

A study on germination and seedling growth of Blackgram (*Vigna mungo* L. Hepper) germplasm against Polyethylene glycol 6000 stress

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Abstract: Seed germination is considered as one of the first and foremost fundamental life stages of a plant, where the success in growth and yield is also depending on this stage. An experiment was conducted in order to study the effect of different concentrations (i.e. 0, 10, 20 and 30 %) of polyethylene glycol (PEG) stress on germination and early growth stages of 29 inbred lines of blackgram (*Vigna mungo* L. Hepper) in Plant Physiology laboratory, Department of Botany and Microbiology, Acharya Nagarjuna University. Different germination indices such as rate of imbibition, germination percent, germination energy, radical length, shoot length, relative water content, protein, nitrogen and biochemical parameters such as proline, peroxidase, polyphenol oxidase, catalase and superoxide dismutase along with drought parameters like percent injury, drought tolerance index was measured. Results showed significant differences among the cultivars at each drought stress level and significant decrease was observed in imbibition percentage, germination, length of radicle and plumule and radicle and plumule dry matter along with biochemical parameters. Among all the test genotypes LBG 17, LBG 764, LBG 752 and LBG 756 showed their efficiency in terms of germination and germination attributes besides showing more biochemical activity to withstand to the drought conditions and suggested to cultivate in soils with less moisture availability.

Keywords: PEG 6000, Blackgram, Water stress, Germination, Drought tolerance index

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I. Introduction

Global climate change, in the form of increasing temperature and decreasing soil moisture should result in the projected decrease in the yield of food crops over the next 50 years (Thomson *et al.*, 2005). The lack of moisture in soil directly affects seed germination and seedling growth of a plant (Bewley and Black, 1994; Larson and Kiemnec, 1997).

Crop establishment depends on an interaction between seedbed environment and seed quality (Khajeh-Hosseini *et al.*, 2003). Among the stages of the plant life cycle seed germination, seedling emergence and seed establishment are the key processes in the survival and growth of any plant species (Almansouri *et al.*, 2001; Delachave and De Pinho, 2003; Hadas, 2004). Germination is regulated by duration of wetting and the amount of moisture in the growth medium (Schutz and Milberg, 1997; Gill *et al.*, 2002).

The water potential that prevents sufficient amount of water for seed germination is critical for any crop. The study of such type of stress condition for any crop plant provides valuable information for agricultural community (Levitt, 1972).

Germinating the seeds in solutions of different water potentials is a convenient method to study the responses of seeds at germination against low water potential (Naylor, 1992). Polyethylene glycol (PEG) compounds have been used to simulate osmotic stress effects *in vitro* to maintain uniform water potentials. PEG 6000 found to be convenient solute to create osmotic stress without causing any toxicity to the plant cells (Verslues *et al.*, 2006). Molecules of PEG 6000 are too small to influence the osmotic potential but large enough not to be absorbed by plant (Carpita *et al.*, 1979). Moreover PEG withdraws water from the cell without entering into apoplast which mimics dry soil (Verslues and Bray, 2004). It is envisaged from the above findings that PEG solution can be used in the laboratory for screening drought tolerance varieties. Therefore, present experiment was designed to screen the germplasm of twenty nine blackgram genotypes against moisture stress at germination and seedling stages under *in vitro* conditions.

Blackgram is one of the important pulse crop and dry seeds are mainly used as a source of vegetable protein by the people of Indian subcontinent, especially in countries like India people used to depend on this pulse crop for their protein diet. The yields of blackgram found to be decrease year by year and the reason is

may be the change in environmental conditions with regular dry spells and less moisture availability. The response of blackgram cultivars to the change in moisture conditions especially during germination and seedling stages is not clearly known. Moreover very few literary supports are available in this regard. So there is an immediate need to study and understand the responses of blackgram cultivars against moisture stress at germination and seedling stage in order to develop drought tolerant genotypes of blackgram.

