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# Comparative behavior of terminal heat tolerant (WH 730) and intolerant (Raj 4014) hexaploid wheat genotypes at germination and growth at early stage under varying temperature regimes

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**Wheat (*Triticum aestivum* L.) is a winter season crop and India holds second rank in production globally. Several reports have supported the facts that heat and cold stress adversely affect the germination, crop growth and development. Terminal heat stress is a major problem for wheat production which severely affects the grain yield. Two wheat genotype *viz.* WH 730 (heat tolerant) and Raj 4014 (heat intolerant) identified from previous field experiments were selected to study the effect of heat and cold treatment on the germination and growth at early stage. High temperature stress was imposed by exposing soaked seeds to 35 and 40°C temperature for 6 h. On the other hand for cold stress, seeds were exposed to 0°C for 2 h. Results indicate that heat treatment at 35°C had no significant effect on germination efficiency of the genotype WH 730, while same was reduced in the genotype Raj 4014 after 24 and 48 h of heat treatment. Germinated seedlings exposed to 35°C showed improved vigour over control seedlings of both genotypes. The genotype WH 730 which was heat tolerant at grain filling stage had longer shoot and root relative to Raj 4014 during growth at early stage (13 days after heat treatment). At 40°C, the germination was poor and both genotypes could not grow under hydroponic conditions. Furthermore, when both genotypes were checked for cold tolerance, null effect was observed on the germination (24 and 48 h post cold treatment) but the growth of the genotype WH 730 at early stage (13 days post cold treatment) was found to be vigorous as compared to Raj 4014 which was drastically reduced by cold treatment and could not hold longer under hydroponic condition. The study showed that the genotype WH 730 which was tolerant to terminal heat stress has also shown tolerance during germination and growth at early stage under both heat and cold stress.**

**Key words:** Heat treatment, cold treatment, hexaploid wheat, hydroponics, tolerance.

## INTRODUCTION

Wheat (*Triticum aestivum* L.) is one of the most important staple food crops of the world. It occupies 17% of the world's crop acreage, feeding about 40% of the world population by providing 20% of total food calories and protein in human nutrition (Gupta et al., 2008). In India, wheat sowing time varies from October to December with temperature range of 10 to 35°C. Apart from genotypic variation, temperature fluctuation plays a crucial role in germination of wheat seeds. Stress affects essentially every aspect of plant growth and development by

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influencing the entire metabolism. The stress level is mainly determined by the degree and duration of stress, developmental stages and time of stress (Gupta and Sheoran, 1983). For heat and cold tolerance, plant involves a number of physiological and biochemical changes *viz.* alteration in membrane structure and function, tissue water content, composition of protein, lipids, primary and secondary metabolites which are tightly regulated by a complex network of multiple genes (Zhang et al., 2005; Kotak et al., 2007; Barnabas et al., 2008; Bohnert et al., 1995). Previous studies have shown that germination depends on the ability of seed to utilize reserve material more efficiently (Rao and Sinha, 1993), and mobilization of seed reserve for germination traits (Penning de Vries et al., 1979). Temperature is a critical factor in germination since it can influence the rate of water absorption and other substrate supply that are necessary for germination, growth and development (Wanjura and Buxtor, 1972; Essemine et al., 2002). Read and Beaten (1963), observed antagonistic relationship between rate of wheat seed germination and low temperature with decreasing soil moisture content without affecting the total germination. The minimum and maximum temperatures can significantly affect the seeds from germination up to maturity stage (Lobell and Ortiz-Monasterio, 2007).

Germination may constitute a precocious investigation for tolerance of plants subjected to heat stress. Hard wheat seeds have a good potential to germinate and develop into seedlings upon exposure to optimal thermal condition (25°C) (Dell' Aquila and Spada, 1994). On the contrary, low and high temperature significantly modulates the velocity and capacity of germination (Bewley and Black, 1982). As a result of complete viability loss, excess temperature treatment leads to death of some seeds and to delayed germination for surviving seeds (Hutchinson, 1944). The normal growth of delayed germinating seeds is unable to withstand adverse condition and become prone to mold infection (Greer, 1953). Several reports have been published to explain the factors responsible for the thermal damage of seeds which includes membrane damage (Thomas 1941), change in chemical composition (Barton 1961), protein degradation, inhibition of protein synthesis, inactivation of enzymes in chlorophyll and mitochondria (Howarth 2005) but no single factor is fully responsible for the thermal damage. Heat stress has an obvious response to decreased synthesis of normal protein which is more pronounced at 40 to 45°C. This decline leads to increased synthesis of a new set of protein known as heat shock protein (Perras and Sarhan, 1989). No single trait fully explains why some wheat cultivars are able to give better yield even in the presence of heat stress.

