing responses to water stress was measured at the physiological level using RWC and water content. After various hours of dehydration, the weight loss rate (WLR) and free proline content were measured.

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MATERIAL AND METHOD

3.1 EXPERIMENTAL PERIOD:

The experiment was conducted during the period from December 2021 to April 2022 under controlled condition.

3.2 LOCATION OF TRIAL:

The suggested study on "Evaluation of drought tolerance in gamma mutagenised wheat lines under controlled conditions." was carried at wheat lab, department of genetics and plant breeding, Institute of Agriculture Sciences (BHU), Varanasi. BHU is located in the south-eastern portion of Varanasi, at 25°15' North latitude and 83° 09' East longitude, at an elevation of around 75.5 meters above mean sea level (MSL). The temperature begins to rise in mid-February and reaches a peak in May-June (mean maximum temperature of 43.6 °C) before gradually declining from July to December-January, with an average lowest temperature of 50 °C. The centre receives around 1100mm of annual rainfall, the most of which occurs between July and September, and the mean relative humidity is 68 percent.

3.3 EXPERIMENTAL MATERIAL:

Drought stress was assessed in this mutant population at the wheat lab, department of genetics and plant breeding, Institute of Agriculture Sciences (BHU), Varanasi

Planting materials:

In this experiment, 9 mutant wheat genotypes along with parent as mentioned in (Table 1) were used as experimental materials. These genotypes were collected from BARC, Trombay, Mumbai.

Table 3.3.1 Wheat genotypes used for evaluation of drought tolerance

| S. No. | Genotypes |
|---------------|------------------|
| 1 | HD2967 |
| 2 | TAW19 |
| 3 | TAW95 |
| 4 | TAW97 |
| 5 | TAW98 |
| 6 | TAW114 |
| 7 | TAW117 |
| 8 | TAW122 |
| 9 | TAW132 |
| 10 | TAW15 |

3.4 EXPERIMENTAL DESIGN:

Seedlings were transplanted in hydroponic culture using a totally randomised design with five replications under control and water stress. Drought was induced in this study using polyethylene glycol 8000 (PEG8000).

3.5 PREPARATION OF HOAGLAND SOLUTION:

For preparation of Hoagland solution six stock solutions were prepared viz, stock A, stock B, stock C, stock D, stock E and stock F. Chemical used for preparation of stock solutions are listed below:

Table 3.2List for preparation of Hoagland solution

| Sl. NO. | Chemical Macro Nutrients | Chemical name | Stock solution (g L ⁻¹) | Volume of stock solution L ⁻¹ of final solution (ml) | Solution |
|---------|---|---------------------------------|-------------------------------------|---|----------|
| 1 | KNO ₃ | Potassium Nitrate | 101.1 | 3 | Stock A |
| 2 | Ca (NO ₃) ₂ .4H ₂ O | Calcium Nitrate Tetrahydrate | 236.16 | 2 | Stock B |
| 3 | NH ₄ H ₂ PO ₄ | Ammonium dihydrogen phosphate | 115.08 | 1 | Stock C |
| 4 | MgSO ₄ .7H ₂ O | Magnesium sulphate heptahydrate | 246.49 | 0.5 | Stock D |
| | Micro Nutrients | | | | |
| 5 | H ₃ BO ₄ | Borane | 1.546 | 0.5 | Stock E |
| 6 | MnSO ₄ .H ₂ O | Manganese sulphate monohydrate | 0.338 | | |
| 7 | ZnSO ₄ .7H ₂ O | Zinc sulphate Heptahydrate | 0.575 | | |
| 8 | CuSO ₄ .5H ₂ O | Copper sulphate pentahydrate | 0.125 | | |
| 9 | H ₂ MoO ₄ .H ₂ O | Molybdic acid monohydrate | 0.081 | | |
| 10 | FeSO ₄ | Ferrous sulphate | 27.85 | 0.5 | Stock F |

After preparation of stock solutions, in one litre of distilled water 7.5ml of half strength of Hoagland solution was used.

3.6 Glassware:

To eliminate contamination, every glassware is cleansed with detergent and autoclaved before use.

3.7 Vertical autoclave:

It is commonly used in labs to sterilise a variety of laboratory equipment. It has a working pressure of 15 psi and a temperature of 121 degrees Celsius that is maintained for 30 minutes.

3.8 Hot Air Oven:

The drying oven technique provides new dry air into the chamber while simultaneously expelling warm damp air, allowing the samples to dry quickly. The samples are dried at 70°C for 24 hours.

3.9 Lux meter:

A lux meter is a tool for measuring light intensity. A light intensity of 250 mol m⁻² s⁻¹ was used in this experiment.

3.10 PEG solution:

Polyethylene glycol is a drought-inducing chemical which is often used to screen drought tolerant genotypes at the seedling stage. PEG alters the osmotic potential of nutrient solution, causing a water stress in the roots of the plants (Lagerwerff *et al.*, 1961; Money, 1989 and Zhu *et al.*, 1997).

PEG 8000 molecules are too tiny to affect osmotic potential, yet large enough to avoid being absorbed by plants and even to permeate intact plant tissues quickly (Carpita *et al.*, 1979).

3.11 LABORATORY EXPERIMENT:

A laboratory experiment was conducted by simulating drought conditions with a 20% polyethylene glycol concentration (PEG-8000). A hydroponic system was

created utilising trays and thermacol with holes ranging from 1-1.5cm in length and breadth, and thermacol was glued with mesh to encourage seedling growth and maintain plants in place. To keep the seeds wet, the experiment was set up using trays filled with water/solution. The solution's PH was set to 6.5 to 7.5, and the relative humidity was set at 65-70 percent. The day/night temperature was 24/22 °C. Cool florescent lights were used to provide light intensity of (250 mol m⁻² s⁻¹) in 10/14 dark and light timing utilising an automated timer (Ayalewet *et al.*, 2015).

PROCEDURE

- Healthy and equal-sized genotype seeds were chosen.
- Seeds were soaked in distilled water overnight (12 hours), then rinsed three times with distilled water the next day.
- The seeds were soaked in a 1 percent sodium hypochlorite solution for 4-5 minutes before being rinsed three times with distilled water.
- Seeds were germinated first in petri dishes coated with whatman paper and kept wet enough to germinate.
- Seeds were cultivated in half strength Hoagland solution for the first 7 days, implanted in a micropipette(1.5cm depth). The nutrition solution was changed on a weekly basis.
- At 14 days, osmotic stress was induced by adding PEG8000 to the nutritional solution at a concentration of 20% for 7 days.
- Throughout the experiment, control seedlings were cultivated in half strength Hoagland solution.
- After one week, replace the stress and control tray solutions with half strength Hoagland solution and grow for four days.
- After the 24th day, harvesting was completed and observations of different parameters were recorded.

3.12 RECORDED OBSERVATION

3.12.1 Length of Shoot:

A measuring scale in cm was used to determine the shoot length of all genotypes of control and treatment seedlings.

3.12.2 Length of Root:

Using a measuring scale in cm, the root length of all genotypes of control and treated seedlings was measured.

3.12.3 Weight of fresh shoots:

The shoot weight of two seedlings was measured and averaged after the root and shoot were separated.

3.12.4 Weight of fresh roots:

Root weight of two seedlings was measured and averaged after root and shoot separation to achieve single plant root weight.

3.12.5 Shoot dry weight:

For two days, all fresh weighed shoot samples were maintained under aluminium foil in a hot air oven at 70 °C. After two days, samples were removed from the oven, weighed, and the dry weight of all samples was calculated in gram.

3.12.6 Root dry weight:

After drying the root in a hot air oven at 70°C and wrapping it in aluminium foil, the dry weight of the root was calculated. After two days, the samples were removed from the oven, weighed, and the weight of all samples was measured in gram.

3.13 Percent damage:

After 25-day percent damage for each genotype was calculated by dividing the length of dried leaf to total length of leaf of the same plant.

$$\text{Percent damage} = \frac{\text{lengthofdriedleaf}}{\text{Totallengthofleaf}} \times 100$$

3.14 INDICES:

3.14.1 Shoot length stress tolerance index (SLSTI%):

After 25-day shoot length stress tolerance index for each genotype was calculated by using formula (Raza *et al.*, 2012)

$$\text{S.L.S.T.I} = \frac{\text{Shootlengthofstressedplant}}{\text{Shootlengthofcontrolplant}} \times 100$$

3.14.2 Root length stress tolerance index (RLSTI%):

After 25-day root length stress tolerance index for each genotype was calculated by using formula (Raza *et al.*, 2012)

$$\text{R.L.S.T.I} = \frac{\text{Rootlengthofstressedplant}}{\text{Rootlengthofcontrolplant}} \times 100$$

3.14.3 Dry matter stress tolerance index of shoot:

$$\text{D.M.S.T.I of shoot} = \frac{\text{Drymatterofstressedshoot}}{\text{Drymatterofcontrolshoot}} \times 100$$

3.14.4 Dry matter stress tolerance index of root:

$$\text{D.M.S.T.I of root} = \frac{\text{Drymatterofstressedroot}}{\text{Drymatterofcontrolroot}} \times 100$$

3.15 Chlorophyll Content(Measured by SPAD Meter)

To determine the greenness or relative chlorophyll content of leaves, the SPAD chlorophyll content was utilised. The metre measures the amount of light (peak wavelength: roughly 650nm; red LED) absorbed by the tissue sample and provides an immediate and non-destructive measurement on the plant. To compensate for the thickness of the leaf, a second peak (peak wavelength: roughly 940nm; infrared LED) was produced in tandem with the infrared LED.

3.16 Statistical Analysis

The data collected for various features was statistically evaluated in order to determine the morpho-physiological character of spring wheat under drought stress. All of the characters' mean values were determined, and analysis of variance and variability were carried out. Data on physiology, chlorophyll, and other parameters were evaluated. Following a test of homogeneity of variances, a combined analysis of variance was performed. Pearson's correlation coefficients (r) were determined individually for the stress and control treatments to reflect the degree of the correlations among agronomic parameters, chlorophyll, and proline content. To discover influential qualities for selection, SPSS was used to perform a principle component analysis (PCA) based on the correlation matrix.

The statistical methods adopted were as follows:

- Analysis of variance for the design of experiment.
 - Selection parameters
- A. Mean
 - B. Range
 - C. Standard deviation
 - D. Standard error
 - E. Correlation coefficient

3.16.1 ANOVA (Analysis of variance):

The measure of variability is variance. The two major goals of analysis of variance are:

- a. Sorting out variation from various sources.
- b. To serve as a foundation for a significance test.

It is based on the following statistical procedure and is used to examine the importance of variation found in various genotypes (**Panse and Sukhatme, 1954**).

Two- way ANOVA:

It is the type of ANOVA that controls what we apply, when we want to investigate the combined effect of two independent factors.

| Source of variation | Degree of freedom | Sum of squares | Mean square | F |
|-----------------------|-------------------|----------------|----------------------------------|-----------------------------|
| Factor A | $k - 1$ | SSA | $MSA = \frac{SSA}{k-1}$ | $F_A = \frac{MSA}{MSE}$ |
| Factor B | $l - 1$ | SSB | $MSB = \frac{SSB}{l-1}$ | $F_B = \frac{MSB}{MSE}$ |
| Interaction AB | $(k - 1)(l - 1)$ | $SSAB$ | $MSAB = \frac{SSAB}{(k-1)(l-1)}$ | $F_{AB} = \frac{MSAB}{MSE}$ |
| Error | $kl(m - 1)$ | SSE | $MSE = \frac{SSE}{kl(m-1)}$ | |
| Total | $klm - 1$ | SST | | |

Abbreviations used:

k = number of levels of factor A

l = number of levels of factor B

kl = number of treatments (each one a combination of a factor A level and a factor B level)

m = number of observations on each treatment

SST= Total sum of square

SSA= Factor A main effect sum of square

SSAB= Interaction sum of square

Test of significance:

MSS due to genotypes were tested against the error variance using F test at 5% and 1% level of significance. If the value of F-calculated is greater than F-tabulated, then the variance between treatments were considered to be significant and vice-versa.