II. Materials And Methods

A total of 29 blackgram genotypes such as LBG17, LBG 402, LBG 611, LBG 623, LBG 645, LBG 648, LBG 685, LBG 709, LBG 716, LBG 723, LBG 728, LBG 729, LBG 730, LBG 731, LBG 734, LBG 735, LBG 736, LBG 738, LBG 741, LBG 747, LBG 748, LBG 749, LBG 750, LBG 751, LBG 752, LBG 754, LBG 756, LBG 759 and LBG 764 were obtained from Regional Agricultural Research Station (RARS), Lam, Guntur. The collected germplasm was subjected to different osmotic potentials (0, 10, 20 and 30 %) of PEG 6000. On third day of experiment data was collected on the following parameters.

Rate of Imbibition (%)

Seeds were surface sterilized by soaking them in a solution of 2.0% aqueous sodium hypochlorite for 15 minutes at room temperature and then rinsed thoroughly with distilled water. Seeds were left imbibed whole the night and the difference between fresh and imbibed values were used to determine the rate of imbibition. The rate of imbibition was measured according Abdul Kadir (2009), and was expressed in percentage (%).

$$\text{Rate of Imbibition} = \frac{W_2 - W_1}{W_1} \times 100$$

Where W_2 = Weight of imbibed seeds

W_1 = Weight of fresh seeds

Germination percent (%)

Percent germination was recorded for every 24 h after the treatment up to 72 h. The seeds with a radical length of more than 2 mm were considered as germinated (Mackay *et al.*, 1995 and Karaguzel *et al.*, 2002). Percentage of germination was measured according Achakzai (2009) and it was expressed in terms of percentage (%).

$$\text{Germination per cent} = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds}} \times 100$$

Germination Energy

A measure of the rapidity of germination was measured by modifying the formula of Yan Li (2008).

$$\text{Germination Energy} = \frac{\text{Number of seeds germinated in PEG concentration in 3 days}}{\text{Total number of seeds}}$$

Radicle and Plumule length

Length of the radical and plumule was measured with the help of a thread and measuring scale from the tip of root or shoot to the collar region of the seed and expressed in centimetres.

Seedling Vigour Index

Seedling vigour index (SVI) was calculated by using the modified formula of Abdul-Baki and Anderson (1973).

$$\text{SVI} = [\text{Seedling length (cm)} \times \text{Germination percentage}]$$

Seedling fresh and dry weight

Seedlings were collected on the final day of experiment and were blotted gently on blotting paper to remove the excess water or polyethylene glycol attached to the seedling. The fresh weights of the seedlings were measured according Afzal *et al.*, (2005). Later the seedlings were kept in hot air oven for about three days at 70°C until the seedling maintains the constant weight, these weights were considered as dry weights of the seedlings (Afzal *et al.*, 2005).

Relative Water Content (RWC)

Relative water content was estimated according Fletcher *et al.*, (1988) on the final day of the experiment. Fresh seedlings were collected and weighed. This was taken as fresh weight (FW). Then the seedlings were dipped in petri dishes containing distilled water and the entire setup was left undisturbedly for four hours. After four hours, seedlings were blotted gently and weighed. This was referred to as the turgid weight (TW). After recording turgid weight, the seedling were dried in an oven at 96°C for four days and the dry weight (DW) was recorded. RWC (%) was calculated by the formula given by Kramer (1983) and was expressed in terms of percentage (%).

$$\text{RWC} = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Turgid weight} - \text{Dry weight}} \times 100$$

Drought tolerance index (DTI)

Stress tolerance index of selected genotypes was evaluated according Fernandez (1992).

$$\text{STI} = 1 - (\text{Ys}/\text{Yp})$$

Where,

Ys = Mean total yield in stress conditions

Yp = Mean total yield in normal conditions

*total yield refers to dry weight of the seedlings.

Percent injury (% I)

Percent injury is the rate of damage occurs to the crop due to fewer water supplies. Percent injury can be measured by using the following formula (Rosielle and Hambling, 1981). Percent injury was expressed in percentage.

$$\text{Percent injury} = \frac{(\text{Yp} - \text{Ys})}{\text{Yp}} \times 100$$

Where

Ys = Mean total yield in stress condition

Yp = Mean total yield in normal condition

*total yield refers to dry weight of the seedlings.