The rapid and uniform field emergence of seed is essential to achieve better growth and high yield. The optimum temperature favors wheat seed to have good germination capacity whereas extreme low and high temperature leads to delay in germination or abolish the germination and early growth due to poor nutrient supply which might be due to poor enzymatic activity at extreme temperature condition. The present study was carried out with an objective to examine the effect of heat and cold treatment on germination and growth at early stage of two wheat cultivars *viz.* WH 730 (heat tolerant) and Raj 4014 (heat intolerant) belonging to north western plain zone (NWPZ) of India.

## MATERIALS AND METHODS

### Experimental materials and treatment condition

The present study was carried out at National Phytotron Facility (NPF), Pusa Campus, New Delhi and National Research Center on Plant Biotechnology (NRCPB), New Delhi, India. Two wheat genotypes *viz.* WH 730 (Kundu et al., 2010) and Raj 4014 (DWR annual report 2010-2011) were selected to examine the effect of heat and cold treatment on germination and growth at early stage. The seeds were procured from the genetic resource unit (GRU), DWR, Karnal, Haryana, India. The pedigree details of the genotypes are given in Table 1. Genotype WH-730 was attributed primarily by its stay green cover and grain-filling under heat stress while Raj-4014 was attributed by high yield but sensitive to heat stress. Seeds of both genotypes were tested under two temperature regimes (35 and 40°C for 6 h) for heat treatment and one temperature (0°C for 2 h) for cold treatment. 25°C was taken as a control throughout the experiment.

### Methods

Seeds were washed twice with deionized water to remove the dust particle followed by soaking in deionized water in beaker and kept overnight (16 h) to mobilize the stored reserve materials to initiate the germination process. Current methodology for the experiment was followed as per Buriro et al. (2011) with slight modification. Soaked seeds were then transferred to 100 × 107 × 20 mm sterile Petri dish, at 10 seeds/Petri dishes, which is manually coated with same size of blotting sheets. The coated Petri dishes were wet prior to heat and cold treatment. The Petri dishes were transferred to defined temperature regime for heat (35 and 40°C) and cold (0°C) treatment in three different preset incubators while respective controls were kept at room temperature (25°C). Heat and cold treatment were given for 6 and 2 h, respectively. Whole experiment was done using three independent biological replications. After heat and cold treatment, each Petri dish was shifted to room temperature (25°C) for the germination to begin. Watering was done regularly as per requirement of the seedlings. Observations were recorded at two stages of germination (24 and 48 h post heat and cold treatment). Four days old seedlings were then transferred to hydroponic condition in Hoagland medium. These seedlings were allowed to grow up to next nine days for recording of the different physiological parameters *viz.* survival rate under hydroponic condition, root and shoot length.

## RESULTS

### Effect of heat treatment on germination efficiency of genotypes WH 730 and Raj 4014

Seed germination and vigorous seedlings are two important pre-requisite for the proper establishment of

**Table 1.** Pedigree details of the genotypes taken for the study.

| Genotype | Source of Genotype | Pedigree |
|---|---|---|
| WH 730 | CCSHAU, Hisar, India | CPAN 2092/Improved Lok-1 |
| Raj 4014 | ARS, Durgapura, Jaipur, India | DL 8025/K 9011 |