3.16.2 Selection Parameters

A. Mean

The mean value of each character was determined by summing up all the observations and dividing them by corresponding number of observations.

$$\bar{X} = \frac{\sum_{i=1}^N Xi}{N}$$

Where,

\bar{X} = mean

$\sum_{i=1}^N Xi$ = Sum of all observations

N = Number of observations

Range:

In statistics, the range of a set of data is the difference between the largest and smallest values. However, in descriptive statistics, this concept of range has a more complex meaning. The range is the size of the smallest interval (statistics) which contains all the data and provides an indication of statistical dispersion. It is measured in the same units as the data. Since it only depends on two of the observations, it is most useful in representing the dispersion of small data sets.

Range = (Highest value of an item in a series) – (Lowest value of an item in a series)

3.16.3 Standard deviation:

Standard deviation is most widely used measure of dispersion of a series and is commonly denoted by the symbol ‘ σ ’ (pronounced as sigma). Standard deviation is defined as the square-root of the average of squares of deviations, when such deviations for the values of individual items in a series are obtained from the arithmetic average.

$$\text{Standard deviation } (\sigma) = \sqrt{\frac{\sum (xi - \mu)^2}{N}}$$

Where,

σ = population standard deviation

N = the size of the population

xi = each value from the population

μ = the population mean

3.16.4 Standard error:

The standard error (SE) of a statistic is the approximate standard deviation of a statistical sample population. The standard error is a statistical term that measures the accuracy with which a sample distribution represents a population by using standard deviation.

$$SE = \frac{\sigma}{\sqrt{n}}$$

Where,

SE = standard error of the sample

σ = sample standard deviation

n = number of samples

3.16.5 Correlation coefficient:

The simple phenotypic correlation coefficients among pairs of characters were calculated according to the formula suggested by Searle (1961).

$$r(X_1X_2) = \frac{\text{Cov}(X_1X_2)}{\sqrt{V(X_1) \cdot V(X_2)}}$$

Where,

X_1 = Character 1

X_2 = Character 2

$r(X_1X_2)$ = Correlation between characters X_1 and X_2

$\text{Cov } X_1X_2$ = Covariance between X_1 and X_2

$V(X_1)$ = Variance of X_1

$V(X_2)$ = Variance of X_2

In the estimation of phenotypic correlation coefficients, phenotypic covariance and variance are considered for calculation.

To test the significance of correlation coefficients, the estimated values were compared with the table value (statistical table by Fisher and Yates, 1963) at $n-2$ degrees of freedom (where n denotes the number of genotypes tested) at 5% and 1% levels of significance.

Scales of correlation coefficients (Searle, 1965)

Very strong = >0.65

Moderately strong = $0.50 - 0.64$

Moderately weak = $0.30 - 0.49$

Very weak = <0.30

3.17 Selection of genotypes for future breeding purpose

According to Singh and Chaudhary (1985), while choosing genotypes for a hybridization programme, the following factors should be taken into account.

- Choosing the cluster from which the parent genotype(s) would be chosen (s)
- Choosing a specific genotype or genotypes from the chosen cluster (s)
- Characters' relative contributions to overall divergence Other key features from genotype performance.

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Plate 1. Transplanting of germinated seedlings



Plate 2. Hydroponics system for screening of wheat genotypes

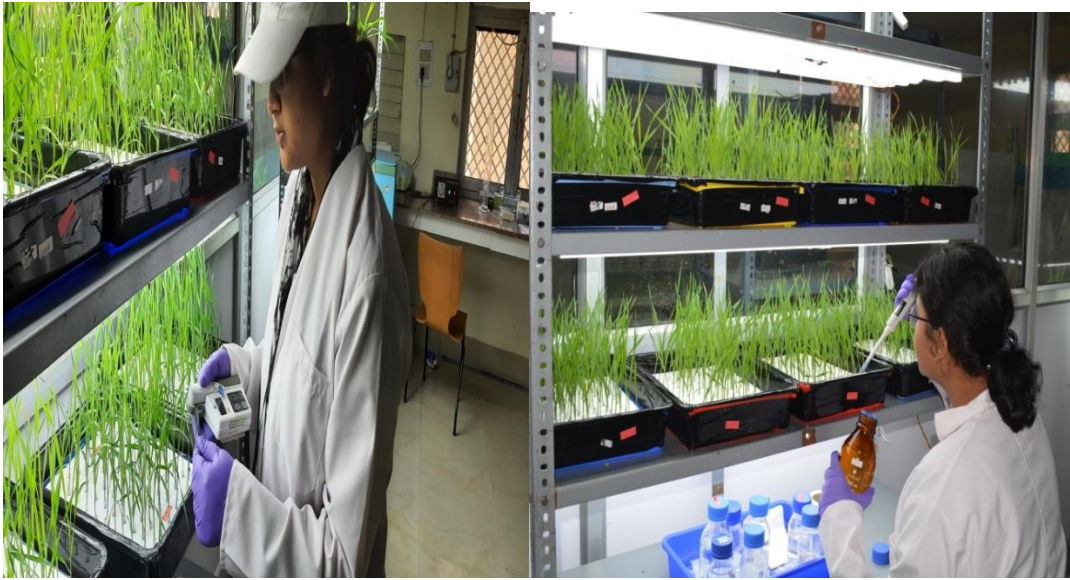


Plate 3. Recording data and changing nutrient solution at every 7th days interval



Plate 4. Harvesting of plants at 24th day after transplanting and recording observations

EXPERIMENTAL FINDINGS

Experiment was conducted under controlled laboratory condition with 10 wheat genotypes (9mutants and one parent) in a CRD (CompletelyRandomised Design) with two replications & repeated twice to check reproducibility of the results. Water stress was induced by using polyethylene glycol (PEG) 8000.The level of genetic diversity as well as the correlation between different genotypes were investigated. The following headings are used to present the findings of this research:

- ANOVA (Analysis of variance)
- Mean and range performance
- Index
 - a) Root length stress tolerance index (STI)
 - b) Shoot length stress tolerance index(STI)
 - c) Dry matter stress tolerance index of shoot(STI)
 - d) Dry matter stress tolerance index of root (STI)
- Correlation coefficient

4.1 ANOVA (Analysis of variance) under osmotic stress and non-stress (control) condition

ANOVA was used to compare several characteristics across ten wheat genotypes, and it found highly significant differences between entries for all of the attributes tested.

On the basis of following parameters data has been recorded:

1. Shoot length(SL)
2. Root length (RL)

3. Chlorophyll content
4. Fresh root weight
5. Fresh weight (g)
 - (a) Shoot
 - (b) Root

ANOVA recorded among 10 wheat genotypes for 5 characters and significant differences among genotype and between genotype was observed with respect to all traits under study including chlorophyll content, shoot length, root length, shoot weight, root weight and root number.

In table(4.1.1), ANOVA for chlorophyll content data shown Fcal values for genotypes (fcal=1.21) and GxE(0.63) interaction is less than Fcrit value [i.e. Fcrit= 2.39] which indicates that there is no significant variation between genotypes and interaction whereas Fcal value (i.e. Fcal = 117.7) of the treatment is more than Fcrit [i.e. Fcrit=4.35] which means that water stress treatment significantly differentiated genotypes from control condition for chlorophyll content.

Table 4.1.1 Analysis of variance table for chlorophyll content

| ANOVA for chlorophyll content | | | | | | |
|-------------------------------|-----------------|-----------|----------|----------|----------|----------|
| Source of Variation | SS | df | MS | F | P-value | F crit |
| Treatment | 593.8446 | 1 | 593.8446 | 117.7028 | 7.9E-10 | 4.351244 |
| Genotype | 55.13047 | 9 | 6.125608 | 1.214125 | 0.340183 | 2.392814 |
| GXE | 28.61573 | 9 | 3.179526 | 0.630197 | 0.758329 | 2.392814 |
| Error | 100.9057 | 20 | 5.045286 | | | |
| Total | 778.4965 | 39 | | | | |

In table(4.1.2), ANOVA for shoot length data shown Fcal values for genotypes (fcal=1.91) and GxE (0.37) interaction is less than Fcrit value [i.e. Fcrit= 2.39] which indicates that there is no significant variation between genotypes and interaction. Whereas Fcal value (i.e. Fcal = 123.7) of the treatment is more than Fcrit [i.e. Fcrit=4.35] which means that water stress treatment significantly differentiated genotypes from control condition for shoot length.

Table 4.1.2 Analysis of variance table for shoot length

| ANOVA for shoot length | | | | | | |
|-------------------------------|----------------|-----------|-----------|----------|----------------|---------------|
| <i>Source of Variation</i> | <i>SS</i> | <i>df</i> | <i>MS</i> | <i>F</i> | <i>P-value</i> | <i>F crit</i> |
| Treatment | 820.4136 | 1 | 820.4136 | 123.1733 | 5.34E-10 | 4.351244 |
| Genotype | 71.82426 | 9 | 7.980473 | 1.198153 | 0.348801 | 2.392814 |
| GXE | 19.00901 | 9 | 2.112112 | 0.317103 | 0.959724 | 2.392814 |
| Error | 133.2129 | 20 | 6.660645 | | | |
| Total | 1044.46 | 39 | | | | |

In table(4.1.3), ANOVA for root length data shown Fcal values for genotypes (fcal=0.27) and GxE (0.12) interaction is less than Fcrit value [i.e. Fcrit= 2.39] which indicates that there is no significant variation between genotypes and interaction of genotypes. Fcal value (i.e. Fcal = 48.7) of the treatment is more than Fcrit [i.e. Fcrit=4.35] which means that water stress treatment significantly differentiated genotypes from control condition for root length.

Table 4.1.3 Analysis of variance table for root length

| ANOVA for root length | | | | | | |
|------------------------------|------------------|-----------|-----------|----------|----------------|---------------|
| <i>Source of Variation</i> | <i>SS</i> | <i>df</i> | <i>MS</i> | <i>F</i> | <i>P-value</i> | <i>F crit</i> |
| Treatment | 996.66013 | 1 | 996.6601 | 48.04572 | 9.91E-07 | 4.351244 |
| Genotype | 251.60739 | 9 | 27.95638 | 1.347685 | 0.275074 | 2.392814 |
| GXE | 343.56241 | 9 | 38.1736 | 1.840224 | 0.122735 | 2.392814 |
| Error | 414.8799 | 20 | 20.744 | | | |
| Total | 2006.7098 | 39 | | | | |

In table(4.1.4), ANOVA for fresh shoot weight data shown that Fcal values for genotypes (fcal=1.91) and GxE (0.37) interaction is less than Fcrit value [i.e. Fcrit= 2.39] recorded which indicates that there is no significant variation between genotypes and interaction. Fcal value (i.e. Fcal = 182.7) of the treatment is more than Fcrit [i.e. Fcrit=4.35] which means that water stress treatment is significantly differentiated genotypes from control condition for fresh shoot weight.