III. Results

Effect of PEG 6000 stress on seed germination, rate of imbibition and germination energy

Percent of seed germination, rate of imbibition and germination energy were severely damaged with increased level of PEG 6000 stress (Table 1). At 10% level of stress, the decrease in germination percent among the cultivars ranged from 1.58% (LBG 731) to 52.9% (LBG 764). PEG stress at 20% level effected germination percent from 3.24% (LBG 730) to 58.33% (LBG 648). Similarly at 30% level PEG stress decreased germination percent from 55.29% (LBG 735) to 99.31% (LBG 764). These observations reveal at increased level (30%) of PEG stress, the varieties viz., LBG 735 and LBG 738 exhibited better tolerance reflected by high germination percent.

The rate of imbibition significantly decreased at 10% level and it ranges from 4.27% in LBG 747 to 23.99% in LBG 645. The rate of imbibition decrease is relatively less (11.39%) in LBG 747 and maximum decrease was noticed (25.96%) in LBG 749 at 20% PEG stress. At 30% PEG stress the percent of decrease in imbibition among the cultivars ranged from 24.03% (in LBG 764) to 42.98% (in LBG 749). Hence, it is observed that under maximum stress, LBG 764, LBG 747 and LBG 730 exhibited more tolerance as expressed in terms of rate imbibition when compared with other cultivars. The germination energy of blackgram cultivars was affected by different levels of PEG stress. There was steep decrease in it when stress level increased from 10% to 20% and to 30%.

The percent of decrease in germination energy is an useful parameter to assess stress tolerance of cultivars. At 30% PEG stress the percent of decrease in germination energy is less (< 50%) in three cultivars (LBG 735, LBG 752 and LBG 756) indicating their high stress tolerance over the other cultivars.

Effect of PEG 6000 stress on relative water content, radical length and plumule length

The PEG stress also effected the relative water content (RWC), radicle length and plumule length of germinating seedlings (Table 2). There is a steady decrease in RWC in all the cultivars with an increase of stress level from 10% to 30%. The decrease in RWC is significant when compared with control. At high level (30%) of PEG stress, the RWC of cultivars ranged from a minimum of 54.32% to a maximum of 86.20%. The

percent of decrease in RWC when compared with corresponding control plants is more in LBG 17 variety (35.44) than in other varieties at 30% stress level. The decrease in RWC is less (11.72 to 14.15) in LBG 738, LBG 754, LBG 731 and LBG 723 and hence considered as stress tolerant cultivars.

The percent decrease in radical length and plumule length of all cultivars at 30% stress was very high (80%-99%) when compared with control plants indicating the severe effect of stress on these parameters (Table 2).

Effect of PEG 6000 stress on seedling vigour index, fresh weight and dry weight

Seedling vigour index, fresh weight and dry weight of seedlings of all the cultivars were recorded under three levels of PEG stress (Table 3). The seedling vigour of LBG 752 and LBG 756 cultivars was more than the other cultivars at 20% and 30% PEG stress levels, indicating their stress tolerance. In these cultivars the percentage of decrease in fresh weight and dry weight was also less when compared with other cultivars (Table 3).

Effect of PEG 6000 stress on seedling injury and drought tolerance

In all cultivars injury percent increased progressively when stress level increased from 10% to 30%, a minimum injury (28%-34%) was observed in the seedlings of LBG 748, LBG 749, LBG 752 and LBG 756 (Table 4).

The drought tolerance index of all genotypes at three levels of stress was calculated and presented in Table 8. LBG 752 and LBG 756 exhibited the highest drought tolerance index both at 10% and 30% level PEG stress (Table 4). The other genotypes exhibited lesser tolerance index than the above mentioned two cultivars.

IV. Discussion

In the present study it was observed that with an increase in water stress (0 – 30%), there was a gradual depletion in rate of water uptake by blackgram seeds of all genotypes. This reduction might be due to the fact that water moves from high potential to low potential due to differences in the free energy content. The gradient of water potential between dry seeds and pure water (0.0 Mpa) decrease rapidly with the addition of any soluble substances such as polyethylene glycol in water. The decrease in water potential gradient between seed and media will prevent the seeds to absorb the desired amount of water (Achakzai, 2009). Similar results were also reported in case of mungbean (Akhter, 1985), a few species of agropyron (Batool, 1988) and maize (Achakzai, 2009).