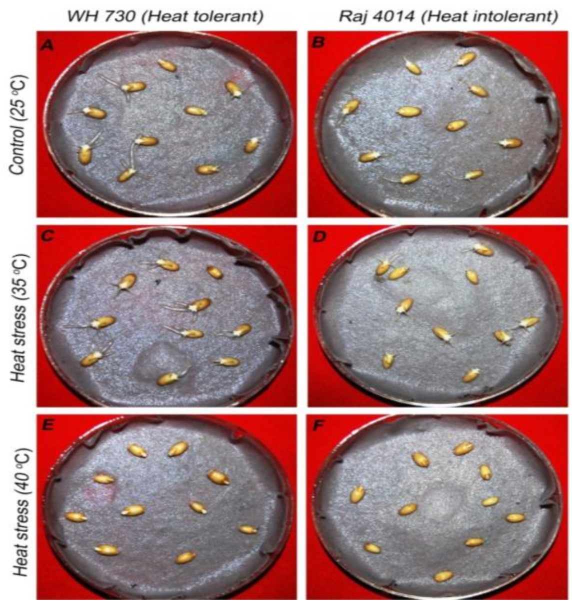


**Figure 1.** Evaluation of germination efficiency and vigour of soaked seeds of genotype WH 730 (heat tolerant) and Raj 4014 (heat intolerant) after 24 h of heat treatment.

wheat crop. Various geographical regions are characterized by different temperature that is why wheat requires a wide range of temperature from germination to maturity. Temperature range of 18 to 25°C is considered ambient for wheat seed germination. Any fluctuation in the ambient temperature may affect the germination and crop stand. This study shows that heat treatment of 35°C had no significant harmful effect on germination efficiency of the genotype WH 730 while same was reduced in the genotype Raj 4014 after 24 and 48 h of treatment. The germinated seeds of both genotypes showed improved vigour over control plants (Figure 1C, D and 2C, D). The mean value of combined germination efficiency of all the replicates was observed to be ~100 and ~90% for the genotype WH 730 and Raj 4014, respectively (Figure 2H). In contrast, when both genotypes were treated with 40°C, the germination was found to be very poor which was less visible at 24 h post treatment but more clearly visible at 48 h post treatment (Figure 1E, F and 2E, F). The mean value of germination efficiency was ~40 and ~7% for WH 730 and Raj 4014, respectively (Figure 2I) but these germinated seeds could not survive for long and died soon after.

### Effect of cold treatment on germination efficiency of genotype WH 730 and Raj 4014

Although wheat is a cold loving plant, sensitivity to cold tolerance depends on the degree and duration of exposure and growth stage. In our study, seeds of both genotypes were exposed to 0°C for 2 h to observe any possible adverse effect on germination and growth at early stage. Our results indicate that relative to their respective control, cold treatment imparted no significant alteration in germination efficiency and vigour of both genotypes after 24 and 48 h of treatment (Figures 3 and 4). The germination efficiency was ~100% for both genotypes (Figure 4F).

### Evaluation of survival rate of heat and cold treated genotypes WH 730 and Raj 4014 under hydroponic condition

As mentioned earlier, after 4 days of germination in Petri dishes, the treated seedlings were transferred under hydroponic condition to grow for the next 9 days. When