Table 4.1.4 Analysis of variance table for shoot weight

| ANOVA for shoot weight | | | | | | |
|-------------------------------|-----------------|-----------|-----------|----------|----------------|---------------|
| <i>Source of Variation</i> | <i>SS</i> | <i>df</i> | <i>MS</i> | <i>F</i> | <i>P-value</i> | <i>F crit</i> |
| Treatment | 10.91302 | 1 | 10.91302 | 182.2282 | 1.65E-11 | 4.351244 |
| Genotype | 1.015793 | 9 | 0.112866 | 1.884661 | 0.114079 | 2.392814 |
| GXE | 0.559038 | 9 | 0.062115 | 1.037216 | 0.445973 | 2.392814 |
| Error | 1.197732 | 20 | 0.059887 | | | |
| Total | 13.68559 | 39 | | | | |

In table(4.1.5), ANOVA for fresh root weight data shown that Fcal values for genotypes (fcal=2.1) and GxE (0.59) interaction is less than Fcrit value [i.e. Fcrit= 2.39] recorded which indicates that there is no significant variation between genotypes and interaction. Fcal value (i.e. Fcal = 154.76) of the treatment is more than Fcrit [i.e. Fcrit=4.35] which means that water stress treatment is significantly differentiated genotypes from control condition for fresh root weight.

Table 4.1.5 Analysis of variance table for root weight

| ANOVA for root weight | | | | | | |
|------------------------------|-----------------|-----------|-----------|----------|----------------|---------------|
| <i>Source of Variation</i> | <i>SS</i> | <i>df</i> | <i>MS</i> | <i>F</i> | <i>P-value</i> | <i>F crit</i> |
| Treatment | 1.674324 | 1 | 1.674324 | 154.7672 | 7.17E-11 | 4.351244 |
| Genotype | 0.204858 | 9 | 0.022762 | 2.104025 | 0.079652 | 2.392814 |
| GXE | 0.057978 | 9 | 0.006442 | 0.595472 | 0.78587 | 2.392814 |
| Error | 0.216367 | 20 | 0.010818 | | | |
| Total | 2.153527 | 39 | | | | |

In table(4.1.6), ANOVA for root number data shown that Fcal values for genotypes (fcal=1.88) and GxE (1.7) interaction is less than Fcrit value [i.e. Fcrit= 2.39] which indicates that there is no significant variation between genotypes and interaction. Fcal value (i.e. Fcal = 42.4) of the treatment is more than Fcrit [i.e. Fcrit=4.35] which means that water stress treatment is significantly differentiated genotypes from control condition for root number.

Table 4.1.6: Analysis of variance table for root number

| ANOVA for root number | | | | | | |
|------------------------------|-----------------|-----------|-----------|----------|----------------|---------------|
| <i>Source of Variation</i> | <i>SS</i> | <i>df</i> | <i>MS</i> | <i>F</i> | <i>P-value</i> | <i>F crit</i> |
| Treatment | 50.31588 | 1 | 50.31588 | 42.41401 | 2.39E-06 | 4.351244 |
| Genotype | 20.09085 | 9 | 2.232317 | 1.881742 | 0.114629 | 2.392814 |
| GXE | 18.23057 | 9 | 2.025619 | 1.707505 | 0.15274 | 2.392814 |
| Error | 23.72607 | 20 | 1.186303 | | | |
| Total | 112.3634 | 39 | | | | |

4.2 Performance (mean and range)

In this experiment, the mean, range, standard deviation, and standard error of 10 wheat genotypes are evaluated. All mutant genotypes are studied and compared with each other as well as under water stress and non-stress condition.

4.2.1 Chlorophyll Content

The mean and range of genotypes, as a result of moisture stress and non-stress conditions on chlorophyll content of mutant genotypes is given in the (Table 4.2.1.1) and graph (4.1)

Under control condition, chlorophyll content of wheat genotypes TAW98, TAW95, TAW122 and TAW15 has the highest chlorophyll content and their mean is 36, 35.6, 36.2 and 33.6 respectively whereas lines with genotype TAW132, TAW19, TAW15, TAW97 and TAW11 exhibit lowest chlorophyll content among all genotypes and mean is 33.5, 33.6, 33, 34.1 and 34.1 while mean of HD2967 (parent) is 25.9.

Under stress condition, chlorophyll content of wheat genotypes TAW97, TAW95, TAW98 and TAW114 has the highest chlorophyll content whose mean is 29.5, 29.1, 27.9 and 27 respectively whereas genotypes TAW19, TAW122, TAW15, TAW117 and TAW132 exhibit lowest chlorophyll content among all with mean 23.9, 25.7, 25.7, 25.6 and 25.8 while mean of HD2967 (parent) is 25.9.

4.2.1.1 Table showing mean and range performance of chlorophyll content of 10 genotypes under stressed condition.

| S.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|-------|----------|-------|-------|-------|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 33.65 | 36.40 | 30.00 | 1.79 | 0.40 |
| 2 | TAW95 | 35.61 | 39.70 | 31.40 | 2.36 | 0.53 |
| 3 | TAW97 | 34.06 | 39.10 | 30.20 | 2.83 | 0.63 |
| 4 | TAW98 | 36.09 | 41.00 | 31.20 | 2.77 | 0.62 |
| 5 | TAW114 | 34.16 | 40.40 | 30.00 | 2.59 | 0.58 |
| 6 | TAW117 | 33.65 | 39.40 | 28.00 | 3.35 | 0.75 |
| 7 | TAW122 | 36.23 | 40.80 | 31.90 | 2.37 | 0.53 |
| 8 | TAW132 | 33.31 | 37.80 | 30.20 | 2.39 | 0.54 |
| 9 | TAW15 | 33.54 | 39.90 | 25.90 | 3.59 | 0.80 |
| 10 | HD2967 | 34.77 | 38.50 | 31.42 | 1.83 | 0.41 |

4.2.1.2 Table showing mean and range performance of chlorophyll content of 10 genotypes under controlled condition.

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|-------|-------|-------|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 23.98 | 29.10 | 15.00 | 4.23 | 0.95 |
| 2 | TAW95 | 29.13 | 37.80 | 21.20 | 4.84 | 1.08 |
| 3 | TAW97 | 29.57 | 37.40 | 23.90 | 3.68 | 0.82 |
| 4 | TAW98 | 27.96 | 33.80 | 22.60 | 3.33 | 0.74 |
| 5 | TAW114 | 27.09 | 37.50 | 21.60 | 3.82 | 0.86 |
| 6 | TAW117 | 25.71 | 32.00 | 15.00 | 4.62 | 1.03 |
| 7 | TAW122 | 25.74 | 30.60 | 11.00 | 4.99 | 1.12 |
| 8 | TAW132 | 25.83 | 32.60 | 18.70 | 3.36 | 0.75 |
| 9 | TAW15 | 25.76 | 31.60 | 19.60 | 2.92 | 0.65 |
| 10 | HD2967 | 25.91 | 27.90 | 22.76 | 1.36 | 0.31 |

4.2.2 Shoot length (cm)

Under control condition, in shoot length of wheat genotypes TAW98, TAW122, TAW95 and TAW114 has highest mean of 33.8, 32.9, 33.6 and 32.3 cm respectively whereas lines with genotype TAW15, TAW19, TAW132, TAW117 and TAW97 exhibits lowest shoot length with their mean 28.5, 31.5, 31.6, 31.7 and 31.8 cm while mean of HD2967 (parent) is 31.1 cm. (Table 4.2.2.1) and graph (4.2)

Under water stress condition, shoot length of wheat genotypes TAW95, TAW132, TAW98 and TAW97 has highest with mean 24.6, 24.62, 24.3 and 24.1 cm respectively whereas lines with genotype TAW15, TAW19, TAW117, TAW114 and TAW122 exhibits lowest shoot length with their mean 20.03, 20.5, 20.8, 22.7 and 23 cm while mean of HD2967 (parent) is 22.1 cm. (table 4.2.2.2)

4.2.2.1 Table showing mean and range performance of 10 genotypes of shoot length under stressed condition.

| Sl.No. | Genotype | Mean | Range | Standard deviation | Standard error |
|--------|----------|------|-------|--------------------|----------------|
|--------|----------|------|-------|--------------------|----------------|

Experimental Findings

| | | | Max | Min | | |
|----|--------|----------|------------|------------|----------|---------|
| 1 | TAW19 | 31.55882 | 39 | 24 | 5.043196 | 1.22315 |
| 2 | TAW95 | 32.625 | 42 | 24 | 5.193303 | 1.16126 |
| 3 | TAW97 | 31.84211 | 38 | 26 | 3.77143 | 0.86523 |
| 4 | TAW98 | 33.875 | 41 | 28 | 3.817808 | 0.85369 |
| 5 | TAW114 | 32.325 | 36 | 26.5 | 2.829706 | 0.63274 |
| 6 | TAW117 | 31.7145 | 38.49 | 22.7 | 4.088687 | 0.91426 |
| 7 | TAW122 | 32.96316 | 38.2 | 26.7 | 3.655469 | 0.83862 |
| 8 | TAW132 | 31.65 | 36.8 | 28.4 | 2.317099 | 0.51812 |
| 9 | TAW15 | 28.55 | 39.2 | 22 | 4.879226 | 1.09103 |
| 10 | HD2967 | 31.31067 | 33.48 | 29.32 | 1.216071 | 0.27192 |

4.2.2.2 Table showing mean and range performance of 10 genotypes of shoot length under controlled condition.

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|-------|-------|-------|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 20.58 | 26.00 | 11.00 | 4.26 | 0.95 |
| 2 | TAW95 | 24.65 | 27.50 | 20.00 | 1.85 | 0.41 |
| 3 | TAW97 | 24.20 | 30.10 | 16.50 | 3.98 | 0.89 |
| 4 | TAW98 | 24.30 | 26.50 | 20.50 | 1.77 | 0.40 |
| 5 | TAW114 | 22.74 | 30.00 | 12.00 | 4.30 | 0.99 |
| 6 | TAW117 | 20.85 | 24.00 | 16.00 | 2.13 | 0.48 |
| 7 | TAW122 | 23.00 | 25.50 | 18.00 | 1.85 | 0.41 |
| 8 | TAW132 | 24.62 | 27.00 | 17.20 | 2.34 | 0.55 |
| 9 | TAW15 | 20.04 | 22.00 | 17.00 | 1.65 | 0.37 |
| 10 | HD2967 | 22.19 | 24.50 | 19.40 | 1.41 | 0.31 |

4.2.3 Root length (in cm)

Under non-stress condition, root length of wheat genotypes TAW132, TAW122, TAW117 and TAW15 has highest with their mean 29.2, 28.8, 28.7 and 27.2 cm respectively. Whereas lines with genotype TAW19, TAW114, TAW98 and TAW95 exhibit lowest root length with their mean 15.1, 16.6, 17.1, 18.3 and 19.8 cm while mean of HD2967 (parent) is 22.5 cm. (table 4.2.3.1) and graph (4.3)

Under water stress condition, in root length of wheat genotypes TAW95, TAW97, TAW98, and TAW132 has highest whose mean 14.4, 13.5, 13.4 and 13.2 cm respectively whereas lines with genotype TAW15, TAW117, TAW114, TAW19 and TAW122 exhibit lowest root length with their mean 9.2, 11.6, 11.8, 11.8 and 12.1 cm while mean of HD2967 (parent) is 11.8 cm. (table 4.2.3.2)

4.2.3.1 Table showing mean and range performance of 10 genotypes of root length under stressed condition (PEG8000)

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|----------|----------|-------|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 15.11765 | 20 | 12 | 2.62518 | 0.6367 |
| 2 | TAW95 | 19.825 | 27 | 12 | 3.89086 | 0.87002 |
| 3 | TAW97 | 17.10526 | 24 | 14 | 2.35454 | 0.54017 |
| 4 | TAW98 | 18.3 | 22.5 | 12 | 3.46182 | 0.77409 |
| 5 | TAW114 | 16.65 | 24 | 12 | 2.61624 | 0.58501 |
| 6 | TAW117 | 28.735 | 44 | 19.6 | 7.81754 | 1.74806 |
| 7 | TAW122 | 28.82105 | 48 | 18.2 | 8.22635 | 1.88725 |
| 8 | TAW132 | 29.25 | 42.2 | 20.6 | 5.90588 | 1.3206 |
| 9 | TAW15 | 27.26 | 40.3 | 18.5 | 6.56838 | 1.46873 |
| 10 | HD2967 | 22.55442 | 31.43333 | 16.28 | 4.58533 | 1.02531 |