A significant delay in the initiation and completion of germination was also observed in PEG treated varieties. The decreased germination in response to water stress is not uniform in all cultivars. This might be due to differences in their range of drought stress tolerance. The reason for decreasing germination with increasing level of stress may be due to water potential and osmotic potential as mediated by solute developed additive effect on the inhibition of seed germination (Bernstein, 1961). The delay in completion of germination is a common response, because seeds require more time to absorb sufficient amount of water, which is vital for the act of initiation of germination (Tesche, 1975; Gul and Allan, 1976 and Almansouri *et al.*, 2001).

In the present study decreased germination energy in treatments of water stress can be due to seed deterioration and degradation of cell membrane (Falleri, 1994). Similarly, Zhu *et al.* (2006) and Ahmadloo *et al.* (2011) also have reported that increase in the osmotic concentration decreased energy of seed germination.

Drought stress decreased the root length it may be due to declining vacuolar K^+ because its accumulation in newly formed vacuoles drives cell expansion (Walker *et al.*, 1998). It is a fact that the drought tolerant accessions had greater shoot and root lengths and biomass production than the sensitive accessions. In this study a reduction in root length was observed with an increase in PEG stress concentration. These results are in accordance with Gamze *et al.* (2005) in pea, in various plant species of wheat and maize (Nayar and Gupta, 2006), in alfalfa (Safarnezad, 2008) and in *Vigna aconitifolia* (Soni *et al.*, 2011).

Relative water content (RWC) was used as a measure of drought. This index may be useful for determining the plant leaf water status (McCaig and Romagosa, 1991). Under osmotic stress a significant reduction in tissue water content of germinated seeds was observed, indicating that these tissues were under stress. Similar observations of decreases in water content under stress conditions were made by Pennypacker *et al.* (1990) in alfalfa Gupta *et al.* (1993) in chickpea, Gill *et al.* (2001) in sorghum, Hsu (2003) in rice and Ravi ranjan and Naik (2011) in pigeonpea. The length of the plumule was progressively decreased with increase in osmotic tension. According to Kramer (1974) the first determined effect due to water deficit was the growth reduction caused by the decline in the cellular expansion. The cellular elongation process and the carbohydrates wall synthesis were very susceptible to water deficit (Wenkert *et al.*, 1978) and the growth decrease was a consequence of the turgescence laying down of those cells (Shalhevet *et al.*, 1995). In the present study plumule length decreased in the entire test varieties in each level of PEG stress. The results are in agreement with the findings of Vahid Jajrami (2009) in wheat, Deshmukh and Dhumal (2011) in sorghum and Burhan *et al.* (2012)

in greengram. Seedling vigour index is more sensitive to water stress. With an increment in PEG concentration SVI was adversely affected and recorded very less at 30% PEG. Gong Ping *et al.* (2000) suggested that the improvement of the vigour index was associated with the enhancement of activated oxygen metabolism in seedlings. PEG stress creates O₂ deficit environment (Mexal *et al.*, 1975) that results in less SVI. Similar results were observed in in pinus (Lo'pez *et al.*, 2009) and in cupressus (Ahmadloo *et al.*, 2011).

In the present study Fresh weight of seedling decreased with an increase in PEG concentration. Fresh root weight found to be minimum in 25% PEG in wheat cultivars (Rauf *et al.*, 2007; Khan *et al.*, 2013) and in sorghum (Bibi *et al.*, 2012). Seedling dry weight showed a similar trend and it decreased with increasing PEG. Similar kind of reduction in root dry weight was reported by Raggi (1992), Anaytullah (2007) in rice, Dezfuli *et al.* (2008) in maize and in cultivars of blackgram (Pratap and Sharma, 2010).

The relative injury rate was a good parameter which illustrates the effect of PEG or water stress on the germination capacity of the inbred-lines. The rate of injury caused by stress was found to be different in the different cultivars of tomato (Ghebremariam *et al.*, 2013). Similar result was reported by Blum (1980) and Majid and Roza (2011) in wheat. There is a significant variation in drought tolerance index among all the genotypes under 10%, 20% and 30% PEG solutions. These values indicated that each genotype showed different responses against drought stress and showed their tolerance capacity. The present study results are in accordance with the reports of Ahamad *et al.* (2009) in sunflower, Datta *et al.* (2011) in wheat, Yugi *et al.* (2012) in rice and Tsago *et al.* (2013) in sorghum.