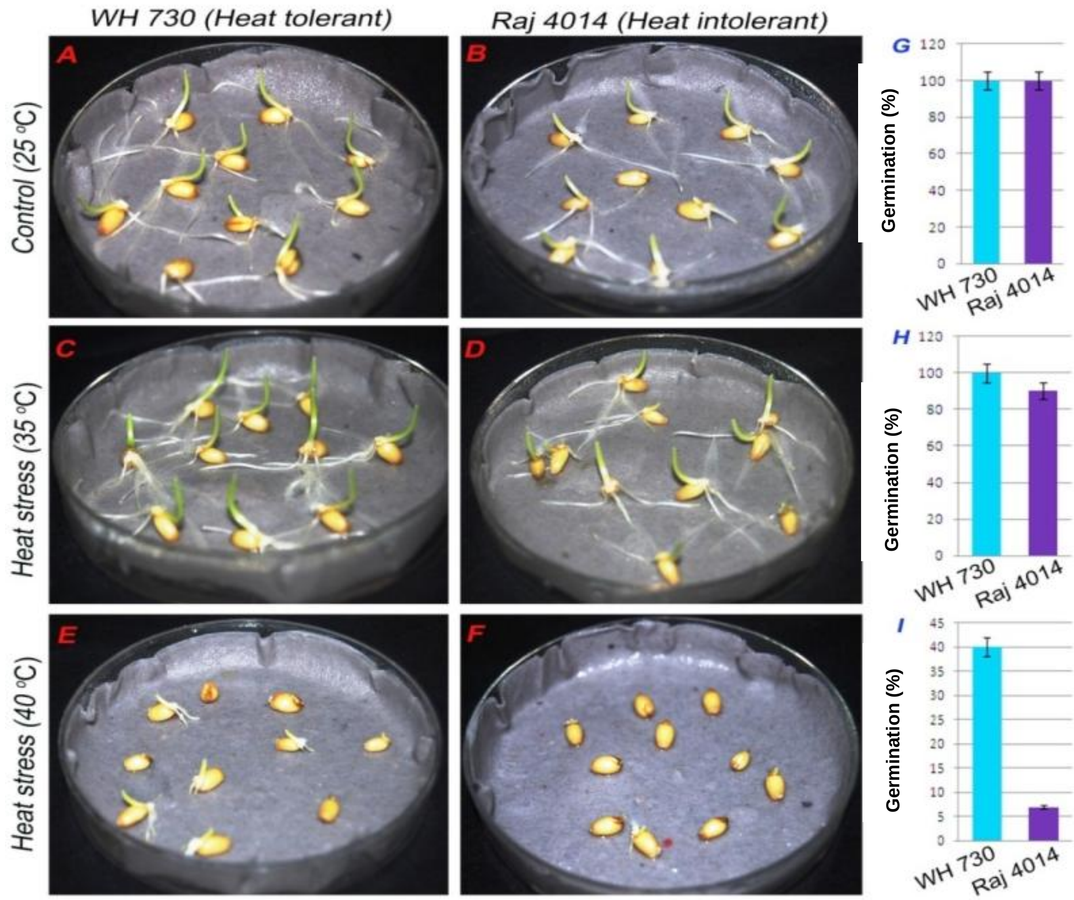


**Figure 2.** Evaluation of germination efficiency and vigour of soaked seeds of genotype WH 730 (heat tolerant) and Raj 4014 (heat intolerant) after 48 h of heat treatment.

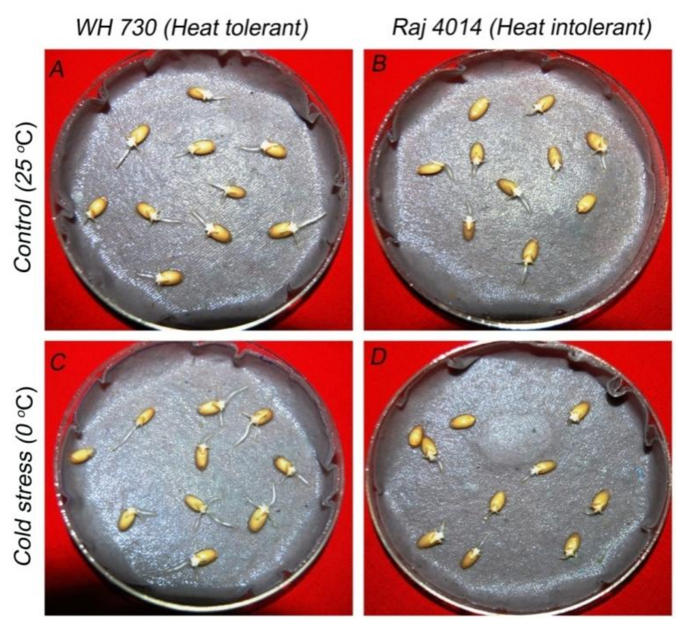


**Figure 3.** Evaluation of germination efficiency and vigour of soaked seeds of genotype WH 730 (heat tolerant) and Raj 4014 (heat intolerant) after 24 h of cold treatment.