4.2.3.2 Table showing mean and range performance of 10 genotypes of root length under controlled condition.

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|-------|-------|-------|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 11.87 | 15.50 | 7.00 | 2.51 | 0.56 |
| 2 | TAW95 | 14.45 | 18.50 | 10.00 | 2.34 | 0.52 |
| 3 | TAW97 | 13.53 | 19.50 | 6.00 | 3.65 | 0.82 |
| 4 | TAW98 | 13.41 | 16.50 | 8.00 | 2.51 | 0.56 |
| 5 | TAW114 | 11.81 | 16.60 | 6.00 | 3.00 | 0.67 |
| 6 | TAW117 | 11.69 | 18.00 | 7.00 | 2.53 | 0.58 |
| 7 | TAW122 | 12.15 | 18.00 | 7.00 | 2.96 | 0.66 |
| 8 | TAW132 | 13.28 | 16.00 | 8.60 | 1.93 | 0.43 |
| 9 | TAW15 | 9.30 | 17.00 | 4.80 | 2.70 | 0.60 |

4.2.3. Fresh weight of shoot(in gm)

Under control condition, in fresh shoot weight of wheat genotypes TAW98, TAW95, TAW132 and TAW122 has highest with mean of 1.88, 1.87, 1.85 and 1.79g respectively whereas lines with genotype TAW19, TAW114, TAW15, TAW117 and TAW97 exhibits lowest fresh shoot weight with mean of 1.07, 1.42, 1.5, 1.55 and 1.54g. While mean of HD2967(parent) is 1.36g. (table 4.2.4.1) and graph (4.4)

Under water stress condition, in fresh shoot weight of wheat genotypes TAW95, TAW114, TAW98 and TAW97 has highest mean of 0.71, 0.7, 0.65 and 0.64g respectively whereas TAW132, TAW15, TAW19, TAW117 and TAW122 exhibits lowest shoot weight with their mean 0.4, 0.41, 0.43, 0.46 and 0.51g while mean of HD2967(parent) has 0.5g. (table 4.2.4.2)

4.2.4.1 Table showing mean and range performance of 10 genotypes of shoot fresh weight stressed condition

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|------|-------|-----|--------------------|----------------|
| | | | Max | Min | | |

| | | | | | | |
|----|--------|------|------|------|------|------|
| 1 | TAW19 | 1.07 | 1.74 | 0.75 | 0.34 | 0.11 |
| 2 | TAW95 | 1.87 | 2.25 | 1.60 | 0.22 | 0.07 |
| 3 | TAW97 | 1.54 | 1.92 | 1.23 | 0.25 | 0.08 |
| 4 | TAW98 | 1.88 | 2.22 | 1.55 | 0.21 | 0.07 |
| 5 | TAW114 | 1.42 | 1.93 | 0.20 | 0.52 | 0.17 |
| 6 | TAW117 | 1.56 | 2.05 | 1.24 | 0.22 | 0.07 |
| 7 | TAW122 | 1.80 | 2.28 | 0.87 | 0.46 | 0.14 |
| 8 | TAW132 | 1.86 | 2.36 | 1.33 | 0.43 | 0.14 |
| 9 | TAW15 | 1.51 | 1.97 | 0.95 | 0.28 | 0.09 |
| 10 | HD2967 | 1.36 | 1.83 | 0.96 | 0.25 | 0.08 |

4.2.4.2 Table showing mean and range performance of 10 genotypes of shoot fresh weight under controlled condition.

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|------|-------|------|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 0.43 | 0.55 | 0.31 | 0.09 | 0.03 |
| 2 | TAW95 | 0.72 | 0.91 | 0.55 | 0.13 | 0.04 |
| 3 | TAW97 | 0.65 | 1.10 | 0.40 | 0.23 | 0.07 |
| 4 | TAW98 | 0.70 | 0.87 | 0.51 | 0.11 | 0.04 |
| 5 | TAW114 | 0.65 | 1.04 | 0.24 | 0.30 | 0.09 |
| 6 | TAW117 | 0.46 | 0.68 | 0.34 | 0.12 | 0.04 |
| 7 | TAW122 | 0.52 | 0.95 | 0.26 | 0.25 | 0.08 |
| 8 | TAW132 | 0.41 | 0.58 | 0.22 | 0.11 | 0.03 |
| 9 | TAW15 | 0.41 | 0.75 | 0.22 | 0.19 | 0.06 |
| 10 | HD2967 | 0.52 | 0.57 | 0.46 | 0.05 | 0.02 |

4.2.4 Fresh weight of root (in gm)

Under non-stress condition, in fresh root weight of wheat genotypes TAW 98,TAW122,TAW95 and TAW132 has highest with mean 0.81,0.8,0.79 and 0.75 respectively whereas genotypes TAW19,TAW15,TAW97,TAW114 and TAW117 exhibits lowest fresh root weight with mean 0.5,0.6,0.7,0.71 and 0.73gwhile mean of HD2967(parent) 0.68g.(table 4.2.5.1) and graph (4.5)

Under water stress condition, fresh root weight of wheat genotypes TAW98, TAW95, TAW15, and TAW114 has highest mean is 0.37, 0.36, 0.33 and 0.32g respectively whereas genotypes TAW19, TAW117, TAW132, TAW97 and TAW122 exhibits lowest fresh root weight with their mean 0.21, 0.27, 0.29, 0.31 and 0.32 g while mean of HD2967 (parent) is 0.29g (table 4.2.5.2).

4.2.5.1 Table showing mean and range performance of 10 genotypes of fresh root weight under stressed condition.

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|------|-------|------|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 0.55 | 0.79 | 0.32 | 0.19 | 0.07 |
| 2 | TAW95 | 0.79 | 0.90 | 0.68 | 0.09 | 0.03 |
| 3 | TAW97 | 0.70 | 0.88 | 0.52 | 0.10 | 0.03 |
| 4 | TAW98 | 0.82 | 0.99 | 0.69 | 0.11 | 0.04 |
| 5 | TAW114 | 0.72 | 0.90 | 0.45 | 0.17 | 0.06 |
| 6 | TAW117 | 0.73 | 0.95 | 0.62 | 0.10 | 0.03 |
| 7 | TAW122 | 0.81 | 1.05 | 0.35 | 0.23 | 0.07 |
| 8 | TAW132 | 0.78 | 1.10 | 0.45 | 0.18 | 0.06 |
| 9 | TAW15 | 0.70 | 0.89 | 0.48 | 0.15 | 0.05 |
| 10 | HD2967 | 0.69 | 0.80 | 0.54 | 0.08 | 0.02 |

4.2.5.2 Table showing mean and range performance of 10 genotypes of fresh root weight under controlled condition

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|------|-------|------|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 0.21 | 0.34 | 0.12 | 0.08 | 0.03 |
| 2 | TAW95 | 0.37 | 0.5 | 0.27 | 0.07 | 0.02 |
| 3 | TAW97 | 0.31 | 0.45 | 0.18 | 0.10 | 0.03 |
| 4 | TAW98 | 0.38 | 0.465 | 0.29 | 0.06 | 0.02 |
| 5 | TAW114 | 0.32 | 0.475 | 0.14 | 0.11 | 0.04 |

| | | | | | | |
|----|--------|------|-------|------|------|------|
| 6 | TAW117 | 0.28 | 0.4 | 0.15 | 0.10 | 0.03 |
| 7 | TAW122 | 0.32 | 0.5 | 0.21 | 0.09 | 0.03 |
| 8 | TAW132 | 0.30 | 0.325 | 0.27 | 0.02 | 0.01 |
| 9 | TAW15 | 0.33 | 0.51 | 0.17 | 0.12 | 0.04 |
| 10 | HD2967 | 0.30 | 0.395 | 0.22 | 0.06 | 0.02 |

4.2.5. Root Number

Under non-stress condition, in root number of wheat genotypes TAW98, TAW95, TAW132 and TAW144 has highest mean 11, 10, 10, 9 and 9 respectively whereas genotypes TAW19, TAW15, TAW117, TAW97 and TAW122 exhibits lowest root number with mean 8, 8.5, 8.8, 9.2 and 9.5 while mean of HD2967 (parent) is 8.9 (table 4.2.5.2) and graph (4.6).

Under water stress condition, in root number of wheat genotypes TAW132, TAW117, TAW122 and TAW15 has highest with mean 8.6, 8.4, 8.5 and 8.1 respectively whereas TAW19, TAW97, TAW98, TAW95 and TAW114 exhibits lowest root number with the mean of 6, 6.4, 6.6, 6.8 and 7.1 while mean of HD2967 (parent) is 6.4 (table 4.2.5.2).

4.2.5.1. Table showing mean and range performance of 10 genotypes of root number under stressed condition.

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|-------|-------|-----|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 8.18 | 11 | 6 | 1.67 | 0.40 |
| 2 | TAW95 | 10.80 | 13 | 9 | 1.36 | 0.30 |
| 3 | TAW97 | 9.21 | 11 | 7 | 0.98 | 0.22 |
| 4 | TAW98 | 11.30 | 14 | 8 | 1.49 | 0.33 |

| | | | | | | |
|----|--------|-------|------|-----|------|------|
| 5 | TAW114 | 9.65 | 13 | 7 | 1.84 | 0.41 |
| 6 | TAW117 | 8.80 | 12 | 6 | 1.64 | 0.37 |
| 7 | TAW122 | 9.53 | 13 | 7 | 1.61 | 0.37 |
| 8 | TAW132 | 10.65 | 13 | 7 | 1.98 | 0.44 |
| 9 | TAW15 | 8.45 | 12 | 6 | 1.64 | 0.37 |
| 10 | HD2967 | 8.94 | 10.5 | 6.5 | 1.13 | 0.26 |

4.2.5.2. Table showing mean and range performance of 10 genotypes of root number under controlled condition.

| Sl.No. | Genotype | Mean | Range | | Standard deviation | Standard error |
|--------|----------|------|-------|-----|--------------------|----------------|
| | | | Max | Min | | |
| 1 | TAW19 | 6 | 8 | 4 | 1.41 | 0.32 |
| 2 | TAW95 | 6.80 | 9 | 4 | 1.24 | 0.28 |
| 3 | TAW97 | 6.45 | 8 | 4 | 1.19 | 0.27 |
| 4 | TAW98 | 6.60 | 9 | 5 | 1.14 | 0.26 |
| 5 | TAW114 | 7.10 | 9 | 5 | 1.12 | 0.25 |
| 6 | TAW117 | 8.45 | 11 | 6 | 1.28 | 0.29 |
| 7 | TAW122 | 8.44 | 13 | 5 | 2.31 | 0.54 |
| 8 | TAW132 | 8.60 | 13 | 6 | 2.30 | 0.52 |
| 9 | TAW15 | 8.15 | 13 | 6 | 2.03 | 0.45 |
| 10 | HD2967 | 6.45 | 9 | 4.5 | 0.91 | 0.20 |

Figure1: Graphical representation of mean performance of chlorophyll content under stress and control condition