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Table 1. Changes in mean germination %, imbibition and germination energy of blackgram cultivars at different osmotic potentials of PEG

Character→ Variety]	Germination (%)				Rate of imbibition (%)				Germination Energy			
	Control	10 %	20%	30%	Control	10 %	20 %	30 %	Control	10 %	20 %	30 %
LBG17	98.00*	86.00 (12.24)*	73.33 (25.17)*	28.00 (71.43)*	61.16*	52.07 (14.86)*	47.34 (22.60)*	37.97 (37.92)	1.00*	0.98 (02.00)*	0.42 (58.00)	0.28*(72.00)
LBG402	72.00	60.00 (16.67)	51.33 (28.71)	22.00 (69.44)*	57.34	47.92 (16.43)	44.49 (22.41)	39.76 (30.66)*	0.72	0.61 (15.28)	0.58 (19.44)*	0.22*(69.44)
LBG611	67.00	44.00 (34.33)	34.33 (48.76)	14.00 (79.10)	56.05	51.66 (07.83)*	47.83 (14.67)*	37.25 (33.34)	0.68	0.44 (35.29)	0.38 (44.12)	0.14 (79.41)
LBG623	77.67	66.67 (14.16)	52.00 (33.05)	06.00 (92.28)	54.33	45.89 (15.53)	42.43 (21.90)	40.06 (26.27)*	0.78	0.66 (15.38)*	0.42 (46.15)	0.06 (92.31)
LBG645	98.67*	76.67 (22.30)*	64.33 (34.80)	06.00 (93.92)	58.18	44.22 (23.99)	43.99 (24.39)	42.21 (27.45)*	1.00*	0.82 (18.00)*	0.78 (22.00)*	0.06 (94.00)
LBG648	76.00	42.33 (44.30)	31.67 (58.33)	04.00 (94.74)	52.14	46.43 (10.95)	40.76 (21.83)	39.26 (24.70)	0.76	0.42 (44.74)	0.28 (63.16)	0.04 (94.74)
LBG685	84.00*	79.67 (05.15)*	65.00 (22.62)	14.33 (82.94)	59.33*	56.35 (05.02)*	48.70 (17.92)*	40.39 (31.92)*	0.83*	0.73 (12.05)*	0.66 (20.49)*	0.24*(71.08)
LBG709	36.00	45.67 (26.86)	41.33 (14.81)	04.33 (87.97)	50.86	44.99 (11.34)	41.73 (17.95)	36.37 (28.49)	0.56	0.43 (23.21)	0.28 (50.00)	0.04 (92.86)
LBG716	76.00	65.67 (13.59)	56.33 (25.88)	18.00 (76.32)*	58.00	51.95 (10.43)*	48.34 (16.66)*	41.97 (27.64)*	0.76	0.66 (13.16)*	0.40 (47.37)	0.18*(76.32)
LBG723	83.67*	70.00 (16.34)	54.00 (35.46)	04.33 (94.82)	56.34	51.26 (09.02)	41.89 (25.65)	33.20 (41.07)	0.84*	0.71 (15.48)*	0.36 (57.14)	0.04 (95.24)
LBG728	68.00	34.33 (49.51)	29.00 (57.35)	12.00 (82.35)	56.08	49.00 (12.62)	41.82 (25.43)	39.73 (29.15)*	0.68	0.34 (60.00)	0.21 (69.12)	0.12 (82.35)
LBG729	70.00	52.33 (25.24)	42.00 (40.00)	25.67 (63.33)*	57.56	50.45 (12.35)	45.47 (21.00)	40.63 (29.41)*	0.70	0.52 (25.71)	0.42 (40.00)	0.26*(62.86)
LBG730	72.00	74.00 (02.78)*	74.33 (03.24)*	22.00 (69.44)*	57.24	52.15 (08.89)*	44.51 (22.24)	43.22 (24.49)*	0.82*	0.74 (09.76)*	0.38 (53.66)	0.22*(73.17)
LBG731	84.00*	82.67 (01.59)*	76.33 (09.13)*	26.00 (69.05)*	60.31*	54.44 (09.73)*	51.76 (14.18)*	36.33 (39.76)	0.84*	0.62 (26.19)*	0.38 (54.76)	0.20*(76.19)