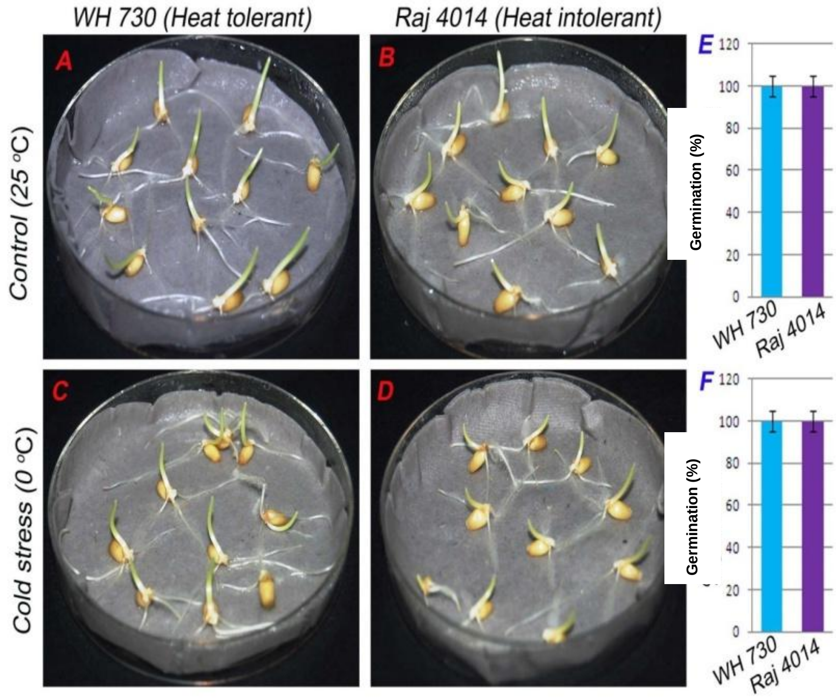


**Figure 4.** Evaluation of germination efficiency and vigour of soaked seeds of genotype WH 730 (heat tolerant) and Raj 4014 (heat intolerant) after 48 h of cold treatment.

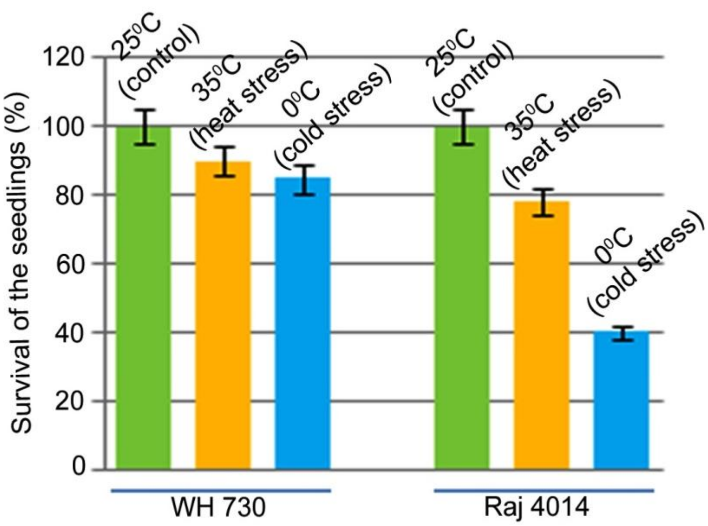


**Figure 5.** Survival rate of heat and cold treated genotype WH 730 and Raj 4014 under hydroponic condition.

germinated seedlings (prior treated with 35°C), ~100%-WH 730 and ~90%-Raj 4014, were transferred to hydroponic condition, we found ~90 and ~78% survival for WH 730 and Raj 4014, respectively. In contrast, cold treatment of 0°C drastically reduced the survival (mean value ~40%) of the heat intolerant genotype Raj 4014 under hydroponic condition, whereas heat tolerant genotype WH 730 was comparatively less affected with mean value of ~85%. Survival of control plants of both genotypes was unaffected with mean value of ~100% under hydroponic condition (Figure 5). Due to poor germination and complete loss of vigour, seeds treated with 40°C could