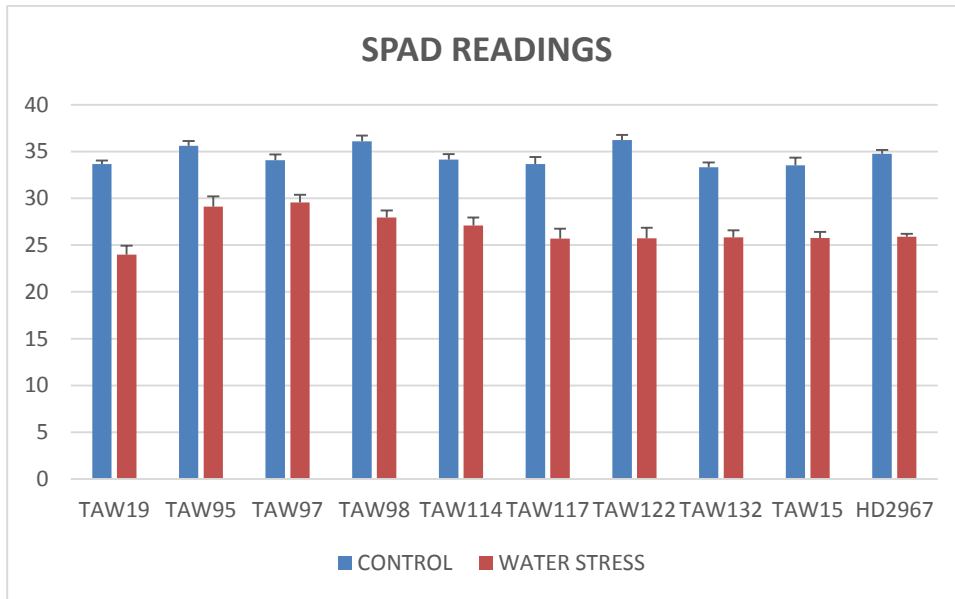


Figure 2: Graphical representation of mean performance of shoot length under stress and control condition

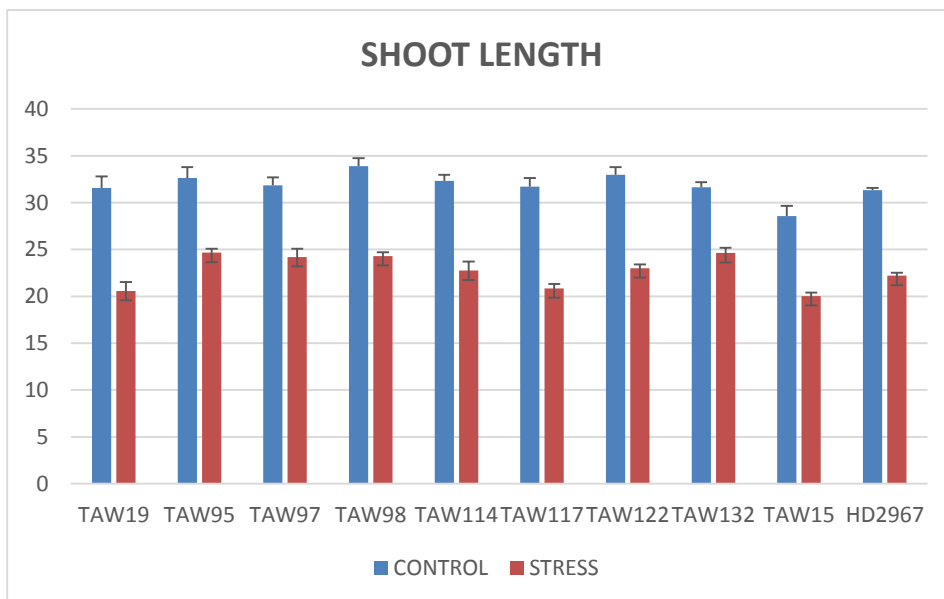


Figure 3: Graphical representation of mean performance of root length under stress and control condition

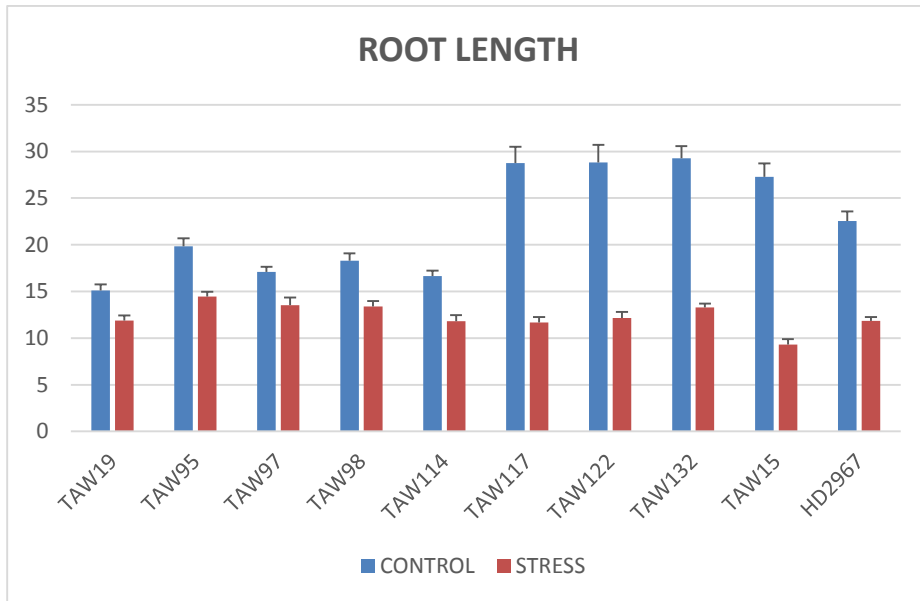


Figure4: Graphical representation of mean performance of shoot fresh weight under stress and control condition

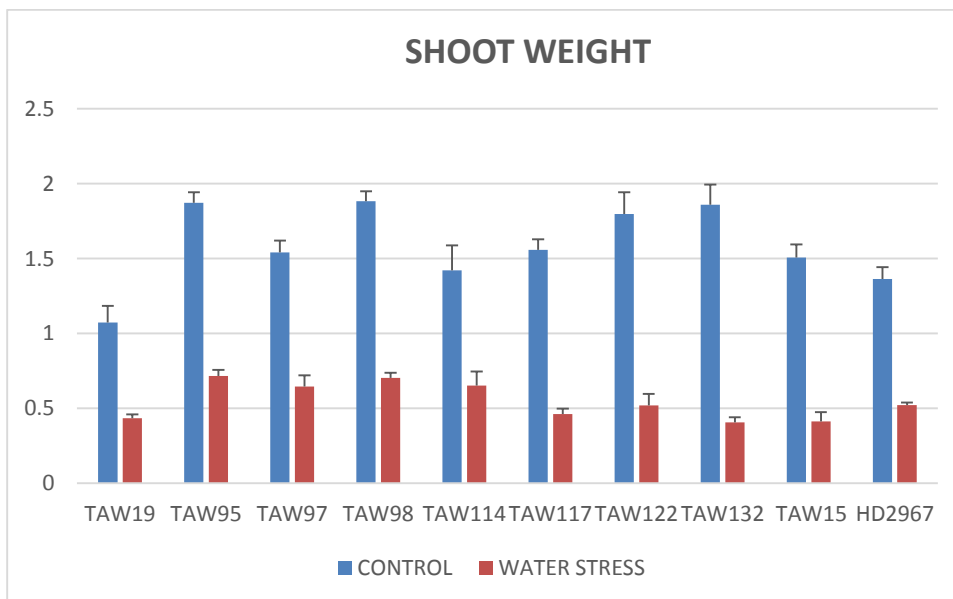


Figure5: Graphical representation of mean performance of root fresh weight under stress and control condition

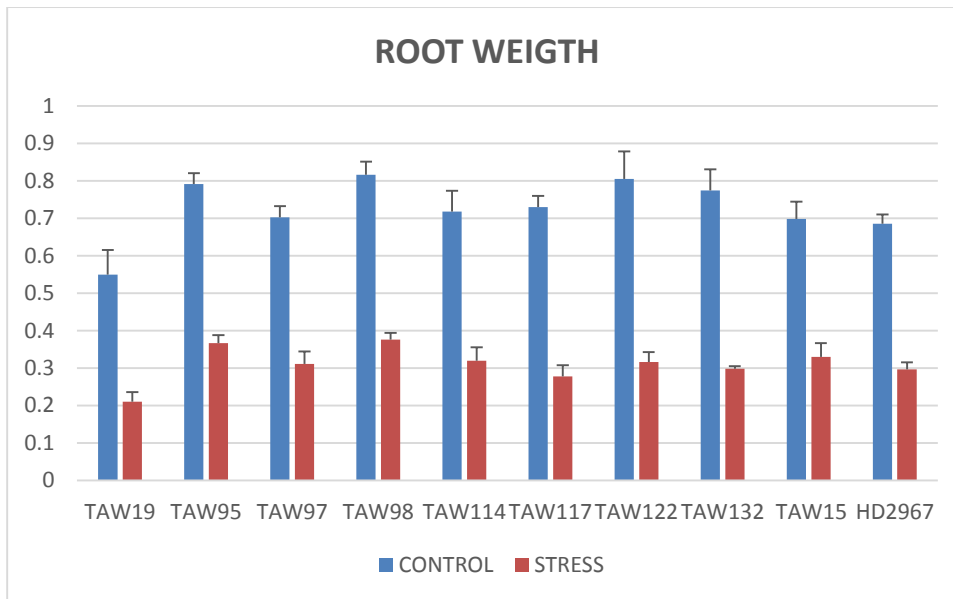
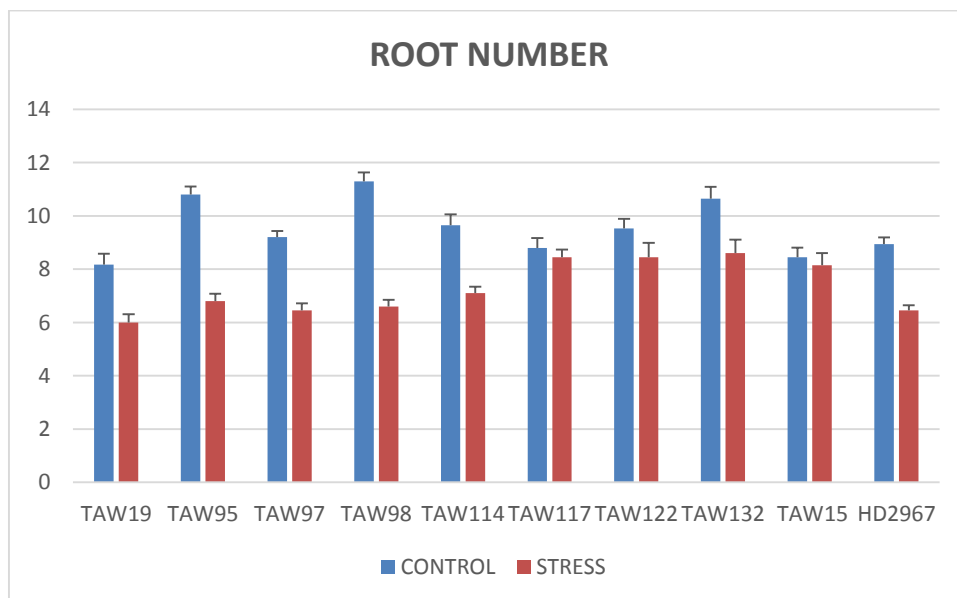


Figure6: Graphical representation of mean performance of root number under stress and controlled condition



4.4 INDICES:

4.4.1 Shoot length stress tolerance index (%)

In stress, at 20 percent PEG (8000) the stress tolerance indices the genotype TAW132, TAW95 and TAW98 with mean 77.8%, 65.04% and 71% has the highest value while genotype TAW117 and TAW19 has the lowest STI with mean 65.04% and 67%.

4.4.2 Root length stress tolerance index (%)

In stress, at 20 percent PEG (8000) the stress tolerance indices the genotype TAW97, TAW19, TAW98 and TAW95 has the highest value with mean 80.3%, 76%, 73% and 72.6% while genotype TAW15 and TAW117 has the lowest STI with mean 34.8% and 42.8%.