LBG734	75.67	64.67 (14.54)	53.67 (29.07)	08.67 (88.54)	56.04	52.35 (06.58)*	47.46 (15.31)*	37.97 (32.24)	0.76	0.64 (15.79)*	0.35 (53.95)	0.08 (89.47)
LBG735	85.00*	67.00 (21.18)	54.33 (36.08)	38.00 (55.29)*	58.42*	49.29 (15.63)	48.57 (16.86)*	39.68 (32.08)*	0.76	0.66 (13.16)*	0.55 (27.63)*	0.38*(50.00)
LBG736	91.33*	72.00 (21.17)*	64.33 (29.56)	04.00 (95.62)	58.70*	52.82 (10.02)*	47.52 (19.05)*	39.98 (31.89)*	0.87*	0.72 (17.24)*	0.60 (31.03)*	0.34*(60.92)
LBG738	80.67	76.00 (05.79)*	73.33 (09.10)*	36.00 (55.37)*	59.13*	51.99 (12.08)*	44.84 (24.17)	40.45 (31.59)*	0.92*	0.76 (17.39)*	0.40 (56.52)	0.36*(60.87)
LBG741	75.00	67.00 (10.67)	51.33 (31.56)	10.00 (86.67)	57.00	50.63 (11.18)	43.38 (23.89)	33.16 (41.82)	0.82*	0.78 (04.88)*	0.67 (18.29)*	0.10 (87.80)
LBG747	86.67*	84.67 (02.31)*	73.00 (15.77)*	04.33 (95.00)	51.03	48.85 (04.27)	45.22 (11.39)	38.62 (24.32)	0.76	0.64 (15.79)*	0.46 (39.47)	0.04 (94.74)
LBG748	88.00*	84.67 (03.78)*	73.00 (17.05)*	26.00 (70.45)*	56.95	51.85 (08.96)*	45.44 (20.21)	42.58 (25.23)*	0.88*	0.72 (18.18)*	0.56 (36.36)*	0.28*(68.18)
LBG749	80.00	76.67 (04.16)*	72.00 (10.00)*	20.00 (75.00)*	60.28*	53.65 (11.00)*	44.63 (25.96)	34.37 (42.98)	0.88*	0.46 (47.73)	0.22 (75.00)	0.02 (97.73)
LBG750	84.00*	74.67 (11.11)*	80.67 (03.96)*	16.33 (80.56)	54.02	48.48 (10.26)	41.82 (22.58)	31.27 (42.11)	0.80*	0.74 (07.50)*	0.34 (57.50)	0.16*(80.00)
LBG751	78.33	74.00 (05.53)*	57.00 (27.23)	08.33 (89.37)	54.54	48.68 (10.74)	43.30 (20.61)	40.46 (25.82)*	0.84*	0.74 (11.90)*	0.26 (69.05)	0.08 (90.48)
LBG752	98.00*	88.00 (10.20)*	77.67 (20.74)*	18.33 (85.38)*	59.21*	54.09 (18.67)*	47.19 (24.02)*	39.62 (41.67)*	0.78	0.68 (38.46)*	0.58 (64.10)*	0.16*(82.05)
LBG754	76.00	63.00 (17.11)	60.33 (20.62)	04.00 (94.74)	54.25	48.45 (10.69)	43.09 (20.57)	39.34 (27.48)	0.99*	0.80 (18.37)*	0.36 (63.27)	0.04 (95.92)
LBG756	83.00*	75.33 (05.94)*	66.00 (20.00)*	17.67 (80.41)*	58.99*	53.50 (11.21)*	46.61 (15.06)*	39.46 (27.82)*	0.53	0.64 (54.72)*	0.56 (50.94)*	0.16*(80.00)
LBG759	70.00	55.67 (20.47)	50.00 (28.57)	08.00 (88.57)	50.37	45.70 (09.27)	41.35 (17.91)	37.91 (24.74)	0.53	0.26 (50.94)	0.21 (60.38)	0.08 (84.91)
LBG764	97.67*	46.00 (52.90)	78.33 (19.80)*	00.67 (99.31)	52.22	43.41 (16.87)	44.72 (14.36)	39.67 (24.03)*	0.70	0.45 (35.71)	0.58 (17.14)*	0.08 (84.91)
Grand Mean	79.63	67.21 (14.91)	59.59 (24.84)	14.51 (81.63)	55.89	49.39 (11.60)	44.66 (20.02)	38.30 (31.32)	0.78	0.61 (23.24)	0.41 (47.10)	0.14 (82.05)
CD	2.77	3.11	5.43	2.60	2.42	2.00	1.28	1.12	0.02	0.01	0.08	0.01
CV (%)	2.30	3.60	4.40	12.10	2.58	3.21	3.21	4.10	1.50	1.40	10.70	6.20