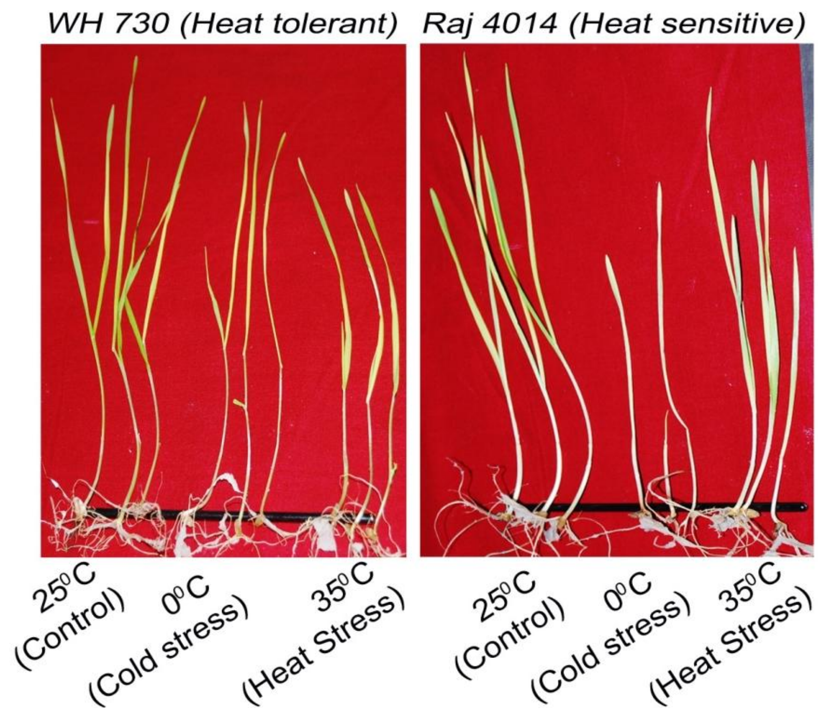


**Figure 6.** Vigour analysis of heat and cold treated genotype WH 730 and Raj 4014 grown hydroponically.

not be transferred under hydroponic condition.

### Phenotyping of heat and cold treated genotypes grown hydroponically

In our investigation made on two wheat genotypes, we found a significant difference in the seedlings vigor and shoot and root length at early stage of growth (13 days after heat and cold treatment). Although heat and cold treated genotypes adjusted under hydroponic condition up to some extent but the vigour was significantly affectted as compared to their control plants. The vigour of control plants is characterized by emergence of healthy secondary leaves in both genotypes as compared to the treatment which showed poor emergence (Figure 6). The mean value of shoot length of the genotype WH 730 was found to be 18.33 cm as compared to its control plant (24.16 cm), however relative to control (20 cm), the mean value for the genotype Raj 4014 was found to be 13.67 cm (Figure 7). When both genotypes under same condition were compared for root length, we could not observe any significant difference relative to their respective control plants.

The mean value of shoot and root length of cold treated genotype WH 730 was 21.83 and 6.7 cm as compared to the control (24.16 and 8.66 cm, respectively) which signifies no significant alteration; however as compare to control plants, cold treatment significantly reduced the mean value of shoot and root length of genotype Raj 4014 which was 9.5 and 4.67 cm, respectively (Figure 7).

## DISCUSSION

Plants have evolved several inbuilt mechanisms to cope up with various abiotic and biotic stresses. Symptoms appear in plants failing to cope up with these stresses. In the present study, three different temperature conditions were chosen to examine their effect on two different physiological parameters viz. germination and growth at early stage of two wheat genotypes. Our results indicate that the exposure of seeds to 35°C could not impart any significant change in germination efficiency (~100%) of the genotype WH-730 while same was reduced (~90%) in Raj 4014 after 24 and 48 h of heat and cold treatment. During growth at early stage (13 days after heat and cold treatment) survival efficiency was found to be ~90 and ~78% for the genotype WH 730 and Raj 4014, respectively. The difference in germination efficiency and survival rate of both genotypes might be due to genotypic variation in the sensitivity of heat and cold tolerance. Improved vigour of both genotypes treated with 35°C could be attributed to improved enzymatic activity of soaked seed which might have increased the reserve mobilization by breaking down the stored starch and giving nutrients to the germinating wheat embryos. In contrast at 40°C, the germination of soaked seeds of both genotypes was very poor which could not be transferred under hydroponic condition. The present study is in accordance with the work of Essemine et al. (2010) in wheat, in which they observed that the wheat seeds exposed to 45°C adversely affected the enzymes involved