4.4.1.1 Table showing SLSI% and RLSI% of 10 genotypes

| Genotype | SLSI% | Genotype | RLSI% |
|-----------------|--------------|-----------------|--------------|
| HD2967 | 72.90 | HD2967 | 72.0 |
| TAW19 | 67.45 | TAW19 | 76.4 |
| TAW95 | 76.18 | TAW95 | 72.6 |
| TAW97 | 75.84 | TAW97 | 80.3 |
| TAW98 | 71.83 | TAW98 | 73.1 |
| TAW114 | 70.87 | TAW114 | 70.3 |
| TAW117 | 65.04 | TAW117 | 42.8 |
| TAW122 | 70.34 | TAW122 | 44.2 |
| TAW132 | 77.81 | TAW132 | 46.2 |
| TAW15 | 71.14 | TAW15 | 34.8 |

4.4.3 Dry matter stress tolerance index of shoot weight (%)

In stress, at 20 percent PEG (8000) the stress tolerance indices the genotype TAW95, TAW98 and TAW114 has the highest value 38.3, 37.2 and 34 % while genotype TAW15 and TAW132 has the lowest STI with mean 25 and 22.8%.

4.4.4 Dry matter stress tolerance index of root weight(%)

In stress, at 20 percent PEG (8000) the stress tolerance indices the genotype TAW114,TAW19, TAW95 and TAW98 has the highest value 47.5%,43.3%,38.3% and 37.2% while genotype TAW122 and TAW15 has the lowest STI with mean 22.9% and 25.5%.

4.4.3.1 Table showing DMSI% of shoot and root

In stress, at 20 percent PEG (8000) the stress tolerance indices the genotype TAW19,TAW114,TAW98 and TAW95 has the highest value with mean 57.03%,48.2%, 45.65% and 45.4% while genotype TAW132 and TAW117 has the lowest STI with mean 35.7% and 37.19%.

| Genotype | DMSI% of shoot | Genotype | DMSI% of root |
|-----------------|-----------------------|-----------------|----------------------|
| HD2967 | 27.9 | HD2967 | 39.95 |
| TAW19 | 43.3 | TAW19 | 57.03 |
| TAW95 | 38.3 | TAW95 | 45.44 |
| TAW97 | 41.7 | TAW97 | 45.17 |
| TAW98 | 37.2 | TAW98 | 45.65 |
| TAW114 | 47.5 | TAW114 | 48.92 |
| TAW117 | 29.9 | TAW117 | 37.19 |
| TAW122 | 28.3 | TAW122 | 38.50 |
| TAW132 | 22.9 | TAW132 | 35.73 |
| TAW15 | 25.5 | TAW15 | 38.19 |

4.5 Correlation studies:

The degree and direction of relationship between the numerous qualities evaluated under stressful conditions is measured using correlation coefficient analysis. Correlation coefficient is valuable tool in crop improvement programme which depicts relationship between several traits. The estimated linear correlation coefficient between the selected parameters is presented in this experiment in the table below (Table 4.5.1 and Table 4.5.2).

4.5.1 Phenotypic correlation under stressed and controlled condition

4.5.1.2 Shoot length

At the phenotypic level, shoot length of stress has showed highly significant positive correlation with root length of stress(0.79**) and strong negative correlation with shoot dry weight(-0.66**)

At the phenotypic level, shoot length of control had a very significant positive association with root length of stress(0.77**)and control(0.6**) only.

4.5.1.3 Root length

In this experimentation, at phenotypic level root length of stress showed highly significant positive correlation with shoot length(0.77**) and strong negative correlation with shoot weight under control (-0.56**) and chlorophyll content (-0.59**) under stress condition.

At the phenotypic level, root length of the control had a highly significant positive correlation with shoot length(0.7**) under control and stress and strong negative correlation with shoot weight(-0.56**) and root weight(-0.6**)

4.5.1.4 Fresh shoot weight

In this experimentation, at phenotypic fresh shoot weight of stress showed highly significant positive correlation with chlorophyll content (0.86**) under stress and shoot weight (0.38**) under control condition.

At the phenotypic level, fresh shoot weight of the control had a highly significant positive correlation with chlorophyll content(0.55**) and shoot weight (1.00**)under control condition.

4.5.1.5 Fresh root weight

In this experimentation, at phenotypic fresh root weight of stress showed highly significant positive correlation with chlorophyll content (0.66**),shoot weight (0.75**)and root weight(0.87**) under stress condition.

At the phenotypic level, fresh root weight ofthe control had a highly significant positive correlation with chlorophyll content (0.57**) and shoot weight(0.95**) under control condition.

Table 4.5.1 Phenotypic correlation between different traits of 10 wheat genotypes

| | SPAD(C) | SPAD(S) | SHOOT WT (C) | SHOOT WT (S) | ROOT WT(C) | ROOT WT(S) | SHOOT LTH(C) | SHOOT LTH(S) | ROOT LTH (C) | ROOT LTH (S) | ROOT NO. (C) | ROOT NO. (S) | SHOOT DRY WT(C) | SHOOT DRY WT(S) | ROOT DRY WT(C) | ROOT DRY WT(S) |
|-----------------|---------|---------|--------------|--------------|------------|------------|--------------|--------------|--------------|--------------|--------------|--------------|-----------------|-----------------|----------------|----------------|
| SPAD(C) | 1.00 | | | | | | | | | | | | | | | |
| SPAD(S) | 0.37 | 1.00* | | | | | | | | | | | | | | |
| SHOOT WT (C) | 0.37 | 0.55** | 1.00* | | | | | | | | | | | | | |
| SHOOT WT (S) | 0.64** | 0.86* | 0.38** | 1.00 | | | | | | | | | | | | |
| ROOT WT(C) | 0.38 | 0.57** | 0.95** | 0.35 | 1.00 | | | | | | | | | | | |
| ROOT WT(S) | 0.66** | 0.86 | 0.75 | 0.87** | 0.75** | 1.00 | | | | | | | | | | |
| SHOOT LTH(C) | -0.15 | 0.25 | -0.09 | 0.20 | -0.17 | 0.08 | 1.00 | | | | | | | | | |
| SHOOT LTH(S) | 0.01 | -0.06 | -0.43 | 0.10 | -0.44 | -0.07 | 0.60* | 1.00 | | | | | | | | |
| ROOT LTH (C) | -0.43 | 0.59* | 0.13 | -0.81** | 0.20 | -0.44 | -0.28 | -0.19 | 1.00 | | | | | | | |
| ROOT LTH (S) | -0.39 | -0.03 | -0.56* | 0.00 | -0.64** | -0.28 | 0.77** | 0.79** | -0.32 | 1.00 | | | | | | |
| ROOT NO. (C) | 0.36 | 0.66** | 0.85** | 0.61** | 0.72** | 0.81** | 0.21 | -0.23 | -0.25 | -0.22 | 1.00 | | | | | |
| ROOT NO. (S) | -0.29 | -0.28 | 0.45 | -0.48* | 0.49* | -0.08 | -0.36 | -0.35 | 0.86** | -0.50* | 0.05 | 1.00 | | | | |
| SHOOT DRY WT(C) | -0.05 | 0.22 | 0.80** | 0.01 | 0.75** | 0.37 | -0.45 | -0.66** | 0.42 | -0.68** | 0.52* | 0.75** | 1.00 | | | |
| SHOOT DRY WT(S) | 0.57* | 0.85** | 0.40 | 0.99** | 0.33* | 0.85** | 0.23 | 0.11 | -0.80** | 0.05 | 0.64** | 0.44 | 0.07 | 1.00 | | |
| ROOT DRY WT(C) | 0.35 | 0.61** | 0.93** | 0.51* | 0.89** | 0.81** | -0.07 | -0.25 | 0.05 | -0.46 | 0.80** | 0.46* | 0.78** | 0.54 | 1.00* | |
| ROOT DRY WT(S) | 0.65** | 0.84** | 0.77** | 0.87** | 0.75** | 0.98** | 0.02 | -0.12 | -0.42 | -0.33 | 0.79** | -0.01 | 0.45 | 0.86 | 0.86** | 1** |

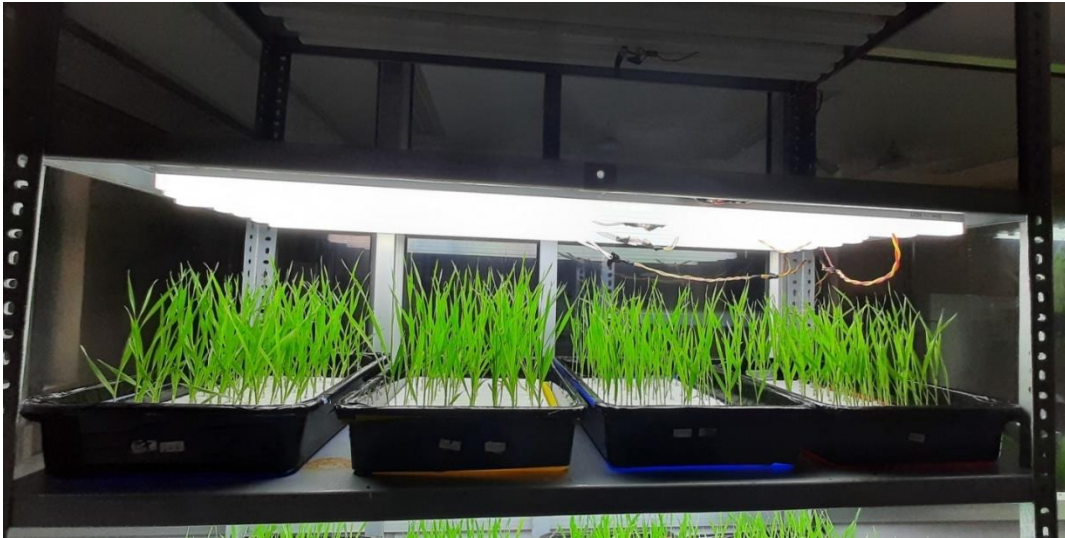


Plate 5: Plants before osmotic stress (at 14th day)



Plate 6: Plants after water stress (24th day)



Plate 7: Root System of genotype TAW9 and TAW98



Plate 8: Comparison of root system of TAW95(2) and TAW98 (3) with HD2967(parent)(1)

DISCUSSION

"Evaluation of drought tolerance in gamma mutagenised wheat genotypes under controlled conditions," is the objective of present experimental study. The screening of drought tolerant lines is based on the genotype, target environment and GXE interaction (Blume 1996). The utilization of a hydroponic system in off-field condition is an innovative approach of doing research throughout the year under controlled condition (Sziraet *al.*, 2008, Hoffmann *et al.*, 2012). The mutant line from BARC were employed in this work for evaluation under water stress and control conditions of wheat genotypes in seedling stage in department of genetics and plant breeding, IAS, BHU.

The genotypes are analysed on the basis of following parameters:

Mean and Range performance

ANOVA (Analysis of variance)

Stress tolerance index

Phenotypic correlation between different traits

5.1 ANOVA (analysis of variance)

In this experiment, 10 wheat genotypes (9 mutants and one parent) were screened for drought tolerance. For all the traits studied under controlled and stressed conditions, such as shoot length (SL), root length (RL), chlorophyll content, fresh root and shoot weight, dry root and shoot, percentage damage and relative water content (RWC), the F_{cal} and P-value of analysis of variance revealed that there is significant difference under control and water stress condition inferring treatment effective in differentiating genotypes in comparison to control condition. Therefore best performing genotypes can be evaluated for further studies (Zarei *et al.*, (2007),

Khakwaniet *al.*,(2011), Baloch *et al.*, (2012), and Ayalewet *al.*, (2015), and many other researcher's found similar conclusion from their respective experimentations.

5.2 Mean and Range performance

As per the research individual genotypic performance indicates the value of the materials and it provides breeders with basic knowledge. The average of several features of accessible genotypes revealed that selecting appropriate genotypes based on the qualities analysed in the materials may be useful. Under stress and control conditions, mutant lines along with their parent evaluated for shoot length (SL),root length (RL),chlorophyll content, fresh root and shoot weight, dry root and shoot,percentage damage and relative water content.(RWC) for preliminary screening of drought tolerant genotype.