*Significant at 5% level; # Values within the brackets represent percent decrease compared to control (without PEG stress)

Table 4. Genotypes differences on percent injury and DTI treated with PEG 6000 during germination

Character→ Variety↓	% Injury			Tolerance index		
	10 %	20 %	30 %	10 %	20 %	30 %
LBG 17	49.08*	50.92*	56.88	0.107*	0.111*	0.124*
LBG 402	25.74*	33.66	43.56	0.026	0.037	0.044
LBG 611	18.57	32.14	60.71*	0.026	0.045	0.085
LBG 623	26.59*	46.82*	63.01*	0.046	0.081*	0.109*
LBG 645	34.12*	41.18	51.18	0.058*	0.070	0.087
LBG 648	38.69*	49.40*	66.67*	0.065*	0.083*	0.112*
LBG 685	02.92	39.42	60.58*	0.004	0.054	0.083
LBG 709	09.82	47.85*	67.48*	0.016	0.078*	0.110*
LBG 716	33.33*	42.06	50.00	0.042	0.053	0.063
LBG 723	38.06*	44.52*	64.52*	0.059*	0.069	0.100*
LBG 728	37.95*	60.51*	70.26*	0.074*	0.118*	0.137*
LBG 729	45.16*	46.45*	60.00*	0.070*	0.072	0.093
LBG 730	17.22	43.71*	58.94*	0.026	0.066	0.089
LBG 731	12.21	27.48	44.27	0.016	0.036	0.058
LBG 734	04.05	43.24*	56.76	0.006	0.058	0.078
LBG 735	16.77	36.65	46.58	0.027	0.059	0.075
LBG 736	20.65	33.15	47.83	0.038	0.061	0.088
LBG 738	20.00	35.15	49.70	0.033	0.058	0.082
LBG 741	05.91	38.17	53.76	0.011	0.071	0.100*
LBG 747	05.94	30.20	43.07	0.012	0.061	0.087
LBG 748	11.18	13.66	28.57	0.018	0.022	0.046
LBG 749	05.43	23.37	33.70	0.010	0.043	0.062
LBG 750	06.67	49.09*	61.82*	0.011	0.081*	0.102*
LBG 751	01.73	50.87*	63.01*	0.009	0.097*	0.118*
LBG 752	04.35	14.29	24.84	0.007	0.040	0.059
LBG 754	11.69	14.94	38.31	0.018	0.023	0.059
LBG 756	03.73	40.96	42.05	0.008	0.023	0.040
LBG 759	41.36*	62.00*	65.00*	0.067*	0.102*	0.132*
LBG 764	47.20*	53.42*	60.87*	0.076*	0.086*	0.098
Grand Mean	21.59	39.49	53.55	0.04	0.07	0.09
CD	2.62	2.89	4.00	0.008	0.005	0.010
CV (%)	3.28	4.51	4.00	9.00	10.20	10.20

*Significant at 5% level

Values within the brackets represent percent decrease compared to control (without PEG stress)

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K. Babu, G. Rosaiah. "A study on germination and seedling growth of Blackgram (*Vigna mungo* L. Hepper) germplasm against Polyethylene glycol 6000 stress." IOSR Journal of Pharmacy and Biological Sciences (IOSR-JPBS) , vol. 12, no. 5, 2017, pp. 90–98.