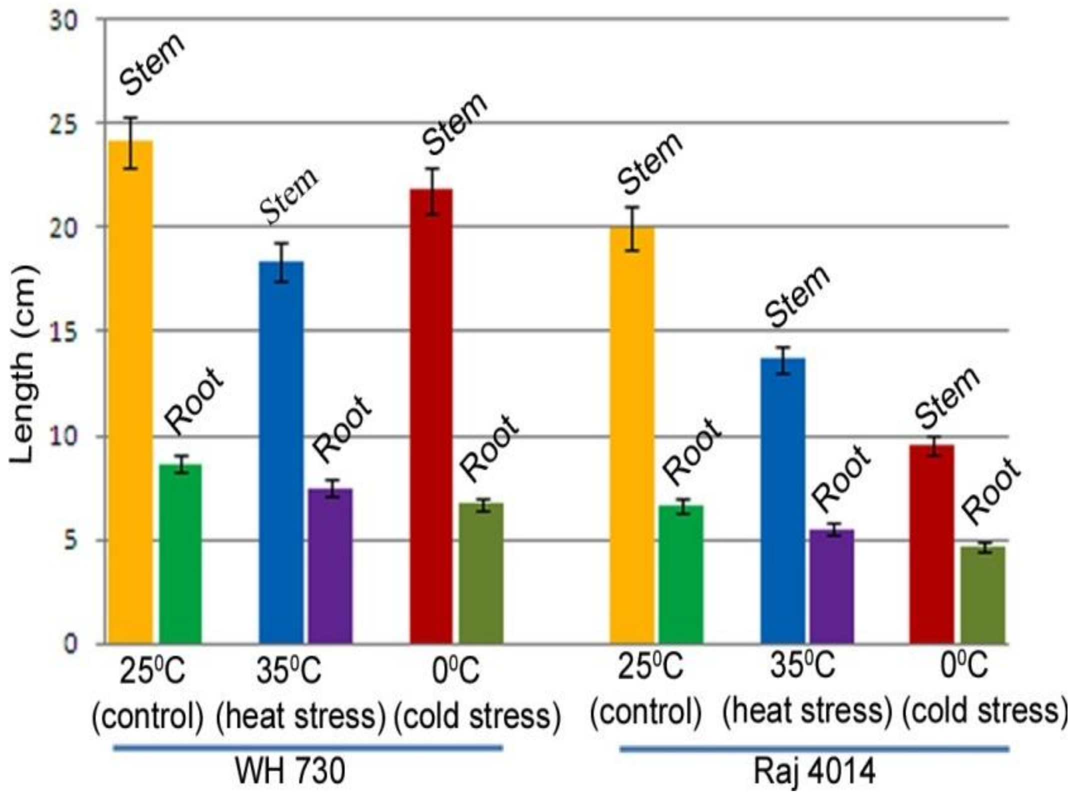


**Figure 7.** Comparison of shoots and root length of heat and cold treated genotypes grown hydroponically.

in starch breakdown and abolishing, giving nutrients to the growing wheat embryos which was less pronounced in germinating seeds treated with 35°C. This result is further supported by Petruzzelli and Toranto (1989) in which they have shown that the rate of reserve mobilizetion and metabolites are decreased in hard wheat seeds to embryo after 4 days of heat treatment which was associated with a loss of seed viability.

In contrast to heat treatment, we observed no significant effect of cold treatment (0°C for 2 h) on the germination (24 and 48 h post treatment) ability of both genotypes while growth at early reatment (13 days after treatment) was severely affected in Raj 4014 (heat intolerant) as compared to WH 730 (heat tolerant). This reduction in root and shoot length and survival rate of Raj 4014 as compared to WH 730 might be due to varietal sensitivity to withstand the cold stress which was undistinguishable at germination stage (24 and 48 h post treatment) but clearly distinguishable during growth at early stage (13 days after treatment). Finally, these results suggest that evaluation of wheat germplasm for heat and cold tolerance could be exploited at germination and growth at early stage.

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## REFERENCES


Annual report (2010-11). Directorate of wheat research, Karnal-132001, (India).

Barnabas B, Jager K, Feher A (2008). The effect of drought and heat stress on reproductive processes in cereals. Plant. Cell. Environ. 31:11-38.