The effect of water stress shown in (table 4.2.2.1) considerably altered the chlorophyll content of a 24 -day-old seedling, and TAW97 has the highest chlorophyll content followed by TAW95 and TAW98 with mean value 29.5,29.1 and 29 whereas TAW19(23.7) has the lowest chlorophyll content among all genotypes as well as parent(HD2967).

Under non-stress conditions, the chlorophyll content was highest in TAW122followed by TAW98 and TAW95with mean 36.2 and 36,35.6whereas TAW 132(33.3) has the lowest chlorophyll content among all genotypes as well as parent (HD2967).

The effect of water stress shown in (table 4.2.3.1) considerably altered the shoot length of a 24 -day-old seedling, and TAW95 has the highest shoot length followed by TAW132 and TAW98with mean 24.65,24.62 and 24.3whereas TAW 15(20.03) has the lowest among genotypes as well as parent (HD2967).

Under non-stress conditions, the shoot length was highest in TAW98 followed by TAW95 with the mean 33.8 and 32.6 whereas TAW15 (28.5) has the lowest among all genotypes as well as parent (HD2967).

The effect of water stress shown in (table 4.2.4.1) considerably altered the root length of a 24 -day-old seedling, and TAW95 has the highest root length followed by TAW97 and TAW98 with mean 114.4,13.5 and 13.4 whereas TAW15(9.1) has the lowest among genotypes as well as parent (HD2967).

Under non-stress conditions, the rot length was highest in TAW132 followed by TAW122 with mean 29.2 and 28.8and whereas TAW19(15.1) has the lowest root length among all genotypes as well as parent (HD2967).

The effect of water stress shown in (table 4.2.5.1) considerably altered the fresh weight of shoot of a 24 -day-old seedling, and TAW95 has the highest fresh weight(g)followed by TAW98 with mean 0.71 and 0.7g respectively whereas TAW132(0.4) has the lowest among genotypes as well as parent (HD2967).

Under non-stress conditions, the fresh weight of shoot(g) was highest in TAW98followed by TAW95 with mean value 1.8 and 1.79 whereas TAW 19(1.07g)has the lowest fresh weight (g) among all genotypes as well as parent (HD2967).

The effect of water stress shown in (table 4.2.6.1) considerably altered the fresh weight of root of a 24 -day-old seedling, and TAW98 has the highest fresh weight(g)followed by TAW95 with the mean 0.37 and 0.36 whereas TAW19(0.21)has the lowest among genotypes as well as parent (HD2967).

Under non-stress conditions, the fresh weight of root(g) was highest in TAW98followed by TAW122 and TAW95 with mean 0.81,0.8 and 0.79whereas TAW19(0.54)has the lowest fresh weight (g) among all genotypes as well as parent (HD2967).

The effect of water stress shown in (table 4.2.7.1) considerably altered the root number of a 24 -day-old seedling, and TAW132 has the highest root number followed by TAW117 with mean 8.6 and 8.4 whereas TAW19(6) has the lowest among genotypes as well as parent (HD2967).

Under non-stress conditions, the root number was highest in TAW98 followed by TAW95 with mean of 11 and 10 respectively and TAW19(8.1) whereas TAW19 has the lowest root number among all genotypes as well as parent (HD2967).

5.3 Stress Tolerance Index (%)

The data for Root Length Stress Tolerance Index (%), Shoot Length Stress Tolerance Index (%), StressTolerance Index (%) of shoot and root.

In this experiment, STI of shoot length TAW132(77.1%) had highest whereas genotype TAW117 (65.04%) had lowest STI among all lines as well as parent HD2967.

In this experiment, STI for root length TAW97(80.3%) had highest whereas genotype TAW15 (34.8%) had lowest STI among all lines as well as parent HD2967.

In this experiment, STI for root weight TAW114(47.5%) had highest whereas genotype TAW15(25.5%) had lowest STI among all lines as well as parent HD2967.

In this experiment, STI of shoot weight TAW19(57%) had highest whereas genotype TAW132 (35.5%) had lowest STI among all lines as well as parent HD2967.

SUMMARY AND CONCLUSION

Wheat (scientific name: *Triticumaestivum L.*) is the world's staple food crop and India's second most significant crop after rice. Its productivity and yield are heavily impacted by the genotypes chosen, as well as the soil and ambient conditions. Drought is now limiting the productivity of wheat. The drought had a considerable impact on plant growth and development, according to the majority of the experts. However, most plants contain a variety of stress-resistance systems. Wheat genotypes are screened on the basis of morpho-physiological parameters to increase our current understanding of stress tolerance. The data taken and analysed will help us to discover some selection criteria that may be effective in producing water stress tolerant genotypes. Keeping in view all of these factors, the current study is titled as “Evaluation of Drought tolerance in gamma mutagenised wheat genotypes under controlled condition”

The experiment is conducted under controlled condition in wheat laboratory, department of genetics and plant breeding, Institute of Agricultural Sciences (IAS), BHU to screen drought tolerant mutant wheat genotype.

The plants are analysed in terms of various traits and parameters along with several replications in a Completely Randomised Designs(CRD).

Polyethylene glycol (PEG), is a chemical inducing water stress under lab conditions and widely used to screen water stress tolerant genotypes at the seedling stage of the crop. Therefore, this experiment is conducted with 9 mutant wheat genotypes along with parent at 20%PEG 8000 stress level (induces osmotic stress condition in roots) at seedling stage and tolerant genotypes are screened for further evaluation.

From this experimentation following inferences were drawn

1. The analysis of variance(ANOVA) showed significant difference for treatment(water stress) and control, indicating that there is substantial variability among the 10 genotypes for treatment and hence selection is effective for screening of promising genotypes for drought tolerance.
2. Mean performance for numerous attributes in stress and non-stress conditions has shown wide range of variability under the present experiment.
3. The studies on correlation coefficient under stressed conditions revealed that fresh shoot weight has a significant positive correlation with shoot length, root length, fresh root weight, dry shoot weight, and dry root weight, whereas under non-stress conditions, fresh shoot weight showed a significant positive correlation with shoot length, root length, fresh root weight, dry shoot weight, and dry root weight. Therefore, root length and dry root weight could be employed for direct selection of drought tolerant genotypes.
4. In a study on inter relationship of stress tolerance index for dry root weight and morphological characters, the genotypes TAW95 and TAW98 has found to be most resilient genotypes, since they performed better than other genotypes as well as their parent under both osmotic stress and control condition and hence can be further evaluated for water stress.

☪

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APPENDIX I

Chlorophyll content under control condition

| TAW19 | TAW95 | TAW97 | TAW98 | TAW114 | TAW117 | TAW122 | TAW132 | TAW15 | HD2967 |
|-----------------|--------------|-----------------|--------------|---------------|--------------|-----------------|--------------|---------------|-----------------|
| 35.9 | 38.4 | 30.8 | 38.7 | 40.4 | 36.3 | 36.8 | 35.7 | 34.6 | 35.54 |
| 34.5 | 31.9 | 39.1 | 39.1 | 33.5 | 39.4 | 38.7 | 30.3 | 33.6 | 36.04 |
| 34.4 | 36 | 37.9 | 39.6 | 33 | 34.2 | 35.6 | 34.5 | 34.7 | 38.5 |
| 35 | 37.1 | 34.2 | 37.3 | 35.2 | 38.4 | 35.7 | 36.5 | 39.9 | 36.48 |
| 35.3 | 31.4 | 33.4 | 39.1 | 30 | 33.1 | 34.9 | 34.3 | 34.4 | 35.5 |
| 31.7 | 37.3 | 34 | 35.3 | 35.6 | 34 | 36.8 | 32.2 | 34.6 | 35.38 |
| 36.4 | 35.8 | 37 | 38.9 | 37.4 | 30.6 | 33.8 | 37.8 | 34 | 35.16 |
| | 35.6 | 36 | 37.3 | 34.6 | 35.2 | 39.4 | 31.5 | 37.4 | 35.64 |
| | 38.1 | 37 | 33.7 | 37.3 | 38.8 | 37.5 | 30.9 | 33 | 36.375 |
| | 34.5 | | 41 | 37 | 33 | | 36.9 | 36.4 | 37.36667 |
| | 37.9 | 36.4 | 38.2 | 34.7 | 28.2 | 35.5 | 36.3 | 36.3 | 31.76 |
| | 39.7 | 37.4 | 35 | 32.3 | 29.3 | 40.8 | 32.1 | 38.2 | 32.7 |
| | 35 | 31 | 34.6 | 34.1 | 36.1 | 34.7 | 30.9 | 31.5 | 31.42 |
| | 36.3 | 33.2 | 34.2 | 31.3 | 35.6 | 37.3 | 32.6 | 29 | 33.8 |
| | 32 | 31.6 | 31.2 | 30.1 | 33.1 | 36.7 | 35.4 | 31.4 | 32.84 |
| | 35 | 30.5 | 34.3 | 32.1 | 31.3 | 31.9 | 32.4 | 37 | 33.46 |
| | 33.8 | 31.3 | 34.8 | 35.4 | 34 | 34.7 | 31.6 | 28.8 | 35.24 |
| | 34.4 | 34.1 | 32.9 | 32.8 | 29.9 | 39.4 | 32.5 | 25.9 | 34.88 |
| | 33.5 | 30.2 | 32.4 | 33.1 | 28 | 36.2 | 30.2 | 30.2 | 33.66 |
| | 38.5 | 32.1 | 34.2 | 33.2 | 34.5 | 32 | 31.6 | 29.8 | 33.74 |
| 33.65294 | 35.61 | 34.06316 | 36.09 | 34.155 | 33.65 | 36.23158 | 33.31 | 33.535 | 34.77408 |

APPENDIX II

Shoot length (cm) under control condition

| | TAW19 | TAW95 | TAW97 | TAW98 | TAW114 | TAW117 | TAW122 | TAW132 | TAW15 | HD2967 |
|----------------|---------|--------|----------|--------|--------|---------|---------|--------|-------|---------|
| 1 | 38 | 37 | 28 | 34 | 32 | 28.7 | 28.3 | 29 | 24.5 | 30.48 |
| 2 | 39 | 41 | 29 | 38 | 30 | 30.7 | 34.5 | 29.7 | 22.6 | 30.36 |
| 3 | 30 | 32 | 31 | 34 | 30 | 33.3 | 34.6 | 29.4 | 26 | 29.68 |
| 4 | 34 | 38 | 31 | 33 | 34 | 22.7 | 35 | 30 | 22 | 33.48 |
| 5 | 30 | 40 | 30 | 33 | 36 | 26.6 | 27 | 35 | 23.3 | 31.34 |
| 6 | 38 | 33 | 31 | 35 | 35 | 33.7 | 26.7 | 28.4 | 25.3 | 29.32 |
| 7 | 27 | 32 | 37 | 40 | 29 | 32 | 28.4 | 29 | 30.4 | 31.9 |
| 8 | 39 | 42 | 29 | 33 | 29 | 29 | 27.3 | 29.4 | 24 | 32.02 |
| 9 | 30 | 30 | 33 | 33 | 33 | 31.3 | 35.2 | 32 | 23.2 | 31.025 |
| | 34 | 24 | 36 | 34 | 35 | 32.8 | | 32 | 26.8 | 29.6667 |
| 1 | 29 | 30 | 38 | 38.5 | 36 | 25.8 | 33.8 | 34.2 | 25.6 | 29.85 |
| 2 | 32 | 31 | 28 | 32 | 34 | 35.1 | 34.6 | 32.4 | 33.4 | 32.175 |
| 3 | 33 | 27 | 32.5 | 30 | 33 | 36.7 | 36 | 32.6 | 32.8 | 33.375 |
| 4 | 24 | 29 | 26 | 28 | 32 | 34 | 38 | 35 | 29.9 | 31.075 |
| 5 | 24 | 28 | 36 | 29 | 36 | 34.7 | 35.4 | 32.3 | 30.7 | 32.15 |
| 6 | 24.5 | 30 | 37 | 29 | 28 | 35.6 | 32.7 | 36.8 | 33.3 | 31.675 |
| 7 | 31 | 28.5 | 28 | 29 | 26.5 | 30.9 | 34.3 | 32.2 | 39.2 | 33.025 |
| 8 | | 29 | 28.5 | 38 | 34 | 35.4 | 38.2 | 32 | 30.6 | 30.75 |
| 9 | | 40 | 36 | 36 | 31 | 26.8 | 31.8 | 31.6 | 35.6 | 31.7667 |
| 10 | | 31 | | 41 | 33 | 38.49 | 34.5 | 30 | 31.8 | 31.1 |
| CONTROL | 31.5588 | 32.625 | 31.84211 | 33.875 | 32.325 | 31.7145 | 32.9632 | 31.65 | 28.55 | 31.3107 |