Barton LV (1961). Seed preservation and longevity. Inter Science Publishers, NY.

Bewley JD and Black M (1982). Viability and longevity: physiology and biochemistry of seeds. Springer Verlag, Berlin-Heidelberg, NY, 2:47.

Bohnert HJ, Nelson DE, Jensen RG (1995). Adaptations to environmental stresses. Plant Cell. 7:1099-1111.

Buriro M, Oad FC, Keerio MI (2011). Wheat seed germination under the influence of temperature regimes. Sarhad J. Agric. 27(4):539-41.

Dell' Aquila A, Spada A (1994). Effects of high and low temperature on protein synthesis pattern of germinating wheat embryos. Plant. Physiol. Biochem. 32:65-73.

Essamine J, Ammar S, Bouzid S (2010). Impact of heat stress on germination and growth in higher plants: Physiological, Biochemical and molecular repercussion and mechanisms of defense. J. Biol. Sci. 10(6): 565-572.

Essemine J, Ammar S, Jbir N, Bouzid S, (2002). Sensivity of two wheat species seeds (*Triticum durum*, var. Karim and *Triticum aestivum*, var. Salamboto heat constraint during germination. Pak. J. Biol. Sci.

10(21):3762-3768.
Greeer EN (1953). Risk in the storage of grain crops in grain drying storage and handling in European countries. Food Agri. Tech. Inform. Serv., Organ. Eur. Econ. Co-op., London. pp.52-60.
Gupta P, Sheoran IS (1983). Response of some enzymes of nitrogen
metabolism to water stress in two species of Brassica. Plant. Physiol. Biochem. 10:5-13.
Gupta PK, Mir RR, Mohan A, Kumar J (2008). Wheat genomics: present status and future prospects. Int. J. plant. Genomics., doi:101155/2008/896451.
Howarth CJ (2005). Genetic improvement of tolerance to high temperature. In: abiotic stresses plant resistance through breeding and molecular approach, Ashraf M and Harris PJC (Eds.). Howarth Press Inc., NY.
Hutchinson JB (1944). The drying of wheat II. The effect of temperature on germination capacity. J. Soc. Chem. Ind., 63:104-107.
Kotak S, Larkindale J, Lee U, von Koskull-Doring P, Vierling E, Scharf KD (2007). Complexity of heat stress response in plants. Curr. Opi. Plant. Biol. 10:310-316.
Kundu S, Singh C, Shoran J, Singh SS (2010). An update on released wheat varieties and registered genetic stocks (*Triticum* L.). Technical bulletin No.13, Directorate of wheat research, Karnal-132001 (India) p. 34.
Lobell DB and Ortiz-Monasterio (2007). Impact of day versus night temperature on spring wheat yields: Acomparison of empirical and CERES model predictions in three locations. Agro. J. 99:469-477.
Penning de Vries FWT, Witlage JM, kremer D (1979). Rate of respiration and increase in structural dry matter in young wheat, rye grass and maize plants in relation to temperature, to water stress and to their sugar content. Ann. Bot. 44:595-609.
Perras M and Sarhan F (1989). Synthesis of freezing tolerance protein in leaves, crown and root during cold acclimation of wheat. Plant. Physiol. 89:577-585.
Petruzzelli M, Sarhan F (1989). Wheat aging: The contribution of embryonic and non-embryonic lesions to loss the seed viability. Physilogia. Plantarum 76:289-294.
Rao DG and Sinha SK (1993). Efficiency of mobilization of seed reserve in sorghum hybrids and their parents as influenced by temperature regimes. Seed Res. 2:97-100.
Read DWL, Beaten JD (1963). Effect of fertilizer, temperature and moisture on germination of wheat. Agro. J. 55: 287-290.
Thomas B (1941). The importance of moisture content in the storage of grains. Z getreidew 28:66.
Wanjura DF and Buxtor DR (1972). Hypocotyl and radicle elongation of cotton as affected by soil environment. Agro. J. 64:431-435.
Zhang Y, Mian MA, Chekhovskiy K, So S, Kupfer D, Lai H, Roe BA (2005). Differential gene expression in Festuca under heat stress conditions. J. Exp. Bot. 56:897-907.