APPENDIX III

Shoot length (cm) under stress condition

| READING | TAW19 | TAW95 | TAW97 | TAW98 | TAW114 | TAW117 | TAW122 | TAW132 | TAW15 | HD2967 |
|---------------|-------|-------|--------|-------|--------|--------|--------|--------|--------|--------|
| 1 | 12.5 | 23 | 20.1 | 20.5 | 20 | 20.2 | 25 | 21.3 | 18.5 | 21.12 |
| 2 | 20.5 | 23 | 16.5 | 22.5 | 18.5 | 22.6 | 25.5 | 26 | 18 | 22.4 |
| 3 | 17.5 | 23.5 | 24.2 | 24 | 21.5 | 22 | 22.5 | 24 | 19.5 | 21.94 |
| 4 | 21 | 22 | 19 | 21 | 22 | 21 | 21.5 | 24.6 | 21 | 21.24 |
| 5 | 15 | 20 | 23 | 26 | 20 | 20 | 23 | 23.4 | 20.7 | 19.4 |
| 6 | 21.5 | 24 | 21 | 22.5 | 22.5 | 17.7 | 18 | 24.5 | 21 | 20.96 |
| 7 | 18.5 | 24.5 | 22 | 23 | 12 | 17.9 | 22.5 | 25.7 | 20.6 | 21.1 |
| 8 | 19 | 25 | 22.5 | 24 | 17.5 | 22 | 23 | 25.9 | 17.3 | 22.1 |
| 9 | 17 | 25 | 18.5 | 26 | 20 | 20.5 | 24 | 25.2 | 21 | 23.05 |
| 10 | 22 | 24.5 | 23.5 | 26 | 21.5 | 20 | 23 | 17.2 | 17.1 | 20 |
| 1 | 11 | 25 | 23 | 25 | 23 | 22 | 25 | 25 | 22 | 23.75 |
| 2 | 23 | 26.5 | 26.5 | 26 | 25.5 | 24 | 23 | 26.8 | 21 | 23.75 |
| 3 | 23 | 24 | 28 | 26.5 | 26.5 | 22 | 23 | 27 | 22 | 22.88 |
| 4 | 25 | 27 | 26 | 26.5 | 26 | 16 | 23 | 25 | 21 | 22.38 |
| 5 | 23.5 | 25 | 29.5 | 25 | 24 | 18 | 24 | 23 | 21 | 23.15 |
| 6 | 22 | 27.5 | 27.5 | 25 | 28 | 21 | 25 | 24 | 19 | 21 |
| 7 | 22.5 | 24.5 | 29.5 | 25 | 26 | 24 | 19 | 25 | 17 | 24.5 |
| 8 | 25 | 27 | 28.5 | 23.5 | 30 | 22 | 23 | 27 | 22 | 21.5 |
| 9 | 26 | 25 | 30.1 | 25 | 26.5 | 23 | 23 | 26 | 21 | 23.25 |
| 10 | 26 | 27 | 25 | 23 | 26.5 | 21 | 24 | | 20 | 24.38 |
| STRESS | 20.58 | 24.65 | 24.195 | 24.3 | 22.737 | 20.845 | 23 | 24.62 | 20.035 | 22.19 |

APPENDIX IV

Root length (cm) under stress condition

| READING | TAW19 | TAW95 | TAW97 | TAW98 | TAW114 | TAW117 | TAW122 | TAW132 | TAW15 | HD2967 |
|---------|--------|-------|--------|--------|--------|--------|--------|--------|-------|--------|
| 1 | 12 | 13.5 | 10.2 | 14.5 | 6 | 10.5 | 14.5 | 14.8 | 7 | 10.72 |
| 2 | 11.5 | 12.5 | 10 | 8 | 9 | 7 | 9 | 12.5 | 7 | 9.58 |
| 3 | 11.5 | 12.5 | 6 | 12.5 | 10 | 12 | 7 | 8.6 | 9.7 | 10.66 |
| 4 | 7.8 | 11.5 | 10 | 12.5 | 10.5 | 10.7 | 18 | 13.9 | 10.5 | 11.08 |
| 5 | 9 | 15 | 8 | 11 | 13 | 12.5 | 10.5 | 14.2 | 8.1 | 10.76 |
| 6 | 10.5 | 13 | 13 | 11 | 12 | 13 | 14.5 | 15.2 | 7.2 | 10.38 |
| 7 | 7 | 12 | 12 | 11.5 | 6.5 | 10.4 | 7.5 | 13.4 | 6.6 | 11.5 |
| 8 | 11.5 | 13 | 12 | 10 | 7 | 11 | 8.5 | 14.6 | 8.6 | 9.925 |
| 9 | 8 | 10 | 11.8 | 11 | 11 | 11.5 | 8 | 10.1 | 12.9 | 8.85 |
| 10 | 11.5 | 12 | 13 | 11 | 10 | 12.5 | 12.5 | 9.8 | 4.8 | 9.8333 |
| 1 | 15.5 | 15 | 15 | 16.5 | 13 | 8 | 14 | 12 | 17 | 11.625 |
| 2 | 15.5 | 15 | 18 | 16 | 14 | 10 | 13 | 12.9 | 9 | 12.625 |
| 3 | 14 | 16 | 19.5 | 15 | 12 | 16 | 14 | 14 | 12 | 13.75 |
| 4 | 12 | 16 | 18 | 14 | 15 | 18 | 13 | 13 | 10 | 14.5 |
| 5 | 14.5 | 16.5 | 16 | 16 | 16.6 | 13 | 14 | 16 | 7 | 15.75 |
| 6 | 13 | 16.5 | 17.5 | 15 | 14.5 | 9 | 15 | 13.9 | 9 | 12.75 |
| 7 | 15 | 18.5 | 14.5 | 15.5 | 13 | 13 | 11 | 14 | 9 | 14 |
| 8 | 11.5 | 15.5 | 18 | 16.2 | 15 | 12 | 12 | 15 | 11 | 13.75 |
| 9 | 14 | 18 | 14.5 | 15.5 | 14 | 12 | 15 | 12.6 | 8.5 | 13 |
| 10 | 12 | 17 | 13.5 | 15.4 | 14 | | 12 | 15 | 11 | 11.75 |
| MEAN | 11.865 | 14.45 | 13.525 | 13.405 | 11.81 | 11.69 | 12.15 | 13.275 | 9.295 | 11.839 |

APPENDIX V

Fresh shoot weight under stress condition

| READING | TAW19 | TAW95 | TAW97 | TAW98 | TAW14 | TAW17 | TAW22 | TAW1 | TAW15 | HD296 |
|---------------------|--------------|-------------|--------------|--------------|---------------|---------------|---------------|---------------|--------------|-----------------|
| 1 | 0.455 | 0.55 | 0.4 | 0.51 | 0.475 | 0.45 | 0.3 | 0.575 | 0.75 | 0.56875 |
| 2 | 0.44 | 0.575 | 0.565 | 0.6 | 0.24 | 0.68 | 0.6 | 0.22 | 0.6 | 0.57 |
| 3 | 0.32 | 0.7 | 0.44 | 0.65 | 0.45 | 0.565 | 0.7 | 0.4 | 0.7 | 0.4575 |
| 4 | 0.33 | 0.56 | 0.51 | 0.6 | 0.415 | 0.595 | 0.95 | 0.475 | 0.32 | 0.545 |
| 5 | 0.494 | 0.7 | 0.405 | 0.75 | 0.355 | 0.459 | 0.85 | 0.345 | 0.375 | 0.573333 |
| 1 | 0.52 | 0.68 | 1.095 | 0.82 | 0.87 | 0.345 | 0.445 | 0.32 | 0.29 | 0.471667 |
| 2 | 0.55 | 0.835 | 0.68 | 0.69 | 0.725 | 0.42 | 0.39 | 0.465 | 0.305 | 0.458333 |
| 3 | 0.475 | 0.91 | 0.905 | 0.77 | 1.005 | 0.425 | 0.435 | 0.54 | 0.26 | 0.515 |
| 4 | 0.435 | 0.825 | 0.615 | 0.87 | 0.95 | 0.335 | 0.27 | 0.335 | 0.305 | 0.54 |
| 5 | 0.31 | 0.82 | 0.84 | 0.765 | 1.04 | 0.345 | 0.255 | 0.38 | 0.22 | |
| WATER STRESS | 0.432 | 0.71 | 0.645 | 0.702 | 0.6525 | 0.4619 | 0.5195 | 0.4055 | 0.412 | 0.5221 |

APPENDIX VI

Fresh root weight under stress condition

| READING | TAW19 | TAW95 | TAW97 | TAW98 | TAW114 | TAW117 | TAW122 | TAW132 | TAW15 | HD2967 |
|---------------|----------------|---------------|--------------|--------------|-------------|---------------|--------------|--------------|-------------|---------------|
| 1 | 0.195 | 0.375 | 0.39 | 0.35 | 0.38 | 0.27 | 0.25 | 0.285 | 0.185 | 0.3175 |
| 2 | 0.185 | 0.43 | 0.44 | 0.435 | 0.335 | 0.21 | 0.325 | 0.325 | 0.27 | 0.2775 |
| 3 | 0.32 | 0.5 | 0.405 | 0.41 | 0.435 | 0.24 | 0.28 | 0.285 | 0.51 | 0.395 |
| 4 | 0.34 | 0.41 | 0.325 | 0.465 | 0.475 | 0.195 | 0.21 | 0.325 | 0.35 | 0.395 |
| 5 | 0.215 | 0.375 | 0.45 | 0.33 | 0.455 | 0.18 | 0.26 | 0.32 | 0.385 | 0.215 |
| 1 | 0.3 | 0.35 | 0.2 | 0.285 | 0.275 | 0.15 | 0.38 | 0.265 | 0.165 | 0.302 |
| 2 | 0.165 | 0.33 | 0.23 | 0.39 | 0.225 | 0.355 | 0.5 | 0.305 | 0.25 | 0.268 |
| 3 | 0.12 | 0.29 | 0.28 | 0.39 | 0.23 | 0.4 | 0.35 | 0.265 | 0.49 | 0.296 |
| 4 | 0.12 | 0.34 | 0.21 | 0.305 | 0.25 | 0.39 | 0.36 | 0.305 | 0.33 | 0.269 |
| 5 | 0.1425 | 0.265 | 0.18 | 0.4 | 0.14 | 0.385 | 0.245 | 0.3 | 0.365 | 0.23 |
| STRESS | 0.21025 | 0.3665 | 0.311 | 0.376 | 0.32 | 0.2775 | 0.316 | 0.298 | 0.33 | 0.2965 |